US 20100124755A1

(19) **United States**

(12) **Patent Application Publication**

(10) **Pub. No.: US 2010/0124755 A1**

(43) **Pub. Date: May 20, 2010**

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(54) **PROSTATE CANCER AND MELANOMA ANTIGENS**

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(21) Appl. No.: **12/629,686**

(22) Filed: **Dec. 2, 2009**

Related U.S. Application Data

(62) Division of application No. 12/033,229, filed on Feb. 19, 2008, now Pat. No. 7,635,753.

(60) Provisional application No. 60/890,590, filed on Feb. 19, 2007.

Publication Classification

(51) **Int. Cl.**
G01N 33/53 (2006.01)

(52) **U.S. Cl.** **435/7.1**

(57) **ABSTRACT**

Methods for identifying a human subject as a candidate for further prostate cancer or melanoma examination are disclosed. Also disclosed are methods for determining whether an immune therapy has elicited a tumor-specific immune response in a prostate cancer or melanoma patient. Further disclosed are kits that can be used to practice the above methods. Methods for identifying candidate compounds for further testing as preventive or therapeutic agents for melanoma are also disclosed.

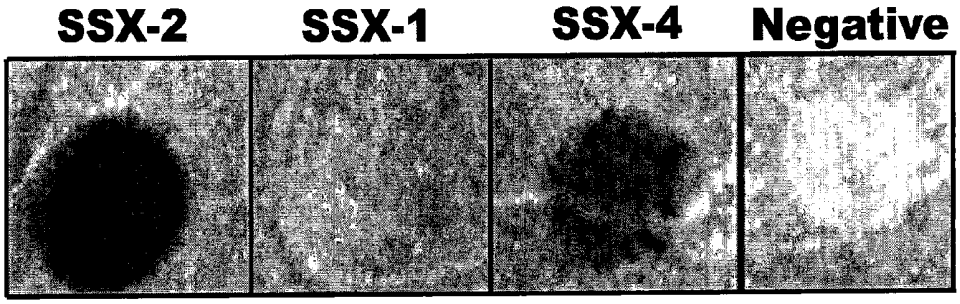


FIG. 1

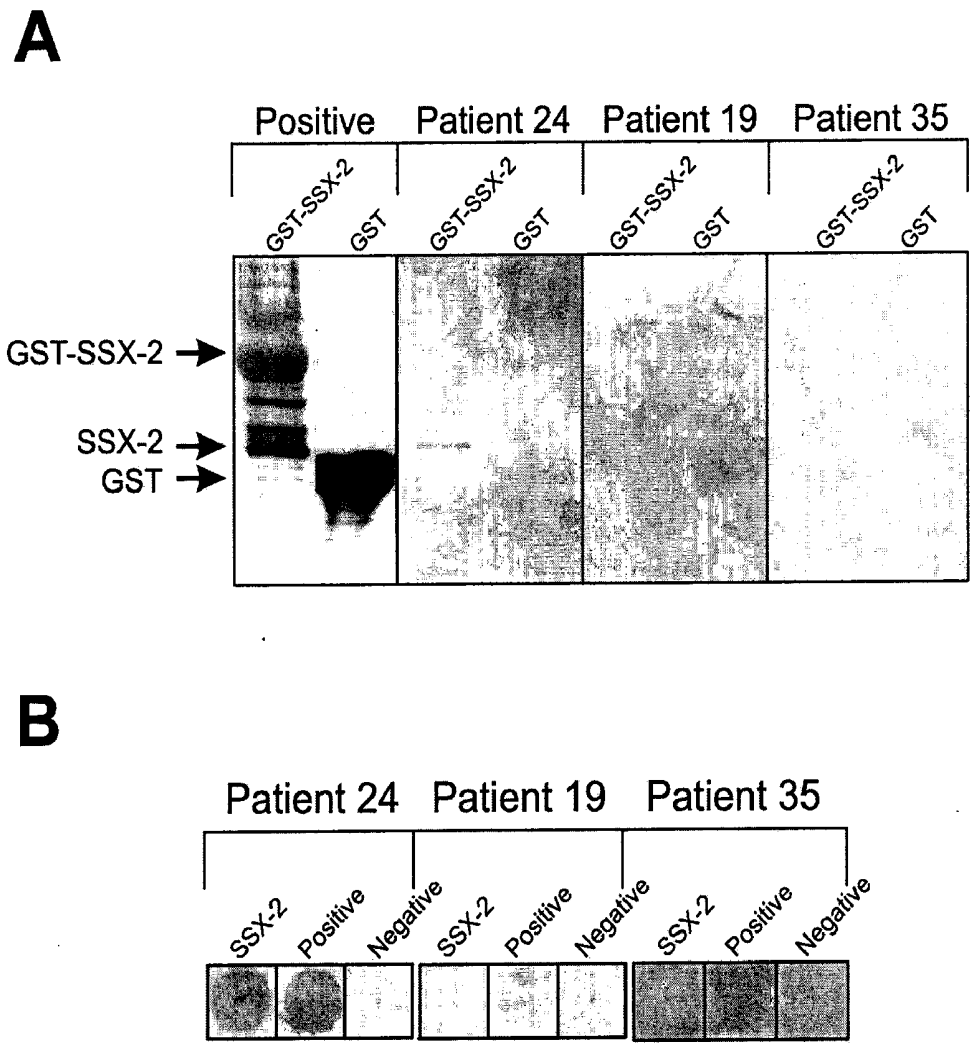

A
Positive
Patient 24
Patient 19
Patient 35
GST-SSX-2
GST
GST-SSX-2
GST
GST-SSX-2
GST
GST-SSX-2
GST
GST-SSX-2
SSX-2
GST
B
Patient 24
Patient 19
Patient 35
SSX-2
Positive
Negative
SSX-2
Positive
Negative
SSX-2
Positive
Negative

FIG. 2

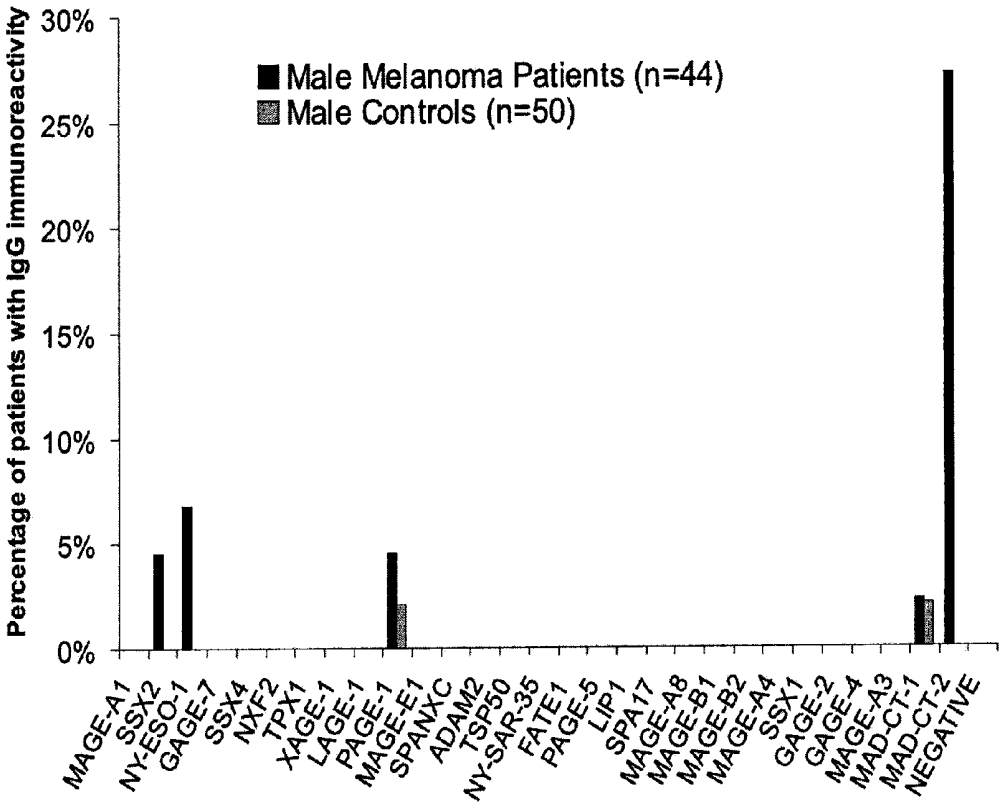

Percentage of patients with IgG immunoreactivity
30%
25%
20%
15%
10%
5%
0%
Male Melanoma Patients (n=44)
Male Controls (n=50)
MAGE-A1
SSX2
NY-ESO-1
GAGE-7
SSX4
NXF2
TPX1
XAGE-1
LAGE-1
PAGE-1
MAGE-E1
SPANXC
ADAM2
TSP50
NY-SAR-35
FATE1
PAGE-5
LIP1
SPA17
MAGE-A8
MAGE-B1
MAGE-B2
MAGE-A4
SSX1
GAGE-2
GAGE-4
MAGE-A3
MAD-CT-1
MAD-CT-2
NEGATIVE

FIG. 3

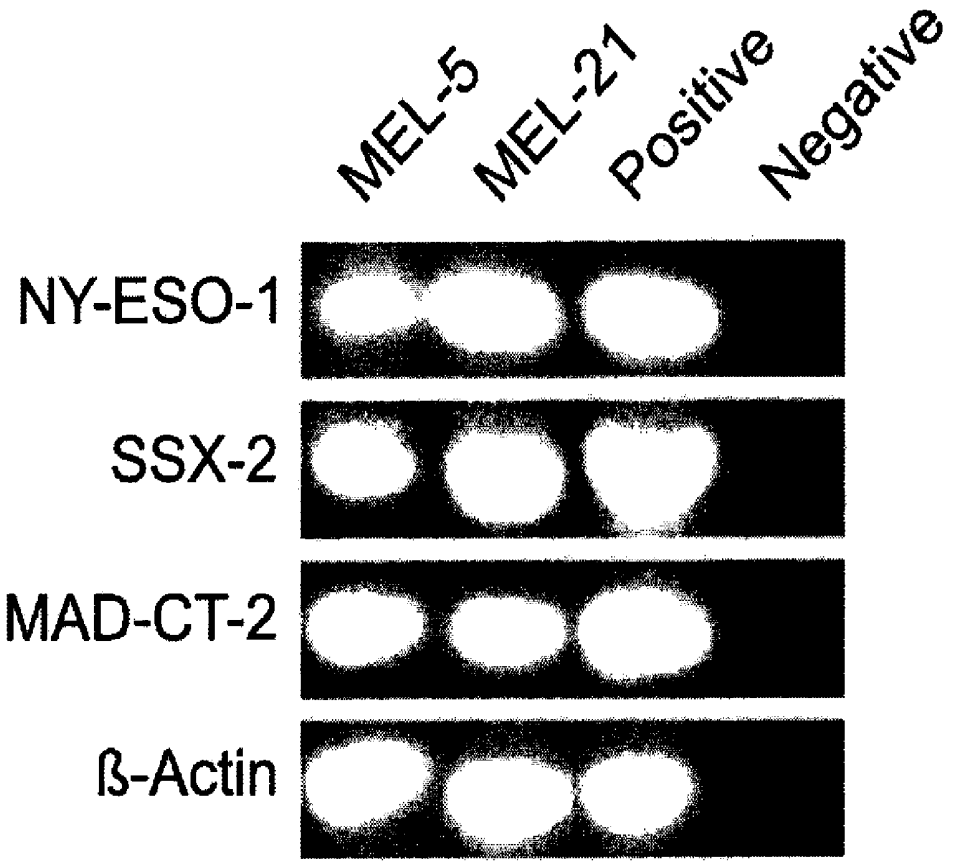


FIG. 4


Relative Densitometric Intensity
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
0.0
-0.5
MAGE-A1
Norm MAGE-A1
SSX2
Norm SSX2
NY-ESO-1
Norm NY-ESO-1
GAGE-7
Norm GAGE-7
SSX4
Norm SSX4
NXF2
Norm NXF2
TPX1
Norm TPX1
XAGE-1
Norm XAGE-1
LAGE-1
Norm LAGE-1
PAGE-1
Norm PAGE-1
MAGE-E1
Norm MAGE-E1
SPANXC
Norm SPANXC
ADAM2
Norm ADAM2
TSP50
Norm TSP50
NY-SAR-35
Norm NY-SAR-35
FATE1
Norm FATE1
PAGE-5
Norm PAGE-5
LIPI
Norm LIPI
SPA17
Norm SPA17
MAGE-A8
Norm MAGE-A8
MAGE-B1
Norm MAGE-B1
MAGE-B2
Norm MAGE-B2
MAGE-A4
Norm MAGE-A4
SSX1
Norm SSX1
GAGE-2
Norm GAGE-2
GAGE-4
Norm GAGE-4
MAGE-A3
Norm MAGE-A3
MADCT-1
Norm MADCT-1
MADCT-2
Norm MADCT-2
Negative
Norm Negative

FIG. 5

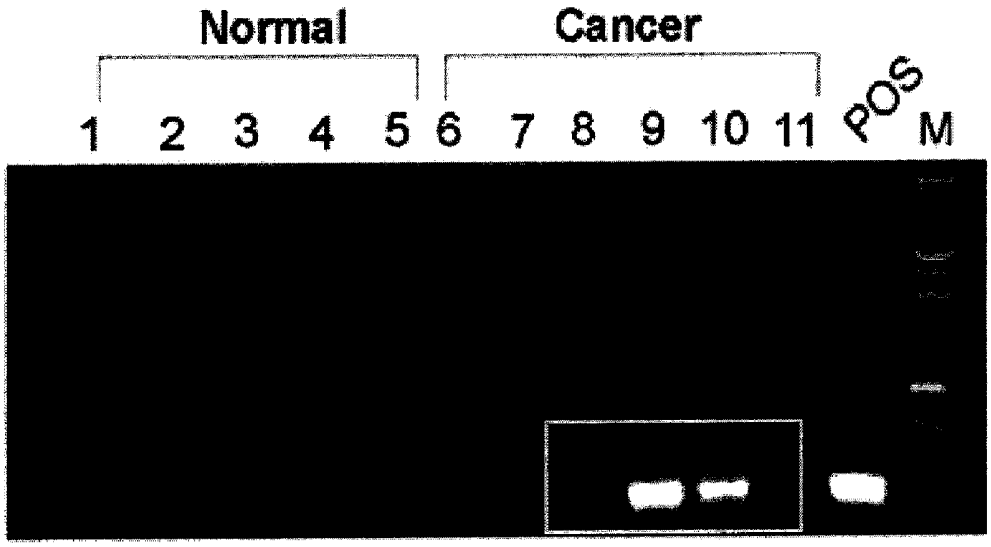


FIG. 6

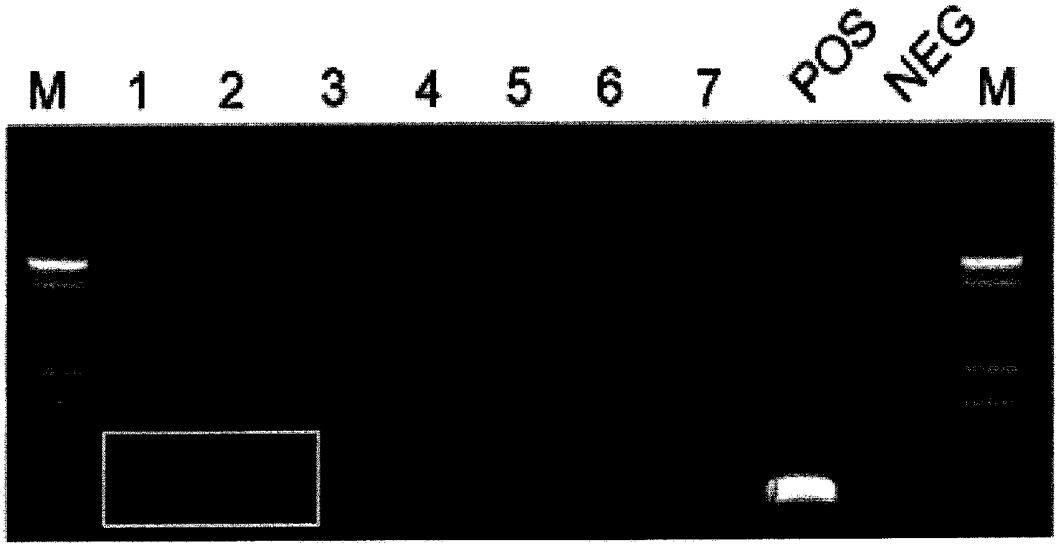


FIG. 7

PROSTATE CANCER AND MELANOMA ANTIGENS

CROSS-REFERENCE TO RELATED APPLICATION

[0001] This application is a divisional of U.S. Utility patent application Ser. No. 12/033,229, filed Feb. 19, 2008, and which claims the benefit of U.S. Provisional Patent Application Ser. No. 60/890,590, filed Feb. 19, 2007. These documents are incorporated herein by reference as if set forth in its entirety.

STATEMENT REGARDING FEDERALLY SPONSORED RESEARCH OR DEVELOPMENT

[0002] This invention was made with United States government support awarded by the following agency: NIH K23 RR16489. The United States has certain rights in this invention.

BACKGROUND

[0003] Immune responses can be elicited to tumor-expressed antigens. For example, several groups have reported the detection of antibody responses to prostate tumor-associated antigens compared with control groups (Wang X, et al., N. Engl. J. Med. 353:1224-1235 (2005); McNeel D, et al., J. Urol. 164:1825-1829 (2000); Minelli A, et al., Anticancer Res. 25:4399-4402 (2005); Bradford T, et al., Urol Oncol. 24:237-242 (2006); and Shi F, et al., Prostate 63:252-258 (2005)). Cancer-testis antigens (CTA) are of particular interest as potential tumor antigens given that their expression is typically restricted to germ cells among normal tissues, but aberrantly expressed in tumor cells (Scanlan M, et al., Cancer Immun. 4:1 (2004)). The absence of MHC class I molecule expression on germ cells makes CTA essentially tumor-specific antigens in terms of potential $CD8^+$ T-cell target antigens (Kowalik I, et al., Andrologia. 21:237-243 (1989)). Many of these CTA were discovered using antibody screening methods including the serological evaluation of recombinant cDNA expression libraries (SEREX) approach (Sahin U, et al., Proc. Natl. Acad. Sci. USA 92:11810-11813 (1005); Hoeppner L, et al., Cancer Immun. 6:1-7 (2006); and Tureci O, et al., Mol. Med. Today 3:342-349 (1997)).

[0004] Several CTA, including members of the MAGE and GAGE families, have been identified as antigens recognized by tumor-specific cytotoxic T-cells (CTL) (Van der Bruggen P, et al., Science 254:1643-1647 (1991); and Van den Eynde B, et al., J. Exp. Med. 182:689-698 (1995)). Detectable immune responses to these antigens are believed to be a result of their ectopic expression in MHC class I-expressing malignant cells. Several CTA have been shown to be recognized by both antibodies and CTL, thus providing validation for the original approach of using antibody screening to identify potential tumor-specific T cell antigens (Jäger E, et al., J. Exp. Med. 187:265-270 (1998); and Monji M, et al., Clin. Cancer Res. 10:6047-6057 (2004)). Some CTA are expressed in several malignancies of different tissue origins (Scanlan et al., supra; and Mashino K, et al., Br. J. Cancer. 85:713-720 (2001)).

BRIEF SUMMARY

[0005] The present invention provides methods for identifying a human subject as a candidate for further prostate cancer or melanoma examination based on certain prostate cancer and melanoma antigens identified by the inventors. Methods of identifying immune responses elicited by an immune therapy directed at prostate cancer or melanoma are also provided. The present invention also provides kits that can be used to practice the above methods. Methods for identifying candidate compounds for further testing as preventive or therapeutic agents for melanoma are also disclosed.

BRIEF DESCRIPTION OF THE SEVERAL VIEWS OF THE DRAWINGS

[0006] FIG. **1** shows an immunoblot (probed with anti-SSX-2/4 specific antibody) that confirmed protein expression. Phage encoding SSX-2, SSX-1, SSX-4 and a negative control (empty phage construct) were spotted directly onto a bacterial lawn. Proteins were transferred to an isopropyl-beta-D-thiogalactopyranoside (IPTG)-infused nitrocellulose membrane, and mouse anti-SSX-2 mAb (clone E3AS, with known reactivity to SSX-2 and SSX-4, but not SSX-1) was used to probe the membrane.

[0007] FIG. **2** shows that high throughput immunoblot (HTI) detected CTA-specific IgG in patient sera. A) Purified glutathione-S-transferase (GST)-SSX-2 and GST were evaluated by Western blot with patient sera (patients 24, 19 or 35) or with monoclonal antibodies (positive) specific for SSX-2 or GST. B) HTI conducted with CTA phage array using sera from patients 24, 19 or 35. Positive refers to phage encoding IgG, and negative refers to empty phage control.

[0008] FIG. **3** shows that patients with melanoma have IgG specific for several CTA. Shown is the percentage of male patients with melanoma (n=44), or male control blood donors without cancer (n=50), with IgG specific for each of the 29 CTA tested.

[0009] FIG. **4** shows that MAD-CT-2, NY-ESO-1 and SSX-2 are expressed in melanoma cell lines. RT-PCR (reverse transcriptase-polymerase chain reaction) with primers specific for NY-ESO-1, SSX-2, MAD-CT-2 or β-actin was conducted using mRNA from cell lines (e.g., MEL-5 and MEL-21), cDNA encoding each of the proteins (positive control), or no DNA template (negative control).

[0010] FIG. **5** shows that HTI identified IgG responses to 4 (i.e., SSX-2, NY-ESO-1, LAGE-1 and NFX2) of 29 known CTA using sera from prostate cancer patients.

[0011] FIG. **6** shows the expression of SSX-2 in 5 normal prostate epithelial cell lines (lanes 1-5: 1. PrEC1; 2. PrEC2; 3. PrEC3; 4. PrEC4; and 5. PZ-HPV7) and 6 prostate cancer cell lines (lanes 6-11: 6. SWPC1; 7. SWPC2; 8. SWNPC2; 9. LAPC4; 10. MDAPCa2b; and 11. MDAPCa2a) by RT-PCR. POS represents a cDNA positive control; whereas M represents molecular weight markers.

[0012] FIG. **7** shows the expression of SSX-2 in prostate cancer metastatic tissues. Prostate cancer metastatic tissue cDNA samples of 7 different prostate cancer patients (lanes 1-7) were analyzed for SSX-2 expression using PCR. Patients 1 and 2 showed relatively strong expression and patients 6 and 7 showed weak expression. POS and NEG are positive and negative controls, respectively. M represents molecular weight markers.

DETAILED DESCRIPTION OF PREFERRED EMBODIMENTS

[0013] The present invention is based on the inventors' identification of a number of antigens to which patients with

prostate cancer or melanoma have developed antibody immune responses. The prostate cancer antigens identified include SSX-2, MAD-Pro-30, MAD-Pro-42, transgelin, ZCWCC3, ACAA1, actinin and NFX2; whereas the melanoma antigens identified include MAD-CT-2, MAD-CT-1 and PAGE-1. The inventors have also found that prostate cancer cells as well as metastatic tissues express several of the above newly identified prostate cancer antigens (e.g., SSX-2). In addition, melanoma cells express several of the newly identified melanoma antigens (e.g., MAD-CT-2). The identification provides new tools for assisting the diagnosis and the detection of recurrence of prostate cancer and melanoma, especially for men. The identification also provides new tools for determining whether an immune therapy elicited a tumor-specific immune response.

[0014] The cancer antigens disclosed herein are known proteins, and their DNA and amino acid sequences are available in the art (see, e.g., Table 1). While it is envisioned that a prostate cancer or melanoma antigen identified by the inventors can be used by itself for the screening of prostate cancer and melanoma, respectively, a panel of more than one antigen is preferred. Other prostate cancer and melanoma antigens known in the art can be included in the panel. For example, a prostate cancer antigen panel may include SSX-2, NY-ESO-1, NFX2, MAD-Pro-22, MAD-Pro-30, MAD-Pro-34, MAD-Pro-42, MAD-CaP-1, MAD-CaP-5, MAD-CaP-15, MAD-CaP-20, MAD-CT-1, MAD-CT-2, MAD-CT-3, MAD-CT-5, transgelin, ZCWCC3, ACAA1, androgen receptor (AR; or ligand-binding domain of AR) and actinin. A smaller prostate cancer antigen panel may include SSX-2, NY-ESO-1, NFX2, MAD-Pro-22, MAD-Pro-30, MAD-Pro-34, MAD-Pro-42, MAD-CaP-1, MAD-CaP-5, MAD-CaP-15, MAD-CaP-20, MAD-CT-1, MAD-CT-2, MAD-CT-3, MAD-CT-5, transgelin, ZCWCC3, ACAA1 and AR (or ligand-binding domain of AR). Another smaller prostate cancer antigen panel may include SSX-2, NY-ESO-1, NFX2, MAD-Pro-22, MAD-Pro-30, MAD-Pro-34, MAD-Pro-42, MAD-CaP-1, MAD-CaP-5, MAD-CaP-15, MAD-CaP-20, MAD-CT-1, MAD-CT-2, MAD-CT-3, MAD-CT-5, transgelin, ACAA1 and AR (or ligand-binding domain of AR). Another smaller prostate cancer antigen panel may include MAD-Pro-34, MAD-Pro-42 and MAD-CT-2. Another smaller prostate cancer antigen panel may include SSX-2, MAD-Pro-30, AR (or ligand-binding domain of AR) and MAD-Pro-22 (PSA). Another smaller prostate cancer antigen panel may include MAD-Pro-30, AR (or ligand-binding domain of AR) and PSA.

[0015] An example of a melanoma antigen panel includes MAD-CT-2, SSX-2, NY-ESO-1, MAD-CT-1 and PAGE-1. A smaller melanoma antigen panel may include MAD-CT-2, SSX-2, NY-ESO-1 and PAGE-1. Another smaller melanoma antigen panel may include MAD-CT-2, SSX-2 and NY-ESO-1.

TABLE 1

Prostate and Melanoma cancer antigens.

Designation Herein	Common Name	GenBank ID
MAD-Pro-22	Prostate specific antigen (PSA)	NM_145864
MAD-Pro-30	Recombination signal binding protein (RBPJK)	NM_015874
MAD-Pro-34	Nucleolar autoantigen; SC65; No55	NM 006455.1
MAD-Pro-42	NY-CO-7/STUB1	NM_005861
MAD-CaP-1	HMG17	NM_005517

TABLE 1-continued

Prostate and Melanoma cancer antigens.

Designation Herein	Common Name	GenBank ID
MAD-CaP-5	KIAA1404 gene product/ZNFX1	NM_021035
MAD-CaP-15	CLL-associated antigen KW-12/ RPL11	NM_000975
MAD-CaP-20	Human bullous pemphigoid antigen	NM_015548
MAD-CT-1	Protamine 2	NM_002762
MAD-CT-2	Hypothetical protein of unknown function FLJ40095/FLJ36144	AK097414
MAD-CT-3	Sorting nexin 13 (SNX13)	NM_015132
MAD-CT-5	Sjogren's syndrome antigen B (autoantigen La)	NM_003142
SSX-2	Synovial sarcoma, X breakpoint 2	Z49105
Transgelin	Transgelin	BC024296
ZCWCC3	Zinc-finger protein CW type with coiled domain 3	NM_015358
ACAA1	Acetyl-coenzyme A acyltransferase 1	NM_001607
AR or AR-LBD	Androgen receptor (AR), or ligand-binding domain of AR	M20132
Actinin	Actinin alpha 1, 2, 4	D89980
NY-ESO-1	NY-ESO-1/CTAG1B	NM_001327
PAGE-1	PAGE1	AF058989
NFX2	NFX2	AF332009
LAGE-1	LAGE-1	AJ223093

[0016] In one aspect, the present invention relates to a method for identifying a human subject as a candidate for further prostate cancer examination. The method includes the step of determining whether the human subject has developed an immune reaction to a prostate cancer antigen selected from SSX-2, MAD-Pro-30, MAD-Pro-42, transgelin, ZCWCC3, ACAA1, actinin and NFX2, wherein the presence of an immune reaction indicates that the human subject is a candidate for further prostate cancer examination. In one embodiment, a prostate cancer antigen panel as disclosed herein is used in the method and whether the human subject has developed an immune reaction to the antigens in the panel is determined. The presence of an immune reaction to any member of the panel indicates that the human subject is a candidate for further prostate cancer examination. For a subject who has already been indicated to have prostate cancer by other tests, the method here can be used to confirm the diagnosis or identify recurrence after treatment.

[0017] The method disclosed above may optionally include the step of subjecting a human subject to further prostate cancer examination if the human subject tests positive for at least one of the prostate cancer antigens. Any known test for assisting the diagnosis of prostate cancer can be used. Examples include serum prostate-specific antigen (PSA) blood test and standard pathological evaluation of prostate tissue specimen obtained from a biopsy. For subjects with a history of treated prostate cancer, radiographic scans can be conducted for detecting recurrent prostate cancer. In one embodiment, prostate or a prostate tissue specimen from the subject is examined for the presence of prostate cancer.

[0018] In a second aspect, the present invention relates to a method for determining whether an immune therapy elicited a tumor-specific immune response in a prostate cancer patient. In one embodiment, the immune therapy is an antigen-specific immune therapy. The new prostate cancer antigens identified herein can serve as targets for antigen-specific immune therapies and whether such an antigen-specific immune therapy has elicited a tumor-specific immune

response can be determined by testing whether a patient has developed an immune reaction to the antigen. An antigen-specific immune therapy may ultimately elicit responses to other antigens (i.e., antigens other than the one the therapy is designed to specifically target). For example, a successful antigen-specific immune therapy causes immune-mediated tumor destruction, leading to recognition of other antigens. Therefore, response to another antigen (other than the one the therapy is designed to specifically target) in an antigen-specific immune therapy indicates indirectly that the therapy elicited an immune response to the targeted antigen and is therefore effective. To practice the method, the prostate cancer antigens disclosed herein can be used individually, in combination with each other, or in combination with other known prostate cancer antigens to determine whether an antigen-specific immune therapy has elicited a tumor-specific immune response. In this regard, whether the patient has developed an immune reaction to a prostate cancer antigen selected from SSX-2, MAD-Pro-30, MAD-Pro-42, transgelin, ZCWCC3, ACAA1, actinin and NFX2 can be analyzed, wherein the presence of an immune reaction indicates that the therapy elicited a tumor-specific immune response. Any panel of prostate cancer antigens disclosed herein can be used for this purpose, wherein the presence of an immune reaction to at least one member of the panel indicates that the therapy elicited an immune response. Preferably, the patient is also tested for immune responses to the antigen or antigens before therapy so that it can be confirmed that an immune response detected after the start of the therapy is elicited by the therapy. The method may optionally include the step of monitoring the status of the prostate cancer in the patient by, e.g., the prostate cancer examination techniques described above.

[0019] In another embodiment of the second aspect, the immune therapy is a non-antigen-specific immune therapy. As used herein, "non-antigen-specific immune therapy" means an immune therapy that does not specifically target a particular antigen or an immune therapy that does not identify the antigen targets. Examples of non-antigen-specific therapies include whole-cell-based therapies such as the GVAX vaccines (granulocyte macrophage colony-stimulating factor-secreting cancer cell immunotherapy) or whole-cell vaccines developed by Onyvax Ltd. (London, England) (Hege K, et al., Int. Rev. Immunol. 25:321-352 (2006); Nemunaitis J, et al., Cancer Gene Ther. 13:555-562 (2006); and Michael A, et al., Clin. Cancer Res. 11:4469-4478 (2005)), cytokine-based therapies such as interleukin-2 or interferon-gamma (King D, et al. J. Clin. Oncol. 22:4463-4473 (2004)) or other immunomodulatory therapies including anti-CTLA-4 therapies used either alone or in combination with another therapy (Thompson R, et al., Urol. Oncol. 24:442-427 (2006); Korman A, et al., Adv. Immunol. 90:297-339 (2006); and Maker A, et al., Ann. Surg. Oncol. 12:1005-1016 (2005)). Traditionally, it has been difficult to assess whether a non-antigen-specific immune therapy generated a tumor-specific immune response because the therapy does not specifically target a particular antigen or the identities of the antigen targets are unknown. The prostate cancer antigens disclosed herein can be used individually, in combination with each other, or in combination with other known prostate cancer antigens to determine whether a non-antigen-specific immune therapy has elicited a tumor-specific immune response. In this regard, whether the patient has developed an immune reaction to a prostate cancer antigen selected from SSX-2, MAD-Pro-30, MAD-Pro-42, transgelin, ZCWCC3, ACAA1, actinin and NFX2 can be analyzed, wherein the presence of an immune reaction indicates that the therapy elicited a tumor-specific immune response. Any panel of prostate cancer antigens disclosed here can be used for this purpose, wherein the presence of an immune reaction to at least one member of the panel indicates that the therapy elicited an immune response. Preferably, the patient is also tested for immune responses to the antigen or antigens before therapy to confirm that an immune response detected after the start of the therapy is elicited by the therapy. The method may optionally include the step of monitoring the status of the prostate cancer in the patient by, e.g., the prostate cancer examination techniques described above.

[0020] In one embodiment, the method is used in clinical trials of antigen-specific or non-antigen-specific immune therapies for determining whether the therapies elicited a tumor-specific immune response.

[0021] One of ordinary skill in the art is more than capable of determining whether a human subject developed an immune reaction to one of the prostate cancer antigens. One way is to determine whether the human subject has produced antibodies to the antigens. For example, one can take a blood sample or blood-derived sample (e.g., a serum sample, a plasma sample or any preparation thereof that preserves the activity of immunoglobulins) from the human subject and test whether it contains antibodies to the antigens. Examples of such tests include enzyme-linked immunosorbent assay (ELISA), Western blot, protein microarray or high-throughput immunoblot analysis (Sreekumar A, et al., J. Natl. Cancer Inst. 96:834-843 (2004)), phage array-type analysis (Wang et al., supra; and Dubovsky J, et al., J. Immunother. 30:675-683 (2007)), and other methods known in the art. Another way is to determine whether the human subject has developed antigen-specific T cells, which is also well-known to one of ordinary skill in the art. For example, antigen stimulation can be used to detect antigen-specific T cells for their ability to proliferate, secrete various cytokines or exhibit cytolytic function (see e.g., Olson B & McNeel D, Prostate 67:1729-1739 (2007)). Other examples include enzyme-linked immunsorbent spot (ELISPOT) assays, fluorescence cell sorting of cytokine-producing cells, and peptide MHC/HLA tetramer assays (see e.g., Hobeika A, et al., J. Immunother. 28:63-72 (2005)). Preferably, circulating antigen-specific T cells are detected as they are the most accessible ones. For example, peripheral blood mononuclear cells (PBMC) can be analyzed for this purpose.

[0022] In a third aspect, the present invention relates to a method for identifying a human subject as a candidate for further melanoma examination. The method includes the step of determining whether the human subject has developed an immune reaction to a melanoma antigen selected from MAD-CT-2, MAD-CT-1 and PAGE-1, wherein the presence of an immune reaction indicates that the human subject is a candidate for further melanoma examination. MAD-CT-2 is a preferred melanoma antigen. In one embodiment, a melanoma antigen panel disclosed herein is used in the method and whether the human subject has developed an immune reaction to the antigens in the panel is determined. The presence of an immune reaction to any member of the panel indicates that the human subject is a candidate for further melanoma examination. For a subject already known to have melanoma by other tests, the method can be used to confirm the diagnosis or suggest recurrent disease.

[0023] The method disclosed above may optionally include the step of subjecting a human subject to further melanoma examination if the human subject is tested positive for at least one of the melanoma antigens. Any known test for assisting the diagnosing of melanoma can be used. For example, a skin specimen from the subject can be examined for the presence of melanoma (e.g., by pathological analysis). Radiographic imaging studies may be employed to evaluate for the presence of metastatic lesions.

[0024] In a fourth aspect, the present invention relates to a method for determining whether an immune therapy elicited a tumor-specific immune response in a melanoma patient. The method is the same as that described above for prostate cancer except that it is practiced with melanoma patients and the melanoma antigens identified by the inventors. In one embodiment, the immune therapy is an antigen-specific immune therapy. In another embodiment, the immune therapy is a non-antigen-specific immune therapy. The melanoma antigens disclosed here can be used individually, in combination with each other, or in combination with other known melanoma antigens to determine whether an immune therapy has elicited a tumor-specific immune response. In this regard, whether the patient has developed an immune reaction to a melanoma antigen selected from MAD-CT-2, MAD-CT-1 and PAGE-1 can be analyzed, wherein the presence of an immune reaction indicates that the therapy has elicited a tumor-specific immune response. Any panel of melanoma antigens disclosed herein can be used for this purpose, wherein the presence of an immune reaction to at least one member of the panel indicates that the therapy has elicited an immune response. Preferably, the patient is also tested for immune responses to the antigen or antigens before therapy so that it can be confirmed that an immune response detected after the start of the therapy is elicited by the therapy. The method may optionally include the step of monitoring the status of melanoma in the patient by, e.g., the melanoma examination techniques described above. In one application, the method is used in clinical trials of antigen-specific or non-antigen-specific immune therapies for determining whether the therapies have elicited a tumor-specific immune response.

[0025] Similar to what has been discussed above with respect to the prostate cancer antigens, it is well within the capability of one of ordinary skill in the art to determine whether a human subject has developed an immune reaction to one of the melanoma antigens, and examples of applicable techniques have been described above in connection with the prostate cancer antigens.

[0026] In a fifth aspect, the present invention relates to yet another method for identifying a human subject as a candidate for further melanoma examination. The method includes the step of determining whether the cells in a region of the subject's skin suspected of being malignant express MAD-CT-2, wherein the expression of MAD-CT-2 indicates that the subject is a candidate for further melanoma examination. The expression of MAD-CT-2 can be determined at either the mRNA level or the protein level and one of ordinary skill in the art is familiar with the techniques for such determination. For example, antibodies directed to an epitope on an antigen can be used to detect the antigen at the protein level, and it is well within the capability of one of ordinary skill in the art to generate such antibodies if not already available. The presence of mRNA for an antigen can be measured using methods for hybridizing nucleic acids, including, without limitation, RNA, DNA and cDNA. Such methods are generally known to those skilled in the art (e.g., RT-PCR amplification, Northern blot and Southern blot).

[0027] Optionally, the method further includes the step of subjecting a human subject who is positive for the expression of MAD-CT-2 to further melanoma examination. For a subject already identified as having melanoma by other tests, the method provided herein can be used to confirm the diagnosis.

[0028] In sixth aspect, the present invention relates a method for identifying candidate compounds for further testing as preventive or therapeutic agents for melanoma. As MAD-CT-2 is expressed in melanoma cells, it can serve as a marker for melanoma drug screening because, presumably, an anti-melanoma agent can bring down the MAD-CT-2 mRNA and protein level in melanoma cells that express this marker. Accordingly, a compound that demonstrates such an activity may be a good candidate for further testing for anti-tumor efficacy. In this regard, animal or human cells that express MAD-CT-2 can be exposed to a test agent, and the effect of the test agent on MAD-CT-2 expression at the mRNA or protein level relative to that of corresponding untreated control cells can be measured, wherein a lower expression than that of the control cells indicates that the agent is a candidate for further testing as preventive or therapeutic agents for melanoma. The animal or human cells that express MAD-CT-2 can contain a DNA sequence encoding MAD-CT-2 under the control of an endogenous promoter such as a native promoter or another promoter following translocation to a new site. Preferably, human or animal melanoma cells that express MAD-CT-2 are used. In one embodiment, such human or animal melanoma cells are those of a cell line.

[0029] In seventh aspect, the invention relates to kits suitable for use in the methods disclosed herein. For the method of identifying a human subject as a candidate for further prostate cancer examination in connection with analyzing whether the subject has developed an immune response to prostate cancer antigens, the kit includes the proteins or suitable fragments thereof of a prostate cancer antigen panel disclosed herein or expression vectors/systems for expressing the proteins or suitable fragments (e.g., a phage system as described in Example 1 below, an expression vector containing a DNA sequence encoding an antigen protein operably linked to a promoter such as a non-native promoter, or a cell containing the expression vector). In this regard, a protein in the panel can be represented by the protein with additional amino acid sequences at one or both of the N- and C-terminal ends, as long as the additional sequences do not affect the function of the proteins in connection with the present invention (i.e., the ability to bind to the corresponding immunoglobulins). The additional amino acid sequences can, but do not have to, assist in the purification, detection, or stabilization of the proteins. Optionally, the kit also includes a positive control, a negative control or both. For example, a positive control may be a composition containing an antibody to one or more of the antigens in the panel (e.g., a blood/serum/plasma preparation from a patient or group of patients known to interact with one or more of the antigens). A negative control for the proteins in the panel and the expression vectors/systems for expressing the proteins may be a non-tumor antigen protein such as a housekeeping protein and an expression

vector/system for expressing the non-tumor antigen. A negative control for the patient's serum or blood-derived sample may be a serum or blood-derived sample from a normal individual that does not interact with any of the proteins in the panel.

[0030] For the method of identifying a human subject as a candidate for further melanoma examination in connection with analyzing whether the subject developed an immune response to melanoma antigens, the kit includes the proteins or suitable fragments thereof of a melanoma antigen panel disclosed herein or expression vectors/systems for expressing the proteins or suitable fragments (e.g., a phage system as described in Example 1 below, an expression vector containing a DNA sequence encoding an antigen protein operably linked to a promoter such as a non-native promoter, or a cell containing the expression vector). In this regard, a protein in the panel can be represented by the protein with additional amino acid sequences at one or both of the N- and C-terminal ends, as long as the additional sequences do not affect the function of the proteins in connection with the present invention (i.e., the ability to bind to the corresponding immunoglobulins). The additional amino acid sequences can, but do not have to, assist in the purification, detection or stabilization of the proteins. Optionally, the kit also includes a positive control, a negative control or both. For example, a positive control may be a composition containing an antibody to one or more of the antigens in the panel (e.g., a blood/serum/plasma preparation from a patient known to interact with one or more of the antigens). A negative control for the proteins in the panel and the expression vectors/systems for expressing the proteins may be a non-tumor antigen protein, such as a housekeeping protein and an expression vector/system for expressing the non-tumor antigen. A negative control for the patient's serum or blood-derived sample may be a serum or blood-derived sample from a normal individual that does not interact with any of the proteins in the panel.

[0031] For the method of identifying a human subject as a candidate for further melanoma examination in connection with analyzing the expression of MAD-CT-2, the kit includes an antibody to MAD-CT-2 or an oligonucleotide set for amplifying and detecting MAD-CT-2 mRNA or cDNA. For example, the oligonucleotide set can contain a pair of PCR primers, preferably RT-PCR primers, which can be used for both amplifying and detecting the mRNA or cDNA. The set may also contain a separate oligonucleotide for detecting the amplified sequence. Optionally, the kit also includes a positive control, a negative control or both. For example, a positive control may be a composition that contains MAD-CT-2 or the corresponding mRNA or cDNA (e.g., melanoma cells or an extract thereof of either a cell line or a patient that are known to express MAD-CT-2). A negative control for the cells from a subject being tested may be cells that do not express MAD-CT-2 (e.g., skin cells or other types of cells from a normal individual). A negative control for the antibody or oligonucleotide set may be an antibody that does not interact with MAD-CT-2 or a set of oligonucleotides that does not amplify and detect the corresponding mRNA/cDNA (e.g., an antibody to a non-tumor antigen protein, such as a housekeeping protein or a set of oligonucleotides that amplifies and detects the mRNA/cDNA of the non-tumor antigen protein).

[0032] Any of the kits described above can optionally contain an instruction manual directing use of the kit according to the method of the present invention.

[0033] The invention will be more fully understood upon consideration of the following non-limiting examples.

Examples

Example 1

Antibody Responses to Cancer-Testis Antigens in Melanoma Patients

[0034] In this example, we show the construction of a panel of 29 CTA in lambda (λ) phage, and implementation of a novel high throughput immunoscreening method using a panel of sera from patients with melanoma (n=44) and volunteer blood donors (n=50). We show that antibody responses occurred in 39% of patients with melanoma to at least one CTA antigen in a defined panel of 5 compared with 4% of controls ($p<0.001$). Moreover, antibody responses to one antigen, MAD-CT-2, occurred in 27% of patients compared with 0/50 controls ($p<0.0001$). We also show that MAD-CT-2 is expressed in melanoma cell lines.

[0035] Materials and Methods.

[0036] Subject Population: Sera were obtained from 44 male patients with metastatic melanoma, mean age 50 years (range 25-78 years). All patients had been treated with primary resection. 19/44 (43%) were treated with prior immunotherapy, and 9/44 (20%) were treated with prior chemotherapy. All subjects gave written institutional review board (IRB)-approved informed consent for their blood products to be used for immunological research. Blood was collected at the University of Wisconsin Hospital and Clinics (Madison, Wis.), and sera were stored in aliquots at −80° C. until used. Control sera were obtained from volunteer male blood donors, mean age 34 years (range 18-57 years), who also gave IRB-approved written informed consent.

[0037] Phage Cloning: Plasmid DNA encoding full-length cDNAs for 29 CTA were either purchased as IMAGE clones (American Type Culture Collection (ATCC); Manassas, Va.), or were obtained from cDNA expression libraries from previous studies (Hoeppner et al., supra; and Stone B, et al., Int. J. Cancer 104:73-84 (2003)). Primers, specific for each CTA, were designed with the purpose of appending a single 5' EcoRI and a single 3' XhoI site for subcloning. In cases where the CTA had an internal EcoRI or XhoI site, an alternate 5' MfeI site or 3' SalI site was used. Table 2 shows the sequence of the primers and IMAGE clone identification for each CTA construct. Polymerase chain reaction (PCR) was performed using template cDNAs, gene specific primers, Taq polymerase (Promega; Madison, Wis.), and 30 amplification cycles optimized with respect to temperature for each primer pair. PCR products were gel purified (Qiaquick Gel Extraction Kit; Qiagen; Valencia, Calif.), digested with the appropriate restriction enzymes, and ligated into λ-phage arms (Lambda ZAP express protocol; Stratagene; La Jolla, Calif.). Phage were amplified by standard methods and sequenced to confirm their identity and to detect any mutations introduced by PCR (Table 2).

TABLE 2

CTA phage construction. Shown are the names and GenBank identifiers for each CTA chosen for analysis. In addition, the 5' and 3' primers used for the gene-specific cDNA PCR amplification and subcloning are shown, and the IMAGE clone identifiers from which the genes were cloned. Variations from the published amino acid sequences identified after final clone sequencing are shown.

Name	GenBank ID	Image Clone ID	5' Primer
MAGE-A1[1]	NM_004988		GGAATTCATGTCTCTTGAGCAGAGGAGTC (SEQ ID NO: 1)
SSX2[2]	BC007343		GGTGCTCAAATACCAGAGAAG * (SEQ ID NO: 3)
NY-ESO-1[3]	AJ003149		GGAATTCCATGCAGGCCGAAGGCCGGGG (SEQ ID NO: 5)
GAG-7[4]	NM_021123		GGAATTCATGAGTTGGCGAGGAAGATCGACC (SEQ ID NO: 7)
SSX4[5]	U90841		GGAATTCATGAACGGAGACGACGCCTTG (SEQ ID NO: 9)
NXF2[6]	BC015020	3921074	CCAATTGATGTGCTCTACTCTAAAGAAGTGTG (SEQ ID NO: 11)
TPX1[7]	BC022011	4826427	GGAATTCAATGGCTTTACTACCGGTGTTGTTTC (SEQ ID NO: 13)
XAGE-1[8]	BC009538	3893227	GGAATTCATGGAGAGCCCCAAAAAGAAGAACC (SEQ ID NO: 15)
LAGE-1[9]	BC002833	3638129	GGAATTCATGCAGGCCGAAGGCCGGGGCAC (SEQ ID NO: 17)
PAGE-1[4]	BC010897	4043535	GGAATTCGATGAGTGCACGAGTGAGATCAAG (SEQ ID NO: 19)
MAGE-E1[10]	BC081566	6292139	CCAATTGCATGGCTGAGGGAAGCTTCAGCGTG (SEQ ID NO: 21)
SPANXC[11]	BC054023	6648369	CCAATTGATGGACAAACAATCCAGTGCCGGCGG (SEQ ID NO23)
ADAM2[12]	BC064547	5744846	GGAATTCATGTGGCGCGTCTTGTTTCTGCTC (SEQ ID NO: 25)
TSP50[13]	BC037775	5272458	GGAATTCATGGGTCGCTGGTGCCAGACCGTC (SEQ ID NO: 27)
NY-SAR-35[14]	BC034320	4836772	GGAATTCATGTCTTCACATAGGAGGAAAGCGAAG (SEQ ID NO: 29)
FATE1[15]	BC022064	4826440	GGAATTCATGGCAGGAGGCCCTCCCAACACC (SEQ ID NO: 31)
PAGE-5[12]	BC009230	3955765	GGATTCGTGATGCAGGCGCCATGGGCC (SEQ ID NO: 33)
LIP1[12]	BC023635	4841470	GGAATTCATGTCTCTACGCTGCGGGGATGCAG (SEQ ID NO: 35)
SPA17[16]	BC032457	5171849	CCAATTGATGTCGATTCCATTCTCCAACACC (SEQ ID NO: 37)
MAGE-A8[1]	BE387798		GGAATTCATGCCTCTTGAGCAGAGGAGTCAG (SEQ ID NO: 39)
MAGE-B1[17]	BE897525		GGAATTCATGCCTCGGGGTCAGAAGAG (SEQ ID NO: 41)
MAGE-B2[17]	BC026071		GGAATTCATGCCTCGTGGTCAGAAGAGTAAG (SEQ ID NO: 43)

TABLE 2-continued

CTA phage construction. Shown are the names and GenBank identifiers for each CTA chosen for analysis. In addition, the 5' and 3' primers used for the gene-specific cDNA PCR amplification and subcloning are shown, and the IMAGE clone identifiers from which the genes were cloned. Variations from the published amino acid sequences identified after final clone sequencing are shown.

MAGE-A4[1]	BC017723		GGAATTCATGTCTTCTGAGCAGAAGAGTC (SEQ ID NO: 45)
SSX1[2]	BC001003	3445470	GGAATTCATGAACGGAGACAACACCTTTG (SEQ ID NO: 47)
GAGE-2[18]	BC069397	7262151	GGAATTCATGAGTTGGCGAGGAAGATCGACC (SEQ ID NO: 49)
GAGE-4[18]	BC069470		GGAATTCATGATTGGGCCTATGCGGCCCGAG (SEQ ID NO: 51)
MAGE-A3[1]	BC016803		*
MAD-CT-1[19]	NM_002762		GAGGAGCCTGAGCGAACG * (SEQ ID NO: 53)
MAD-CT-2[19]	AK097414 (SEQ ID NO: 55)		GAGGATATGAGATCAGAAAGAGAAG *

Name	3' Primer	Sequence variation:
MAGE-A1[1]	CCGCTCGAGCTCAGACTCCCTCTTCCTCCTC (SEQ ID NO: 2)	
SSX2[2]	CTTTGGGTCCAGATCTCTCGTG * (SEQ ID NO: 4)	
NY-ESO-1[3]	CCGCTCGAGCTTAGCGCCTCTGCCCTGAGGG (SEQ ID NO: 6)	G to V mutation at a.a.#41
GAG-7[4]	CCGCTCGAGTTACACTGTGAGCTTTTCACC (SEQ ID NO: 8)	
SSX4[5]	CCGCTCGAGCTACTCGTCATCTTCCTCAGGG (SEQ ID NO: 10)	
NXF2[6]	CCGCTCGAGTTAGGAGATTTGCTTGAAGGCCTCTG (SEQ ID NO: 12)	
TPX1[7]	CCGCTCGAGTCAGTAAATTTTGTTCTCACATAGG (SEQ ID NO: 14)	
XAGE-1[8]	CCGCTCGAGTTAAACTTGTTGCTCTTCACCTG (SEQ ID NO: 16)	
LAGE-1[9]	CCGCTCGAGCTAAATGAGAGGGGCAGAGAACATC (SEQ ID NO: 18)	
PAGE-1[4]	CCGCTCGAGTTATGGCTGCCCATCCCTGCTTC (SEQ ID NO: 20)	
MAGE-E1[10]	CCGCTCGAGTCAACGGTGCTGGATCCAGGAG (SEQ ID NO: 22)	
SPANXC[11]	CCGCTCGAGCTACTTTGCAGGTATTTCACATTATTTC (SEQ ID NO: 24)	Missing last 7 a.a.'s
ADAM2[12]	CGGCTCGAGACTACCCTTTAGGTTCACTCTCAC (SEQ ID NO: 26)	
TSP50[13]	CCGCTCGAGTCAGAGGGCAGCAAGGAGG (SEQ ID NO: 28)	

TABLE 2-continued

CTA phage construction. Shown are the names and GenBank identifiers for each CTA chosen for analysis. In addition, the 5' and 3' primers used for the gene-specific cDNA PCR amplification and subcloning are shown, and the IMAGE clone identifiers from which the genes were cloned. Variations from the published amino acid sequences identified after final clone sequencing are shown.

NY-SAR-35[14]	CCGCTCGAGCTACTCGTCACCATGTTCCTCAC (SEQ ID NO: 30)
FATE1[15]	CCGCTCGAGATCACTGGTTCATCCACAGCCAC (SEQ ID NO: 32)
PAGE-5[12]	CCGCTCGAGCTATAGTTGCCCTTCACCTGCTTGG (SEQ ID NO: 34)
LIP1[12]	CCGCTCGAGTTTTAGAGGTCTTTTGTTTTTCTTTTAGCC (SEQ ID NO: 36)
SPA17[16]	CCGCTCGAGTCACTTGTTTTCCTCTTTTTCCTC (SEQ ID NO: 38)
MAGE-A8[1]	CCGCTCGAGACTCACTCTTCCCCCTCTCTCAA (SEQ ID NO: 40)
MAGE-B1[17]	ACGCGTCGACTCACATGGGGTGGGAGGACCTG (SEQ ID NO: 42)
MAGE-B2[17]	CCGCTCGAGCTCAGACTCCGGCTTTCTCTTC (SEQ ID NO: 44)
MAGE-A4[1]	CCGCTCGAGCTCAGACTCCCTCTTCCTCCT (SEQ ID NO: 46)
SSX1[2]	CCGCTCGAGTTACTCGTCATCTTCCTCAGGG (SEQ ID NO: 48)
GAGE-2[18]	CCGCTCGAGTTAACACTGTGATTGCTTTTCACCTTC (SEQ ID NO: 50)
GAGE-4[18]	CCGCTCGAGTTAACACTGTGATTGCCCTTCACCTTC (SEQ ID NO: 52)
MAGE-A3[1]	*
MAD-CT-1[19]	GGATTCTTAGTGCCTTCTGCATGTTCTCTT * (SEQ ID NO: 54)
MAD-CT-2[19]	TCCACACTGCCAGTGTGGCTCAT * (SEQ ID NO: 56)

* Phage encoding MAGE-A3 were obtained from previous unpublished studies.

[1]De Plaen E et al. Immunogenetics 1994, 40: 360-369.
[2]Crew AJ et al. Embo J. 1995, 14: 2333-2340.
[3]Chen YT et al. Proc. Natl. Acad. Sci. U.S.A. 1997, 94: 1914-1918.
[4]Chen ME et al. J Biol Chem. 1998, 273: 17618-17625.
[5]Gure AO et al. Int J Cancer 1997, 72: 965-971.
[6]Loriot A et al. Int J Cancer 2003, 105: 371-376.
[7]Kasahara M et al. Immunogenetics 1989, 29: 61-63.
[8]Brinkmann U et al. Cancer Res. 1999, 59: 1445-1448.
[9]Chen YT et al. Proc Natl Acad Sci U S A. 1998, 95: 6919-6923.
[10]Sasaki M et al. Cancer Res. 2001, 61: 4809-4814.
[11]Zendman AJ et al. Cancer Res. 1999, 59: 6223-6229.
[12]Scanlan MJ et al. Int J Cancer 2002, 98: 485-492.
[13]Yuan L et al. Cancer Res. 1999, 59: 3215-3221.
[14]Lee SY et al. Proc Natl Acad Sci U S A. 2003, 100: 2651-2656.
[15]Dong XY et al. Br J Cancer 2003, 89: 291-297.
[16]Lim SH et al. Blood 2001, 97: 1508-1510.
[17]Lurquin C et al. Genomics 1997, 46: 397-408.
[18]Van den Eynde B et al. J Exp Med. 1995, 182: 689-698.
[19]Hoeppner LH et al. Cancer Immun. 2006, 6: 1-7.

[0038] Reverse transcriptase-PCR (RT-PCR): *E. coli* cultures (XL-1 blue MRF strain; Stratagene) were transduced with 10^6 pfu of individual phage and grown overnight in LB media+20 mM $MgSO_4$+0.2% maltose+2.5 mM isopropyl-

beta-D-thiogalactopyranoside (IPTG) at 37° C. Total RNA was prepared from centrifugally pelleted cultures (RNeasy Mini Columns; Qiagen). RT-PCR reactions were conducted using the Qiagen One-Step RT-PCR Kit (Qiagen) and transcript-specific primers (T7 Promoter and T3 Promoter; Invitrogen; Carlsbad, Calif.). To control for the possible amplification of contaminating genomic or phage DNA in the total RNA preparations, duplicate reactions were first heated to 100° C. for 10 minutes immediately prior to the normal RT-PCR reaction to attenuate reverse transcriptase activity. PCR amplification reactions were resolved on agarose gels and the size of the amplified transcript confirmed by comparison with DNA size markers (1 Kb ladder; Promega).

[0039] High Throughput Immunoblot: XL-1 blue MRF *E. coli* were grown overnight at 31° C. in LB medium supplemented with 20 mM $MgSO_4$ and 0.2% maltose. Cultured cells were then collected by centrifugation, resuspended in 10 mM $MgSO_4$, and poured in top agarose (LB broth/10 mM $MgSO_4$/0.2% maltose/0.7% agarose) over LB agar in Omniwell plates (Nunc). 9,000 pfu of individual phage were then spotted in a 0.9 μl volume in replicates onto multiple bacterial agar lawns using a liquid handling robot (Beckman; Biomek FX; Fullerton, Calif.). Spotted plates were allowed to sit undisturbed for 15 minutes at room temperature, and then overlaid with nitrocellulose membranes impregnated with 10 mM IPTG. Plates were incubated overnight at 37° C. The next day, filters were gently peeled from bacterial lawns and washed twice in TBST (50 mM Tris pH 7.2, 100 mM NaCl, 0.5% Tween-20) for 5 minutes and once in TBS (50 mM Tris pH 7.2, 100 mM NaCl) for an additional 5 minutes. The filters were then blocked in TBS+1% BSA for 1 hour shaking at room temperature.

[0040] Membranes were then probed with human serum preadsorbed to *E. coli* proteins and the empty phage construct diluted 1:100 in blocking solution at 4° C. overnight. Following this, the membranes were again washed twice in TBST and once in TBS, and human IgG was detected with anti-human IgG-AP (Sigma; St. Louis, Mo.) diluted 1:25,000 in blocking solution for 1 hour shaking at room temperature. The filters were again washed and then developed with 0.3 mg/ml nitro-blue tetrazolium chloride (NBT)+0.15 mg/ml 5-bromo-4-chloro-3'-indolyphosphate p-toluidine salt (BCIP). After development, filters were washed with deionized water, and then dried prior to evaluation. Immunoreactive plaques were recorded for each filter by visual comparison with an internal positive (phage encoding human IgG) and negative (empty phage encoding beta-galactosidase) control plaques. A plaque was defined as positive if replicate plaques were read as positive and there was accordance among at least six of nine independent observers, to reduce the possibility of subjective interpretation (Dunphy E, et al., J. Clin. Immunol. 24:492-501 (2004)). Comparison of immunoreactive spots among subject groups was made by chi-square analysis.

[0041] Western blot: Amino-terminal glutathione-S-transferase (GST) linked SSX-2 and GST (control) were purified from IPTG-induced overnight *E. coli* (Rosetta Gami expression strain) cultures transformed with pET41b plasmid (EMD Biosciences; San Diego, Calif.) containing full length SSX-2 cloned in frame with the ATG start codon. Purified proteins were stored at −80° C. and were thawed immediately before mixing 1:1 with 2×SDS Laemmli's loading buffer (0.04 M Tris pH 6.8, 12% glycerol, 1.25% sodium docecyl sulfate (SDS), 3% β-mercaptoethanol, 0.06% bromophenol blue) and boiling for 10 minutes at 100° C. The proteins were then resolved on 15% SDS-polyacrylamide gels and were electrophoretically transferred to nitrocellulose membranes. Membranes were then probed, using standard immunoblot techniques, with preadsorbed patient sera diluted 1:200 in blocking solution or protein-specific antibodies. Detection antibodies included goat anti-SSX-2 polyclonal antibody (N-16; Santa Cruz Biotechnology; Santa Cruz, Calif.), and anti-GST monoclonal antibody (rabbit A5800; Invitrogen; Carlsbad, Calif.).

[0042] Results.

[0043] Cloning and validation of phage encoding CTA: We wished to develop a tool permitting simultaneous analysis of antibody responses to multiple antigens within a single serum sample. Given the ease of λ phage cloning and expression compared with standard methods of protein purification, we chose to use a phage expression array methodology, similar to what we have previously described (Dunphy E J et al. J. Clin. Immunol. 2004, 24:492-501). Consequently, cDNA encoding 25 unique CTA were amplified by PCR, ligated into λ phage arms, packaged into phage particles, and sequenced to confirm their identity. In addition, phage encoding SSX-2, MAGE-A3, MAD-CT-1 and MAD-CT-2 were available from prior studies (Hoeppner et al., supra; Crew A, et al., Embo. J. 14:2333-2340 (1995); and De Plaen E, et al., Immunogenetics 40:360-369 (1994), each of which is incorporated herein by reference as if set forth in its entirety). Gene transcription from each CTA phage construct was evaluated using RT-PCR reactions on total RNA purified from phage-transduced *E. coli.* In all cases, CTA phage produced mRNA transcripts of the correct predicted size. Similarly, immunoblot confirmed protein expression of CTA for which antibody reagents were available. For example, FIG. **1** demonstrates that a monoclonal antibody recognizing SSX2 and SSX4 could identify protein expression from phage-transduced *E. coli.*

[0044] Antigen-specific IgG to a panel of CTA were detected by high throughput immunoblot (HTI): Phage encoding individual CTA were directly spotted onto bacterial lawns grown in top agarose using a Biomek FX liquid handling robot. Optimization studies showed that a volume of 0.9 μl of 10^4 pfu/μl phage produced reproducibly dense plaques. After plating and transfer to nitrocellulose membranes, the filters were probed with human sera that had been preadsorbed for antibodies to *E. coli* and diluted 1:100. Specifically, sera were obtained from 44 male patients with metastatic melanoma, and 50 male controls without cancer, and used to probe 94 individual membranes. Human IgG was then detected and visualized as described. Within each spotted array, phage encoding human IgG were included as a positive control, and empty phage were included as a negative control. In our hands, by using the liquid handling robot to uniformly array phage, we could easily screen at least 100 sera at the same time and accomplish the entire screening in three days.

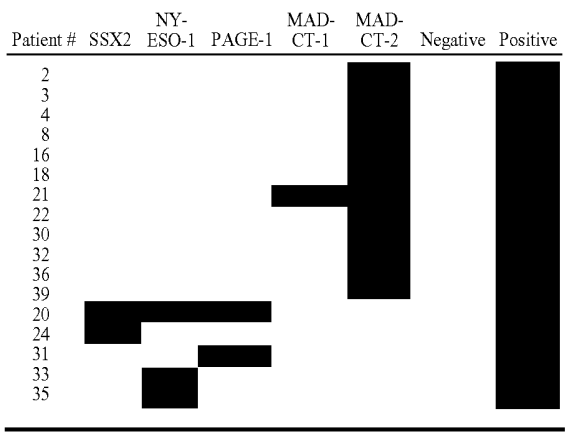
[0045] Given the presence of internal positive and negative control phage plaques on each membrane, several methods were initially evaluated to objectively evaluate the final immunoblot membranes in an automated fashion. These methods included using fluorescent analysis of membranes following the use of fluorescent-tagged secondary antibodies, and automated densitometric analysis following colorimetric staining (not shown). Unfortunately, given variable backgrounds due to the *E. coli* bacterial lawns, these methods were unreliable and frequently "missed" plaques that were clearly immunoreactive, and false positive plaques were often observed. Consequently, we found that visual inspection was more accurate. To eliminate subjectivity from this evaluation, and reduce the possibility of false positive interpretation, each filter was reviewed and scored by a panel of independent readers. Plaques were scored as immunoreactive if duplicate plaques were each scored positive by the same observer, and if there was concordance among at least 6 of 9 independent observers, similar to what we have previously reported (Dunphy E, et al., J. Clin. Immunol. 24:492-501 (2004)). A subset of sera was re-evaluated in similar fashion to confirm the reproducibility of the findings, and immunoreactive plaques were similar to those observed in the initial screening (data not shown).

[0046] The presence of IgG specific for SSX-2 was confirmed by Western blot analysis: Western blot analysis was used when possible to confirm responses to CTA identified by HTI. As shown in FIG. **2**A, IgG specific for SSX-2 were detectable by Western blot using sera from patient 24, a patient found by HTI to have IgG specific for SSX-2 (FIG. **2**B). Conversely, no response was detectable using sera from patients 19 or 35 from whom no detectable HTI response to SSX-2 was detected, indicating that HTI screening provided data consistent with that by Western blot analysis.

[0047] Patients with melanoma have frequent antibody responses to SSX-2, NY-ESO-1, and MAD-CT-2: As shown in FIG. **3**, IgG responses to SSX-2 and NY-ESO-1 were detected in 2/44 (5%, p=0.13) and 3/44 (7%, p=0.061) patients, respectively, compared with 0/50 male controls. In addition, IgG responses to MAD-CT-2 were identified in 12/44 (27%) of patients compared with 0/50 controls (p<0.001). IgG responses were also detected at lower frequencies to PAGE-1 and MAD-CT-1. Overall, IgG responses to at least one of the CTA were detected in 17/44 (39%) of male patients with melanoma, compared with 2/50 (4%) of controls (p<0.001) (FIG. **3** and Table 3). There was no apparent association with whether patients had been previously treated with immunotherapy, as 7/17 patients with antibody responses had been previously treated with immunotherapy (41%) compared with 12/27 patients who had no CTA-specific antibody responses (44%, p=0.83). IgG responses to MAD-CT-2 were detected in patients who did not demonstrate reactivity to SSX-2 or NY-ESO-1 (Table 3). This did not appear to be due to differences in patterns of gene expression, as mRNA encoding all three gene products were detectable in two different melanoma cell lines tested (FIG. **4**). The identification of MAD-CT-2 transcripts in melanoma cell lines, however, demonstrates that it is a melanoma CTA.

TABLE 3

IgG responses to at least one CTA were detectable in multiple patients with melanoma. Shown are the patients with serum IgG specific for each CTA (shaded box) arrayed against the CTA tested.



Example 2

Antibody Responses to Cancer-Testis Antigens in Prostate Cancer Patients

[0048] Using techniques similar to those described in Example 1 above or an ELISA assay, the inventors have found that antibody responses occurred at a higher rate in prostate cancer patients than in control individuals for the following antigens: SSX-2, NY-ESO-1, LAGE-1, NFX2, MAD-Pro-22, MAD-Pro-30, MAD-Pro-34, MAD-Pro-42, MAD-CaP-1, MAD-CaP-5, MAD-CaP-15, MAD-CaP-20, MAD-CT-1, MAD-CT-2, MAD-CT-3, MAD-CT-5, transgelin, ZCWCC3, ACAA1, AR (or ligand-binding domain of AR) and actinin. The results are summarized in Table 4 with the results obtained by either ELISA assays (labeled) or using the techniques described in Example 1. The ELISA assays were conducted using purified antigen proteins which were probed with sera from prostate cancer patients or normal individuals followed by detection of antigen-antibody interaction using enzyme-conjugated anti-human Ig antibody and the corresponding enzyme substrate.

[0049] FIG. **5** shows a specific example of the studies conducted with the techniques described in Example 1 in which SSX-2, NY-ESO-1, LAGE-1, and NFX2 were found to be expressed at a higher rate in prostate cancer patients than in control individuals.

[0050] Using RT-PCR and SSX-2 specific antibody, the inventors found that SSX-2 was expressed in at least three prostate cancer cell lines: LAPC4 (FIG. **6**), MDAPCa2b (FIG. **6**), and LNCap (data not shown). The inventors also found that SSX-2 was expressed in prostate cancer metastatic tissue samples (FIG. **7**).

TABLE 4

	Antigen-specific IgG Responses	
Antigen	Patients with Prostate Cancer	Control Male Blood Donors
SSX-2	1/100	0/50
NY-ESO-1	3/100	0/50
MAD-Pro-22	16/100	9/64
(PSA)	22/200 by ELISA	3/100 by ELISA
MAD-Pro-30	18/100	3/64
MAD-Pro-34	5/100	0/64
MAD-Pro-42	5/100	0/64
MAD-CaP-1	2/100	0/50
MAD-CaP-5	5/100	1/50
MAD-CaP-15	3/100	0/50
MAD-CaP-20	5/100	1/50
MAD-CT-1	5/109	1/52
MAD-CT-2	3/109	0/52
MAD-CT-3	2/109	0/52
MAD-CT-5	5/109	1/52
Transgelin	2/27	0/25
ZCWCC3	5/26	4/25
ACAA1	13/26	9/25
AR LBD	18/105 by ELISA	0/41 by ELISA
Actinin	1/1	

[0051] Although the invention has been described in connection with specific embodiments, it is understood that the invention is not limited to such specific embodiments but encompasses all such modifications and variations apparent to a skilled artisan that fall within the scope of the appended claims.

SEQUENCE LISTING

<160> NUMBER OF SEQ ID NOS: 70

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<213> ORGANISM: ARTIFICIAL SEQUENCE
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ggaattccat gcaggccgaa ggccgggg 28

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<210> SEQ ID NO 13
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ccaattgatg gacaaacaat ccagtgccgg cgg 33

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<400> SEQUENCE: 24

ccgctcgagc tactttgcag gtatttcaca ttatttc 37

<210> SEQ ID NO 25
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<212> TYPE: DNA
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<210> SEQ ID NO 26
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<213> ORGANISM: ARTIFICIAL SEQUENCE
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<223> OTHER INFORMATION: PCR PRIMER

<400> SEQUENCE: 26

cggctcgaga ctacccttta ggttcactct cac 33

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<212> TYPE: DNA
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<220> FEATURE:
<223> OTHER INFORMATION: PCR PRIMER

<400> SEQUENCE: 27

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<400> SEQUENCE: 28

ccgctcgagt cagagggcag caaggagg 28

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ggaattcatg gcaggaggcc ctcccaacac c 31

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ccgctcgaga tcactggttc atccacagcc ac 32

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ggaattcgtg atgcaggcgc catgggcc 28

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<212> TYPE: DNA
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<400> SEQUENCE: 35

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<212> TYPE: DNA
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<220> FEATURE:
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<210> SEQ ID NO 37
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ccaattgatg tcgattccat tctccaacac c 31

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<212> TYPE: DNA
<213> ORGANISM: ARTIFICIAL SEQUENCE
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ccgctcgagt cacttgtttt cctctttttc ctc 33

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<220> FEATURE:
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<212> TYPE: DNA
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ccgctcgaga ctcactcttc cccctctctc aa 32

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<210> SEQ ID NO 42
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acgcgtcgac tcacatgggg tgggaggacc tg 32

<210> SEQ ID NO 43
<211> LENGTH: 31
<212> TYPE: DNA
<213> ORGANISM: ARTIFICIAL SEQUENCE
<220> FEATURE:
<223> OTHER INFORMATION: PCR PRIMER

<400> SEQUENCE: 43

ggaattcatg cctcgtggtc agaagagtaa g 31

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<210> SEQ ID NO 45
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<212> TYPE: DNA
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ggaattcatg agttggcgag gaagatcgac c 31

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<400> SEQUENCE: 50

ccgctcgagt taacactgtg attgcttttc accttc 36

<210> SEQ ID NO 51
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<212> TYPE: DNA

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<213> ORGANISM: ARTIFICIAL SEQUENCE
<220> FEATURE:
<223> OTHER INFORMATION: PCR PRIMER

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ccgctcgagt taacactgtg attgcccttc accttc 36

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<400> SEQUENCE: 53

gaggagcctg agcgaacg 18

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<211> LENGTH: 31
<212> TYPE: DNA
<213> ORGANISM: ARTIFICIAL SEQUENCE
<220> FEATURE:
<223> OTHER INFORMATION: PCR PRIMER

<400> SEQUENCE: 54

ggaattctta gtgccttctg catgttctct t 31

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<212> TYPE: DNA
<213> ORGANISM: ARTIFICIAL SEQUENCE
<220> FEATURE:
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gaggatatga gatcagaaag agaag 25

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tccacactgc cagtgtggct cat 23

<210> SEQ ID NO 57
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<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: MAD-Pro-22

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<400> SEQUENCE: 57

Met Trp Val Pro Val Val Phe Leu Thr Leu Ser Val Thr Trp Ile Gly
1 5 10 15

Glu Arg Gly His Gly Trp Gly Asp Ala Gly Glu Gly Ala Ser Pro Asp
20 25 30

Cys Gln Ala Glu Ala Leu Ser Pro Pro Thr Gln His Pro Ser Pro Asp
35 40 45

Arg Glu Leu Gly Ser Phe Leu Ser Leu Pro Ala Pro Leu Gln Ala His
50 55 60

Thr Pro Ser Pro Ser Ile Leu Gln Gln Ser Ser Leu Pro His Gln Val
65 70 75 80

Pro Ala Pro Ser His Leu Pro Gln Asn Phe Leu Pro Ile Ala Gln Pro
85 90 95

Ala Pro Cys Ser Gln Leu Leu Tyr
100

<210> SEQ ID NO 58
<211> LENGTH: 487
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: MAD-Pro-30

<400> SEQUENCE: 58

Met Ala Pro Val Val Thr Gly Lys Phe Gly Glu Arg Pro Pro Pro Lys
1 5 10 15

Arg Leu Thr Arg Glu Ala Met Arg Asn Tyr Leu Lys Glu Arg Gly Asp
20 25 30

Gln Thr Val Leu Ile Leu His Ala Lys Val Ala Gln Lys Ser Tyr Gly
35 40 45

Asn Glu Lys Arg Phe Phe Cys Pro Pro Pro Cys Val Tyr Leu Met Gly
50 55 60

Ser Gly Trp Lys Lys Lys Lys Glu Gln Met Glu Arg Asp Gly Cys Ser
65 70 75 80

Glu Gln Glu Ser Gln Pro Cys Ala Phe Ile Gly Ile Gly Asn Ser Asp
85 90 95

Gln Glu Met Gln Gln Leu Asn Leu Glu Gly Lys Asn Tyr Cys Thr Ala
100 105 110

Lys Thr Leu Tyr Ile Ser Asp Ser Asp Lys Arg Lys His Phe Met Leu
115 120 125

Ser Val Lys Met Phe Tyr Gly Asn Ser Asp Asp Ile Gly Val Phe Leu
130 135 140

Ser Lys Arg Ile Lys Val Ile Ser Lys Pro Ser Lys Lys Lys Gln Ser
145 150 155 160

Leu Lys Asn Ala Asp Leu Cys Ile Ala Ser Gly Thr Lys Val Ala Leu
165 170 175

Phe Asn Arg Leu Arg Ser Gln Thr Val Ser Thr Arg Tyr Leu His Val
180 185 190

Glu Gly Gly Asn Phe His Ala Ser Ser Gln Gln Trp Gly Ala Phe Phe
195 200 205

Ile His Leu Leu Asp Asp Asp Glu Ser Glu Gly Glu Glu Phe Thr Val
210 215 220

Arg Asp Gly Tyr Ile His Tyr Gly Gln Thr Val Lys Leu Val Cys Ser

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225                 230                 235                 240

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

Gln Thr Ala Leu Leu Asp Ala Asp Asp Pro Val Ser Gln Leu His Lys
260                 265                 270

Cys Ala Phe Tyr Leu Lys Asp Thr Glu Arg Met Tyr Leu Cys Leu Ser
275                 280                 285

Gln Glu Arg Ile Ile Gln Phe Gln Ala Thr Pro Cys Pro Lys Glu Pro
290                 295                 300

Asn Lys Glu Met Ile Asn Asp Gly Ala Ser Trp Thr Ile Ile Ser Thr
305                 310                 315                 320

Asp Lys Ala Glu Tyr Thr Phe Tyr Glu Gly Met Gly Pro Val Leu Ala
325                 330                 335

Pro Val Thr Pro Val Pro Val Val Glu Ser Leu Gln Leu Asn Gly Gly
340                 345                 350

Gly Asp Val Ala Met Leu Glu Leu Thr Gly Gln Asn Phe Thr Pro Asn
355                 360                 365

Leu Arg Val Trp Phe Gly Asp Val Glu Ala Glu Thr Met Tyr Arg Cys
370                 375                 380

Gly Glu Ser Met Leu Cys Val Val Pro Asp Ile Ser Ala Phe Arg Glu
385                 390                 395                 400

Gly Trp Arg Trp Val Arg Gln Pro Val Gln Val Pro Val Thr Leu Val
405                 410                 415

Arg Asn Asp Gly Ile Ile Tyr Ser Thr Ser Leu Thr Phe Thr Tyr Thr
420                 425                 430

Pro Glu Pro Gly Pro Arg Pro His Cys Ser Ala Ala Gly Ala Ile Leu
435                 440                 445

Arg Ala Asn Ser Ser Gln Val Pro Pro Asn Glu Ser Asn Thr Asn Ser
450                 455                 460

Glu Gly Ser Tyr Thr Asn Ala Ser Thr Asn Ser Thr Ser Val Thr Ser
465                 470                 475                 480

Ser Thr Ala Thr Val Val Ser
485


<210> SEQ ID NO 59
<211> LENGTH: 919
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: AR or AR-LBD

<400> SEQUENCE: 59

Met Glu Val Gln Leu Gly Leu Gly Arg Val Tyr Pro Arg Pro Pro Ser
1               5                   10                  15

Lys Thr Tyr Arg Gly Ala Phe Gln Asn Leu Phe Gln Ser Val Arg Glu
20                  25                  30

Val Ile Gln Asn Pro Gly Pro Arg His Pro Glu Ala Ala Ser Ala Ala
35                  40                  45

Pro Pro Gly Ala Ser Leu Leu Leu Leu Gln Gln Gln Gln Gln Gln Gln
50                  55                  60

Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Gln Glu Thr
65                  70                  75                  80

Ser Pro Arg Gln Gln Gln Gln Gln Gly Glu Asp Gly Ser Pro Gln
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85                  90                  95

Ala His Arg Arg Gly Pro Thr Gly Tyr Leu Val Leu Asp Glu Glu Gln
100                 105                 110

Gln Pro Ser Gln Pro Gln Ser Ala Leu Glu Cys His Pro Glu Arg Gly
115                 120                 125

Cys Val Pro Glu Pro Gly Ala Ala Val Ala Ala Ser Lys Gly Leu Pro
130                 135                 140

Gln Gln Leu Pro Ala Pro Pro Asp Glu Asp Asp Ser Ala Ala Pro Ser
145                 150                 155                 160

Thr Leu Ser Leu Leu Gly Pro Thr Phe Pro Gly Leu Ser Ser Cys Ser
165                 170                 175

Ala Asp Leu Lys Asp Ile Leu Ser Glu Ala Ser Thr Met Gln Leu Leu
180                 185                 190

Gln Gln Gln Gln Gln Glu Ala Val Ser Glu Gly Ser Ser Ser Gly Arg
195                 200                 205

Ala Arg Glu Ala Ser Gly Ala Pro Thr Ser Ser Lys Asp Asn Tyr Leu
210                 215                 220

Gly Gly Thr Ser Thr Ile Ser Asp Asn Ala Lys Glu Leu Cys Lys Ala
225                 230                 235                 240

Val Ser Val Ser Met Gly Leu Gly Val Glu Ala Leu Glu His Leu Ser
245                 250                 255

Pro Gly Glu Gln Leu Arg Gly Asp Cys Met Tyr Ala Pro Leu Leu Gly
260                 265                 270

Val Pro Pro Ala Val Arg Pro Thr Pro Cys Ala Pro Leu Ala Glu Cys
275                 280                 285

Lys Gly Ser Leu Leu Asp Asp Ser Ala Gly Lys Ser Thr Glu Asp Thr
290                 295                 300

Ala Glu Tyr Ser Pro Phe Lys Gly Gly Tyr Thr Lys Gly Leu Glu Gly
305                 310                 315                 320

Glu Ser Leu Gly Cys Ser Gly Ser Ala Ala Ala Gly Ser Ser Gly Thr
325                 330                 335

Leu Glu Leu Pro Ser Thr Leu Ser Leu Tyr Lys Ser Gly Ala Leu Asp
340                 345                 350

Glu Ala Ala Ala Tyr Gln Ser Arg Asp Tyr Tyr Asn Phe Pro Leu Ala
355                 360                 365

Leu Ala Gly Pro Pro Pro Pro Pro Pro Pro His Pro His Ala Arg
370                 375                 380

Ile Lys Leu Glu Asn Pro Leu Asp Tyr Gly Ser Ala Trp Ala Ala Ala
385                 390                 395                 400

Ala Ala Gln Cys Arg Tyr Gly Asp Leu Ala Ser Leu His Gly Ala Gly
405                 410                 415

Ala Ala Gly Pro Gly Ser Gly Ser Pro Ser Ala Ala Ala Ser Ser Ser
420                 425                 430

Trp His Thr Leu Phe Thr Ala Glu Glu Gly Gln Leu Tyr Gly Pro Cys
435                 440                 445

Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly Gly
450                 455                 460

Gly Gly Gly Gly Gly Gly Gly Gly Glu Ala Gly Ala Val Ala Pro Tyr
465                 470                 475                 480

Gly Tyr Thr Arg Pro Pro Gln Gly Leu Ala Gly Gln Glu Ser Asp Phe
485                 490                 495
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Thr Ala Pro Asp Val Trp Tyr Pro Gly Gly Met Val Ser Arg Val Pro
500                 505                 510

Tyr Pro Ser Pro Thr Cys Val Lys Ser Glu Met Gly Pro Trp Met Asp
515                 520                 525

Ser Tyr Ser Gly Pro Tyr Gly Asp Met Arg Leu Glu Thr Ala Arg Asp
530                 535                 540

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

Ile Cys Gly Asp Glu Ala Ser Gly Cys His Tyr Gly Ala Leu Thr Cys
565                 570                 575

Gly Ser Cys Lys Val Phe Phe Lys Arg Ala Ala Glu Gly Lys Gln Lys
580                 585                 590

Tyr Leu Cys Ala Ser Arg Asn Asp Cys Thr Ile Asp Lys Phe Arg Arg
595                 600                 605

Lys Asn Cys Pro Ser Cys Arg Leu Arg Lys Cys Tyr Glu Ala Gly Met
610                 615                 620

Thr Leu Gly Ala Arg Lys Leu Lys Lys Leu Gly Asn Leu Lys Leu Gln
625                 630                 635                 640

Glu Glu Gly Glu Ala Ser Ser Thr Thr Ser Pro Thr Glu Glu Thr Thr
645                 650                 655

Gln Lys Leu Thr Val Ser His Ile Glu Gly Tyr Glu Cys Gln Pro Ile
660                 665                 670

Phe Leu Asn Val Leu Glu Ala Ile Glu Pro Gly Val Val Cys Ala Gly
675                 680                 685

His Asp Asn Asn Gln Pro Asp Ser Phe Ala Ala Leu Leu Ser Ser Leu
690                 695                 700

Asn Glu Leu Gly Glu Arg Gln Leu Val His Val Val Lys Trp Ala Lys
705                 710                 715                 720

Ala Leu Pro Gly Phe Arg Asn Leu His Val Asp Asp Gln Met Ala Val
725                 730                 735

Ile Gln Tyr Ser Trp Met Gly Leu Met Val Phe Ala Met Gly Trp Arg
740                 745                 750

Ser Phe Thr Asn Val Asn Ser Arg Met Leu Tyr Phe Ala Pro Asp Leu
755                 760                 765

Val Phe Asn Glu Tyr Arg Met His Lys Ser Arg Met Tyr Ser Gln Cys
770                 775                 780

Val Arg Met Arg His Leu Ser Gln Glu Phe Gly Trp Leu Gln Ile Thr
785                 790                 795                 800

Pro Gln Glu Phe Leu Cys Met Lys Ala Leu Leu Leu Phe Ser Ile Ile
805                 810                 815

Pro Val Asp Gly Leu Lys Asn Gln Lys Phe Phe Asp Glu Leu Arg Met
820                 825                 830

Asn Tyr Ile Lys Glu Leu Asp Arg Ile Ile Ala Cys Lys Arg Lys Asn
835                 840                 845

Pro Thr Ser Cys Ser Arg Arg Phe Tyr Gln Leu Thr Lys Leu Leu Asp
850                 855                 860

Ser Val Gln Pro Ile Ala Arg Glu Leu His Gln Phe Thr Phe Asp Leu
865                 870                 875                 880

Leu Ile Lys Ser His Met Val Ser Val Asp Phe Pro Glu Met Met Ala
885                 890                 895
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Glu Ile Ile Ser Val Gln Val Pro Lys Ile Leu Ser Gly Lys Val Lys
900 905 910

Pro Ile Tyr Phe His Thr Gln
915

<210> SEQ ID NO 60
<211> LENGTH: 1309
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: SSX-2
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (955)..(958)
<223> OTHER INFORMATION: n is a, c, g, or t
<220> FEATURE:
<221> NAME/KEY: misc_feature
<222> LOCATION: (968)..(968)
<223> OTHER INFORMATION: n is a, c, g, or t

<400> SEQUENCE: 60

actttctctc tctttcgatt cttccatact cagagtacgc acggtctgat tttctctttg 60

gattcttcca aaatcagagt cagactgctc ccggtgccat gaacggagac gacgcctttg 120

caaggagacc cacggttggt gctcaaatac cagagaagat ccaaaaggcc ttcgatgata 180

ttgccaaata cttctctaag gaagagtggg aaaagatgaa agcctcggag aaaatcttct 240

atgtgtatat gaagagaaag tatgaggcta tgactaaact aggtttcaag gccaccctcc 300

caccttttcat gtgtaataaa cgggccgaag acttccaggg gaatgatttg gataatgacc 360

ctaaccgtgg gaatcaggtt gaacgtcctc agatgacttt cggcaggctc cagggaatct 420

ccccgaagat catgcccaag aagccagcag aggaaggaaa tgattcggag gaagtgccag 480

aagcatctgg cccacaaaat gatgggaaag agctgtgccc cccgggaaaa ccaactacct 540

ctgagaagat tcacgagaga tctggaccca aaaggggga acatgcctgg acccacagac 600

tgcgtgagag aaaacagctg gtgatttatg aagagatcag cgaccctgag gaagatgacg 660

agtaactccc ctcagggata cgacacatgc ccatgatgag aagcagaacg tggtgacctt 720

tcacgaacat gggcatggct gcggacccct cgtcatcagg tgcatagcaa gtgaaagcaa 780

gtgttcacaa cagtgaaaag ttgagcgtca ttttttcttag tgtgccaaga gttcgatgtt 840

agcgtttacg ttgtattttc ttacactgtg tcattctgtt agatactaac attttcattg 900

atgacgcaag ccatacttaa tgcatatttt ggtttgggta tccatgaacc taccnnnnga 960

aaccaagnat tgccggttac ctctgcatgg accagcatta ccctcctctc tccccagatg 1020

tgactactga ggcagttctg agtgtttaat ttcagatttt ttcctctgca tttacacaca 1080

cacgacacaa accacaccac acacacacac acacacacac acacacacac acacacacca 1140

agtaccagta taagcatctg ccatctgctt ttcccattgc catgcgtcct ggtcaagctc 1200

ccctcactct gtttcctggt cagcatgtac tcccctcatc cgattcccct gtagcagtca 1260

ctgcacagtt aataaacctt tgcaaacgtt aaaaaaaaaa aaaaaaaaa 1309

<210> SEQ ID NO 61
<211> LENGTH: 303
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: MAD-Pro-42

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<400> SEQUENCE: 61

Met Lys Gly Lys Glu Glu Lys Glu Gly Gly Ala Arg Leu Gly Ala Gly
1 5 10 15

Gly Gly Ser Pro Glu Lys Ser Pro Ser Ala Gln Glu Leu Lys Glu Gln
20 25 30

Gly Asn Arg Leu Phe Val Gly Arg Lys Tyr Pro Glu Ala Ala Ala Cys
35 40 45

Tyr Gly Arg Ala Ile Thr Arg Asn Pro Leu Val Ala Val Tyr Tyr Thr
50 55 60

Asn Arg Ala Leu Cys Tyr Leu Lys Met Gln Gln His Glu Gln Ala Leu
65 70 75 80

Ala Asp Cys Arg Arg Ala Leu Glu Leu Asp Gly Gln Ser Val Lys Ala
85 90 95

His Phe Phe Leu Gly Gln Cys Gln Leu Glu Met Glu Ser Tyr Asp Glu
100 105 110

Ala Ile Ala Asn Leu Gln Arg Ala Tyr Ser Leu Ala Lys Glu Gln Arg
115 120 125

Leu Asn Phe Gly Asp Asp Ile Pro Ser Ala Leu Arg Ile Ala Lys Lys
130 135 140

Lys Arg Trp Asn Ser Ile Glu Glu Arg Arg Ile His Gln Glu Ser Glu
145 150 155 160

Leu His Ser Tyr Leu Ser Arg Leu Ile Ala Ala Glu Arg Glu Arg Glu
165 170 175

Leu Glu Glu Cys Gln Arg Asn His Glu Gly Asp Glu Asp Asp Ser His
180 185 190

Val Arg Ala Gln Gln Ala Cys Ile Glu Ala Lys His Asp Lys Tyr Met
195 200 205

Ala Asp Met Asp Glu Leu Phe Ser Gln Val Asp Glu Lys Arg Lys Lys
210 215 220

Arg Asp Ile Pro Asp Tyr Leu Cys Gly Lys Ile Ser Phe Glu Leu Met
225 230 235 240

Arg Glu Pro Cys Ile Thr Pro Ser Gly Ile Thr Tyr Asp Arg Lys Asp
245 250 255

Ile Glu Glu His Leu Gln Arg Val Gly His Phe Asp Pro Val Thr Arg
260 265 270

Ser Pro Leu Thr Gln Glu Gln Leu Ile Pro Asn Leu Ala Met Lys Glu
275 280 285

Val Ile Asp Ala Phe Ile Ser Glu Asn Gly Trp Val Glu Asp Tyr
290 295 300

<210> SEQ ID NO 62
<211> LENGTH: 2166
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: Transgelin

<400> SEQUENCE: 62

cttttggaag tctcttcaga caaacactgg agagaaggca cagcacccta gggtgacatc 60

agtggacagg tcagtttcct gttcctgttc ccagccaccc ctgtctgtct gcccagggac 120

ctacctgcct ggcccactcc tgctgccacc cctctccatg agtgggactc ctgagcagtg 180

ccaggcccag gccctcaggg tggcagttgc tggatggggc caggcttccc attccctggc 240

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aggcagactc ttggctctgg aagatccctg ggggactctg acttcttggt tactcaggct 300

actccgtaag ggtagcctta cgagagcgta agggcatgag gggaagtcga tgggtagacc 360

aggtgtctct gatttctctg aggaaatgca gcttctttgc caagaactct tgggctttgg 420

tggcttcgtc gcccttgcct ggggtgggcg agttttagtc tcttcagctg cagaccagaa 480

gctgagctga cctgaaaaga gctggagttc tggagttcgg gcttccaggc cagggcctgc 540

ttctgggtga gagtgggagg taatggttgc ttgaggcaga ggcagtaaac cctcacaccc 600

gcagctggac aaaggcacca gctggctgta ttctgtgggg gcaatgtggc ttctggaagg 660

tctctccagt ggctctgtag ctggtcttgg caaagaatgt tctagaccaa gggttgtcaa 720

actacagctc atgggccaaa tccagcctgc tgtctgcttt tgtaagtaaa gttttattgg 780

aacaaagcca tgttcatttg ttacatattg tctatggctg ctttcacact gcaacagcag 840

agaccatgtg tcctgcaaag cttaaaatat gttctgtctt atcctttaca aaaacagttt 900

actgacccct gctctagacc tccaaagacc tggtatcctc tttcctggtg ttcaagttca 960

gagaggtgtc tggagtaggg gctgagcctg gcctggatgg gcagtgctgt gacaagtgtc 1020

taggatggcc gggtatcctg cacagagcta gaaggctgcc tggcacgggt gaaagcagag 1080

ctgctccctg accctctgcc cctccctcct ccaccctggc ctgctttagc tttccccaga 1140

catggccaac aagggtcctt cctatggcat gagccgcgaa gtgcagtcca aaatcgagaa 1200

gaagtatgac gaggagctgg aggagcggct ggtggagtgg atcatagtgc agtgtggccc 1260

tgatgtgggc cgcccagacc gtgggcgctt gggcttccag gtctggctga agaatggcgt 1320

gattctgagc aagctggtga acagcctgta ccctgatggc tccaagccgg tgaaggtgcc 1380

cgagaaccca ccctccatgg tcttcaagca gatggagcag gtggctcagt tcctgaaggc 1440

ggctgaggac tatggggtca tcaagactga catgttccag actgttgacc tctttgaagg 1500

caaagacatg gcagcagtgc agaggaccct gatggctttg ggcagcttgg cagtgaccaa 1560

gaatgatggg cactaccgtg gagatcccaa ctggtttatg aagaaagcgc aggagcataa 1620

gagggaattc acagagagcc agctgcagga gggaaagcat gtcattggcc ttcagatggg 1680

cagcaacaga ggggcctccc aggccggcat gacaggctac ggacgacctc ggcagatcat 1740

cagttagagc ggagagggct agccctgagc ccggccctcc cccagctcct tggctgcagc 1800

catcccgctt agcctgcctc acccacaccc gtgtggtacc ttcagccctg gccaagcttt 1860

gaggctctgt cactgagcaa tggtaactgc acctgggcag ctcctccctg tgcccccagc 1920

ctcagcccaa cttcttaccc gaaagcatca ctgccttggc ccctccctcc cggctgcccc 1980

catcacctct actgtctcct ccctgggcta agcaggggag aagcgggctg ggggtagcct 2040

ggatgtgggc caagtccact gtcctccttg gcggcaaaag cccattgaag aagaaccagc 2100

ccagcctgcc ccctatcttg tcctggaata tttttggggt tggaactcaa aaaaaaaaaa 2160

aaaaaa 2166

<210> SEQ ID NO 63
<211> LENGTH: 939
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: ZCWCC3

<400> SEQUENCE: 63

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```
Met Ala Ala Gln Pro Pro Arg Gly Ile Arg Leu Ser Ala Leu Cys Pro
1               5                   10                  15

Lys Phe Leu His Thr Asn Ser Thr Ser His Thr Trp Pro Phe Ser Ala
20                  25                  30

Val Ala Glu Leu Ile Asp Asn Ala Tyr Asp Pro Asp Val Asn Ala Lys
35                  40                  45

Gln Ile Trp Ile Asp Lys Thr Val Ile Asn Asp His Ile Cys Leu Thr
50                  55                  60

Phe Thr Asp Asn Gly Asn Gly Met Thr Ser Asp Lys Leu His Lys Met
65                  70                  75                  80

Leu Ser Phe Gly Phe Ser Asp Lys Val Thr Met Asn Gly His Val Pro
85                  90                  95

Val Gly Leu Tyr Gly Asn Gly Phe Lys Ser Gly Ser Met Arg Leu Gly
100                 105                 110

Lys Asp Ala Ile Val Phe Thr Lys Asn Gly Glu Ser Met Ser Val Gly
115                 120                 125

Leu Leu Ser Gln Thr Tyr Leu Glu Val Ile Lys Ala Glu His Val Val
130                 135                 140

Val Pro Ile Val Ala Phe Asn Lys His Arg Gln Met Ile Asn Leu Ala
145                 150                 155                 160

Glu Ser Lys Ala Ser Leu Ala Ala Ile Leu Glu His Ser Leu Phe Ser
165                 170                 175

Thr Glu Gln Lys Leu Leu Ala Glu Leu Asp Ala Ile Ile Gly Lys Lys
180                 185                 190

Gly Thr Arg Ile Ile Ile Trp Asn Leu Arg Ser Tyr Lys Asn Ala Thr
195                 200                 205

Glu Phe Asp Phe Glu Lys Asp Lys Tyr Asp Ile Arg Ile Pro Glu Asp
210                 215                 220

Leu Asp Glu Ile Thr Gly Lys Lys Gly Tyr Lys Lys Gln Glu Arg Met
225                 230                 235                 240

Asp Gln Ile Ala Pro Glu Ser Asp Tyr Ser Leu Arg Ala Tyr Cys Ser
245                 250                 255

Ile Leu Tyr Leu Lys Pro Arg Met Gln Ile Ile Leu Arg Gly Gln Lys
260                 265                 270

Val Lys Thr Gln Leu Val Ser Lys Ser Leu Ala Tyr Ile Glu Arg Asp
275                 280                 285

Val Tyr Arg Pro Lys Phe Leu Ser Lys Thr Val Arg Ile Thr Phe Gly
290                 295                 300

Phe Asn Cys Arg Asn Lys Asp His Tyr Gly Ile Met Met Tyr His Arg
305                 310                 315                 320

Asn Arg Leu Ile Lys Ala Tyr Glu Lys Val Gly Cys Gln Leu Arg Ala
325                 330                 335

Asn Asn Met Gly Val Gly Val Val Gly Ile Ile Glu Cys Asn Phe Leu
340                 345                 350

Lys Pro Thr His Asn Lys Gln Asp Phe Asp Tyr Thr Asn Glu Tyr Arg
355                 360                 365

Leu Thr Ile Thr Ala Leu Gly Glu Lys Leu Asn Asp Tyr Trp Asn Glu
370                 375                 380

Met Lys Val Lys Lys Asn Thr Glu Tyr Pro Leu Asn Leu Pro Val Glu
385                 390                 395                 400
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Asp Ile Gln Lys Arg Pro Asp Gln Thr Trp Val Gln Cys Asp Ala Cys
405                 410                 415

Leu Lys Trp Arg Lys Leu Pro Asp Gly Met Asp Gln Leu Pro Glu Lys
420                 425                 430

Trp Tyr Cys Ser Asn Asn Pro Asp Pro Gln Phe Arg Asn Cys Glu Val
435                 440                 445

Pro Glu Glu Pro Glu Asp Glu Asp Leu Val His Pro Thr Tyr Glu Lys
450                 455                 460

Thr Tyr Lys Lys Thr Asn Lys Glu Lys Phe Arg Ile Arg Gln Pro Glu
465                 470                 475                 480

Met Ile Pro Arg Ile Asn Ala Glu Leu Leu Phe Arg Pro Thr Ala Leu
485                 490                 495

Ser Thr Pro Ser Phe Ser Ser Pro Lys Glu Ser Val Pro Arg Arg His
500                 505                 510

Leu Ser Glu Gly Thr Asn Ser Tyr Ala Thr Arg Leu Leu Asn Asn His
515                 520                 525

Gln Val Pro Pro Gln Ser Glu Pro Glu Ser Asn Ser Leu Lys Arg Arg
530                 535                 540

Leu Ser Thr Arg Ser Ser Ile Leu Asn Ala Lys Asn Arg Arg Leu Ser
545                 550                 555                 560

Ser Gln Phe Glu Asn Ser Val Tyr Lys Gly Asp Asp Asp Glu Asp
565                 570                 575

Val Ile Ile Leu Glu Glu Asn Ser Thr Pro Lys Pro Ala Val Asp His
580                 585                 590

Asp Ile Asp Met Lys Ser Glu Gln Ser His Val Glu Gln Gly Gly Val
595                 600                 605

Gln Val Glu Phe Val Gly Asp Ser Glu Pro Cys Gly Gln Thr Gly Ser
610                 615                 620

Thr Ser Thr Ser Ser Ser Arg Cys Asp Gln Gly Asn Thr Ala Ala Thr
625                 630                 635                 640

Gln Thr Glu Val Pro Ser Leu Val Val Lys Lys Glu Glu Thr Val Glu
645                 650                 655

Asp Glu Ile Asp Val Arg Asn Asp Ala Val Ile Leu Pro Ser Cys Val
660                 665                 670

Glu Ala Glu Ala Lys Ile His Glu Thr Gln Glu Thr Thr Asp Lys Ser
675                 680                 685

Ala Asp Asp Ala Gly Cys Gln Leu Gln Glu Leu Arg Asn Gln Leu Leu
690                 695                 700

Leu Val Thr Glu Glu Lys Glu Asn Tyr Lys Arg Gln Cys His Met Phe
705                 710                 715                 720

Thr Asp Gln Ile Lys Val Leu Gln Gln Arg Ile Leu Glu Met Asn Asp
725                 730                 735

Lys Tyr Val Lys Lys Glu Thr Cys His Gln Ser Thr Glu Thr Asp Ala
740                 745                 750

Val Phe Leu Leu Glu Ser Ile Asn Gly Lys Ser Glu Ser Pro Asp His
755                 760                 765

Met Val Ser Gln Tyr Gln Gln Ala Leu Glu Glu Ile Glu Arg Leu Lys
770                 775                 780

Lys Gln Cys Ser Ala Leu Gln His Val Lys Ala Glu Cys Ser Gln Cys
785                 790                 795                 800

Ser Asn Asn Glu Ser Lys Ser Glu Met Asp Glu Met Ala Val Gln Leu
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805                 810                 815

Asp Asp Val Phe Arg Gln Leu Asp Lys Cys Ser Ile Glu Arg Asp Gln
820                 825                 830

Tyr Lys Ser Glu Val Glu Leu Leu Glu Met Glu Lys Ser Gln Ile Arg
835                 840                 845

Ser Gln Cys Glu Glu Leu Lys Thr Glu Val Glu Gln Leu Lys Ser Thr
850                 855                 860

Asn Gln Gln Thr Ala Thr Asp Val Ser Thr Ser Ser Asn Ile Glu Glu
865                 870                 875                 880

Ser Val Asn His Met Asp Gly Glu Ser Leu Lys Leu Arg Ser Leu Arg
885                 890                 895

Val Asn Val Gly Gln Leu Leu Ala Met Ile Val Pro Asp Leu Asp Leu
900                 905                 910

Gln Gln Val Asn Tyr Asp Val Asp Val Val Asp Glu Ile Leu Gly Gln
915                 920                 925

Val Val Glu Gln Met Ser Glu Ile Ser Ser Thr
930                 935


<210> SEQ ID NO 64
<211> LENGTH: 424
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: ACAA1

<400> SEQUENCE: 64

Met Gln Arg Leu Gln Val Val Leu Gly His Leu Arg Gly Pro Ala Asp
1               5                   10                  15

Ser Gly Trp Met Pro Gln Ala Ala Pro Cys Leu Ser Gly Ala Pro Gln
20                  25                  30

Ala Ser Ala Ala Asp Val Val Val Val His Gly Arg Arg Thr Ala Ile
35                  40                  45

Cys Arg Ala Gly Arg Gly Gly Phe Lys Asp Thr Thr Pro Asp Glu Leu
50                  55                  60

Leu Ser Ala Val Met Thr Ala Val Leu Lys Asp Val Asn Leu Arg Pro
65                  70                  75                  80

Glu Gln Leu Gly Asp Ile Cys Val Gly Asn Val Leu Gln Pro Gly Ala
85                  90                  95

Gly Ala Ile Met Ala Arg Ile Ala Gln Phe Leu Ser Asp Ile Pro Glu
100                 105                 110

Thr Val Pro Leu Ser Thr Val Asn Arg Gln Cys Ser Ser Gly Leu Gln
115                 120                 125

Ala Val Ala Ser Ile Ala Gly Gly Ile Arg Asn Gly Ser Tyr Asp Ile
130                 135                 140

Gly Met Ala Cys Gly Val Glu Ser Met Ser Leu Ala Asp Arg Gly Asn
145                 150                 155                 160

Pro Gly Asn Ile Thr Ser Arg Leu Met Glu Lys Glu Lys Ala Arg Asp
165                 170                 175

Cys Leu Ile Pro Met Gly Ile Thr Ser Glu Asn Val Ala Glu Arg Phe
180                 185                 190

Gly Ile Ser Arg Glu Lys Gln Asp Thr Phe Ala Leu Ala Ser Gln Gln
195                 200                 205

Lys Ala Ala Arg Ala Gln Ser Lys Gly Cys Phe Gln Ala Glu Ile Val
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210                 215                 220

Pro Val Thr Thr Thr Val His Asp Asp Lys Gly Thr Lys Arg Ser Ile
225                 230                 235                 240

Thr Val Thr Gln Asp Glu Gly Ile Arg Pro Ser Thr Thr Met Glu Gly
245                 250                 255

Leu Ala Lys Leu Lys Pro Ala Phe Lys Lys Asp Gly Ser Thr Thr Ala
260                 265                 270

Gly Asn Ser Ser Gln Val Ser Asp Gly Ala Ala Ala Ile Leu Leu Ala
275                 280                 285

Arg Arg Ser Lys Ala Glu Glu Leu Gly Leu Pro Ile Leu Gly Val Leu
290                 295                 300

Arg Ser Tyr Ala Val Val Gly Val Pro Pro Asp Ile Met Gly Ile Gly
305                 310                 315                 320

Pro Ala Tyr Ala Ile Pro Val Ala Leu Gln Lys Ala Gly Leu Thr Val
325                 330                 335

Ser Asp Val Asp Ile Phe Glu Ile Asn Glu Ala Phe Ala Ser Gln Ala
340                 345                 350

Ala Tyr Cys Val Glu Lys Leu Arg Leu Pro Pro Glu Lys Val Asn Pro
355                 360                 365

Leu Gly Gly Ala Val Ala Leu Gly His Pro Leu Gly Cys Thr Gly Ala
370                 375                 380

Arg Gln Val Ile Thr Leu Leu Asn Glu Leu Lys Arg Arg Gly Lys Arg
385                 390                 395                 400

Ala Tyr Gly Val Val Ser Met Cys Ile Gly Thr Gly Met Gly Ala Ala
405                 410                 415

Ala Val Phe Glu Tyr Pro Gly Asn
420


<210> SEQ ID NO 65
<211> LENGTH: 884
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: Actinin

<400> SEQUENCE: 65

Met Gly Asp Tyr Met Ala Gln Glu Asp Asp Trp Asp Arg Asp Leu Leu
1               5                  10                  15

Leu Asp Pro Ala Trp Glu Lys Gln Gln Arg Lys Thr Phe Thr Ala Trp
20                  25                  30

Cys Asn Ser His Leu Arg Lys Ala Gly Thr Gln Ile Glu Asn Ile Asp
35                  40                  45

Glu Asp Phe Arg Asp Gly Leu Lys Leu Met Leu Leu Leu Glu Val Ile
50                  55                  60

Ser Gly Glu Arg Leu Pro Lys Pro Glu Arg Gly Lys Met Arg Val His
65                  70                  75                  80

Lys Ile Asn Asn Val Asn Lys Ala Leu Asp Phe Ile Ala Ser Lys Gly
85                  90                  95

Val Lys Leu Val Ser Ile Gly Ala Glu Glu Ile Val Asp Gly Asn Ala
100                 105                 110

Lys Met Thr Leu Gly Met Ile Trp Thr Ile Ile Leu Arg Phe Ala Ile
115                 120                 125

Gln Asp Ile Ser Val Glu Glu Thr Ser Ala Lys Glu Gly Leu Leu Leu
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130                 135                 140

Trp Cys Gln Arg Lys Thr Ala Pro Tyr Lys Asn Val Asn Val Gln Asn
145                 150                 155                 160

Phe His Ile Ser Trp Lys Asp Gly Leu Ala Phe Asn Ala Leu Ile His
165                 170                 175

Arg His Arg Pro Glu Leu Ile Glu Tyr Asp Lys Leu Arg Lys Asp Asp
180                 185                 190

Pro Val Thr Asn Leu Asn Asn Ala Phe Glu Val Ala Glu Lys Tyr Leu
195                 200                 205

Asp Ile Pro Lys Met Leu Asp Ala Glu Asp Ile Val Asn Thr Ala Arg
210                 215                 220

Pro Asp Glu Lys Ala Ile Met Thr Tyr Val Ser Ser Phe Tyr His Ala
225                 230                 235                 240

Phe Ser Gly Ala Gln Lys Ala Glu Thr Ala Ala Asn Arg Ile Cys Lys
245                 250                 255

Val Leu Ala Val Asn Gln Glu Asn Glu His Leu Met Glu Asp Tyr Glu
260                 265                 270

Lys Leu Ala Ser Asp Leu Leu Glu Trp Ile Arg Arg Thr Ile Pro Trp
275                 280                 285

Leu Glu Asp Arg Val Pro Gln Lys Thr Ile Gln Glu Met Gln Gln Lys
290                 295                 300

Leu Glu Asp Phe Arg Asp Tyr Arg Arg Val His Lys Pro Pro Lys Val
305                 310                 315                 320

Gln Glu Lys Cys Gln Leu Glu Ile Asn Phe Asn Thr Leu Gln Thr Lys
325                 330                 335

Leu Arg Leu Ser Asn Arg Pro Ala Phe Met Pro Ser Glu Gly Lys Met
340                 345                 350

Val Ser Asp Ile Asn Asn Gly Trp Gln His Leu Glu Gln Ala Glu Lys
355                 360                 365

Gly Tyr Glu Glu Trp Leu Leu Asn Glu Ile Arg Arg Leu Glu Arg Leu
370                 375                 380

Asp His Leu Ala Glu Lys Phe Arg Gln Lys Ala Ser Ile His Glu Ala
385                 390                 395                 400

Trp Thr Asp Gly Lys Glu Ala Met Leu Lys His Arg Asp Tyr Glu Thr
405                 410                 415

Ala Thr Leu Ser Asp Ile Lys Ala Leu Ile Arg Lys His Glu Ala Phe
420                 425                 430

Glu Ser Asp Leu Ala Ala His Gln Asp Arg Val Glu Gln Ile Ala Ala
435                 440                 445

Ile Ala Gln Glu Leu Asn Glu Leu Asp Tyr Tyr Asp Ser His Asn Val
450                 455                 460

Asn Thr Arg Cys Gln Lys Ile Cys Asp Gln Trp Asp Ala Leu Gly Ser
465                 470                 475                 480

Leu Thr His Ser Arg Arg Glu Ala Leu Glu Lys Thr Glu Lys Gln Leu
485                 490                 495

Glu Ala Ile Asp Gln Leu His Leu Glu Tyr Ala Lys Arg Ala Ala Pro
500                 505                 510

Phe Asn Asn Trp Met Glu Ser Ala Met Glu Asp Leu Gln Asp Met Phe
515                 520                 525

Ile Val His Thr Ile Glu Glu Ile Glu Gly Leu Ile Ser Ala His Asp
530                 535                 540
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Gln Phe Lys Ser Thr Leu Pro Asp Ala Asp Arg Glu Arg Glu Ala Ile
545                 550                 555                 560

Leu Ala Ile His Lys Glu Ala Gln Arg Ile Ala Glu Ser Asn His Ile
565                 570                 575

Lys Leu Ser Gly Ser Asn Pro Tyr Thr Thr Val Thr Pro Gln Ile Ile
580                 585                 590

Asn Ser Lys Trp Glu Lys Val Gln Gln Leu Val Pro Lys Arg Asp His
595                 600                 605

Ala Leu Leu Glu Glu Gln Ser Lys Gln Gln Ser Asn Glu His Leu Arg
610                 615                 620

Arg Gln Phe Ala Ser Gln Ala Asn Val Val Gly Pro Trp Ile Gln Thr
625                 630                 635                 640

Lys Met Glu Glu Ile Gly Arg Ile Ser Ile Glu Met Asn Gly Thr Leu
645                 650                 655

Glu Asp Gln Leu Ser His Leu Lys Gln Tyr Glu Arg Ser Ile Val Asp
660                 665                 670

Tyr Lys Pro Asn Leu Asp Leu Leu Glu Gln Gln His Gln Leu Ile Gln
675                 680                 685

Glu Ala Leu Ile Phe Asp Asn Lys His Thr Asn Tyr Thr Met Glu His
690                 695                 700

Ile Arg Val Gly Trp Glu Gln Leu Leu Thr Thr Ile Ala Arg Thr Ile
705                 710                 715                 720

Asn Glu Val Glu Asn Gln Ile Leu Thr Arg Asp Ala Lys Gly Ile Ser
725                 730                 735

Gln Glu Gln Met Gln Glu Phe Arg Ala Ser Phe Asn His Phe Asp Lys
740                 745                 750

Asp His Gly Gly Ala Leu Gly Pro Glu Glu Phe Lys Ala Cys Leu Ile
755                 760                 765

Ser Leu Gly Tyr Asp Val Glu Asn Asp Arg Gln Gly Glu Ala Glu Phe
770                 775                 780

Asn Arg Ile Met Ser Leu Val Asp Pro Asn His Ser Gly Leu Val Thr
785                 790                 795                 800

Phe Gln Ala Phe Ile Asp Phe Met Ser Arg Glu Thr Thr Asp Thr Asp
805                 810                 815

Thr Ala Asp Gln Val Ile Ala Ser Phe Lys Val Leu Ala Gly Asp Lys
820                 825                 830

Asn Phe Ile Thr Ala Glu Glu Leu Arg Arg Glu Leu Pro Pro Asp Gln
835                 840                 845

Ala Glu Tyr Cys Ile Ala Arg Met Ala Pro Tyr Gln Gly Pro Asp Ala
850                 855                 860

Val Pro Gly Ala Leu Asp Tyr Lys Ser Phe Ser Thr Ala Leu Tyr Gly
865                 870                 875                 880

Glu Ser Asp Leu


<210> SEQ ID NO 66
<211> LENGTH: 1024
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: NFX2

<400> SEQUENCE: 66
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```
Met Ala Glu Ala Pro Pro Val Ser Gly Thr Phe Lys Phe Asn Thr Asp
1               5                  10                  15

Ala Ala Glu Phe Ile Pro Gln Glu Lys Lys Asn Ser Gly Leu Asn Cys
20                  25                  30

Gly Thr Gln Arg Arg Leu Asp Ser Asn Arg Ile Gly Arg Arg Asn Tyr
35                  40                  45

Ser Ser Pro Pro Pro Cys His Leu Ser Arg Gln Val Pro Tyr Asp Glu
50                  55                  60

Ile Ser Ala Val His Gln His Ser Tyr His Pro Ser Gly Ser Lys Pro
65                  70                  75                  80

Lys Ser Gln Gln Thr Ser Phe Gln Ser Ser Pro Cys Asn Lys Ser Pro
85                  90                  95

Lys Ser His Gly Leu Gln Asn Gln Pro Trp Gln Lys Leu Arg Asn Glu
100                 105                 110

Lys His His Ile Arg Val Lys Lys Ala Gln Ser Leu Ala Glu Gln Thr
115                 120                 125

Ser Asp Thr Ala Gly Leu Glu Ser Ser Thr Arg Ser Glu Ser Gly Thr
130                 135                 140

Asp Leu Arg Glu His Ser Pro Ser Glu Ser Glu Lys Glu Val Val Gly
145                 150                 155                 160

Ala Asp Pro Arg Gly Ala Lys Pro Lys Lys Ala Thr Gln Phe Val Tyr
165                 170                 175

Ser Tyr Gly Arg Gly Pro Lys Val Lys Gly Lys Leu Lys Cys Glu Trp
180                 185                 190

Ser Asn Arg Thr Thr Pro Lys Pro Glu Asp Ala Gly Pro Glu Ser Thr
195                 200                 205

Lys Pro Val Gly Val Phe His Pro Asp Ser Ser Glu Ala Ser Ser Arg
210                 215                 220

Lys Gly Val Leu Asp Gly Tyr Gly Ala Arg Arg Asn Glu Gln Arg Arg
225                 230                 235                 240

Tyr Pro Gln Lys Arg Pro Pro Trp Glu Val Glu Gly Ala Arg Pro Arg
245                 250                 255

Pro Gly Arg Asn Pro Pro Lys Gln Glu Gly His Arg His Thr Asn Ala
260                 265                 270

Gly His Arg Asn Asn Met Gly Pro Ile Pro Lys Asp Asp Leu Asn Glu
275                 280                 285

Arg Pro Ala Lys Ser Thr Cys Asp Ser Glu Asn Leu Ala Val Ile Asn
290                 295                 300

Lys Ser Ser Arg Arg Val Asp Gln Glu Lys Cys Thr Val Arg Arg Gln
305                 310                 315                 320

Asp Pro Gln Val Val Ser Pro Phe Ser Arg Gly Lys Gln Asn His Val
325                 330                 335

Leu Lys Asn Val Glu Thr His Thr Gly Ser Leu Ile Glu Gln Leu Thr
340                 345                 350

Thr Glu Lys Tyr Glu Cys Met Val Cys Cys Glu Leu Val Arg Val Thr
355                 360                 365

Ala Pro Val Trp Ser Cys Gln Ser Cys Tyr His Val Phe His Leu Asn
370                 375                 380

Cys Ile Lys Lys Trp Ala Arg Ser Pro Ala Ser Gln Ala Asp Gly Gln
385                 390                 395                 400

Ser Gly Trp Arg Cys Pro Ala Cys Gln Asn Val Ser Ala His Val Pro
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-continued

405 410 415

Asn Thr Tyr Thr Cys Phe Cys Gly Lys Val Lys Asn Pro Glu Trp Ser
420 425 430

Arg Asn Glu Ile Pro His Ser Cys Gly Glu Val Cys Arg Lys Lys Gln
435 440 445

Pro Gly Gln Asp Cys Pro His Ser Cys Asn Leu Leu Cys His Pro Gly
450 455 460

Pro Cys Pro Pro Cys Pro Ala Phe Met Thr Lys Thr Cys Glu Cys Gly
465 470 475 480

Arg Thr Arg His Thr Val Arg Cys Gly Gln Ala Val Ser Val His Cys
485 490 495

Ser Asn Pro Cys Glu Asn Ile Leu Asn Cys Gly Gln His Gln Cys Ala
500 505 510

Glu Leu Cys His Gly Gly Gln Cys Gln Pro Cys Gln Ile Ile Leu Asn
515 520 525

Gln Val Cys Tyr Cys Gly Ser Thr Ser Arg Asp Val Leu Cys Gly Thr
530 535 540

Asp Val Gly Lys Ser Asp Gly Phe Gly Asp Phe Ser Cys Leu Lys Ile
545 550 555 560

Cys Gly Lys Asp Leu Lys Cys Gly Asn His Thr Cys Ser Gln Val Cys
565 570 575

His Pro Gln Pro Cys Gln Gln Cys Pro Arg Leu Pro Gln Leu Val Arg
580 585 590

Cys Cys Pro Cys Gly Gln Thr Pro Leu Ser Gln Leu Leu Glu Leu Gly
595 600 605

Ser Ser Ser Arg Lys Thr Cys Met Asp Pro Val Pro Ser Cys Gly Lys
610 615 620

Val Cys Gly Lys Pro Leu Pro Cys Gly Ser Leu Asp Phe Ile His Thr
625 630 635 640

Cys Glu Lys Leu Cys His Glu Gly Asp Cys Gly Pro Cys Ser Arg Thr
645 650 655

Ser Val Ile Ser Cys Arg Cys Ser Phe Arg Thr Lys Glu Leu Pro Cys
660 665 670

Thr Ser Leu Lys Ser Glu Asp Ala Thr Phe Met Cys Asp Lys Arg Cys
675 680 685

Asn Lys Lys Arg Leu Cys Gly Arg His Lys Cys Asn Glu Ile Cys Cys
690 695 700

Val Asp Lys Glu His Lys Cys Pro Leu Ile Cys Gly Arg Lys Leu Arg
705 710 715 720

Cys Gly Leu His Arg Cys Glu Glu Pro Cys His Arg Gly Asn Cys Gln
725 730 735

Thr Cys Trp Gln Ala Ser Phe Asp Glu Leu Thr Cys His Cys Gly Ala
740 745 750

Ser Val Ile Tyr Pro Pro Val Pro Cys Gly Thr Arg Pro Pro Glu Cys
755 760 765

Thr Gln Thr Cys Ala Arg Val His Glu Cys Asp His Pro Val Tyr His
770 775 780

Ser Cys His Ser Glu Glu Lys Cys Pro Pro Cys Thr Phe Leu Thr Gln
785 790 795 800

Lys Trp Cys Met Gly Lys His Glu Phe Arg Ser Asn Ile Pro Cys His
805 810 815

-continued

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Leu Val Asp Ile Ser Cys Gly Leu Pro Cys Ser Ala Thr Leu Pro Cys
820                 825                 830

Gly Met His Lys Cys Gln Arg Leu Cys His Lys Gly Glu Cys Leu Val
835                 840                 845

Asp Glu Pro Cys Lys Gln Pro Cys Thr Thr Pro Arg Ala Asp Cys Gly
850                 855                 860

His Pro Cys Met Ala Pro Cys His Thr Ser Ser Pro Cys Pro Val Thr
865                 870                 875                 880

Ala Cys Lys Ala Lys Val Glu Leu Gln Cys Glu Cys Gly Arg Arg Lys
885                 890                 895

Glu Met Val Ile Cys Ser Glu Ala Ser Ser Thr Tyr Gln Arg Ile Ala
900                 905                 910

Ala Ile Ser Met Ala Ser Lys Ile Thr Asp Met Gln Leu Gly Gly Ser
915                 920                 925

Val Glu Ile Ser Lys Leu Ile Thr Lys Lys Glu Val His Gln Ala Arg
930                 935                 940

Leu Glu Cys Asp Glu Glu Cys Ser Ala Leu Glu Arg Lys Lys Arg Leu
945                 950                 955                 960

Ala Glu Ala Phe His Ile Ser Glu Asp Ser Asp Pro Phe Asn Ile Arg
965                 970                 975

Ser Ser Gly Ser Lys Phe Ser Asp Ser Leu Lys Glu Asp Ala Arg Lys
980                 985                 990

Asp Leu Lys Phe Val Ser Asp Val  Glu Lys Glu Met Glu  Thr Leu Val
995                 1000                1005

Glu Ala  Val Asn Lys Val Glu  Val Glu Thr Ser His  Trp Thr Phe
1010                1015                1020

Leu
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<210> SEQ ID NO 67
<211> LENGTH: 1799
<212> TYPE: DNA
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: MAD-CT-2

<400> SEQUENCE: 67

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tgatgtggcc ccaaccccac ctccctcccc accccatgat gtcagaaaaa accagacaga    120

acaaattggc tgaggccaag aaaaagttta cagactatcg tcagtggaac attgctggtg    180

ttggtaccgg agcaactgac accaaaaaga agaaaataaa tcatggcact aaccctgaga    240

caaccacttc ggggggctgc cactcgcctg aggatacaca acagaaccga gcgcagctga    300

aagaagaaaa gaagatgcgg gagcaggaag atgtggagac aggaggagag gctgcaggag    360

caggagaagc agatgtggga gcaggaggag aagatgcggg atcaggagca gaagatgtgg    420

gaccaggagg agaggatgtg ggagcaggac gagaggctgc ggcagaagga ggagagaatg    480

cgggagcaga agatgtggca gcaggtggag aagatgcggg aggagaagaa gacgcaggag    540

caggagaaga agacatggga ccaggagaag atgcgagagg aggagagcat gcgggagcgg    600

gagaagaaga tgcgggagga ggaggagatg atgcgggagc aggaggagaa gatgcaggag    660

caggaagaaa agatgcagga gcaggaggag gagatgtggg agcaggagga gaagatgtgg    720
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gagcaggaag agaagatgtg ggagcagcag aggctaccgg aacagaagga gaggctgtgg 780

gaacacgaga agatgcagga gcaggagaag atatgggagc aggaggagaa gatgcgggac 840

caggaggaga agatgcgggg ccaggaggag aagatgcggg ggcaggagga gaagatgcgg 900

gggcaggagg agaagatgtg ggggcaggag gagaagatgt gggggcagga ggagaagatg 960

tggggccagg aggagaagat gtgggggcag gaggagaaga tgtggggcca ggaggagaag 1020

atgcgggggg caggaggaga agatgcgggg gcaggaggag aagatgcggg gccaggagga 1080

gaagatgcgg gggcaggagg aggagatgcg gggggcagga ggagaagatg cgggggggcag 1140

gaggagaaga tgcgggggac aggaggagaa gatgcggggg ccaggaggag aagatgcggg 1200

agcaggagga gaagatgcgg gagcaggagg agaagatgca gggccaggag gagaagatgc 1260

gggagcagga ggagaagatg cggggccagg aggagaagat gcgggagcag gaggagaaga 1320

tgcggggcca ggaggagaag atgtggggcc aggaggagaa gatgtggggg caggaggaga 1380

agatgtgggg gcaggaggag atgatgcgag agaaggagga gaggatatga gatcagaaag 1440

agaagatgca ggagaggctg ccagagcacg aggagcggtg ctcagagccc tgcctccctc 1500

cctccaaagt tctttgtaat atgagccaca ctggcagtgt ggagcctgca ggaggagagg 1560

ctggggaggg ttctccgcag gacaacccca ctgcacagga gatcatgcag ctgttttgtg 1620

ggatgaagaa cgcccagcag tgcccaggat taggcagtac ctcctgcatc ccattcttct 1680

accgaggaga caagagaaag atgaagatca tcaatatcta aaagttggca ctgtcaacaa 1740

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<210> SEQ ID NO 68
<211> LENGTH: 102
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: MAD-CT-1

<400> SEQUENCE: 68

Met Val Arg Tyr Arg Val Arg Ser Leu Ser Glu Arg Ser His Glu Val
1 5 10 15

Tyr Arg Gln Gln Leu His Gly Gln Glu Gln Gly His His Gly Gln Glu
20 25 30

Glu Gln Gly Leu Ser Pro Glu His Val Glu Val Tyr Glu Arg Thr His
35 40 45

Gly Gln Ser His Tyr Arg Arg Arg His Cys Ser Arg Arg Arg Leu His
50 55 60

Arg Ile His Arg Arg Gln His Arg Ser Cys Arg Arg Arg Lys Arg Arg
65 70 75 80

Ser Cys Arg His Arg Arg Arg His Arg Arg Gly Cys Arg Thr Arg Lys
85 90 95

Arg Thr Cys Arg Arg His
100

<210> SEQ ID NO 69
<211> LENGTH: 146
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: PAGE-1

<400> SEQUENCE: 69

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Met Gly Phe Leu Arg Arg Leu Ile Tyr Arg Arg Arg Pro Met Ile Tyr
1 5 10 15

Val Glu Ser Ser Glu Glu Ser Ser Asp Glu Gln Pro Asp Glu Val Glu
20 25 30

Ser Pro Thr Gln Ser Gln Asp Ser Thr Pro Ala Glu Glu Arg Glu Asp
35 40 45

Glu Gly Ala Ser Ala Ala Gln Gly Gln Glu Pro Glu Ala Asp Ser Gln
50 55 60

Glu Leu Val Gln Pro Lys Thr Gly Cys Glu Pro Gly Asp Gly Pro Asp
65 70 75 80

Thr Lys Arg Val Cys Leu Arg Asn Glu Glu Gln Met Lys Leu Pro Ala
85 90 95

Glu Gly Pro Glu Pro Glu Ala Asp Ser Gln Glu Gln Val His Pro Lys
100 105 110

Thr Gly Cys Glu Arg Gly Asp Gly Pro Asp Val Gln Glu Leu Gly Leu
115 120 125

Pro Asn Pro Glu Glu Val Lys Thr Pro Glu Glu Asp Glu Gly Gln Ser
130 135 140

Gln Pro
145

<210> SEQ ID NO 70
<211> LENGTH: 180
<212> TYPE: PRT
<213> ORGANISM: Homo sapiens
<220> FEATURE:
<223> OTHER INFORMATION: NY-ESO-1

<400> SEQUENCE: 70

Met Gln Ala Glu Gly Arg Gly Thr Gly Gly Ser Thr Gly Asp Ala Asp
1 5 10 15

Gly Pro Gly Gly Pro Gly Ile Pro Asp Gly Pro Gly Gly Asn Ala Gly
20 25 30

Gly Pro Gly Glu Ala Gly Ala Thr Gly Gly Arg Gly Pro Arg Gly Ala
35 40 45

Gly Ala Ala Arg Ala Ser Gly Pro Gly Gly Gly Ala Pro Arg Gly Pro
50 55 60

His Gly Gly Ala Ala Ser Gly Leu Asn Gly Cys Cys Arg Cys Gly Ala
65 70 75 80

Arg Gly Pro Glu Ser Arg Leu Leu Glu Phe Tyr Leu Ala Met Pro Phe
85 90 95

Ala Thr Pro Met Glu Ala Glu Leu Ala Arg Arg Ser Leu Ala Gln Asp
100 105 110

Ala Pro Pro Leu Pro Val Pro Gly Val Leu Leu Lys Glu Phe Thr Val
115 120 125

Ser Gly Asn Ile Leu Thr Ile Arg Leu Thr Ala Ala Asp His Arg Gln
130 135 140

Leu Gln Leu Ser Ile Ser Ser Cys Leu Gln Gln Leu Ser Leu Leu Met
145 150 155 160

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Trp Ile Thr Gln Cys Phe Leu Pro Val Phe Leu Ala Gln Pro Pro Ser
165                 170                 175

Gly Gln Arg Arg
180
```

1. A method for identifying a human subject as a candidate for further prostate cancer examination comprising the step of:

determining whether the human subject has developed an immune reaction to a prostate cancer antigen selected from SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof, MAD-Pro-30 (SEQ ID NO: 58), MAD-Pro-42 (SEQ ID NO: 61), transgelin encoded by SEQ ID NO: 62 or a conservatively modified variant thereof, ZCWCC3 (SEQ ID NO: 63), ACAA1 (SEQ ID NO: 64), actinin (SEQ ID NO: 65), and NFX2 (SEQ ID NO: 66) wherein the presence of an immune reaction indicates that the human subject is a candidate for further prostate cancer examination.

2. The method of claim **1**, wherein the antigen is selected from SSX-2 encoded by a nucleic acid comprising SEQ ID NO: 60 or a conservatively modified variant thereof, MAD-Pro-30 (SEQ ID NO: 58), MAD-Pro-42 (SEQ ID NO: 61), transgelin (SEQ ID NO: 62), ZCWCC3 (SEQ ID NO: 63), and ACAA1 (SEQ ID NO: 64).

3. The method of claim **1**, wherein the antigen is MAD-Pro-30 (SEQ ID NO: 58).

4. The method of claim **3**, wherein whether the human subject developed an immune reaction to antigens in an antigen panel is determined, and wherein the panel comprises MAD-Pro-30 (SEQ ID NO: 58), SSX-2 encoded by a nucleic acid comprising the nucleotide sequence of SEQ ID NO: 60 or a conservatively modified variant thereof, androgen receptor (AR) ligand binding domain (SEQ ID NO: 59), and MAD-Pro-22 (PSA) (SEQ ID NO: 57).

5. The method of claim **1**, wherein the development of an immune reaction is determined by testing whether a blood sample from the human subject contains an antibody to an antigen.

6. A method for determining whether an immune therapy has elicited a tumor-specific immune response in a prostate cancer patient, the method comprising the steps of:

providing an immune therapy to a prostate cancer patient; and

determining whether the patient developed an immune reaction to a prostate cancer antigen selected from SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof, MAD-Pro-30 (SEQ ID NO: 58), MAD-Pro-42 (SEQ ID NO: 61), transgelin encoded by the nucleotide sequence of SEQ ID NO: 62 or a conservatively modified variant thereof, ZCWCC3 (SEQ ID NO: 63), ACAA1 (SEQ ID NO: 64), actinin (SEQ ID NO: 65), and NFX2 (SEQ NO: 66), wherein the presence of an immune reaction indicates that the therapy has elicited a tumor-specific immune response.

7. The method of claim **6**, wherein the antigen is selected from SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof, MAD-Pro-30 (SEQ ID NO: 58), MAD-Pro-42 (SEQ ID NO: 61), transgelin encoded by SEQ ID NO: 62 or a conservatively modified variant thereof, ZCWCC3 (SEQ ID NO: 63), and ACAA1 (SEQ ID NO: 64).

8. The method of claim **6**, wherein the antigen is MAD-Pro-30 (SEQ ID NO: 58).

9. The method of claim **8**, wherein whether the prostate cancer patient developed an immune reaction to antigens in an antigen panel is determined, and wherein the panel comprises MAD-Pro-30 (SEQ ID NO: 58), SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof, androgen receptor (AR) ligand binding domain (SEQ ID NO: 59), and MAD-Pro-22 (PSA) (SEQ ID NO: 57).

10. The method of claim **6**, wherein the immune therapy is a non-antigen-specific immune therapy.

11. The method of claim **6**, wherein the development of an immune reaction is determined by testing whether a blood sample from the prostate cancer patient contains an antibody to an antigen.

12. (canceled)

13. A method for identifying a human subject as a candidate for further melanoma examination comprising the step of:

determining whether the human subject has developed an immune reaction to a melanoma antigen selected from MAD-CT-2 encoded by SEQ ID NO: 67 or a conservatively modified variant thereof, MAD-CT-1 (SEQ ID NO: 68), and PAGE-1 (SEQ ID NO: 69), wherein the presence of an immune reaction indicates that the human subject is a candidate for further melanoma examination.

14. The method of claim **13**, wherein the antigen is MAD-CT-2.

15. The method of claim **14**, wherein whether the human subject developed an immune reaction to antigens in an antigen panel is determined, and wherein the panel comprises MAD-CT-1 (SEQ ID NO: 68), PAGE-1 (SEQ ID NO: 69), SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof, and NY-ESO-1 (SEQ ID NO: 70).

16. The method of claim **13**, wherein the development of an immune reaction is determined by testing whether a blood sample from the human subject contains an antibody to an antigen.

17. A method for determining whether an immune therapy has elicited a tumor-specific immune response in a melanoma patient, the method comprising the steps of:

providing an immune therapy to a melanoma patient; and

determining whether the patient developed an immune reaction to a melanoma antigen selected from MAD-CT-2 encoded by SEQ ID NO: 67 or a conservatively modified variant thereof, MAD-CT-1 (SEQ ID NO: 68), and PAGE-1 (SEQ ID NO: 69), wherein the presence of an immune reaction indicates that the therapy has elicited a tumor-specific immune response.

18. The method of claim **17**, wherein the antigen is MAD-CT-2 encoded by SEQ ID NO: 67 or a conservatively modified variant thereof.

19. The method of claim **18**, wherein whether the melanoma patient developed an immune reaction to antigens in an antigen panel is determined, and wherein the panel comprises MAD-CT-1 (SEQ ID NO: 68), PAGE-1 (SEQ ID NO: 69), SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof, and NY-ESO-1 (SEQ ID NO: 70).

20. The method of claim **17**, wherein the immune therapy is a non-antigen-specific immune therapy.

21. The method of claim **17**, wherein the development of an immune reaction is determined by testing whether a blood sample from the melanoma patient contains an antibody to an antigen.

22. A method for identifying a human subject as a candidate for further melanoma examination comprising the step of:

determining whether the cells in a region of the subject's skin suspected of being malignant express MAD-CT-2 encoded by SEQ ID NO: 67 or a conservatively modified variant thereof, wherein the expression of MAD-CT-2 indicates that the subject is a candidate for further melanoma examination.

23. A method for identifying candidate compounds for further testing as preventive or therapeutic agents for melanoma, the method comprising the steps of:

providing cells that express MAD-CT-2 encoded by SEQ ID NO: 67 or a conservatively modified variant thereof;

exposing the cells to a test compound;

determining the expression of level of MAD-CT-2 in the exposed cells; and

comparing the expression level in the exposed cells to that of corresponding control cells that are not exposed to the test compound, wherein a lower expression level than that in the control cells indicates that the compound is a candidate for further testing as a preventive or therapeutic agent for melanoma.

24. A kit comprising:

a first polypeptide that comprises MAD-CT-2 encoded by SEQ ID NO: 67 or a conservatively modified variant thereof;

a second polypeptide that comprises SSX-2 encoded by SEQ ID NO: 60 or a conservatively modified variant thereof; and

a third polypeptide that comprises NY-ESO-1 (SEQ ID NO: 70).

* * * * *

专利名称(译)	前列腺癌和黑色素瘤抗原		
公开(公告)号	US20100124755A1	公开(公告)日	2010-05-20
申请号	US12/629686	申请日	2009-12-02
[标]申请(专利权)人(译)	MCNEEL DOUGLAS摹 邓菲EDWARDĴ DUBOVSKY贾森 HOEPPNER LUKE ^ h		
申请(专利权)人(译)	MCNEEL DOUGLAS摹 邓菲EDWARDĴ DUBOVSKY贾森 HOEPPNER LUKE ^ h		
当前申请(专利权)人(译)	MCNEEL DOUGLAS摹 邓菲EDWARDĴ DUBOVSKY贾森 HOEPPNER LUKE ^ h		
[标]发明人	MCNEEL DOUGLAS G DUNPHY EDWARD J DUBOVSKY JASON A HOEPPNER LUKE H		
发明人	MCNEEL, DOUGLAS G. DUNPHY, EDWARD J. DUBOVSKY, JASON A. HOEPPNER, LUKE H.		
IPC分类号	G01N33/53		
CPC分类号	G01N33/5743 G01N33/57488 G01N33/57434 A61P37/00		
优先权	60/890590 2007-02-19 US		
外部链接	Espacenet USPTO		

摘要(译)

公开了用于鉴定人受试者作为进一步前列腺癌或黑素瘤检查的候选者的方法。还公开了用于确定免疫疗法是否已在前列腺癌或黑素瘤患者中引发肿瘤特异性免疫应答的方法。进一步公开了可用于实施上述方法的试剂盒。还公开了鉴定用于进一步测试作为黑素瘤的预防或治疗剂的候选化合物的方法。