METHODS AND COMPOSITIONS FOR ASSESSING IMMUNE RESPONSE IN MURINE TUMOR MODELS

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority to application PCT/CN2020/094595, filed June 5, 2020, the disclosure of which is incorporated herein by reference.

FIELD OF THE INVENTION

[0002] The present invention generally relates to molecular biology, cancer biology and animal models.

BACKGROUND

[0003] Cancer is a heterogenous disease and highly related to the immunological system. The significance of fully understanding the interactions between cancer and immunity in cancer therapeutics has been broadly noted, particularly for immune therapies. However, only 20-30% of cancer patients respond to immunotherapies and there is also the added complexity of inducing immune-related adverse effects and auto-immune reactions. Being able to predict who will benefit from immunotherapies could spare treatment costs and patient health.

[0004] Experimental murine models derived from a variety of biological technologies are essential preclinical model systems. Molecular pathology techniques, such as gene expression biomarkers, are widely used to systemically profile and characterize these models. So far, however, there has been no robust and cost-effective assay designed for murine preclinical immune-oncology research to characterize tumor-immune interactions at a gene expression level.

SUMMARY OF INVENTION

[0005] In one aspect, the present disclosure provides a method for assessing immune response in a murine model. In one embodiment, the method comprises: obtaining a sample from the murine model; detecting the expression level of a panel of genes in the sample, wherein the panel comprises at least 50 genes selected from the group disclosed herein; comparing the expression level of the panel to a reference expression level; and evaluating the immune response in the murine model.

[0006] In certain embodiments, the murine model has a tumor. In certain embodiments, the sample is a tumor tissue.

[0007] In certain embodiments, the murine model been treated with a therapy. In certain embodiments, the therapy is an immune therapy.

[0008] In certain embodiments, the expression of the panel of genes is detected using next-generation sequencing (NGS).

[0009] In certain embodiments, the immune response comprises activation of an immune cell selected from the group consisting of B-cell, dendritic cell, macrophage, monocyte, natural killer cell, CD4+ T cell and CD8+ T cell.

[0010] In another aspect, the present disclosure provides a kit for assessing immune response in a murine model. In certain embodiments, the kit comprises primers for detecting the expression of a panel of genes in a sample from the murine model, wherein the panel comprises at least 50 genes selected from the group disclosed herein.

[0011] In another aspect, the present disclosure provides a microarray for assessing immune response in a murine model. In certain embodiments, the microarray comprises probes for detecting the expression level of a panel of genes in a sample from the murine model, wherein the panel comprises at least 50 genes selected from the group disclosed herein.

[0012] In yet another aspect, the present disclosure provides a non-transitory computer readable medium having instructions stored thereon, wherein the instructions, when executed by a processor, cause the processor to: retrieving the expression level of a panel of genes in the sample, wherein the panel comprises at least 50 genes selected from the group disclosed herein; comparing the expression level of the panel to a reference expression level; and evaluating the immune response in the murine model.

BRIEF DESCRIPTION OF THE DRAWINGS

[0013] The following drawings form part of the present specification and are included to further demonstrate certain aspects of the present disclosure. The disclosure may be better understood by reference to one or more of these drawings in combination with the detailed description of specific embodiments presented herein.

[0014] **FIG. 1A-1B** show the results of cell sorting for B cells (**FIG. 1A**) and T cells (**FIG. 1B**) from non-tumor bearing mice spleens from validation panel.

[0015] **FIG. 2** shows the quality assessment by comparing murine immune-oncology panel target gene and non-target gene expression levels in validation panels.

[0016] **FIG. 3** shows the single -sample gene set enrichment analysis (ssGSEA) on murine immune-oncology panel signatures grouped in 50 categories demonstrates high consistency of biomarkers in validation panels.

[0017] **FIG. 4** show the estimated cell fractions in validation panels using murine pan-cancer gene expression panel-based signatures.

DETAILED DESCRIPTION OF THE INVENTION

[0018] Before the present disclosure is described in greater detail, it is to be understood that this disclosure is not limited to particular embodiments described, and as such may, of course, vary. It is also to be understood that the terminology used herein is for the purpose of describing particular embodiments only, and is not intended to be limiting, since the scope of the present disclosure will be limited only by the appended claims.

[0019] 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 belongs. Although any methods and materials similar or equivalent to those described herein can also be used in the practice or testing of the present disclosure, the preferred methods and materials are now described.

[0020] All publications and patents cited in this specification are herein incorporated by reference as if each individual publication or patent were specifically and individually indicated to be incorporated by reference and are incorporated herein by reference to disclose and describe the methods and/or materials in connection with which the publications are cited. The citation of any publication is for its disclosure prior to the filing date and should not be construed as an admission that the present disclosure is not entitled to antedate such publication by virtue of prior disclosure. Further, the dates of publication provided could be different from the actual publication dates that may need to be independently confirmed.

[0021] As will be apparent to those of skill in the art upon reading this disclosure, each of the individual embodiments described and illustrated herein has discrete components and features which may be readily separated from or combined with the features of any of the other several embodiments without departing from the scope or spirit of the present disclosure. Any recited method can be carried out in the order of events recited or in any other order that is logically possible.

[0022] Definitions

[0023] The following definitions are provided to assist the reader. Unless otherwise defined, all terms of art, notations and other scientific or medical terms or terminology used herein are intended to have the meanings commonly understood by those of skill in the chemical and medical arts. In some cases, terms with commonly understood meanings are defined herein for clarity and/or for ready reference, and the inclusion of such definitions herein should not necessarily be construed to represent a substantial difference over the definition of the term as generally understood in the art.

[0024] As used herein, the singular forms "a", "an" and "the" include plural references unless the context clearly dictates otherwise.

[0025] The term "amount" or "level" refers to the quantity of a polynucleotide of interest or a polypeptide of interest present in a sample. Such quantity may be expressed in the absolute terms, i.e., the total quantity of the polynucleotide or polypeptide in the sample, or in the relative terms, i.e., the concentration of the polynucleotide or polypeptide in the sample.

[0026] As used herein, the term "cancer" or "tumor" refers to any diseases involving an abnormal cell growth and include all stages and all forms of the disease that affects any tissue, organ or cell in the body. The term includes all known cancers and neoplastic conditions, whether characterized as malignant, benign, soft tissue, or solid, and cancers of all stages and grades including pre- and post-metastatic cancers. In general, cancers can be categorized according to the tissue or organ from which the cancer is located or originated and morphology of cancerous tissues and cells. As used herein, cancer types include, without limitation, acute lymphoblastic leukemia (ALL), acute myeloid leukemia, adrenocortical carcinoma, anal cancer, astrocytoma, childhood cerebellar or cerebral, basal-cell carcinoma, bile duct cancer, bladder cancer, bone tumor, brain cancer, cerebellar astrocytoma, cerebral astrocytoma/malignant glioma, ependymoma, medulloblastoma, supratentorial primitive neuroectodermal tumors, visual pathway and hypothalamic glioma, breast cancer, Burkitt's lymphoma, cervical cancer, chronic lymphocytic leukemia, chronic myelogenous leukemia, colon cancer, emphysema, endometrial cancer, ependymoma, esophageal cancer, Ewing's sarcoma, retinoblastoma, gastric (stomach) cancer, glioma, head and neck cancer, heart cancer, Hodgkin lymphoma, islet cell carcinoma (endocrine pancreas), Kaposi sarcoma, kidney cancer (renal cell cancer), laryngeal cancer, leukaemia, liver cancer, lung cancer, neuroblastoma, non-Hodgkin lymphoma, ovarian cancer, pancreatic cancer, pharyngeal cancer, prostate cancer, rectal cancer, renal cell carcinoma (kidney cancer), retinoblastoma,

Ewing family of tumors, skin cancer, stomach cancer, testicular cancer, throat cancer, thyroid cancer, vaginal cancer.

[0027] A "cell", as used herein, can be prokaryotic or eukaryotic. A prokaryotic cell includes, for example, bacteria. A eukaryotic cell includes, for example, a fungus, a plant cell, and an animal cell. The types of an animal cell (e.g., a mammalian cell or a human cell) includes, for example, a cell from circulatory/immune system or organ (e.g., a B cell, a T cell (cytotoxic T cell, natural killer T cell, regulatory T cell, T helper cell), a natural killer cell, a granulocyte (e.g., basophil granulocyte, an eosinophil granulocyte, a neutrophil granulocyte and a hypersegmented neutrophil), a monocyte or macrophage, a red blood cell (e.g., reticulocyte), a mast cell, a thrombocyte or megakaryocyte, and a dendritic cell); a cell from an endocrine system or organ (e.g., a thyroid cell (e.g., thyroid epithelial cell, parafollicular cell), a parathyroid cell (e.g., parathyroid chief cell, oxyphil cell), an adrenal cell (e.g., chromaffin cell), and a pineal cell (e.g., pinealocyte)); a cell from a nervous system or organ (e.g., a glioblast (e.g., astrocyte and oligodendrocyte), a microglia, a magnocellular neurosecretory cell, a stellate cell, a boettcher cell, and a pituitary cell (e.g., gonadotrope, corticotrope, thyrotrope, somatotrope, and lactotroph)); a cell from a respiratory system or organ (e.g., a pneumocyte (a type I pneumocyte and a type II pneumocyte), a clara cell, a goblet cell, an alveolar macrophage); a cell from circular system or organ (e.g., myocardiocyte and pericyte); a cell from digestive system or organ (e.g., a gastric chief cell, a parietal cell, a goblet cell, a paneth cell, a G cell, a D cell, an ECL cell, an I cell, a K cell, an S cell, an enteroendocrine cell, an enterochromaffin cell, an APUD cell, a liver cell (e.g., a hepatocyte and Kupffer cell)); a cell from integumentary system or organ (e.g., a bone cell (e.g., an osteoblast, an osteocyte, and an osteoclast), a teeth cell (e.g., a cementoblast, and an ameloblast), a cartilage cell (e.g., a chondroblast and a chondrocyte), a skin/hair cell (e.g., a trichocyte, a keratinocyte, and a melanocyte (Nevus cell)), a muscle cell (e.g., myocyte), an adipocyte, a fibroblast, and a tendon cell), a cell from urinary system or organ (e.g., a podocyte, a juxtaglomerular cell, an intraglomerular mesangial cell, an extraglomerular mesangial cell, a kidney proximal tubule brush border cell, and a macula densa cell), and a cell from reproductive system or organ (e.g., a spermatozoon, a Sertoli cell, a leydig cell, an ovum, an oocyte). A cell can be normal, healthy cell; or a diseased or unhealthy cell (e.g., a cancer cell). A cell further includes a mammalian zygote or a stem cell which include an embryonic stem cell, a fetal stem cell, an induced pluripotent stem cell, and an adult stem cell. A stem cell is a cell that is capable of undergoing cycles of cell division while maintaining an undifferentiated state and differentiating into specialized cell types. A stem cell can be an

omnipotent stem cell, a pluripotent stem cell, a multipotent stem cell, an oligopotent stem cell and a unipotent stem cell, any of which may be induced from a somatic cell. A stem cell may also include a cancer stem cell. A mammalian cell can be a rodent cell, e.g., a mouse, rat, hamster cell. A mammalian cell can be a lagomorpha cell, e.g., a rabbit cell. A mammalian cell can also be a primate cell, e.g., a human cell. In certain examples, the cells are those used for mass bioproduction, e.g., CHO cells.

[0028] The term "complementarity" refers to the ability of a nucleic acid to form hydrogen bond(s) with another nucleic acid sequence by either traditional Watson-Crick or other non- traditional types. A percent complementarity indicates the percentage of residues in a nucleic acid molecule which can form hydrogen bonds (e.g., Watson-Crick base pairing) with a second nucleic acid sequence (e.g., 5, 6, 7, 8, 9, 10 out of 10 being 50%, 60%>, 70%>, 80%>, 90%, and 100% complementary). "Perfectly complementary" means that all the contiguous residues of a nucleic acid sequence will hydrogen bond with the same number of contiguous residues in a second nucleic acid sequence. "Substantially complementary" as used herein refers to a degree of complementarity that is at least 60%, 65%, 70%, 75%, 80%, 85%, 90%, 95%. 97%, 98%, 99%, or 100% over a region of 8, 9, 10, 11, 12, 13, 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25, 30, 35, 40, 45, 50, or more nucleotides, or refers to two nucleic acids that hybridize under stringent conditions.

[0029] It is noted that in this disclosure, terms such as "comprises", "comprised", "comprising", "contains", "containing" and the like have the meaning attributed in United States Patent law; they are inclusive or open-ended and do not exclude additional, un-recited elements or method steps. Terms such as "consisting essentially of" and "consists essentially of" have the meaning attributed in United States Patent law; they allow for the inclusion of additional ingredients or steps that do not materially affect the basic and novel characteristics of the claimed invention. The terms "consists of" and "consisting of" have the meaning ascribed to them in United States Patent law; namely that these terms are close ended.

[0030] The terms "determining," "assessing," "assaying," "measuring" and "detecting" can be used interchangeably and refer to both quantitative and semi-quantitative determinations. Where either a quantitative and semi-quantitative determination is intended, the phrase "determining a level" of a polynucleotide or polypeptide of interest or "detecting" a polynucleotide or polypeptide of interest can be used.

[0031] The term "hybridizing" refers to the binding, duplexing, or hybridizing of a nucleic acid molecule preferentially to a particular nucleotide sequence under stringent conditions. The term "stringent conditions" refers to conditions under which a probe will

hybridize preferentially to its target subsequence, and to a lesser extent to, or not at all to, other sequences in a mixed population (e.g., a cell lysate or DNA preparation from a tissue biopsy). A "stringent hybridization" and "stringent hybridization wash conditions" in the context of nucleic acid hybridization (e.g., as in array, microarray, Southern or northern hybridizations) are sequence dependent, and are different under different environmental parameters. An extensive guide to the hybridization of nucleic acids is found in, e.g., Tijssen *Laboratory Techniques in Biochemistry and Molecular Biology—Hybridization with Nucleic Acid Probes part I, Ch. 2, "Overview of principles of hybridization and the strategy of nucleic acid probe assays,"* (1993) Elsevier, N.Y. Generally, highly stringent hybridization and wash conditions are selected to be about 5° C lower than the thermal melting point (Tm) for the specific sequence at a defined ionic strength and pH. The Tm is the temperature (under defined ionic strength and pH) at which 50% of the target sequence hybridizes to a perfectly matched probe. Very stringent conditions are selected to be equal to the Tm for a particular probe. An example of stringent hybridization conditions for hybridization of complementary nucleic acids which have more than 100 complementary residues on an array or on a filter in a Southern or northern blot is 42° C. using standard hybridization solutions (see, e.g., Sambrook and Russell *Molecular Cloning: A Laboratory Manual (3rd ed.) Vol. 1-3* (2001) Cold Spring Harbor Laboratory, Cold Spring Harbor Press, NY). An example of highly stringent wash conditions is 0.15 M NaCl at 72° C for about 15 minutes. An example of stringent wash conditions is a 0.2×SSC wash at 65° C for 15 minutes. Often, a high stringency wash is preceded by a low stringency wash to remove background probe signal. An example medium stringency wash for a duplex of, e.g., more than 100 nucleotides, is 1×SSC at 45° C for 15 minutes. An example of a low stringency wash for a duplex of, e.g., more than 100 nucleotides, is 4×SSC to 6×SSC at 40° C for 15 minutes.

[0032] The term "murine model", as used herein a mouse used to study the development and progression of a disease or disorder, such as cancer, and to test treatments before they are given to human.

[0033] The term "nucleic acid" and "polynucleotide" are used interchangeably and refer to a polymeric form of nucleotides of any length, either deoxyribonucleotides or ribonucleotides, or analogs thereof. Polynucleotides may have any three-dimensional structure, and may perform any function, known or unknown. Non-limiting examples of polynucleotides include a gene, a gene fragment, exons, introns, messenger RNA (mRNA), transfer RNA, ribosomal RNA, ribozymes, cDNA, shRNA, single-stranded short or long

RNAs, recombinant polynucleotides, branched polynucleotides, plasmids, vectors, isolated DNA of any sequence, control regions, isolated RNA of any sequence, nucleic acid probes, and primers. The nucleic acid molecule may be linear or circular.

[0034] The term "oligonucleotide" refers to a nucleic acid sequence of at least about five nucleotides to about 500 nucleotides (e.g. 5, 6, 7, 8, 9, 10, 12, 15, 18, 20, 21, 22, 25, 30, 35, 40, 45, 50, 55, 60, 65, 70, 75, 80, 85, 90, 100, 125, 150, 175, 200, 250, 300, 350, 400, 450 or 500 nucleotides). In some embodiments, for example, an oligonucleotide can be from about 15 nucleotides to about 30 nucleotides, or about 20 nucleotides to about 25 nucleotides, which can be used, for example, as a primer in a polymerase chain reaction (PCR) amplification assay and/or as a probe in a hybridization assay or in a microarray. Oligonucleotides of this invention can be natural or synthetic, e.g., DNA, RNA, PNA, LNA, modified backbones, etc., as are well known in the art.

[0035] The term "sample" or "biological sample" used herein refers to any cell, tissue, organoid or any other sample that contains one or more nucleic acid molecule(s) of interest. In certain embodiments, the sample is a cell (e.g., normal cell, cancer cell, cell line), a tissue (e.g., a normal tissue, a cancer tissue, a xenograft or allograft tissue), an organoid, etc.

[0036] The term "substrate" when used in the context of an array refers to material capable of supporting associated assay components (e.g., assay regions, cells, test compounds, etc.). Examples of substrates include, but are not limited to glass, Si-based materials, functionalized polystyrene, functionalized polyethylene-glycol, functionalized organic polymers, nitrocellulose or nylon membranes, paper, cotton, and materials suitable for synthesis. Substrates need not be flat and include any type of shape including spherical shapes (e.g., beads). Materials attached to a substrate may be attached to any portion of the substrate (e.g., may be attached to an interior portion of a porous substrate material). Preferred embodiments of the present technology have nucleic acid probes attached to a substrate. A nucleic acid probe is "attached" to a substrate when it is associated with the substrate through a non-random chemical or physical interaction. In some preferred embodiments, the attachment is through a covalent bond, e.g., as provided by a linker.

[0037] The term "tumor sample" includes a biological sample or a sample from a biological source that contains one or more tumor cells. Biological samples include samples from body fluids, e.g., blood, plasma, serum, or urine, or samples derived, e.g., by biopsy, from cells, tissues or organs, preferably tumor tissue suspected to include or essentially consist of cancer cells.

[0038] Gene Panels for Assessing Immune Response

[0039] Malignant tumors do not only contain cancer cells. Normal cells form the body also infiltrate tumors. For example, a variety of immune cells infiltrating tumors can help detect and kill cancer cells, which has important impact on tumor progression and response to therapy. While gene expression levels are often studied in tumors, the proportion of different cell type, such as immune cells in the tumor, is rarely studied at the gene expression level.

[0040] The methods and compositions described herein are based, in part, on the discovery of a group of genes that can be used to identify and assess the immune response, such as the proportion of various immune cells in tumors. In certain embodiment, the tumors are obtained in a murine tumor model. In certain embodiments, the genes are selected based on the genomic data from non-tumor bearing mice as well as tumor bearing mice. In certain embodiments, the group of genes can be used to identify at least 8 different immune cell types, including B-cell, dendritic cell, macrophage, monocyte, natural killer cell, CD4+ T cell and CD8+ T cell. In certain embodiments, the group of genes covers surface markers and transcriptomic biomarkers for immune system, the key pathways at the interface of the tumor, tumor microenvironment, and immune response, as well as internal reference genes for data normalization.

[0041] In certain embodiments, the group of genes used herein are selected from the group as shown in **Table 1**.

[0042] Methods of Detecting the Expression Level of Gene Panel

[0043] In one aspect, the present disclosure provides a method for assessing immune response in a murine model. In certain embodiments, the method comprises: obtaining a sample from the murine model; detecting the expression level of a panel of genes in the sample, wherein the panel comprises at least 50, 100, 150, 200, 250, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000 genes selected from the group as shown in **Table 1**; comparing the expression level of the panel to a reference expression level; and evaluating the immune response in the murine model.

[0044] In certain embodiments, the expression level of the gene panel can be detected in the RNA (e.g., mRNA) level using proper methods known in the art, including, without limitation, amplification assay, hybridization assay and sequencing assay.

[0045] *Amplification assay*

[0046] A nucleic acid amplification assay involves copying a target nucleic acid (e.g. DNA or RNA), thereby increasing the number of copies of the amplified nucleic acid sequence. Amplification may be exponential or linear. Exemplary nucleic acid amplification methods include, but are not limited to, amplification using the polymerase chain reaction ("PCR", see U.S. Patents 4,683,195 and 4,683,202; PCR Protocols: A Guide To Methods And Applications (Innis et al., eds, 1990)), reverse transcriptase polymerase chain reaction (RT-PCR), quantitative real-time PCR (qRT-PCR); quantitative PCR, such as TaqMan®, nested PCR, ligase chain reaction (See Abravaya, K., et al., Nucleic Acids Research, 23:675-682, (1995), branched DNA signal amplification (see, Urdea, M. S., et al., AIDS, 7 (suppl 2):S11-S14, (1993), amplifiable RNA reporters, Q-beta replication (see Lizardi et al., *Biotechnology* (1988) 6: 1197), transcription-based amplification (see, Kwoh et al., *Proc. Natl. Acad. Sci. USA* (1989) 86: 1173-1177), boomerang DNA amplification, strand displacement activation, cycling probe technology, self-sustained sequence replication (Guatelli et al., *Proc. Natl. Acad. Sci. USA* (1990) 87:1874-1878), rolling circle replication (U.S. Patent No. 5,854,033), isothermal nucleic acid sequence based amplification (NASBA), and serial analysis of gene expression (SAGE).

[0047] In certain embodiments, the nucleic acid amplification assay is a PCR-based method. PCR is initiated with a pair of primers that hybridize to the target nucleic acid sequence to be amplified, followed by elongation of the primer by polymerase which synthesizes the new strand using the target nucleic acid sequence as a template and dNTPs as building blocks. Then the new strand and the target strand are denatured to allow primers to bind for the next cycle of extension and synthesis. After multiple amplification cycles, the total number of copies of the target nucleic acid sequence can increase exponentially. When the nucleic acid obtained from a sample is RNA, the amplification step may optionally comprise a reverse transcription step to produce cDNA of the RNA in the sample. The cDNA is then amplified using the primers to allow detection of expression level of the genes of interest.

[0048] In certain embodiments, intercalating agents that produce a signal when intercalated in double stranded DNA may be used. Exemplary agents include SYBR GREEN™ and SYBR GOLD™. Since these agents are not template-specific, it is assumed that the signal is generated based on template-specific amplification. This can be confirmed by monitoring signal as a function of temperature because melting point of template sequences will generally be much higher than, for example, primer-dimers, etc.

[0049] In certain embodiments, a detectably labeled primer or a detectably labeled probe can be used, to allow detection of the mRNA (or cDNA reverse transcribed from mRNA) of the gene of interest corresponding to that primer or probe. In certain embodiments, multiple labeled primers or labeled probes with different detectable labels can be used to allow simultaneous detection of the expression of multiple gene of interest.

[0050] *Hybridization assay*

[0051] Nucleic acid hybridization assays use probes to hybridize to the target nucleic acid, thereby allowing detection of the target nucleic acid. Non-limiting examples of hybridization assay include Northern blotting, Southern blotting, in situ hybridization, microarray analysis, and multiplexed hybridization-based assays.

[0052] In certain embodiments, the probes for hybridization assay are detectably labeled. In certain embodiments, the nucleic acid-based probes for hybridization assay are unlabeled. Such unlabeled probes can be immobilized on a solid support, such as a microarray, and can hybridize to the target nucleic acid molecules which are detectably labeled.

[0053] In certain embodiments, hybridization assays can be performed by isolating the nucleic acids (e.g., RNA or DNA), separating the nucleic acids (e.g., by gel electrophoresis) followed by transfer of the separated nucleic acid on suitable membrane filters (e.g. nitrocellulose filters), where the probes hybridize to the target nucleic acids and allows detection. See, for example, Molecular Cloning: A Laboratory Manual, J. Sambrook et al., eds., 2nd edition, Cold Spring Harbor Laboratory Press, 1989, Chapter 7. The hybridization of the probe and the target nucleic acid can be detected or measured by methods known in the art. For example, autoradiographic detection of hybridization can be performed by exposing hybridized filters to photographic film.

[0054] In some embodiments, hybridization assays can be performed on microarrays. Microarrays provide a method for the simultaneous measurement of the levels of large numbers of target nucleic acid molecules. The target nucleic acids can be RNA, DNA, cDNA reverse transcribed from mRNA, or chromosomal DNA. The target nucleic acids can be allowed to hybridize to a microarray comprising a substrate having multiple immobilized nucleic acid probes arrayed at a density of up to several million probes per square centimeter of the substrate surface. The RNA or DNA in the sample is hybridized to complementary probes on the array and then detected by laser scanning. Hybridization intensities for each probe on the array are determined and converted to a quantitative value representing relative

levels of the RNA or DNA. See, U.S. Patent Nos. 6,040,138, 5,800,992 and 6,020,135, 6,033,860, and 6,344,316.

[0055] Techniques for the synthesis of these arrays using mechanical synthesis methods are described in, e.g., U.S. Patent No. 5,384,261. Although a planar array surface is often employed the array may be fabricated on a surface of virtually any shape or even a multiplicity of surfaces. Arrays may be peptides or nucleic acids on beads, gels, polymeric surfaces, fibers such as fiber optics, glass or any other appropriate substrate, see U.S. Patent Nos. 5,770,358, 5,789,162, 5,708,153, 6,040,193 and 5,800,992. Arrays may be packaged in such a manner as to allow for diagnostics or other manipulation of an all-inclusive device. Useful microarrays are also commercially available, for example, microarrays from Affymetrix, from Nano String Technologies, QuantiGene 2.0 Multiplex Assay from Panomics.

[0056] *Sequencing methods*

[0057] Sequencing methods useful in the measurement of the expression of the gene panel involves sequencing of the target nucleic acid. Any sequencing known in the art can be used to detect the expression of the gene of interest. In general, sequencing methods can be categorized to traditional or classical methods and high throughput sequencing (next generation sequencing). Traditional sequencing methods include Maxam-Gilbert sequencing (also known as chemical sequencing) and Sanger sequencing (also known as chain-termination methods).

[0058] High throughput sequencing, or next generation sequencing, by using methods distinguished from traditional methods, such as Sanger sequencing, is highly scalable and able to sequence the entire genome or transcriptome at once. High throughput sequencing involves sequencing-by-synthesis, sequencing-by-ligation, and ultra-deep sequencing (such as described in Marguiles et al., *Nature* 437 (7057): 376-80 (2005)). Sequence-by-synthesis involves synthesizing a complementary strand of the target nucleic acid by incorporating labeled nucleotide or nucleotide analog in a polymerase amplification. Immediately after or upon successful incorporation of a label nucleotide, a signal of the label is measured and the identity of the nucleotide is recorded. The detectable label on the incorporated nucleotide is removed before the incorporation, detection and identification steps are repeated. Examples of sequence-by-synthesis methods are known in the art, and are described for example in U.S. Pat. No. 7,056,676, U.S. Pat. No. 8,802,368 and U.S. Pat. No. 7,169,560, the contents of which are incorporated herein by reference. Sequencing-by-synthesis may be performed on a solid surface (or a microarray or a chip) using fold-back PCR and anchored primers. Target

nucleic acid fragments can be attached to the solid surface by hybridizing to the anchored primers, and bridge amplified. This technology is used, for example, in the Illumina® sequencing platform.

[0059] Pyrosequencing involves hybridizing the target nucleic acid regions to a primer and extending the new strand by sequentially incorporating deoxynucleotide triphosphates corresponding to the bases A, C, G, and T (U) in the presence of a polymerase. Each base incorporation is accompanied by release of pyrophosphate, converted to ATP by sulfurylase, which drives synthesis of oxyluciferin and the release of visible light. Since pyrophosphate release is equimolar with the number of incorporated bases, the light given off is proportional to the number of nucleotides adding in any one step. The process is repeated until the entire sequence is determined.

[0060] In certain embodiments, the expression of the genes of interest is detected by whole transcriptome shotgun sequencing (RNA sequencing). The method of RNA sequencing has been described (see Wang Z, Gerstein M and Snyder M, *Nature Review Genetics* (2009) 10:57-63; Maher CA et al., *Nature* (2009) 458:97-101; Kukurba K & Montgomery SB, Cold Spring Harbor Protocols (2015) 2015(11): 951-969). In certain embodiment, samples subject to the RNA sequencing are pre-treated to enrich the expression product of the genes of interest, e.g., using oligonucleotide-based hybridization/capture techniques. In certain embodiments, the probes used in the hybridization/capture techniques have a length ranging from 20-200 nucleotides, 20-500 nucleotides, 20-1000 nucleotides, 20-2000 nucleotides, 20-5000 nucleotides, or 20-7000 nucleotides.

[0061] Comparing with a Reference Level

[0062] In certain embodiments, the methods disclosed herein include a step of comparing the detected expression level of the gene panel to a reference expression level.

[0063] The term "reference expression level" refers to a level of expression that is representative of a reference sample. In certain embodiments, the reference sample is obtained from a healthy (e.g., non-tumor bearing) subject or tissue. In certain embodiments, the reference sample is a cancer or tumor tissue. In certain embodiments, the reference sample is obtained from a subject or tissue in which the immune response is activated, e.g., by the tumor or therapeutics (e.g., treated by an anti-tumor therapy, e.g., an anti-immune checkpoint antibody, such as ani-PD-1, anti-PD-L1). In certain embodiments, the reference expression level is obtained using the same or comparable measurement method or assay as used in the detection of the expression level of the target gene in the test sample.

[0064] In certain embodiments, the reference expression level can be predetermined. For example, the reference expression level can be calculated or generalized based on measurements of the expression level of the target gene in a collection of general cancer or tumor samples or tissues from a tumor of the same type. For another example, the reference expression level can be based on statistics of the expression level of the target gene generally observed in an average cancer or tumor samples from a general cancer or tumor population.

[0065] In certain embodiments, the comparing step in the method provided herein involves determining the difference between the detected expression level and the reference expression level. The difference from the reference expression level can be elevation or reduction.

[0066] In certain embodiments, the difference from the reference expression level is further compared with a threshold. In certain embodiments, a threshold can be set by statistical methods, such that if the difference from the reference expression level reaches the threshold, such difference can be considered statistically significant. Useful statistical analysis methods are described in L. D. Fisher & G. vanBelle, Biostatistics: A Methodology for the Health Sciences (Wiley-Interscience, NY, 1993). Statistically significance can be determined based on confidence ("p") values, which can be calculated using an unpaired 2-tailed t test. A p value less than or equal to, for example, 0.1, 0.05, 0.025, or 0.01 usually can be used to indicated statistical significance. Confidence intervals and p-values can be determined by methods well-known in the art. See, e.g., Dowdy and Wearden, Statistics for Research, John Wiley & Sons, New York, 1983.

[0067] Evaluation of anti-tumor therapy

[0068] In another aspect, the present disclosure further provides a method to evaluate a treatment in a murine model based on the expression level of a panel of genes as disclosed herein. In certain embodiments, the method comprises administering a treatment to a murine model; obtaining a sample from the murine model; detecting the expression level of a panel of genes in the sample, wherein the panel comprises at least 50 genes selected from the group as shown in **Table 1**; comparing the expression level of the panel to a reference expression level; and evaluating the immune response in the murine model.

[0069] In certain embodiments, the treatment involves an anti-hormonal agent, including natural or synthetic organic or peptide compounds that act to regulate or inhibit hormone action on tumors.

[0070] In certain embodiments, the treatment involves an angiogenesis inhibitor, including VEGFR inhibitors, integrin receptor antagonists and integrin antagonists.

[0071] In certain embodiments, the treatment involves a tumor cell pro-apoptotic or apoptosis-stimulating agent.

[0072] In certain embodiments, the treatment involves a signal transduction inhibitor, such as an erbB2 receptor inhibitors, inhibitors of other protein tyrosine-kinases; ras inhibitors; raf inhibitors; MEK inhibitors; mTOR inhibitors; cyclin dependent kinase inhibitors; protein kinase C inhibitors; and PDK-1 inhibitors.

[0073] In certain embodiments, the treatment involves an anti-proliferative agent. Anti-proliferative agents include, for example: inhibitors of the enzyme farnesyl protein transferase and inhibitors of the receptor tyrosine kinase PDGFR.

[0074] In certain embodiments, the treatment involves a cancer immunotherapy agent, such as an antibody specifically binding to an immune checkpoint. Immune checkpoints include, for example: A2AR, B7.1, B7.2, B7-H2, B7-H3, B7-H4, B7-H6, BTLA, CD48, CD160, CD244, CTLA-4, ICOS, LAG-3, LILRB1, LILRB2, LILRB4, OX40, PD-1, PD-L1, PD-L2, SIRPalpha (CD47), TIGIT, TIM-3, TIM-1, TIM-4, and VISTA.

[0075] Kits and Microarrays

[0076] In another aspect, the present disclosure provides kits for use in the methods described above. The kits may comprise any or all of the reagents to perform the methods described herein. In certain embodiments, the kit comprises primers for detecting the expression of a panel of genes in a sample from the murine model. In certain embodiments, the panel comprises at least 50, 100, 150, 200, 250, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000 genes selected from the group as shown in Table 1.

[0077] In certain embodiments, the kit further comprises an agent for amplifying the expression product, e.g., mRNA, of the gene panel using the primers. In addition, the kits may include instructional materials containing directions (i.e., protocols) for the practice of the methods provided herein. While the instructional materials typically comprise written or printed materials they are not limited to such. Any medium capable of storing such instructions and communicating them to an end user is contemplated by this invention. Such media include, but are not limited to electronic storage media (e.g., magnetic discs, tapes, cartridges, chips), optical media (e.g., CD ROM), and the like. Such media may include addresses to internet sites that provide such instructional materials.

[0078] In another aspect, the present disclosure provides oligonucleotide probes for detecting the expression level of a panel of genes in a sample from the murine model. In certain embodiments, the panel comprises at least 50, 100, 150, 200, 250, 300, 350, 400, 450, 500, 550, 600, 650, 700, 750, 800, 850, 900, 950, 1000 genes selected from the group as shown in Table 1.

[0079] In certain embodiments, the probes are attached to a solid support, such as an array slide or chip, e.g., as described in Eds., Bowtell and Sambrook *DNA Microarrays: A Molecular Cloning Manual* (2003) Cold Spring Harbor Laboratory Press. Construction of such devices are well known in the art, for example as described in US Patents and Patent Publications U.S. Patent No. 5,837,832; PCT application W095/11995; U.S. Patent No. 5,807,522; US Patent Nos. 7,157,229, 7,083,975, 6,444,175, 6,375,903, 6,315,958, 6,295,153, and 5,143,854, 2007/0037274, 2007/0140906, 2004/0126757, 2004/0110212, 2004/0110211, 2003/0143550, 2003/0003032, and 2002/0041420. Nucleic acid arrays are also reviewed in the following references: *Biotechnol Annu Rev* (2002) 8:85-101; Sosnowski et al. *Psychiatr Genet* (2002)12(4): 181-92; Heller, *Annu Rev Biomed Eng* (2002) 4: 129-53; Kolchinsky et al., *Hum. Mutat* (2002) 19(4):343-60; and McGail et al., *Adv Biochem Eng Biotechnol* (2002) 77:21-42.

[0080] A microarray can be composed of a large number of unique, single-stranded polynucleotides, usually either synthetic antisense polynucleotides or fragments of cDNAs, fixed to a solid support. Typical polynucleotides are preferably about 6-60 nucleotides in length, more preferably about 15-30 nucleotides in length, and most preferably about 18-25 nucleotides in length. For certain types of arrays or other detection kits/systems, it may be preferable to use oligonucleotides that are only about 7-20 nucleotides in length. In other types of arrays, such as arrays used in conjunction with chemiluminescent detection technology, preferred probe lengths can be, for example, about 15-80 nucleotides in length, preferably about 50-70 nucleotides in length, more preferably about 55-65 nucleotides in length, and most preferably about 60 nucleotides in length.

[0081] Techniques for the synthesis of these arrays using mechanical synthesis methods are described in, e.g., U.S. Patent No. 5,384,261. Although a planar array surface is often employed the array may be fabricated on a surface of virtually any shape or even a multiplicity of surfaces. Arrays may also be nucleic acids on beads, gels, polymeric surfaces, fibers such as fiber optics, glass or any other appropriate substrate, see U.S. Patent Nos. 5,770,358, 5,789,162, 5,708,153, 6,040,193 and 5,800,992. Arrays may be packaged in such a manner as to allow for diagnostics or other manipulation of an all-inclusive device.

[0082] The probes and primers necessary for practicing the present invention can be synthesized and labeled using well known techniques. Oligonucleotides used as probes and primers may be chemically synthesized according to the solid phase phosphoramidite triester method first described by Beaucage and Caruthers, *Tetrahedron Letts.* (1981) 22: 1859-1862, using an automated synthesizer, as described in Needham-Van Devanter et al, Nucleic Acids Res. (1984) 12:6159-6168.

[0083] Computer-implemented Methods, Systems and Devices

[0084] Any of the methods described herein may be totally or partially performed with a computer system including one or more processors, which can be configured to perform the steps. Thus, embodiments are directed to computer systems configured to perform the steps of any of the methods described herein, potentially with different components performing a respective step or a respective group of steps. Although presented as numbered steps, steps of methods herein can be performed at a same time or in a different order. Additionally, portions of these steps may be used with portions of other steps from other methods. Also, all or portions of a step may be optional. Any of the steps of any of the methods can be performed with modules, circuits, or other means for performing these steps.

[0085] Any of the computer systems mentioned herein may utilize any suitable number of subsystems. In some embodiments, a computer system includes a single computer apparatus, where the subsystems can be the components of the computer apparatus. In other embodiments, a computer system can include multiple computer apparatuses, each being a subsystem, with internal components. The subsystems can be interconnected via a system bus. Additional subsystems include, for examples, a printer, keyboard, storage device(s), monitor, which is coupled to display adapter, and others. Peripherals and input/output (I/O) devices, which couple to I/O controller, can be connected to the computer system by any number of means known in the art, such as serial port. For example, serial port or external interface (e.g., Ethernet, Wi-Fi, etc.) can be used to connect computer system to a wide area network such as the Internet, a mouse input device, or a scanner. The interconnection via system bus allows the central processor to communicate with each subsystem and to control the execution of instructions from system memory or the storage device(s) (e.g., a fixed disk, such as a hard drive or optical disk), as well as the exchange of information between subsystems. The system memory and/or the storage device(s) may embody a computer readable medium. Any of the data mentioned herein can be output from one component to another component and can be output to the user.

[0086] A computer system can include a plurality of the same components or subsystems, e.g., connected together by external interface or by an internal interface. In some embodiments, computer systems, subsystem, or apparatuses can communicate over a network. In such instances, one computer can be considered a client and another computer a server, where each can be part of a same computer system. A client and a server can each include multiple systems, subsystems, or components.

[0087] It should be understood that any of the embodiments of the present disclosure can be implemented in the form of control logic using hardware (e.g., an application specific integrated circuit or field programmable gate array) and/or using computer software with a generally programmable processor in a modular or integrated manner. As used herein, a processor includes a multi-core processor on a same integrated chip, or multiple processing units on a single circuit board or networked. Based on the disclosure and teachings provided herein, a person of ordinary skill in the art will know and appreciate other ways and/or methods to implement embodiments of the present disclosure using hardware and a combination of hardware and software.

[0088] Any of the software components or functions described in this application may be implemented as software code to be executed by a processor using any suitable computer language such as, for example, Java, C++ or Perl using, for example, conventional or object-oriented techniques. The software code may be stored as a series of instructions or commands on a computer readable medium for storage and/or transmission, suitable media include random access memory (RAM), a read only memory (ROM), a magnetic medium such as a hard-drive or a floppy disk, or an optical medium such as a compact disk (CD) or DVD (digital versatile disk), flash memory, and the like. The computer readable medium may be any combination of such storage or transmission devices.

[0089] Such programs may also be encoded and transmitted using carrier signals adapted for transmission via wired, optical, and/or wireless networks conforming to a variety of protocols, including the Internet. As such, a computer readable medium according to an embodiment of the present invention may be created using a data signal encoded with such programs. Computer readable media encoded with the program code may be packaged with a compatible device or provided separately from other devices (e.g., via Internet download). Any such computer readable medium may reside on or within a single computer product (e.g. a hard drive, a CD, or an entire computer system), and may be present on or within different computer products within a system or network. A computer system may include a monitor, printer, or other suitable display for providing any of the results mentioned herein to a user.

[0090] The following examples are provided to better illustrate the claimed invention and are not to be interpreted as limiting the scope of the invention. All specific compositions, materials, and methods described below, in whole or in part, fall within the scope of the present invention. These specific compositions, materials, and methods are not intended to limit the invention, but merely to illustrate specific embodiments falling within the scope of the invention. One skilled in the art may develop equivalent compositions, materials, and methods without the exercise of inventive capacity and without departing from the scope of the invention. It will be understood that many variations can be made in the procedures herein described while still remaining within the bounds of the present invention. It is the intention of the inventors that such variations are included within the scope of the invention.

EXAMPLE 1

[0091] This example illustrates the identification of murine pan-cancer gene expression panel for immuno-oncology and tumor microenvironment studies

[0092] Materials and Methods

[0093] Marker genes were identified by combining public and in-house reference genomic data for non-tumor bearing mice as well as tumor-bearing mouse models. Oligonucleotide-based hybridization/capture techniques, analogous to exome sequencing of targeting specific regions, are established on Illumina Novaseq and BGI MGIseq platforms. The results were further verified using PCR assays and eight control panels with murine immune cells and tumor cell lines.

[0094] Results

[0095] Based on the immune signature analysis, the inventors selected 1080 genes (see **Table 1**) that cover surface markers and transcriptomic biomarkers for immune system, the key pathways at the interface of the tumor, tumor microenvironment, and immune response, as well as internal reference genes for data normalization. The validation panels showed good consistency of gene expression in respective cell types (see **FIGS. 2** and **3**). The inventors also estimated the cell fractions in validation panels based on the expression of the gene panel (see **FIG. 4**).

[0096] The murine immune-oncology NGS panel can be used to characterize tumor-immune interactions more efficiently and in a cost-effective manner for preclinical studies, which will provide an enhanced insight into the tumor microenvironment, especially following treatment with immuno-therapy, with high-content data that has the potential to better understand the lack of response versus response, which ultimately could translate more

effectively into the clinic as well as for early identification of immunotherapy companion diagnostics.

[0097] While the disclosure has been particularly shown and described with reference to specific embodiments (some of which are preferred embodiments), it should be understood by those having skill in the art that various changes in form and detail may be made therein without departing from the spirit and scope of the present disclosure as disclosed herein.

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000030724	Cd19	ENSMUSG00000036887	C1qa
ENSMUSG00000026447	Pik3c2b	ENSMUSG00000036856	Wnt4
ENSMUSG00000026616	Cr2	ENSMUSG00000058908	Pla2g2a
ENSMUSG00000020183	Cpm	ENSMUSG00000028914	Casp9
ENSMUSG00000095105	Edaradd	ENSMUSG00000028599	Tnfrsf1b
ENSMUSG00000051022	Hs3st1	ENSMUSG00000028602	Tnfrsf8
ENSMUSG00000005540	Fcer2a	ENSMUSG00000028991	Mtor
ENSMUSG00000044694	2010007H06Rik	ENSMUSG00000039936	Pik3cd
ENSMUSG00000002007	Srpk3	ENSMUSG00000063524	Eno1
ENSMUSG00000079445	B3gnt7	ENSMUSG00000028965	Tnfrsf9
ENSMUSG00000057337	Chst3	ENSMUSG00000024793	Tnfrsf25
ENSMUSG00000057098	Ebf1	ENSMUSG00000042333	Tnfrsf14
ENSMUSG00000017400	Stac2	ENSMUSG00000029075	Tnfrsf4
ENSMUSG00000001027	Scn4a	ENSMUSG00000041954	Tnfrsf18
ENSMUSG00000104213	Ighd	ENSMUSG00000035692	Isg15
ENSMUSG00000022416	Cacna1i	ENSMUSG00000040274	Cdk6
ENSMUSG00000042514	Klhl14	ENSMUSG00000002944	Cd36
ENSMUSG00000037071	Scd1	ENSMUSG00000042453	Reln
ENSMUSG00000050556	Kcnb1	ENSMUSG00000025746	Il6
ENSMUSG00000014030	Pax5	ENSMUSG00000025747	Tyms
ENSMUSG00000052271	Bhlha15	ENSMUSG00000029145	Eif2b4
ENSMUSG00000071226	Cecr2	ENSMUSG00000029084	Cd38
ENSMUSG00000030256	Bhlhe41	ENSMUSG00000029086	Prom1
ENSMUSG00000030313	Dennd5b	ENSMUSG00000044827	Tlr1
ENSMUSG00000023274	Cd4	ENSMUSG00000062960	Kdr
ENSMUSG00000001444	Tbx21	ENSMUSG00000029371	Cxcl5
ENSMUSG00000015619	Gata3	ENSMUSG00000029373	Pf4
ENSMUSG00000028150	Rorc	ENSMUSG00000029379	Cxcl3
ENSMUSG00000039521	Foxp3	ENSMUSG00000029375	Cxcl15
ENSMUSG00000026011	Ctla4	ENSMUSG00000029380	Cxcl1
ENSMUSG00000057329	Bcl2	ENSMUSG00000058427	Cxcl2
ENSMUSG00000026581	Sell	ENSMUSG00000029378	Areg
ENSMUSG00000019982	Myb	ENSMUSG00000029417	Cxcl9
ENSMUSG00000048756	Foxo3	ENSMUSG00000060183	Cxcl11
ENSMUSG00000038151	Prdm1	ENSMUSG00000023078	Cxcl13
ENSMUSG00000037868	Egr2	ENSMUSG00000046709	Mapk10
ENSMUSG00000019942	Cdk1	ENSMUSG00000042190	Cmklr1
ENSMUSG00000044199	S1pr4	ENSMUSG00000041827	Oasl1
ENSMUSG00000020297	Nsg2	ENSMUSG00000029556	Hnf1a
ENSMUSG00000054404	Slfn5	ENSMUSG00000029602	Rasal1
ENSMUSG00000020644	Id2	ENSMUSG00000032690	Oas2
ENSMUSG00000015437	Gzmb	ENSMUSG00000032661	Oas3

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000022346	Myc	ENSMUSG00000052776	Oas1a
ENSMUSG00000075602	Ly6a	ENSMUSG00000043733	Ptpn11
ENSMUSG00000000028	Cdc45	ENSMUSG00000025534	Gusb
ENSMUSG00000026872	Zeb2	ENSMUSG00000049551	Fzd9
ENSMUSG00000027720	Il2	ENSMUSG00000070464	Ccl26
ENSMUSG00000045092	S1pr1	ENSMUSG00000004814	Ccl24
ENSMUSG00000062585	Cnr2	ENSMUSG00000025856	Pdgfa
ENSMUSG00000007872	Id3	ENSMUSG00000079109	Pms2
ENSMUSG00000029178	Klf3	ENSMUSG00000029648	Flt1
ENSMUSG00000030103	Bhlhe40	ENSMUSG00000066551	Hmgb1
ENSMUSG00000030156	Cd69	ENSMUSG00000041147	Brca2
ENSMUSG00000055148	Klf2	ENSMUSG00000009376	Met
ENSMUSG00000032294	Pkm	ENSMUSG00000010797	Wnt2
ENSMUSG00000052336	Cx3cr1	ENSMUSG00000029771	Irf5
ENSMUSG00000050232	Cxcr3	ENSMUSG00000057137	Tmem140
ENSMUSG00000053977	Cd8a	ENSMUSG00000038507	Parp12
ENSMUSG00000003134	Tbc1d8	ENSMUSG00000029915	Clec5a
ENSMUSG00000054702	Ap1s3	ENSMUSG00000029687	Ezh2
ENSMUSG00000038179	Slamf7	ENSMUSG00000054435	Gimap4
ENSMUSG00000039783	Kmo	ENSMUSG00000047867	Gimap6
ENSMUSG00000004665	Cnn2	ENSMUSG00000029798	Herc6
ENSMUSG00000000861	Bcl11a	ENSMUSG00000025889	Snca
ENSMUSG00000011256	Adam19	ENSMUSG00000049093	Il23r
ENSMUSG00000037944	Ccr7	ENSMUSG00000053044	Cd8b1
ENSMUSG00000047415	Gpr68	ENSMUSG00000030045	Mrpl19
ENSMUSG00000021298	Gpr132	ENSMUSG00000000628	Hk2
ENSMUSG00000022504	Ciita	ENSMUSG00000034245	Hdac11
ENSMUSG00000052013	Btla	ENSMUSG00000033933	Vhl
ENSMUSG00000067341	H2-Eb2	ENSMUSG00000000440	Pparg
ENSMUSG00000073409	H2-Q6	ENSMUSG00000061353	Cxcl12
ENSMUSG00000000673	Haao	ENSMUSG00000030170	Wnt5b
ENSMUSG00000024789	Jak2	ENSMUSG00000004446	Bid
ENSMUSG00000076441	Ass1	ENSMUSG00000030111	A2m
ENSMUSG00000026875	Traf1	ENSMUSG00000030142	Clec4e
ENSMUSG00000035000	Dpp4	ENSMUSG00000008845	Cd163
ENSMUSG00000027583	Zbtb46	ENSMUSG00000023456	Tpi1
ENSMUSG00000070691	Runx3	ENSMUSG00000030124	Lag3
ENSMUSG00000039899	Fgl2	ENSMUSG00000038213	Tapbpl
ENSMUSG00000005672	Kit	ENSMUSG00000030336	Cd27
ENSMUSG00000037905	Bri3bp	ENSMUSG00000000184	Ccnd2
ENSMUSG00000042817	Flt3	ENSMUSG00000079299	Klrb1
ENSMUSG00000067610	Klri1	ENSMUSG00000079293	Clec7a

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000074227	Spint2	ENSMUSG00000030162	Olr1
ENSMUSG00000039062	Anpep	ENSMUSG00000030165	Klrd1
ENSMUSG00000030643	Rab30	ENSMUSG00000079853	Klra1
ENSMUSG00000006362	Cbfa2t3	ENSMUSG00000030246	Ldhb
ENSMUSG00000032322	Pstpip1	ENSMUSG00000030268	Bcat1
ENSMUSG00000066456	Hmgn3	ENSMUSG00000030265	Kras
ENSMUSG00000047678	Gpr82	ENSMUSG00000058818	Pirb
ENSMUSG00000050921	P2ry10	ENSMUSG00000055541	Lair1
ENSMUSG00000052688	Rab7b	ENSMUSG00000070873	Lilra5
ENSMUSG00000034675	Dbn1	ENSMUSG00000004371	Il11
ENSMUSG00000025279	Dnase1l3	ENSMUSG00000049130	C5ar1
ENSMUSG00000090877	Hspa1b	ENSMUSG00000002083	Bbc3
ENSMUSG00000091971	Hspa1a	ENSMUSG00000001918	Slc1a5
ENSMUSG00000028654	Mycl	ENSMUSG00000053228	Ceacam3
ENSMUSG00000031951	Tmem231	ENSMUSG00000040987	Mill2
ENSMUSG00000039452	Snx22	ENSMUSG00000002983	Relb
ENSMUSG00000060509	Xcr1	ENSMUSG00000062300	Nectin2
ENSMUSG00000031355	Arhgap6	ENSMUSG00000040525	Cblc
ENSMUSG00000025938	Slco5a1	ENSMUSG00000040511	Pvr
ENSMUSG00000025964	Adam23	ENSMUSG00000003379	Cd79a
ENSMUSG00000039395	Mreg	ENSMUSG00000002602	Axl
ENSMUSG00000005338	Cadm3	ENSMUSG00000030603	Psmc4
ENSMUSG00000020926	Adam11	ENSMUSG00000002068	Ccne1
ENSMUSG00000021087	Rtn1	ENSMUSG00000039013	Siglecf
ENSMUSG00000004562	Arhgef40	ENSMUSG00000004612	Nkg7
ENSMUSG00000044309	Apol7c	ENSMUSG00000008193	Spib
ENSMUSG00000003352	Cacnb3	ENSMUSG00000038644	Pold1
ENSMUSG00000075269	Bex6	ENSMUSG00000003184	Irf3
ENSMUSG00000024124	Prss30	ENSMUSG00000003873	Bax
ENSMUSG00000015468	Notch4	ENSMUSG00000063229	Ldha
ENSMUSG00000028327	Stra6l	ENSMUSG00000030605	Mfge8
ENSMUSG00000029101	Rgs12	ENSMUSG00000030528	Blm
ENSMUSG00000029581	Fscn1	ENSMUSG00000001741	Il16
ENSMUSG00000031486	Adgra2	ENSMUSG00000015709	Arnt2
ENSMUSG00000005947	Itgae	ENSMUSG00000015957	Wnt11
ENSMUSG00000025779	Ly96	ENSMUSG00000070436	Serpinh1
ENSMUSG00000026155	Smap1	ENSMUSG00000070427	Il18bp
ENSMUSG00000026117	Zap70	ENSMUSG00000030966	Trim21
ENSMUSG00000026073	Il1r2	ENSMUSG00000030790	Adm
ENSMUSG00000062939	Stat4	ENSMUSG00000034990	Otoa
ENSMUSG00000026104	Stat1	ENSMUSG00000030695	Aldoa
ENSMUSG00000026103	Gls	ENSMUSG00000042492	Tbc1d10b

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000026029	Casp8	ENSMUSG00000030830	Itgal
ENSMUSG00000026012	Cd28	ENSMUSG00000030789	Itgax
ENSMUSG00000026009	Icos	ENSMUSG00000054555	Adam12
ENSMUSG00000026180	Cxcr2	ENSMUSG00000031004	Mki67
ENSMUSG00000026177	Slc11a1	ENSMUSG00000054612	Mgmt
ENSMUSG00000026167	Wnt10a	ENSMUSG00000078566	Bnip3
ENSMUSG00000006538	Ihh	ENSMUSG00000060591	Ifitm2
ENSMUSG00000026213	Stk11ip	ENSMUSG00000025491	Ifitm1
ENSMUSG00000043230	Fam124b	ENSMUSG00000054065	Pkp3
ENSMUSG00000026166	Ccl20	ENSMUSG00000025499	Hras
ENSMUSG00000048126	Col6a3	ENSMUSG00000025498	Irf7
ENSMUSG00000007805	Twist2	ENSMUSG00000037664	Cdkn1c
ENSMUSG00000026313	Hdac4	ENSMUSG00000031077	Fadd
ENSMUSG00000026285	Pdcd1	ENSMUSG00000070348	Ccnd1
ENSMUSG00000067006	Serpinb5	ENSMUSG00000040197	Cd209e
ENSMUSG00000026390	Marco	ENSMUSG00000023235	Ccl25
ENSMUSG00000045382	Cxcr4	ENSMUSG00000031465	Angpt2
ENSMUSG00000026420	Il24	ENSMUSG00000031537	Ikbkb
ENSMUSG00000016529	Il10	ENSMUSG00000031548	Sfrp1
ENSMUSG00000070645	Ren1	ENSMUSG00000031551	Ido1
ENSMUSG00000026429	Ube2t	ENSMUSG00000031565	Fgfr1
ENSMUSG00000026395	Ptprc	ENSMUSG00000031490	Eif4ebp1
ENSMUSG00000032487	Ptgs2	ENSMUSG00000031529	Tnks
ENSMUSG00000026479	Lamc2	ENSMUSG00000031639	Tlr3
ENSMUSG00000026700	Tnfsf4	ENSMUSG00000031628	Casp3
ENSMUSG00000066755	Tnfsf18	ENSMUSG00000031627	Irf2
ENSMUSG00000000817	Fasl	ENSMUSG00000031520	Vegfc
ENSMUSG00000026582	Sele	ENSMUSG00000036246	Gmip
ENSMUSG00000026580	Selp	ENSMUSG00000031849	Comp
ENSMUSG00000038463	Olfml2b	ENSMUSG00000056204	Pgpep1
ENSMUSG00000070524	Fcrlb	ENSMUSG00000031834	Pik3r2
ENSMUSG00000026656	Fcgr2b	ENSMUSG00000031805	Jak3
ENSMUSG00000059089	Fcgr4	ENSMUSG00000031712	Il15
ENSMUSG00000004709	Cd244a	ENSMUSG00000055994	Nod2
ENSMUSG00000004707	Ly9	ENSMUSG00000056608	Chd9
ENSMUSG00000015355	Cd48	ENSMUSG00000031666	Rbl2
ENSMUSG00000038147	Cd84	ENSMUSG00000074151	Nlrc5
ENSMUSG00000005339	Fcer1a	ENSMUSG00000031779	Ccl22
ENSMUSG00000039997	Ifi203	ENSMUSG00000031778	Cx3cl1
ENSMUSG00000039748	Exo1	ENSMUSG00000031780	Ccl17
ENSMUSG00000079164	Tlr5	ENSMUSG00000031672	Got2
ENSMUSG00000039239	Tgfb2	ENSMUSG00000031673	Cdh11

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000026605	Cenpf	ENSMUSG00000031871	Cdh5
ENSMUSG00000026630	Batf3	ENSMUSG00000096188	Cmtm4
ENSMUSG00000026628	Atf3	ENSMUSG00000069922	Ces3a
ENSMUSG00000026639	Lamb3	ENSMUSG00000031897	Psmb10
ENSMUSG00000016194	Hsd11b1	ENSMUSG00000000303	Cdh1
ENSMUSG00000019768	Esr1	ENSMUSG00000040010	Slc7a5
ENSMUSG00000046916	Myct1	ENSMUSG00000032815	Fanca
ENSMUSG00000079685	Ulbp1	ENSMUSG00000025888	Casp1
ENSMUSG00000019850	Tnfaip3	ENSMUSG00000050578	Mmp13
ENSMUSG00000071369	Map3k5	ENSMUSG00000049723	Mmp12
ENSMUSG00000019970	Sgk1	ENSMUSG00000043089	Mmp1a
ENSMUSG00000019987	Arg1	ENSMUSG00000018623	Mmp7
ENSMUSG00000019843	Fyn	ENSMUSG00000032000	Birc3
ENSMUSG00000019916	P4ha1	ENSMUSG00000031925	Maml2
ENSMUSG00000020101	Vsir	ENSMUSG00000049307	Fut4
ENSMUSG00000037012	Hk1	ENSMUSG00000031928	Mre11a
ENSMUSG00000020178	Adora2a	ENSMUSG00000004099	Dnmt1
ENSMUSG00000000290	Itgb2	ENSMUSG00000037405	Icam1
ENSMUSG00000000732	Icosl	ENSMUSG00000032174	Icam5
ENSMUSG00000020312	Shc2	ENSMUSG00000036777	Anln
ENSMUSG00000020325	Fstl3	ENSMUSG00000032125	Robo4
ENSMUSG00000035673	Sbno2	ENSMUSG00000047880	Cxcr5
ENSMUSG00000020167	Tcf3	ENSMUSG00000032089	Il10ra
ENSMUSG00000035242	Oaz1	ENSMUSG00000039542	Ncam1
ENSMUSG00000004934	Pias4	ENSMUSG00000039217	Il18
ENSMUSG00000069515	Lyz1	ENSMUSG00000034218	Atm
ENSMUSG00000020184	Mdm2	ENSMUSG00000032298	Neil1
ENSMUSG00000025407	Gli1	ENSMUSG00000035914	Cd276
ENSMUSG00000040280	Ndufa4l2	ENSMUSG00000032375	Aph1b
ENSMUSG00000040033	Stat2	ENSMUSG00000032366	Tpm1
ENSMUSG00000025358	Cdk2	ENSMUSG00000032204	Aqp9
ENSMUSG00000025354	Dnajc14	ENSMUSG00000032344	Cgas
ENSMUSG00000002129	Sf3a1	ENSMUSG00000032420	Nt5e
ENSMUSG00000034394	Lif	ENSMUSG00000032356	Rasgrf1
ENSMUSG00000018654	Ikzf1	ENSMUSG00000032374	Plod2
ENSMUSG00000020122	Egfr	ENSMUSG00000045322	Tlr9
ENSMUSG00000057967	Fgf18	ENSMUSG00000032596	Uba7
ENSMUSG00000020399	Havcr2	ENSMUSG00000032498	Mlh1
ENSMUSG00000020380	Rad50	ENSMUSG00000046785	Epm2aip1
ENSMUSG00000018899	Irf1	ENSMUSG00000047898	Ccr4
ENSMUSG00000018906	P4ha2	ENSMUSG00000032434	Cmtm6
ENSMUSG00000018916	Csf2	ENSMUSG00000032440	Tgfbr2

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000009900	Wnt3a	ENSMUSG00000032508	Myd88
ENSMUSG00000032691	Nlrp3	ENSMUSG00000006932	Ctnnb1
ENSMUSG00000020538	Srebf1	ENSMUSG00000029530	Ccr9
ENSMUSG00000019505	Ubb	ENSMUSG00000049103	Ccr2
ENSMUSG00000020901	Pik3r5	ENSMUSG00000079227	Ccr5
ENSMUSG00000059552	Trp53	ENSMUSG00000015340	Cybb
ENSMUSG00000018774	Cd68	ENSMUSG00000037358	Dipk2b
ENSMUSG00000089669	Tnfsf13	ENSMUSG00000005696	Sh2d1a
ENSMUSG00000097328	Tnfsf12	ENSMUSG00000031119	Gpc4
ENSMUSG00000005198	Polr2a	ENSMUSG00000031132	Cd40lg
ENSMUSG00000000317	Bcl6b	ENSMUSG00000031137	Fgf13
ENSMUSG00000018920	Cxcl16	ENSMUSG00000033343	Magea4
ENSMUSG00000017390	Aldoc	ENSMUSG00000004221	Ikbkg
ENSMUSG00000020826	Nos2	ENSMUSG00000031400	G6pdx
ENSMUSG00000020716	Nf1	ENSMUSG00000035725	Prkx
ENSMUSG00000020676	Ccl11	ENSMUSG00000035427	Mageb4
ENSMUSG00000009185	Ccl8	ENSMUSG00000031304	Il2rg
ENSMUSG00000020702	Ccl1	ENSMUSG00000057439	Kir3dl2
ENSMUSG00000035042	Ccl5	ENSMUSG00000031424	Kir3dl1
ENSMUSG00000019122	Ccl9	ENSMUSG00000031274	Col4a5
ENSMUSG00000018927	Ccl6	ENSMUSG00000094196	Magea3
ENSMUSG00000000982	Ccl3	ENSMUSG00000096644	Magea1
ENSMUSG00000018930	Ccl4	ENSMUSG00000040522	Tlr8
ENSMUSG00000034329	Brip1	ENSMUSG00000026573	Xcl1
ENSMUSG00000020516	Rps6kb1	ENSMUSG00000020027	Socs2
ENSMUSG00000007646	Rad51c	ENSMUSG00000050222	Il17d
ENSMUSG00000000120	Ngfr	ENSMUSG00000022015	Tnfsf11
ENSMUSG00000071415	Rpl23	ENSMUSG00000000489	Pdgfb
ENSMUSG00000062312	Erbb2	ENSMUSG00000039481	Nrtn
ENSMUSG00000018168	Ikzf3	ENSMUSG00000002603	Tgfb1
ENSMUSG00000038067	Csf3	ENSMUSG00000045826	Ptprcap
ENSMUSG00000001552	Jup	ENSMUSG00000076431	Sox4
ENSMUSG00000004040	Stat3	ENSMUSG00000029705	Cux1
ENSMUSG00000010358	Ifi35	ENSMUSG00000008496	Pou2f2
ENSMUSG00000017146	Brca1	ENSMUSG00000041515	Irf8
ENSMUSG00000008855	Hdac5	ENSMUSG00000026189	Pecr
ENSMUSG00000034757	Tmub2	ENSMUSG00000056220	Pla2g4a
ENSMUSG00000020689	Itgb3	ENSMUSG00000026473	Glul
ENSMUSG00000020694	Tlk2	ENSMUSG00000026471	Mr1
ENSMUSG00000001029	Icam2	ENSMUSG00000019818	Cd164
ENSMUSG00000020717	Pecam1	ENSMUSG00000020432	Tcn2
ENSMUSG00000050965	Prkca	ENSMUSG00000020604	Arsg

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000034652	Cd300a	ENSMUSG00000050103	Agmo
ENSMUSG00000017716	Birc5	ENSMUSG00000035711	Dok3
ENSMUSG00000025583	Rptor	ENSMUSG00000021477	Ctsl
ENSMUSG00000025163	Cd7	ENSMUSG00000021710	Nln
ENSMUSG00000020649	Rrm2	ENSMUSG00000041707	Tmem273
ENSMUSG00000020641	Rsad2	ENSMUSG00000037824	Tspan14
ENSMUSG00000063632	Sox11	ENSMUSG00000064373	Selenop
ENSMUSG00000020573	Pik3cg	ENSMUSG00000081534	Slc48a1
ENSMUSG00000035799	Twist1	ENSMUSG00000022788	Fgd4
ENSMUSG00000073079	Srp54a	ENSMUSG00000000326	Comt
ENSMUSG00000021025	Nfkbia	ENSMUSG00000000127	Fer
ENSMUSG00000045930	Clec14a	ENSMUSG00000005803	Sqor
ENSMUSG00000021109	Hif1a	ENSMUSG00000014361	Mertk
ENSMUSG00000021125	Arg2	ENSMUSG00000027900	Dram2
ENSMUSG00000021253	Tgfb3	ENSMUSG00000039005	Tlr4
ENSMUSG00000021179	Nrde2	ENSMUSG00000028497	Hacd4
ENSMUSG00000057963	Itpk1	ENSMUSG00000028859	Csf3r
ENSMUSG00000064215	Ifi27	ENSMUSG00000019055	Plod1
ENSMUSG00000066366	Serpina1a	ENSMUSG00000029759	Pon3
ENSMUSG00000041359	Tcl1	ENSMUSG00000061758	Akr1b10
ENSMUSG00000001729	Akt1	ENSMUSG00000029925	Tbxas1
ENSMUSG00000002799	Jag2	ENSMUSG00000029998	Pcyox1
ENSMUSG00000025321	Itgb8	ENSMUSG00000030272	Camk1
ENSMUSG00000021303	Gng4	ENSMUSG00000040466	Blvrb
ENSMUSG00000021319	Sfrp4	ENSMUSG00000003363	Pld3
ENSMUSG00000016477	E2f3	ENSMUSG00000030761	Myo7a
ENSMUSG00000021408	Ripk1	ENSMUSG00000030894	Tpp1
ENSMUSG00000021367	Edn1	ENSMUSG00000007891	Ctsd
ENSMUSG00000021379	Id4	ENSMUSG00000042870	Tom1
ENSMUSG00000021457	Syk	ENSMUSG00000031903	Pla2g15
ENSMUSG00000021464	Ror2	ENSMUSG00000016534	Lamp2
ENSMUSG00000021474	Sfxn1	ENSMUSG00000044583	Tlr7
ENSMUSG00000021508	Cxcl14	ENSMUSG00000059498	Fcgr3
ENSMUSG00000021540	Smad5	ENSMUSG00000051439	Cd14
ENSMUSG00000052957	Gas1	ENSMUSG00000026131	Dst
ENSMUSG00000069805	Fbp1	ENSMUSG00000047793	Sned1
ENSMUSG00000021577	Sdha	ENSMUSG00000019944	Rhobtb1
ENSMUSG00000021614	Vcan	ENSMUSG00000034714	Ttyh2
ENSMUSG00000021678	F2rl1	ENSMUSG00000022272	Myo10
ENSMUSG00000021650	Ptcd2	ENSMUSG00000052942	Glis3
ENSMUSG00000041431	Ccnb1	ENSMUSG00000028184	Adgrl2
ENSMUSG00000041417	Pik3r1	ENSMUSG00000034853	Acot11

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000042417	Ccno	ENSMUSG00000042476	Abcb4
ENSMUSG00000042284	Itga1	ENSMUSG00000023079	Gtf2ird1
ENSMUSG00000074715	Ccl28	ENSMUSG00000030064	Frmd4b
ENSMUSG00000025278	Flnb	ENSMUSG00000030409	Dmpk
ENSMUSG00000021806	Nid2	ENSMUSG00000034656	Cacna1a
ENSMUSG00000021994	Wnt5a	ENSMUSG00000032014	Oaf
ENSMUSG00000021794	Glud1	ENSMUSG00000006014	Prg4
ENSMUSG00000041445	Mmrn2	ENSMUSG00000069919	Hba-a1
ENSMUSG00000021831	Ero1l	ENSMUSG00000069917	Hba-a2
ENSMUSG00000022193	Psmb5	ENSMUSG00000020773	Trim47
ENSMUSG00000040618	Pck2	ENSMUSG00000006356	Crip2
ENSMUSG00000002325	Irf9	ENSMUSG00000095079	Igha
ENSMUSG00000022221	Ripk3	ENSMUSG00000076612	Ighg2c
ENSMUSG00000022156	Gzme	ENSMUSG00000076613	Ighg2b
ENSMUSG00000054509	Parp4	ENSMUSG00000023031	Cela1
ENSMUSG00000021974	Fgf9	ENSMUSG00000083282	Ctsf
ENSMUSG00000014453	Blk	ENSMUSG00000060224	Pyroxd2
ENSMUSG00000045731	Pnoc	ENSMUSG00000032890	Rims3
ENSMUSG00000022051	Bnip3l	ENSMUSG00000067149	Jchain
ENSMUSG00000014813	Stc1	ENSMUSG00000076569	Igkv5-39
ENSMUSG00000034205	Loxl2	ENSMUSG00000030084	Plxna1
ENSMUSG00000022105	Rb1	ENSMUSG00000073940	Hbb-bt
ENSMUSG00000055737	Ghr	ENSMUSG00000052305	Hbb-bs
ENSMUSG00000079105	C7	ENSMUSG00000025511	Tspan4
ENSMUSG00000039942	Ptger4	ENSMUSG00000025151	Maged1
ENSMUSG00000022150	Dab2	ENSMUSG00000005087	Cd44
ENSMUSG00000050310	Rictor	ENSMUSG00000046841	Ckap4
ENSMUSG00000005268	Prlr	ENSMUSG00000044986	Tst
ENSMUSG00000022309	Angpt1	ENSMUSG00000024659	Anxa1
ENSMUSG00000063727	Tnfrsf11b	ENSMUSG00000074570	Cass4
ENSMUSG00000022419	Deptor	ENSMUSG00000074340	Ovgp1
ENSMUSG00000079018	Ly6c1	ENSMUSG00000106671	Gm42900
ENSMUSG00000033576	Apol6	ENSMUSG00000056091	St3gal5
ENSMUSG00000071713	Csf2rb	ENSMUSG00000030413	Pglyrp1
ENSMUSG00000068227	Il2rb	ENSMUSG00000009628	Tex15
ENSMUSG00000018169	Mfng	ENSMUSG00000009633	G0s2
ENSMUSG00000033006	Sox10	ENSMUSG00000000204	Slfn4
ENSMUSG00000058099	Nfam1	ENSMUSG00000025161	Slc16a3
ENSMUSG00000036106	Prr5	ENSMUSG00000021360	Gcnt2
ENSMUSG00000022382	Wnt7b	ENSMUSG00000022564	Grina
ENSMUSG00000022615	Tymp	ENSMUSG00000001227	Sema6b
ENSMUSG00000022451	Twf1	ENSMUSG00000058624	Gda

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000033065	Pfkm	ENSMUSG00000086564	Cd101
ENSMUSG00000023050	Map3k12	ENSMUSG00000051397	Tacstd2
ENSMUSG00000039457	Ppl	ENSMUSG00000033720	Sfxn5
ENSMUSG00000038037	Socs1	ENSMUSG00000030340	Scnn1a
ENSMUSG00000022496	Tnfrsf17	ENSMUSG00000004609	Cd33
ENSMUSG00000049502	Dtx3l	ENSMUSG00000041268	Dmxl2
ENSMUSG00000022906	Parp9	ENSMUSG00000073489	Ifi204
ENSMUSG00000022901	Cd86	ENSMUSG00000038188	Scarf1
ENSMUSG00000002847	Pla1a	ENSMUSG00000017417	Plxdc1
ENSMUSG00000075122	Cd80	ENSMUSG00000079017	Ifi27l2a
ENSMUSG00000071552	Tigit	ENSMUSG00000006360	Crip1
ENSMUSG00000022667	Cd200r1	ENSMUSG00000076614	Ighg1
ENSMUSG00000022657	Cd96	ENSMUSG00000022587	Ly6e
ENSMUSG00000055447	Cd47	ENSMUSG00000024675	Ms4a4c
ENSMUSG00000022876	Samsn1	ENSMUSG00000041488	Stx3
ENSMUSG00000022967	Ifnar1	ENSMUSG00000035273	Hpse
ENSMUSG00000000386	Mx1	ENSMUSG00000038387	Rras
ENSMUSG00000023830	Igf2r	ENSMUSG00000026193	Fn1
ENSMUSG00000014773	Dll1	ENSMUSG00000026365	Cfh
ENSMUSG00000014767	Tbp	ENSMUSG00000026536	Ifi211
ENSMUSG00000045551	Fpr1	ENSMUSG00000039109	F13a1
ENSMUSG00000079700	Fpr3	ENSMUSG00000022584	Ly6c2
ENSMUSG00000055839	Elob	ENSMUSG00000079419	Ms4a6c
ENSMUSG00000024173	Tpsab1	ENSMUSG00000024679	Ms4a6d
ENSMUSG00000024182	Axin1	ENSMUSG00000026796	Fam129b
ENSMUSG00000046711	Hmga1	ENSMUSG00000048058	Ldlrad3
ENSMUSG00000023067	Cdkn1a	ENSMUSG00000027435	Cd93
ENSMUSG00000024002	Brd4	ENSMUSG00000027994	Mcub
ENSMUSG00000002289	Angptl4	ENSMUSG00000034557	Zfyve9
ENSMUSG00000024308	Tapbp	ENSMUSG00000040964	Arhgef10l
ENSMUSG00000061232	H2-K1	ENSMUSG00000029322	Plac8
ENSMUSG00000024330	Col11a2	ENSMUSG00000038156	Spon1
ENSMUSG00000098241	H2-Pa	ENSMUSG00000031444	F10
ENSMUSG00000037649	H2-DMa	ENSMUSG00000014846	Tppp3
ENSMUSG00000096727	Psmb9	ENSMUSG00000047409	Ctdspl
ENSMUSG00000037321	Tap1	ENSMUSG00000030786	Itgam
ENSMUSG00000024338	Psmb8	ENSMUSG00000022074	Tnfrsf10b
ENSMUSG00000024339	Tap2	ENSMUSG00000022965	Ifngr2
ENSMUSG00000041538	H2-Ob	ENSMUSG00000002897	Il17ra
ENSMUSG00000034786	Gpsm3	ENSMUSG00000030341	Tnfrsf1a
ENSMUSG00000024371	C2	ENSMUSG00000030745	Il21r
ENSMUSG00000024399	Ltb	ENSMUSG00000004043	Stat5a

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000073411	H2-D1	ENSMUSG00000009739	Pou6f1
ENSMUSG00000079507	H2-Q1	ENSMUSG00000022521	Crebbp
ENSMUSG00000091705	H2-Q2	ENSMUSG00000030067	Foxp1
ENSMUSG00000067235	H2-Q10	ENSMUSG00000022582	Ly6g
ENSMUSG00000038762	Abcf1	ENSMUSG00000015533	Itga2
ENSMUSG00000067212	H2-T23	ENSMUSG00000067750	Khdc1a
ENSMUSG00000016206	H2-M3	ENSMUSG00000037202	Prf1
ENSMUSG00000023947	Nfkbie	ENSMUSG00000023132	Gzma
ENSMUSG00000023951	Vegfa	ENSMUSG00000030114	Klrg1
ENSMUSG00000034165	Ccnd3	ENSMUSG00000030361	Klrb1a
ENSMUSG00000042265	Trem1	ENSMUSG00000030325	Klrb1c
ENSMUSG00000023992	Trem2	ENSMUSG00000079298	Klrb1b
ENSMUSG00000000708	Kat2b	ENSMUSG00000050241	Klre1
ENSMUSG00000047123	Ticam1	ENSMUSG00000030149	Klrk1
ENSMUSG00000035678	Tnfsf9	ENSMUSG00000062524	Ncr1
ENSMUSG00000019489	Cd70	ENSMUSG00000045087	S1pr5
ENSMUSG00000004730	Adgre1	ENSMUSG00000032446	Eomes
ENSMUSG00000032796	Lama1	ENSMUSG00000026121	Sema4c
ENSMUSG00000024079	Eif2ak2	ENSMUSG00000104117	Gm20743
ENSMUSG00000045394	Epcam	ENSMUSG00000045827	Serpinb9
ENSMUSG00000024151	Msh2	ENSMUSG00000104876	Trdc
ENSMUSG00000005370	Msh6	ENSMUSG00000078202	Nrarp
ENSMUSG00000024232	Bambi	ENSMUSG00000043102	Qrfp
ENSMUSG00000024235	Map3k8	ENSMUSG00000028214	Gem
ENSMUSG00000024238	Zeb1	ENSMUSG00000032135	Mcam
ENSMUSG00000036904	Fzd8	ENSMUSG00000059901	Adamts14
ENSMUSG00000024290	Rock1	ENSMUSG00000022114	Spry2
ENSMUSG00000024304	Cdh2	ENSMUSG00000043932	Klri2
ENSMUSG00000059898	Dsc3	ENSMUSG00000079852	Klra4
ENSMUSG00000024382	Ercc3	ENSMUSG00000089727	Klra8
ENSMUSG00000024379	Tslp	ENSMUSG00000033024	Klra9
ENSMUSG00000005871	Apc	ENSMUSG00000067591	Klra3
ENSMUSG00000044201	Cdc25c	ENSMUSG00000032899	Styk1
ENSMUSG00000038418	Egr1	ENSMUSG00000020900	Myh10
ENSMUSG00000051486	Pcdhb11	ENSMUSG00000075033	Nxpe3
ENSMUSG00000024454	Hdac3	ENSMUSG00000023805	Synj2
ENSMUSG00000024427	Spry4	ENSMUSG00000026950	Neb
ENSMUSG00000024620	Pdgfrb	ENSMUSG00000027546	Atp9a
ENSMUSG00000024621	Csf1r	ENSMUSG00000085028	Slc2a4rg-ps
ENSMUSG00000033871	Ppargc1b	ENSMUSG00000051444	Bbs12
ENSMUSG00000024892	Pcx	ENSMUSG00000091575	2010016I18Rik
ENSMUSG00000006464	Bbs1	ENSMUSG00000030319	Cand2

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000024912	Fosl1	ENSMUSG00000055675	Kbtbd11
ENSMUSG00000024910	Ctsw	ENSMUSG00000032221	Mns1
ENSMUSG00000024927	Rela	ENSMUSG00000056418	BC043934
ENSMUSG00000024959	Bad	ENSMUSG00000070942	Il1rl2
ENSMUSG00000024962	Vegfb	ENSMUSG00000009350	Mpo
ENSMUSG00000024669	Cd5	ENSMUSG00000017493	Igfbp4
ENSMUSG00000101389	Ms4a4a	ENSMUSG00000004558	Ndrg2
ENSMUSG00000024677	Ms4a6b	ENSMUSG00000028644	Ermap
ENSMUSG00000024680	Ms4a2	ENSMUSG00000028943	Espn
ENSMUSG00000039982	Dtx4	ENSMUSG00000029359	Tesc
ENSMUSG00000016496	Cd274	ENSMUSG00000086894	Gm15708
ENSMUSG00000016498	Pdcd1lg2	ENSMUSG00000029869	Ephb6
ENSMUSG00000024806	Mlana	ENSMUSG00000090958	Lrrc32
ENSMUSG00000024810	Il33	ENSMUSG00000035239	Neu3
ENSMUSG00000024868	Dkk1	ENSMUSG00000045659	Plekha7
ENSMUSG00000013663	Pten	ENSMUSG00000012889	Podnl1
ENSMUSG00000071573	Rnls	ENSMUSG00000031933	Izumo1r
ENSMUSG00000024778	Fas	ENSMUSG00000039384	Dusp10
ENSMUSG00000045932	Ifit2	ENSMUSG00000021699	Pde4d
ENSMUSG00000074896	Ifit3	ENSMUSG00000033022	Cdo1
ENSMUSG00000034459	Ifit1	ENSMUSG00000047604	Frat2
ENSMUSG00000024989	Cep55	ENSMUSG00000100826	Snhg14
ENSMUSG00000025001	Hells	ENSMUSG00000050382	Kif7
ENSMUSG00000048120	Entpd1	ENSMUSG00000069920	B3gnt9
ENSMUSG00000025190	Got1	ENSMUSG00000035202	Lars2
ENSMUSG00000025199	Chuk	ENSMUSG00000002006	Pdzd4
ENSMUSG00000025225	Nfkb2	ENSMUSG00000041757	Plekha6
ENSMUSG00000025064	Col17a1	ENSMUSG00000025359	Pmel
ENSMUSG00000025025	Mxi1	ENSMUSG00000051166	Eml5
ENSMUSG00000034765	Dusp5	ENSMUSG00000057132	Rpgrip1
ENSMUSG00000025782	Taf3	ENSMUSG00000035235	Trim13
ENSMUSG00000026773	Pfkfb3	ENSMUSG00000052005	Gm9864
ENSMUSG00000026712	Mrc1	ENSMUSG00000060227	Casc4
ENSMUSG00000026981	Il1rn	ENSMUSG00000089417	Gm22009
ENSMUSG00000026923	Notch1	ENSMUSG00000033295	Ptprf
ENSMUSG00000062647	Rpl7a	ENSMUSG00000105742	Gm42748
ENSMUSG00000026918	Brd3	ENSMUSG00000064023	Klk8
ENSMUSG00000026837	Col5a1	ENSMUSG00000051504	Siglech
ENSMUSG00000026835	Fcnb	ENSMUSG00000025104	Hdgfl3
ENSMUSG00000026874	Hc	ENSMUSG00000001986	Gria3
ENSMUSG00000053475	Tnfaip6	ENSMUSG00000005763	Cd247
ENSMUSG00000026834	Acvr1c	ENSMUSG00000026770	Il2ra

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000000392	Fap	ENSMUSG00000002033	Cd3g
ENSMUSG00000026896	Ifih1	ENSMUSG00000032094	Cd3d
ENSMUSG00000027111	Itga6	ENSMUSG00000032093	Cd3e
ENSMUSG00000006494	Pdk1	ENSMUSG00000026358	Rgs1
ENSMUSG00000075271	Ttc30a1	ENSMUSG00000042684	Npl
ENSMUSG00000027009	Itga4	ENSMUSG00000020377	Ltc4s
ENSMUSG00000027087	Itgav	ENSMUSG00000001123	Lgals9
ENSMUSG00000044338	Aplnr	ENSMUSG00000035385	Ccl2
ENSMUSG00000002109	Ddb2	ENSMUSG00000035373	Ccl7
ENSMUSG00000027193	Api5	ENSMUSG00000035352	Ccl12
ENSMUSG00000040152	Thbs1	ENSMUSG00000021665	Hexb
ENSMUSG00000027323	Rad51	ENSMUSG00000022912	Pros1
ENSMUSG00000027314	Dll4	ENSMUSG00000024190	Dusp1
ENSMUSG00000027242	Wdr76	ENSMUSG00000003541	Ier3
ENSMUSG00000060802	B2m	ENSMUSG00000024672	Ms4a7
ENSMUSG00000027360	Hdc	ENSMUSG00000027293	Ehd4
ENSMUSG00000027368	Dusp2	ENSMUSG00000027199	Gatm
ENSMUSG00000027399	Il1a	ENSMUSG00000015852	Fcrls
ENSMUSG00000027398	Il1b	ENSMUSG00000015947	Fcgr1
ENSMUSG00000037902	Sirpa	ENSMUSG00000028195	Ccn1
ENSMUSG00000027322	Siglec1	ENSMUSG00000015243	Abca1
ENSMUSG00000027358	Bmp2	ENSMUSG00000028494	Plin2
ENSMUSG00000027276	Jag1	ENSMUSG00000029484	Anxa3
ENSMUSG00000074743	Thbd	ENSMUSG00000029304	Spp1
ENSMUSG00000007659	Bcl2l1	ENSMUSG00000040552	C3ar1
ENSMUSG00000003283	Hck	ENSMUSG00000002985	Apoe
ENSMUSG00000001403	Ube2c	ENSMUSG00000048779	P2ry6
ENSMUSG00000017737	Mmp9	ENSMUSG00000031451	Gas6
ENSMUSG00000017652	Cd40	ENSMUSG00000031659	Adcy7
ENSMUSG00000042821	Snai1	ENSMUSG00000043953	Ccrl2
ENSMUSG00000056501	Cebpb	ENSMUSG00000025804	Ccr1
ENSMUSG00000027544	Nfatc2	ENSMUSG00000025283	Sat1
ENSMUSG00000040289	Hey1	ENSMUSG00000055170	Ifng
ENSMUSG00000078780	Gm5150	ENSMUSG00000014599	Csf1
ENSMUSG00000001865	Cpa3	ENSMUSG00000031750	Il34
ENSMUSG00000039304	Tnfsf10	ENSMUSG00000026070	Il18r1
ENSMUSG00000037661	Gpr160	ENSMUSG00000026068	Il18rap
ENSMUSG00000027665	Pik3ca	ENSMUSG00000020009	Ifngr1
ENSMUSG00000074637	Sox2	ENSMUSG00000018341	Il12rb2
ENSMUSG00000027793	Ccna1	ENSMUSG00000030167	Klrc1
ENSMUSG00000036362	P2ry13	ENSMUSG00000048521	Cxcr6
ENSMUSG00000028011	Tdo2	ENSMUSG00000013089	Etv5

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000027995	Tlr2	ENSMUSG00000045991	Onecut2
ENSMUSG00000028076	Cd1d1	ENSMUSG00000059325	Hopx
ENSMUSG00000004885	Crabp2	ENSMUSG00000025929	Il17a
ENSMUSG00000027947	Il6ra	ENSMUSG00000041872	Il17f
ENSMUSG00000056054	S100a8	ENSMUSG00000004791	Pgf
ENSMUSG00000056071	S100a9	ENSMUSG00000041324	Inhba
ENSMUSG00000038642	Ctss	ENSMUSG00000027718	Il21
ENSMUSG00000027878	Notch2	ENSMUSG00000010142	Tnfrsf13b
ENSMUSG00000051076	Vtcn1	ENSMUSG00000022797	Tfrc
ENSMUSG00000027863	Cd2	ENSMUSG00000027164	Traf6
ENSMUSG00000027852	Nras	ENSMUSG00000027864	Ptgfrn
ENSMUSG00000032902	Slc16a1	ENSMUSG00000019256	Ahr
ENSMUSG00000027840	Wnt2b	ENSMUSG00000021356	Irf4
ENSMUSG00000027966	Col11a1	ENSMUSG00000072889	Nfxl1
ENSMUSG00000027962	Vcam1	ENSMUSG00000032238	Rora
ENSMUSG00000033377	Palmd	ENSMUSG00000000869	Il4
ENSMUSG00000028017	Egf	ENSMUSG00000031497	Tnfsf13b
ENSMUSG00000028163	Nfkb1	ENSMUSG00000003420	Fcgrt
ENSMUSG00000028268	Gbp3	ENSMUSG00000034266	Batf
ENSMUSG00000040264	Gbp2b	ENSMUSG00000056749	Nfil3
ENSMUSG00000028270	Gbp2	ENSMUSG00000024140	Epas1
ENSMUSG00000005034	Prkacb	ENSMUSG00000024401	Tnf
ENSMUSG00000028224	Nbn	ENSMUSG00000037104	Socs5
ENSMUSG00000041135	Ripk2	ENSMUSG00000034855	Cxcl10
ENSMUSG00000028284	Map3k7	ENSMUSG00000048480	Cxcr1
ENSMUSG00000028444	Cntfr	ENSMUSG00000026321	Tnfrsf11a
ENSMUSG00000073889	Il11ra1	ENSMUSG00000023206	Il15ra
ENSMUSG00000073888	Ccl27a	ENSMUSG00000026942	Traf2
ENSMUSG00000071005	Ccl19	ENSMUSG00000060477	Irak2
ENSMUSG00000094686	Ccl21a	ENSMUSG00000032012	Nectin1
ENSMUSG00000035683	Melk	ENSMUSG00000025997	Ikzf2
ENSMUSG00000007613	Tgfbr1	ENSMUSG00000022528	Hes1
ENSMUSG00000028362	Tnfsf8	ENSMUSG00000003545	Fosb
ENSMUSG00000095498	Ifna1	ENSMUSG00000072109	A530040E14Rik
ENSMUSG00000044303	Cdkn2a	ENSMUSG00000112023	Lilr4b
ENSMUSG00000073802	Cdkn2b	ENSMUSG00000112148	Lilrb4a
ENSMUSG00000028530	Jak1	ENSMUSG00000029580	Actb
ENSMUSG00000028518	Prkaa2	ENSMUSG00000061577	Adgrg5
ENSMUSG00000028716	Pdzk1ip1	ENSMUSG00000057666	Gapdh
ENSMUSG00000028678	Kif2c	ENSMUSG00000105646	Gm30211
ENSMUSG00000006398	Cdc20	ENSMUSG00000049932	H2ax
ENSMUSG00000033191	Tie1	ENSMUSG00000069265	H3c1

Table 1. Murine Pan-Cancer Gene Expression Panel for Immuno-Oncology and Tumor Microenvironment Study

Ensemble ID	Gene	Ensemble ID	Gene
ENSMUSG00000028645	Slc2a1	ENSMUSG00000062070	Pgk1
ENSMUSG00000042677	Zc3h12a	ENSMUSG00000034487	Poglut3
ENSMUSG00000000409	Lck	ENSMUSG00000071866	Ppia
ENSMUSG00000028580	Pum1	ENSMUSG00000109713	Pvrig
ENSMUSG00000007880	Arid1a	ENSMUSG00000050379	Septin6
ENSMUSG00000037157	Il22ra1	ENSMUSG00000024349	Sting1
ENSMUSG00000036905	C1qb	ENSMUSG00000022285	Ywhaz