

(19) **DANMARK**

(10) **DK/EP 3147373 T3**



(12)

Oversættelse af europæisk patentskrift

Patent- og
Varemærkestyrelsen

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- (51) Int.Cl.: **C 12 Q 1/6886 (2018.01)**
- (45) Oversættelsen bekendtgjort den: **2019-08-12**
- (80) Dato for Den Europæiske Patentmyndigheds bekendtgørelse om meddelelse af patentet: **2019-05-15**
- (86) Europæisk ansøgning nr.: **16195347.6**
- (86) Europæisk indleveringsdag: **2011-07-25**
- (87) Den europæiske ansøgnings publiceringsdag: **2017-03-29**
- (30) Prioritet: **2010-07-27 US 368217 P** **2010-11-16 US 414310 P**
2011-05-12 US 201161485536 P
- (62) Stamansøgningsnr: **15152517.7**
- (84) Designerede stater: **AL AT BE BG CH CY CZ DE DK EE ES FI FR GB GR HR HU IE IS IT LI LT LU LV MC MK MT NL NO PL PT RO RS SE SI SK SM TR**
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- (54) Benævnelse: **FREMANGSMÅDE TIL ANVENDELSE AF GENEKSPRESSION TIL AT BESTEMME PROGNOSEN FOR PROSTATACANCER**
- (56) Fremdragne publikationer:
WO-A2-2004/108896
WO-A2-2005/008213
WO-A2-2008/067065
WO-A2-2010/056993
PERNER S ET AL: "TMPRSS2-ETS gene fusion in prostate cancer", DER UROLOGE, AUSGABE A ; ZEITSCHRIFT FÜR KLINISCHE UND PRAKTISCHE UROLOGIE ORGAN DER DEUTSCHEN GESELLSCHAFT FÜR UROLOGIE, SPRINGER, BERLIN, DE, vol. 46, no. 7, 8 July 2007 (2007-07-08), pages 754-760, XP019535650, ISSN: 1433-0563, DOI: 10.1007/S00120-007-1347-0

Fortsættes ...

DESCRIPTION

TECHNICAL FIELD

[0001] The present disclosure relates to molecular diagnostic assays that provide information concerning methods to use gene expression profiles to determine prognostic information for cancer patients. Specifically, the present disclosure provides genes and microRNAs, the expression levels of which may be used to determine the likelihood that a prostate cancer patient will experience a local or distant cancer recurrence.

INTRODUCTION

[0002] Prostate cancer is the most common solid malignancy in men and the second most common cause of cancer-related death in men in North America and the European Union (EU). In 2008, over 180,000 patients will be diagnosed with prostate cancer in the United States alone and nearly 30,000 will die of this disease. Age is the single most important risk factor for the development of prostate cancer, and applies across all racial groups that have been studied. With the aging of the U.S. population, it is projected that the annual incidence of prostate cancer will double by 2025 to nearly 400,000 cases per year.

[0003] Since the introduction of prostate-specific antigen (PSA) screening in the 1990's, the proportion of patients presenting with clinically evident disease has fallen dramatically such that patients categorized as "low risk" now constitute half of new diagnoses today. PSA is used as a tumor marker to determine the presence of prostate cancer as high PSA levels are associated with prostate cancer. Despite a growing proportion of localized prostate cancer patients presenting with low-risk features such as low stage (T1) disease, greater than 90% of patients in the US still undergo definitive therapy, including prostatectomy or radiation. Only about 15% of these patients would develop metastatic disease and die from their prostate cancer, even in the absence of definitive therapy. A. Bill-Axelson, et al., J Nat'l Cancer Inst. 100(16):1144-1154 (2008). Therefore, the majority of prostate cancer patients are being over-treated.

[0004] Estimates of recurrence risk and treatment decisions in prostate cancer are currently based primarily on PSA levels and/or tumor stage. Although tumor stage has been demonstrated to have significant association with outcome sufficient to be included in pathology reports, the College of American Pathologists Consensus Statement noted that variations in approach to the acquisition, interpretation, reporting, and analysis of this information exist. C. Compton, et al., Arch Pathol Lab Med 124:979-992 (2000). As a consequence, existing pathologic staging methods have been criticized as lacking reproducibility and therefore may provide imprecise estimates of individual patient risk.

[0005] WO 2010/056993 relates to biomarkers for prognosis of prostate cancer.

[0006] WO 2008/067065 relates to proteins for cancer prognosis.

SUMMARY

[0007] The invention is defined in the claims.

[0008] This application discloses molecular assays that involve measurement of expression level(s) of one or more genes, gene subsets, microRNAs, or one or more microRNAs in combination with one or more genes or gene subsets, from a biological sample obtained from a prostate cancer patient, and analysis of the measured expression levels to provide information concerning the likelihood of cancer recurrence. For example, the likelihood of cancer recurrence could be described in terms of a score based on clinical or biochemical recurrence-free interval.

[0009] In addition, this application discloses molecular assays that involve measurement of expression level(s) of one or more genes, gene subsets, microRNAs, or one or more microRNAs in combination with one or more genes or gene subsets, from a biological sample obtained to identify a risk classification for a prostate cancer patient. For example, patients may be stratified using expression level(s) of one or more genes or microRNAs associated, positively or negatively, with cancer recurrence or death from cancer, or with a prognostic factor. In an exemplary embodiment, the prognostic factor is Gleason pattern.

[0010] The biological sample may be obtained from standard methods, including surgery, biopsy, or bodily fluids. It may comprise tumor tissue or cancer cells, and, in some cases, histologically normal tissue, e.g., histologically normal tissue adjacent the tumor tissue. In exemplary embodiments, the biological sample is positive or negative for a TMPRSS2 fusion.

[0011] In exemplary embodiments, expression level(s) of one or more genes and/or microRNAs that are associated, positively or negatively, with a particular clinical outcome in prostate cancer are used to determine prognosis and appropriate therapy. The genes disclosed herein may be used alone or arranged in functional gene subsets, such as cell adhesion/migration, immediate-early stress response, and extracellular matrix-associated. Each gene subset comprises the genes disclosed herein, as well as genes that are co-expressed with one or more of the disclosed genes. The calculation may be performed on a computer, programmed to execute the gene expression analysis. The microRNAs disclosed herein may also be used alone or in combination with any one or more of the microRNAs and/or genes disclosed.

[0012] In exemplary embodiments, the molecular assay may involve expression levels for at least two genes. The genes, or gene subsets, may be weighted according to strength of association with prognosis or tumor microenvironment. In another exemplary embodiment, the molecular assay may involve expression levels of at least one gene and at least one microRNA. The gene-microRNA combination may be selected based on the likelihood that the gene-microRNA combination functionally interact.

BRIEF DESCRIPTION OF THE DRAWING

[0013] Figure 1 shows the distribution of clinical and pathology assessments of biopsy Gleason score, baseline PSA level, and clinical T-stage.

DEFINITIONS

[0014] Unless defined otherwise, technical and scientific terms used herein have the same meaning as commonly understood by one of ordinary skill in the art to which this invention belongs. Singleton et al., Dictionary of Microbiology and Molecular Biology 2nd ed., J. Wiley & Sons (New York, NY 1994), and March, Advanced Organic Chemistry Reactions, Mechanisms and Structure 4th ed., John Wiley & Sons (New York, NY 1992), provide one skilled in the art with a general guide to many of the terms used in the present application.

[0015] The terms "tumor" and "lesion" as used herein, refer to all neoplastic cell growth and proliferation, whether malignant or benign, and all pre-cancerous and cancerous cells and tissues. Those skilled in the art will realize that a tumor tissue sample may comprise multiple biological elements, such as one or more cancer cells, partial or fragmented cells, tumors in various stages, surrounding histologically normal-appearing tissue, and/or macro or micro-dissected tissue.

[0016] The terms "cancer" and "cancerous" refer to or describe the physiological condition in mammals that is typically characterized by unregulated cell growth. Examples of cancer in the present disclosure include cancer of the urogenital tract, such as prostate cancer.

[0017] The "pathology" of cancer includes all phenomena that compromise the well-being of the patient. This includes, without limitation, abnormal or uncontrollable cell growth, metastasis, interference with the normal functioning of neighboring cells, release of cytokines or other secretory products at abnormal levels, suppression or aggravation of inflammatory or immunological response, neoplasia, premalignancy, malignancy, invasion of surrounding or distant tissues or organs, such as lymph nodes, etc.

[0018] As used herein, the term "prostate cancer" is used interchangeably and in the broadest sense refers to all stages and all forms of cancer arising from the tissue of the prostate gland.

[0019] According to the tumor, node, metastasis (TNM) staging system of the American Joint Committee on Cancer (AJCC), AJCC Cancer Staging Manual (7th Ed., 2010), the various stages of prostate cancer are defined as follows: Tumor: T1: clinically inapparent tumor not palpable or visible by imaging, T1a: tumor incidental histological finding in 5% or less of tissue resected, T1b: tumor incidental histological finding in more than 5% of tissue resected, T1c: tumor identified by needle biopsy; T2: tumor confined within prostate, T2a: tumor involves one half of one lobe or less, T2b: tumor involves more than half of one lobe, but not both lobes, T2c: tumor involves both lobes; T3: tumor extends through the prostatic capsule, T3a: extracapsular extension (unilateral or bilateral), T3b: tumor invades seminal vesicle(s); T4: tumor is fixed or invades adjacent structures other than seminal vesicles (bladder neck, external sphincter, rectum, levator muscles,

or pelvic wall). Node: NO: no regional lymph node metastasis; N1: metastasis in regional lymph nodes. Metastasis: M0: no distant metastasis; M1: distant metastasis present.

[0020] The Gleason Grading system is used to help evaluate the prognosis of men with prostate cancer. Together with other parameters, it is incorporated into a strategy of prostate cancer staging, which predicts prognosis and helps guide therapy. A Gleason "score" or "grade" is given to prostate cancer based upon its microscopic appearance. Tumors with a low Gleason score typically grow slowly enough that they may not pose a significant threat to the patients in their lifetimes. These patients are monitored ("watchful waiting" or "active surveillance") over time. Cancers with a higher Gleason score are more aggressive and have a worse prognosis, and these patients are generally treated with surgery (e.g., radical prostatectomy) and, in some cases, therapy (e.g., radiation, hormone, ultrasound, chemotherapy). Gleason scores (or sums) comprise grades of the two most common tumor patterns. These patterns are referred to as Gleason patterns 1-5, with pattern 1 being the most well-differentiated. Most have a mixture of patterns. To obtain a Gleason score or grade, the dominant pattern is added to the second most prevalent pattern to obtain a number between 2 and 10. The Gleason Grades include: G1: well differentiated (slight anaplasia) (Gleason 2-4); G2: moderately differentiated (moderate anaplasia) (Gleason 5-6); G3-4: poorly differentiated/undifferentiated (marked anaplasia) (Gleason 7-10).

[0021] Stage groupings: Stage I: T1a N0 M0 G1; Stage II: (T1a N0 M0 G2-4) or (T1b, c, T1, T2, N0 M0 Any G); Stage III: T3 N0 M0 Any G; Stage IV: (T4 N0 M0 Any G) or (Any T N1 M0 Any G) or (Any T Any N M1 Any G).

[0022] As used herein, the term "tumor tissue" refers to a biological sample containing one or more cancer cells, or a fraction of one or more cancer cells. Those skilled in the art will recognize that such biological sample may additionally comprise other biological components, such as histologically appearing normal cells (e.g., adjacent the tumor), depending upon the method used to obtain the tumor tissue, such as surgical resection, biopsy, or bodily fluids.

[0023] As used herein, the term "AUA risk group" refers to the 2007 updated American Urological Association (AUA) guidelines for management of clinically localized prostate cancer, which clinicians use to determine whether a patient is at low, intermediate, or high risk of biochemical (PSA) relapse after local therapy.

[0024] As used herein, the term "adjacent tissue (AT)" refers to histologically "normal" cells that are adjacent a tumor. For example, the AT expression profile may be associated with disease recurrence and survival.

[0025] As used herein "non-tumor prostate tissue" refers to histologically normal-appearing tissue adjacent a prostate tumor.

[0026] Prognostic factors are those variables related to the natural history of cancer, which influence the recurrence rates and outcome of patients once they have developed cancer. Clinical parameters that have been associated with a worse prognosis include, for example, increased tumor stage, PSA level at presentation, and Gleason grade or pattern. Prognostic factors are

frequently used to categorize patients into subgroups with different baseline relapse risks.

[0027] The term "prognosis" is used herein to refer to the likelihood that a cancer patient will have a cancer-attributable death or progression, including recurrence, metastatic spread, and drug resistance, of a neoplastic disease, such as prostate cancer. For example, a "good prognosis" would include long term survival without recurrence and a "bad prognosis" would include cancer recurrence.

[0028] As used herein, the term "expression level" as applied to a gene refers to the normalized level of a gene product, e.g. the normalized value determined for the RNA expression level of a gene or for the polypeptide expression level of a gene.

[0029] The term "gene product" or "expression product" are used herein to refer to the RNA (ribonucleic acid) transcription products (transcripts) of the gene, including mRNA, and the polypeptide translation products of such RNA transcripts. A gene product can be, for example, an unspliced RNA, an mRNA, a splice variant mRNA, a microRNA, a fragmented RNA, a polypeptide, a post-translationally modified polypeptide, a splice variant polypeptide, etc.

[0030] The term "RNA transcript" as used herein refers to the RNA transcription products of a gene, including, for example, mRNA, an unspliced RNA, a splice variant mRNA, a microRNA, and a fragmented RNA.

[0031] The term "microRNA" is used herein to refer to a small, non-coding, single-stranded RNA of ~18 - 25 nucleotides that may regulate gene expression. For example, when associated with the RNA-induced silencing complex (RISC), the complex binds to specific mRNA targets and causes translation repression or cleavage of these mRNA sequences.

[0032] Unless indicated otherwise, each gene name used herein corresponds to the Official Symbol assigned to the gene and provided by Entrez Gene (URL: www.ncbi.nlm.nih.gov/sites/entrez) as of the filing date of this application.

[0033] The terms "correlated" and "associated" are used interchangeably herein to refer to the association between two measurements (or measured entities). The disclosure provides genes, gene subsets, microRNAs, or microRNAs in combination with genes or gene subsets, the expression levels of which are associated with tumor stage. For example, the increased expression level of a gene or microRNA may be positively correlated (positively associated) with a good or positive prognosis. Such a positive correlation may be demonstrated statistically in various ways, e.g. by a cancer recurrence hazard ratio less than one. In another example, the increased expression level of a gene or microRNA may be negatively correlated (negatively associated) with a good or positive prognosis. In that case, for example, the patient may experience a cancer recurrence.

[0034] The terms "good prognosis" or "positive prognosis" as used herein refer to a beneficial clinical outcome, such as long-term survival without recurrence. The terms "bad prognosis" or "negative prognosis" as used herein refer to a negative clinical outcome, such as cancer

recurrence.

[0035] The term "risk classification" means a grouping of subjects by the level of risk (or likelihood) that the subject will experience a particular clinical outcome. A subject may be classified into a risk group or classified at a level of risk based on the methods of the present disclosure, e.g. high, medium, or low risk. A "risk group" is a group of subjects or individuals with a similar level of risk for a particular clinical outcome.

[0036] The term "long-term" survival is used herein to refer to survival for a particular time period, e.g., for at least 5 years, or for at least 10 years.

[0037] The term "recurrence" is used herein to refer to local or distant recurrence (i.e., metastasis) of cancer. For example, prostate cancer can recur locally in the tissue next to the prostate or in the seminal vesicles. The cancer may also affect the surrounding lymph nodes in the pelvis or lymph nodes outside this area. Prostate cancer can also spread to tissues next to the prostate, such as pelvic muscles, bones, or other organs. Recurrence can be determined by clinical recurrence detected by, for example, imaging study or biopsy, or biochemical recurrence detected by, for example, sustained follow-up prostate-specific antigen (PSA) levels ≥ 0.4 ng/mL or the initiation of salvage therapy as a result of a rising PSA level.

[0038] The term "clinical recurrence-free interval (cRFI)" is used herein as time (in months) from surgery to first clinical recurrence or death due to clinical recurrence of prostate cancer. Losses due to incomplete follow-up, other primary cancers or death prior to clinical recurrence are considered censoring events; when these occur, the only information known is that up through the censoring time, clinical recurrence has not occurred in this subject. Biochemical recurrences are ignored for the purposes of calculating cRFI.

[0039] The term "biochemical recurrence-free interval (bRFI)" is used herein to mean the time (in months) from surgery to first biochemical recurrence of prostate cancer. Clinical recurrences, losses due to incomplete follow-up, other primary cancers, or death prior to biochemical recurrence are considered censoring events.

[0040] The term "Overall Survival (OS)" is used herein to refer to the time (in months) from surgery to death from any cause. Losses due to incomplete follow-up are considered censoring events. Biochemical recurrence and clinical recurrence are ignored for the purposes of calculating OS.

[0041] The term "Prostate Cancer-Specific Survival (PCSS)" is used herein to describe the time (in years) from surgery to death from prostate cancer. Losses due to incomplete follow-up or deaths from other causes are considered censoring events. Clinical recurrence and biochemical recurrence are ignored for the purposes of calculating PCSS.

[0042] The term "upgrading" or "upstaging" as used herein refers to a change in Gleason grade from 3+3 at the time of biopsy to 3+4 or greater at the time of radical prostatectomy (RP), or Gleason grade 3+4 at the time of biopsy to 4+3 or greater at the time of RP, or seminal vesical

involvement (SVI), or extracapsular involvement (ECE) at the time of RP.

[0043] In practice, the calculation of the measures listed above may vary from study to study depending on the definition of events to be considered censored.

[0044] The term "microarray" refers to an ordered arrangement of hybridizable array elements, e.g. oligonucleotide or polynucleotide probes, on a substrate.

[0045] The term "polynucleotide" generally refers to any polyribonucleotide or polydeoxyribonucleotide, which may be unmodified RNA or DNA or modified RNA or DNA. Thus, for instance, polynucleotides as defined herein include, without limitation, single- and double-stranded DNA, DNA including single- and double-stranded regions, single- and double-stranded RNA, and RNA including single- and double-stranded regions, hybrid molecules comprising DNA and RNA that may be single-stranded or, more typically, double-stranded or include single- and double-stranded regions. In addition, the term "polynucleotide" as used herein refers to triple-stranded regions comprising RNA or DNA or both RNA and DNA. The strands in such regions may be from the same molecule or from different molecules. The regions may include all of one or more of the molecules, but more typically involve only a region of some of the molecules. One of the molecules of a triple-helical region often is an oligonucleotide. The term "polynucleotide" specifically includes cDNAs. The term includes DNAs (including cDNAs) and RNAs that contain one or more modified bases. Thus, DNAs or RNAs with backbones modified for stability or for other reasons, are "polynucleotides" as that term is intended herein. Moreover, DNAs or RNAs comprising unusual bases, such as inosine, or modified bases, such as tritiated bases, are included within the term "polynucleotides" as defined herein. In general, the term "polynucleotide" embraces all chemically, enzymatically and/or metabolically modified forms of unmodified polynucleotides, as well as the chemical forms of DNA and RNA characteristic of viruses and cells, including simple and complex cells.

[0046] The term "oligonucleotide" refers to a relatively short polynucleotide, including, without limitation, single-stranded deoxyribonucleotides, single- or double-stranded ribonucleotides, RNA/DNA hybrids and double-stranded DNAs. Oligonucleotides, such as single-stranded DNA probe oligonucleotides, are often synthesized by chemical methods, for example using automated oligonucleotide synthesizers that are commercially available. However, oligonucleotides can be made by a variety of other methods, including in vitro recombinant DNA-mediated techniques and by expression of DNAs in cells and organisms.

[0047] The term "Ct" as used herein refers to threshold cycle, the cycle number in quantitative polymerase chain reaction (qPCR) at which the fluorescence generated within a reaction well exceeds the defined threshold, i.e. the point during the reaction at which a sufficient number of amplicons have accumulated to meet the defined threshold.

[0048] The term "Cp" as used herein refers to "crossing point." The Cp value is calculated by determining the second derivatives of entire qPCR amplification curves and their maximum value. The Cp value represents the cycle at which the increase of fluorescence is highest and where the logarithmic phase of a PCR begins.

[0049] The terms "threshold" or "thresholding" refer to a procedure used to account for non-linear relationships between gene expression measurements and clinical response as well as to further reduce variation in reported patient scores. When thresholding is applied, all measurements below or above a threshold are set to that threshold value. Non-linear relationship between gene expression and outcome could be examined using smoothers or cubic splines to model gene expression in Cox PH regression on recurrence free interval or logistic regression on recurrence status. D. Cox, Journal of the Royal Statistical Society, Series B 34:187-220 (1972). Variation in reported patient scores could be examined as a function of variability in gene expression at the limit of quantitation and/or detection for a particular gene.

[0050] As used herein, the term "amplicon," refers to pieces of DNA that have been synthesized using amplification techniques, such as polymerase chain reactions (PCR) and ligase chain reactions.

[0051] "Stringency" of hybridization reactions is readily determinable by one of ordinary skill in the art, and generally is an empirical calculation dependent upon probe length, washing temperature, and salt concentration. In general, longer probes require higher temperatures for proper annealing, while shorter probes need lower temperatures. Hybridization generally depends on the ability of denatured DNA to re-anneal when complementary strands are present in an environment below their melting temperature. The higher the degree of desired homology between the probe and hybridizable sequence, the higher the relative temperature which can be used. As a result, it follows that higher relative temperatures would tend to make the reaction conditions more stringent, while lower temperatures less so. For additional details and explanation of stringency of hybridization reactions, see Ausubel et al., Current Protocols in Molecular Biology (Wiley Interscience Publishers, 1995).

[0052] "Stringent conditions" or "high stringency conditions", as defined herein, typically: (1) employ low ionic strength and high temperature for washing, for example 0.015 M sodium chloride/0.0015 M sodium citrate/0.1% sodium dodecyl sulfate at 50°C; (2) employ during hybridization a denaturing agent, such as formamide, for example, 50% (v/v) formamide with 0.1% bovine serum albumin/0.1% Ficoll/0.1% polyvinylpyrrolidone/50mM sodium phosphate buffer at pH 6.5 with 750 mM sodium chloride, 75 mM sodium citrate at 42°C; or (3) employ 50% formamide, 5 x SSC (0.75 M NaCl, 0.075 M sodium citrate), 50 mM sodium phosphate (pH 6.8), 0.1% sodium pyrophosphate, 5 x Denhardt's solution, sonicated salmon sperm DNA (50 µg/ml), 0.1% SDS, and 10% dextran sulfate at 42°C, with washes at 42°C in 0.2 x SSC (sodium chloride/sodium citrate) and 50% formamide, followed by a high-stringency wash consisting of 0.1 x SSC containing EDTA at 55°C.

[0053] "Moderately stringent conditions" may be identified as described by Sambrook et al., Molecular Cloning: A Laboratory Manual, New York: Cold Spring Harbor Press, 1989, and include the use of washing solution and hybridization conditions (e.g., temperature, ionic strength and %SDS) less stringent than those described above. An example of moderately stringent conditions is overnight incubation at 37°C in a solution comprising: 20% formamide, 5 x SSC (150 mM NaCl, 15 mM trisodium citrate), 50 mM sodium phosphate (pH 7.6), 5 x Denhardt's solution, 10% dextran sulfate, and 20 mg/ml denatured sheared salmon sperm DNA, followed by washing the

filters in 1 x SSC at about 37-50°C. The skilled artisan will recognize how to adjust the temperature, ionic strength, etc. as necessary to accommodate factors such as probe length and the like.

[0054] The terms "splicing" and "RNA splicing" are used interchangeably and refer to RNA processing that removes introns and joins exons to produce mature mRNA with continuous coding sequence that moves into the cytoplasm of an eukaryotic cell.

[0055] The terms "co-express" and "co-expressed", as used herein, refer to a statistical correlation between the amounts of different transcript sequences across a population of different patients. Pairwise co-expression may be calculated by various methods known in the art, e.g., by calculating Pearson correlation coefficients or Spearman correlation coefficients. Co-expressed gene cliques may also be identified using graph theory. An analysis of co-expression may be calculated using normalized expression data. A gene is said to be co-expressed with a particular disclosed gene when the expression level of the gene exhibits a Pearson correlation coefficient greater than or equal to 0.6.

[0056] A "computer-based system" refers to a system of hardware, software, and data storage medium used to analyze information. The minimum hardware of a patient computer-based system comprises a central processing unit (CPU), and hardware for data input, data output (e.g., display), and data storage. An ordinarily skilled artisan can readily appreciate that any currently available computer-based systems and/or components thereof are suitable for use in connection with the methods of the present disclosure. The data storage medium may comprise any manufacture comprising a recording of the present information as described above, or a memory access device that can access such a manufacture.

[0057] To "record" data, programming or other information on a computer readable medium refers to a process for storing information, using any such methods as known in the art. Any convenient data storage structure may be chosen, based on the means used to access the stored information. A variety of data processor programs and formats can be used for storage, e.g. word processing text file, database format, etc.

[0058] A "processor" or "computing means" references any hardware and/or software combination that will perform the functions required of it. For example, a suitable processor may be a programmable digital microprocessor such as available in the form of an electronic controller, mainframe, server or personal computer (desktop or portable). Where the processor is programmable, suitable programming can be communicated from a remote location to the processor, or previously saved in a computer program product (such as a portable or fixed computer readable storage medium, whether magnetic, optical or solid state device based). For example, a magnetic medium or optical disk may carry the programming, and can be read by a suitable reader communicating with each processor at its corresponding station.

[0059] As used herein, the terms "active surveillance" and "watchful waiting" mean closely monitoring a patient's condition without giving any treatment until symptoms appear or change. For example, in prostate cancer, watchful waiting is usually used in older men with other medical

problems and early-stage disease.

[0060] As used herein, the term "surgery" applies to surgical methods undertaken for removal of cancerous tissue, including pelvic lymphadenectomy, radical prostatectomy, transurethral resection of the prostate (TURP), excision, dissection, and tumor biopsy/removal. The tumor tissue or sections used for gene expression analysis may have been obtained from any of these methods.

[0061] As used herein, the term "therapy" includes radiation, hormonal therapy, cryosurgery, chemotherapy, biologic therapy, and high-intensity focused ultrasound.

[0062] As used herein, the term "TMPRSS fusion" and "TMPRSS2 fusion" are used interchangeably and refer to a fusion of the androgen-driven TMPRSS2 gene with the ERG oncogene, which has been demonstrated to have a significant association with prostate cancer. S. Perner, et al., *Urologe A*. 46(7):754-760 (2007); S.A. Narod, et al., *Br J Cancer* 99(6):847-851 (2008). As used herein, positive TMPRSS fusion status indicates that the TMPRSS fusion is present in a tissue sample, whereas negative TMPRSS fusion status indicates that the TMPRSS fusion is not present in a tissue sample. Experts skilled in the art will recognize that there are numerous ways to determine TMPRSS fusion status, such as real-time, quantitative PCR or high-throughput sequencing. See, e.g., K. Mertz, et al., *Neoplasia* 9(3):200-206 (2007); C. Maher, *Nature* 458(7234):97-101 (2009).

GENE EXPRESSION METHODS USING GENES, GENE SUBSETS, AND MICRORNAS

[0063] The present disclosure provides molecular assays that involve measurement of expression level(s) of one or more genes, gene subsets, microRNAs, or one or more microRNAs in combination with one or more genes or gene subsets, from a biological sample obtained from a prostate cancer patient, and analysis of the measured expression levels to provide information concerning the likelihood of cancer recurrence.

[0064] The present disclosure further provides methods to classify a prostate tumor based on expression level(s) of one or more genes and/or microRNAs. The disclosure further provides genes and/or microRNAs that are associated, positively or negatively, with a particular prognostic outcome. In exemplary embodiments, the clinical outcomes include cRFI and bRFI. In another embodiment, patients may be classified in risk groups based on the expression level(s) of one or more genes and/or microRNAs that are associated, positively or negatively, with a prognostic factor. In an exemplary embodiment, that prognostic factor is Gleason pattern.

[0065] Various technological approaches for determination of expression levels of the disclosed genes and microRNAs are set forth in this specification, including, without limitation, RT-PCR, microarrays, high-throughput sequencing, serial analysis of gene expression (SAGE) and Digital Gene Expression (DGE), which will be discussed in detail below. In particular aspects, the expression level of each gene or microRNA may be determined in relation to various features of the expression products of the gene including exons, introns, protein epitopes and protein activity.

[0066] The expression level(s) of one or more genes and/or microRNAs may be measured in tumor tissue. For example, the tumor tissue may be obtained upon surgical removal or resection of the tumor, or by tumor biopsy. The tumor tissue may be or include histologically "normal" tissue, for example histologically "normal" tissue adjacent to a tumor. The expression level of genes and/or microRNAs may also be measured in tumor cells recovered from sites distant from the tumor, for example circulating tumor cells, body fluid (e.g., urine, blood, blood fraction, etc.).

[0067] The expression product that is assayed can be, for example, RNA or a polypeptide. The expression product may be fragmented. For example, the assay may use primers that are complementary to target sequences of an expression product and could thus measure full transcripts as well as those fragmented expression products containing the target sequence. Further information is provided in Table A (inserted in specification prior to claims).

[0068] The RNA expression product may be assayed directly or by detection of a cDNA product resulting from a PCR-based amplification method, e.g., quantitative reverse transcription polymerase chain reaction (qRT-PCR). (See e.g., U.S. Patent No. 7,587,279). Polypeptide expression product may be assayed using immunohistochemistry (IHC). Further, both RNA and polypeptide expression products may also be assayed using microarrays.

CLINICAL UTILITY

[0069] Prostate cancer is currently diagnosed using a digital rectal exam (DRE) and Prostate-specific antigen (PSA) test. If PSA results are high, patients will generally undergo a prostate tissue biopsy. The pathologist will review the biopsy samples to check for cancer cells and determine a Gleason score. Based on the Gleason score, PSA, clinical stage, and other factors, the physician must make a decision whether to monitor the patient, or treat the patient with surgery and therapy.

[0070] At present, clinical decision-making in early stage prostate cancer is governed by certain histopathologic and clinical factors. These include: (1) tumor factors, such as clinical stage (e.g. T1, T2), PSA level at presentation, and Gleason grade, that are very strong prognostic factors in determining outcome; and (2) host factors, such as age at diagnosis and co-morbidity. Because of these factors, the most clinically useful means of stratifying patients with localized disease according to prognosis has been through multifactorial staging, using the clinical stage, the serum PSA level, and tumor grade (Gleason grade) together. In the 2007 updated American Urological Association (AUA) guidelines for management of clinically localized prostate cancer, these parameters have been grouped to determine whether a patient is at low, intermediate, or high risk of biochemical (PSA) relapse after local therapy. I. Thompson, et al., Guideline for the management of clinically localized prostate cancer, J Urol. 177(6):2106-31 (2007).

[0071] Although such classifications have proven to be helpful in distinguishing patients with localized disease who may need adjuvant therapy after surgery/radiation, they have less ability to discriminate between indolent cancers, which do not need to be treated with local therapy, and

aggressive tumors, which require local therapy. In fact, these algorithms are of increasingly limited use for deciding between conservative management and definitive therapy because the bulk of prostate cancers diagnosed in the PSA screening era now present with clinical stage T1c and PSA ≤ 10 ng/mL.

[0072] Patients with T1 prostate cancer have disease that is not clinically apparent but is discovered either at transurethral resection of the prostate (TURP, T1a, T1b) or at biopsy performed because of an elevated PSA (> 4 ng/mL, T1c). Approximately 80% of the cases presenting in 2007 are clinical T1 at diagnosis. In a Scandinavian trial, OS at 10 years was 85% for patients with early stage prostate cancer (T1/T2) and Gleason score ≤ 7 , after radical prostatectomy.

[0073] Patients with T2 prostate cancer have disease that is clinically evident and is organ confined; patients with T3 tumors have disease that has penetrated the prostatic capsule and/or has invaded the seminal vesicles. It is known from surgical series that clinical staging underestimates pathological stage, so that about 20% of patients who are clinically T2 will be pT3 after prostatectomy. Most of patients with T2 or T3 prostate cancer are treated with local therapy, either prostatectomy or radiation. The data from the Scandinavian trial suggest that for T2 patients with Gleason grade ≤ 7 , the effect of prostatectomy on survival is at most 5% at 10 years; the majority of patients do not benefit from surgical treatment at the time of diagnosis. For T2 patients with Gleason > 7 or for T3 patients, the treatment effect of prostatectomy is assumed to be significant but has not been determined in randomized trials. It is known that these patients have a significant risk (10-30%) of recurrence at 10 years after local treatment, however, there are no prospective randomized trials that define the optimal local treatment (radical prostatectomy, radiation) at diagnosis, which patients are likely to benefit from neo-adjuvant/adjuvant androgen deprivation therapy, and whether treatment (androgen deprivation, chemotherapy) at the time of biochemical failure (elevated PSA) has any clinical benefit.

[0074] Accurately determining Gleason scores from needle biopsies presents several technical challenges. First, interpreting histology that is "borderline" between Gleason pattern is highly subjective, even for urologic pathologists. Second, incomplete biopsy sampling is yet another reason why the "predicted" Gleason score on biopsy does not always correlate with the actual "observed" Gleason score of the prostate cancer in the gland itself. Hence, the accuracy of Gleason scoring is dependent upon not only the expertise of the pathologist reading the slides, but also on the completeness and adequacy of the prostate biopsy sampling strategy. T. Stamey, Urology 45:2-12 (1995). The gene/microRNA expression assay and associated information provided by the practice of the methods disclosed herein provide a molecular assay method to facilitate optimal treatment decision-making in early stage prostate cancer. An exemplary embodiment provides genes and microRNAs, the expression levels of which are associated (positively or negatively) with prostate cancer recurrence. For example, such a clinical tool would enable physicians to identify T2/T3 patients who are likely to recur following definitive therapy and need adjuvant treatment.

[0075] In addition, the methods disclosed herein may allow physicians to classify tumors, at a molecular level, based on expression level(s) of one or more genes and/or microRNAs that are

significantly associated with prognostic factors, such as Gleason pattern and TMPRSS fusion status. These methods would not be impacted by the technical difficulties of intra-patient variability, histologically determining Gleason pattern in biopsy samples, or inclusion of histologically normal appearing tissue adjacent to tumor tissue. Multi-analyte gene/microRNA expression tests can be used to measure the expression level of one or more genes and/or microRNAs involved in each of several relevant physiologic processes or component cellular characteristics. The methods disclosed herein may group the genes and/or microRNAs. The grouping of genes and microRNAs may be performed at least in part based on knowledge of the contribution of those genes and/or microRNAs according to physiologic functions or component cellular characteristics, such as in the groups discussed above. Furthermore, one or more microRNAs may be combined with one or more genes. The gene-microRNA combination may be selected based on the likelihood that the gene-microRNA combination functionally interact. The formation of groups (or gene subsets), in addition, can facilitate the mathematical weighting of the contribution of various expression levels to cancer recurrence. The weighting of a gene/microRNA group representing a physiological process or component cellular characteristic can reflect the contribution of that process or characteristic to the pathology of the cancer and clinical outcome.

[0076] Optionally, the methods disclosed may be used to classify patients by risk, for example risk of recurrence. Patients can be partitioned into subgroups (e.g., tertiles or quartiles) and the values chosen will define subgroups of patients with respectively greater or lesser risk.

[0077] The utility of a disclosed gene marker in predicting prognosis may not be unique to that marker. An alternative marker having an expression pattern that is parallel to that of a disclosed gene may be substituted for, or used in addition to, that co-expressed gene or microRNA. Due to the co-expression of such genes or microRNAs, substitution of expression level values should have little impact on the overall utility of the test. The closely similar expression patterns of two genes or microRNAs may result from involvement of both genes or microRNAs in the same process and/or being under common regulatory control in prostate tumor cells. The present disclosure thus contemplates the use of such co-expressed genes, gene subsets, or microRNAs as substitutes for, or in addition to, genes of the present disclosure.

METHODS OF ASSAYING EXPRESSION LEVELS OF A GENE PRODUCT

[0078] The methods and compositions of the present disclosure will employ, unless otherwise indicated, conventional techniques of molecular biology (including recombinant techniques), microbiology, cell biology, and biochemistry, which are within the skill of the art. Exemplary techniques are explained fully in the literature, such as, "Molecular Cloning: A Laboratory Manual", 2nd edition (Sambrook et al., 1989); "Oligonucleotide Synthesis" (M.J. Gait, ed., 1984); "Animal Cell Culture" (R.I. Freshney, ed., 1987); "Methods in Enzymology" (Academic Press, Inc.); "Handbook of Experimental Immunology", 4th edition (D.M. Weir & C.C. Blackwell, eds., Blackwell Science Inc., 1987); "Gene Transfer Vectors for Mammalian Cells" (J.M. Miller & M.P. Calos, eds., 1987); "Current Protocols in Molecular Biology" (F.M. Ausubel et al., eds., 1987); and "PCR: The Polymerase Chain Reaction", (Mullis et al., eds., 1994).

[0079] Methods of gene expression profiling include methods based on hybridization analysis of polynucleotides, methods based on sequencing of polynucleotides, and proteomics-based methods. Exemplary methods known in the art for the quantification of RNA expression in a sample include northern blotting and in situ hybridization (Parker & Barnes, *Methods in Molecular Biology* 106:247-283 (1999)); RNase protection assays (Hod, *Biotechniques* 13:852-854 (1992)); and PCR-based methods, such as reverse transcription PCT (RT-PCR) (Weis et al., *Trends in Genetics* 8:263-264 (1992)). Antibodies may be employed that can recognize sequence-specific duplexes, including DNA duplexes, RNA duplexes, and DNA-RNA hybrid duplexes or DNA-protein duplexes. Representative methods for sequencing-based gene expression analysis include Serial Analysis of Gene Expression (SAGE), and gene expression analysis by massively parallel signature sequencing (MPSS).

Reverse Transcriptase PCR (RT-PCR)

[0080] Typically, mRNA or microRNA is isolated from a test sample. The starting material is typically total RNA isolated from a human tumor, usually from a primary tumor. Optionally, normal tissues from the same patient can be used as an internal control. Such normal tissue can be histologically-appearing normal tissue adjacent a tumor. mRNA or microRNA can be extracted from a tissue sample, e.g., from a sample that is fresh, frozen (e.g. fresh frozen), or paraffin-embedded and fixed (e.g. formalin-fixed).

[0081] General methods for mRNA and microRNA extraction are well known in the art and are disclosed in standard textbooks of molecular biology, including Ausubel et al., *Current Protocols of Molecular Biology*, John Wiley and Sons (1997). Methods for RNA extraction from paraffin embedded tissues are disclosed, for example, in Rupp and Locker, *Lab Invest.* 56:A67 (1987), and De Andrés et al., *BioTechniques* 18:42044 (1995). In particular, RNA isolation can be performed using a purification kit, buffer set and protease from commercial manufacturers, such as Qiagen, according to the manufacturer's instructions. For example, total RNA from cells in culture can be isolated using Qiagen RNeasy mini-columns. Other commercially available RNA isolation kits include MasterPure™ Complete DNA and RNA Purification Kit (EPICENTRE®, Madison, WI), and Paraffin Block RNA Isolation Kit (Ambion, Inc.). Total RNA from tissue samples can be isolated using RNA Stat-60 (Tel-Test). RNA prepared from tumor can be isolated, for example, by cesium chloride density gradient centrifugation.

[0082] The sample containing the RNA is then subjected to reverse transcription to produce cDNA from the RNA template, followed by exponential amplification in a PCR reaction. The two most commonly used reverse transcriptases are avilo myeloblastosis virus reverse transcriptase (AMV-RT) and Moloney murine leukemia virus reverse transcriptase (MMLV-RT). The reverse transcription step is typically primed using specific primers, random hexamers, or oligo-dT primers, depending on the circumstances and the goal of expression profiling. For example, extracted RNA can be reverse-transcribed using a GeneAmp RNA PCR kit (Perkin Elmer, CA, USA), following the manufacturer's instructions. The derived cDNA can then be used as a template in the subsequent PCR reaction.

[0083] PCR-based methods use a thermostable DNA-dependent DNA polymerase, such as a Taq DNA polymerase. For example, TaqMan® PCR typically utilizes the 5'-nuclease activity of Taq or Tth polymerase to hydrolyze a hybridization probe bound to its target amplicon, but any enzyme with equivalent 5' nuclease activity can be used. Two oligonucleotide primers are used to generate an amplicon typical of a PCR reaction product. A third oligonucleotide, or probe, can be designed to facilitate detection of a nucleotide sequence of the amplicon located between the hybridization sites the two PCR primers. The probe can be detectably labeled, e.g., with a reporter dye, and can further be provided with both a fluorescent dye, and a quencher fluorescent dye, as in a Taqman® probe configuration. Where a Taqman® probe is used, during the amplification reaction, the Taq DNA polymerase enzyme cleaves the probe in a template-dependent manner. The resultant probe fragments disassociate in solution, and signal from the released reporter dye is free from the quenching effect of the second fluorophore. One molecule of reporter dye is liberated for each new molecule synthesized, and detection of the unquenched reporter dye provides the basis for quantitative interpretation of the data.

[0084] TaqMan® RT-PCR can be performed using commercially available equipment, such as, for example, high-throughput platforms such as the ABI PRISM 7700 Sequence Detection System® (Perkin-Elmer-Applied Biosystems, Foster City, CA, USA), or Lightcycler (Roche Molecular Biochemicals, Mannheim, Germany). In a preferred embodiment, the procedure is run on a LightCycler® 480 (Roche Diagnostics) real-time PCR system, which is a microwell plate-based cyclor platform.

[0085] 5'-Nuclease assay data are commonly initially expressed as a threshold cycle (" C_T "). Fluorescence values are recorded during every cycle and represent the amount of product amplified to that point in the amplification reaction. The threshold cycle (C_T) is generally described as the point when the fluorescent signal is first recorded as statistically significant. Alternatively, data may be expressed as a crossing point (" C_p "). The C_p value is calculated by determining the second derivatives of entire qPCR amplification curves and their maximum value. The C_p value represents the cycle at which the increase of fluorescence is highest and where the logarithmic phase of a PCR begins.

[0086] To minimize errors and the effect of sample-to-sample variation, RT-PCR is usually performed using an internal standard. The ideal internal standard gene (also referred to as a reference gene) is expressed at a quite constant level among cancerous and non-cancerous tissue of the same origin (i.e., a level that is not significantly different among normal and cancerous tissues), and is not significantly affected by the experimental treatment (i.e., does not exhibit a significant difference in expression level in the relevant tissue as a result of exposure to chemotherapy), and expressed at a quite constant level among the same tissue taken from different patients. For example, reference genes useful in the methods disclosed herein should not exhibit significantly different expression levels in cancerous prostate as compared to normal prostate tissue. RNAs frequently used to normalize patterns of gene expression are mRNAs for the housekeeping genes glyceraldehyde-3-phosphate-dehydrogenase (GAPDH) and β -actin. Exemplary reference genes used for normalization comprise one or more of the following genes: AAMP, ARF1, ATP5E, CLTC, GPS1, and PGK1. Gene expression measurements can be normalized relative to the mean of one or more (e.g., 2, 3, 4, 5, or more) reference genes.

Reference-normalized expression measurements can range from 2 to 15, where a one unit increase generally reflects a 2-fold increase in RNA quantity.

[0087] Real time PCR is compatible both with quantitative competitive PCR, where internal competitor for each target sequence is used for normalization, and with quantitative comparative PCR using a normalization gene contained within the sample, or a housekeeping gene for RT-PCR. For further details see, e.g. Held et al., *Genome Research* 6:986-994 (1996).

[0088] The steps of a representative protocol for use in the methods of the present disclosure use fixed, paraffin-embedded tissues as the RNA source. For example, mRNA isolation, purification, primer extension and amplification can be performed according to methods available in the art. (see, e.g., Godfrey et al. *J. Molec. Diagnostics* 2: 84-91 (2000); Specht et al., *Am. J. Pathol.* 158: 419-29 (2001)). Briefly, a representative process starts with cutting about 10 μ m thick sections of paraffin-embedded tumor tissue samples. The RNA is then extracted, and protein and DNA depleted from the RNA-containing sample. After analysis of the RNA concentration, RNA is reverse transcribed using gene specific primers followed by RT-PCR to provide for cDNA amplification products.

Design of Intron-Based PCR Primers and Probes

[0089] PCR primers and probes can be designed based upon exon or intron sequences present in the mRNA transcript of the gene of interest. Primer/probe design can be performed using publicly available software, such as the DNA BLAT software developed by Kent, W.J., *Genome Res.* 12(4):656-64 (2002), or by the BLAST software including its variations.

[0090] Where necessary or desired, repetitive sequences of the target sequence can be masked to mitigate non-specific signals. Exemplary tools to accomplish this include the Repeat Masker program available on-line through the Baylor College of Medicine, which screens DNA sequences against a library of repetitive elements and returns a query sequence in which the repetitive elements are masked. The masked intron sequences can then be used to design primer and probe sequences using any commercially or otherwise publicly available primer/probe design packages, such as Primer Express (Applied Biosystems); MGB assay-by-design (Applied Biosystems); Primer3 (Steve Rozen and Helen J. Skaletsky (2000) Primer3 on the WWW for general users and for biologist programmers. See S. Rrawetz, S. Misener, *Bioinformatics Methods and Protocols: Methods in Molecular Biology*, pp. 365-386 (Humana Press)).

[0091] Other factors that can influence PCR primer design include primer length, melting temperature (T_m), and G/C content, specificity, complementary primer sequences, and 3'-end sequence. In general, optimal PCR primers are generally 17-30 bases in length, and contain about 20-80%, such as, for example, about 50-60% G+C bases, and exhibit T_m 's between 50 and 80 $^{\circ}$ C, e.g. about 50 to 70 $^{\circ}$ C.

[0092] For further guidelines for PCR primer and probe design see, e.g. Dieffenbach, CW. et al, "General Concepts for PCR Primer Design" in: *PCR Primer, A Laboratory Manual*, Cold Spring

Harbor Laboratory Press, New York, 1995, pp. 133-155; Innis and Gelfand, "Optimization of PCRs" in: PCR Protocols, A Guide to Methods and Applications, CRC Press, London, 1994, pp. 5-11; and Plasterer, T.N. Primerselect: Primer and probe design. Methods Mol. Biol. 70:520-527 (1997).

[0093] Table A provides further information concerning the primer, probe, and amplicon sequences associated with the Examples disclosed herein.

MassARRAY® System

[0094] In MassARRAY-based methods, such as the exemplary method developed by Sequenom, Inc. (San Diego, CA) following the isolation of RNA and reverse transcription, the obtained cDNA is spiked with a synthetic DNA molecule (competitor), which matches the targeted cDNA region in all positions, except a single base, and serves as an internal standard. The cDNA/competitor mixture is PCR amplified and is subjected to a post-PCR shrimp alkaline phosphatase (SAP) enzyme treatment, which results in the dephosphorylation of the remaining nucleotides. After inactivation of the alkaline phosphatase, the PCR products from the competitor and cDNA are subjected to primer extension, which generates distinct mass signals for the competitor- and cDNA-derived PCR products. After purification, these products are dispensed on a chip array, which is pre-loaded with components needed for analysis with matrix-assisted laser desorption ionization time-of-flight mass spectrometry (MALDI-TOF MS) analysis. The cDNA present in the reaction is then quantified by analyzing the ratios of the peak areas in the mass spectrum generated. For further details see, e.g. Ding and Cantor, Proc. Natl. Acad. Sci. USA 100:3059-3064 (2003).

Other PCR-based Methods

[0095] Further PCR-based techniques that can find use in the methods disclosed herein include, for example, BeadArray® technology (Illumina, San Diego, CA; Oliphant et al., Discovery of Markers for Disease (Supplement to Biotechniques), June 2002; Ferguson et al., Analytical Chemistry 72:5618 (2000)); BeadArray for Detection of Gene Expression® (BADGE), using the commercially available Luminex 100 LabMAP® system and multiple color-coded microspheres (Luminex Corp., Austin, TX) in a rapid assay for gene expression (Yang et al., Genome Res. 11:1888-1898 (2001)); and high coverage expression profiling (HiCEP) analysis (Fukumura et al., Nucl. Acids. Res. 31(16) e94 (2003).

Microarrays

[0096] Expression levels of a gene or microArray of interest can also be assessed using the microarray technique. In this method, polynucleotide sequences of interest (including cDNAs and oligonucleotides) are arrayed on a substrate. The arrayed sequences are then contacted under

conditions suitable for specific hybridization with detectably labeled cDNA generated from RNA of a test sample. As in the RT-PCR method, the source of RNA typically is total RNA isolated from a tumor sample, and optionally from normal tissue of the same patient as an internal control or cell lines. RNA can be extracted, for example, from frozen or archived paraffin-embedded and fixed (e.g. formalin-fixed) tissue samples.

[0097] For example, PCR amplified inserts of cDNA clones of a gene to be assayed are applied to a substrate in a dense array. Usually at least 10,000 nucleotide sequences are applied to the substrate. For example, the microarrayed genes, immobilized on the microchip at 10,000 elements each, are suitable for hybridization under stringent conditions. Fluorescently labeled cDNA probes may be generated through incorporation of fluorescent nucleotides by reverse transcription of RNA extracted from tissues of interest. Labeled cDNA probes applied to the chip hybridize with specificity to each spot of DNA on the array. After washing under stringent conditions to remove non-specifically bound probes, the chip is scanned by confocal laser microscopy or by another detection method, such as a CCD camera. Quantitation of hybridization of each arrayed element allows for assessment of corresponding RNA abundance.

[0098] With dual color fluorescence, separately labeled cDNA probes generated from two sources of RNA are hybridized pair wise to the array. The relative abundance of the transcripts from the two sources corresponding to each specified gene is thus determined simultaneously. The miniaturized scale of the hybridization affords a convenient and rapid evaluation of the expression pattern for large numbers of genes. Such methods have been shown to have the sensitivity required to detect rare transcripts, which are expressed at a few copies per cell, and to reproducibly detect at least approximately two-fold differences in the expression levels (Skena et al, Proc. Natl. Acad. Sci. USA 93(2): 106-149 (1996)). Microarray analysis can be performed by commercially available equipment, following manufacturer's protocols, such as by using the Affymetrix GenChip® technology, or Incyte's microarray technology.

Serial Analysis of Gene Expression (SAGE)

[0099] Serial analysis of gene expression (SAGE) is a method that allows the simultaneous and quantitative analysis of a large number of gene transcripts, without the need of providing an individual hybridization probe for each transcript. First, a short sequence tag (about 10-14 bp) is generated that contains sufficient information to uniquely identify a transcript, provided that the tag is obtained from a unique position within each transcript. Then, many transcripts are linked together to form long serial molecules, that can be sequenced, revealing the identity of the multiple tags simultaneously. The expression pattern of any population of transcripts can be quantitatively evaluated by determining the abundance of individual tags, and identifying the gene corresponding to each tag. For more details see, e.g. Velculescu et al., Science 270:484-487 (1995); and Velculescu et al., Cell 88:243-51 (1997).

Gene Expression Analysis by Nucleic Acid Sequencing

[0100] Nucleic acid sequencing technologies are suitable methods for analysis of gene expression. The principle underlying these methods is that the number of times a cDNA sequence is detected in a sample is directly related to the relative expression of the RNA corresponding to that sequence. These methods are sometimes referred to by the term Digital Gene Expression (DGE) to reflect the discrete numeric property of the resulting data. Early methods applying this principle were Serial Analysis of Gene Expression (SAGE) and Massively Parallel Signature Sequencing (MPSS). See, e.g., S. Brenner, et al., *Nature Biotechnology* 18(6):630-634 (2000). More recently, the advent of "next-generation" sequencing technologies has made DGE simpler, higher throughput, and more affordable. As a result, more laboratories are able to utilize DGE to screen the expression of more genes in more individual patient samples than previously possible. See, e.g., J. Marioni, *Genome Research* 18(9): 1509-1517 (2008); R. Morin, *Genome Research* 18(4):610-621 (2008); A. Mortazavi, *Nature Methods* 5(7):621-628 (2008); N. Cloonan, *Nature Methods* 5(7):613-619 (2008).

Isolating RNA from Body Fluids

[0101] Methods of isolating RNA for expression analysis from blood, plasma and serum (see, e.g., K. Enders, et al., *Clin Chem* 48,1647-53 (2002) (and references cited therein) and from urine (see, e.g., R. Boom, et al., *J Clin Microbiol.* 28, 495-503 (1990) and references cited therein) have been described.

Immunohistochemistry

[0102] Immunohistochemistry methods are also suitable for detecting the expression levels of genes and applied to the method disclosed herein. Antibodies (e.g., monoclonal antibodies) that specifically bind a gene product of a gene of interest can be used in such methods. The antibodies can be detected by direct labeling of the antibodies themselves, for example, with radioactive labels, fluorescent labels, hapten' labels such as, biotin, or an enzyme such as horse radish peroxidase or alkaline phosphatase. Alternatively, unlabeled primary antibody can be used in conjunction with a labeled secondary antibody specific for the primary antibody. Immunohistochemistry protocols and kits are well known in the art and are commercially available.

Proteomics

[0103] The term "proteome" is defined as the totality of the proteins present in a sample (e.g. tissue, organism, or cell culture) at a certain point of time. Proteomics includes, among other things, study of the global changes of protein expression in a sample (also referred to as "expression proteomics"). Proteomics typically includes the following steps: (1) separation of individual proteins in a sample by 2-D gel electrophoresis (2-D PAGE); (2) identification of the individual proteins recovered from the gel, e.g. by mass spectrometry or N- terminal sequencing, and (3) analysis of the data using bioinformatics.

General Description of the mRNA/microRNA Isolation, Purification and Amplification

[0104] The steps of a representative protocol for profiling gene expression using fixed, paraffin-embedded tissues as the RNA source, including mRNA or microRNA isolation, purification, primer extension and amplification are provided in various published journal articles. (See, e.g., T.E. Godfrey, et al., J. Molec. Diagnostics 2: 84-91 (2000); K. Specht et al., Am. J. Pathol. 158: 419-29 (2001), M. Cronin, et al., Am J Pathol 164:35-42 (2004)). Briefly, a representative process starts with cutting a tissue sample section (e.g. about 10 μ m thick sections of a paraffin-embedded tumor tissue sample). The RNA is then extracted, and protein and DNA are removed. After analysis of the RNA concentration, RNA repair is performed if desired. The sample can then be subjected to analysis, e.g., by reverse transcribed using gene specific promoters followed by RT-PCR.

STATISTICAL ANALYSIS OF EXPRESSION LEVELS IN IDENTIFICATION OF GENES AND MICRORNAs

[0105] One skilled in the art will recognize that there are many statistical methods that may be used to determine whether there is a significant relationship between a parameter of interest (e.g., recurrence) and expression levels of a marker gene/microRNA as described here. In an exemplary embodiment, the present invention provides a stratified cohort sampling design (a form of case-control sampling) using tissue and data from prostate cancer patients. Selection of specimens was stratified by T stage (T1, T2), year cohort (<1993, $\geq$ 1993), and prostatectomy Gleason Score (low/intermediate, high). All patients with clinical recurrence were selected and a sample of patients who did not experience a clinical recurrence was selected. For each patient, up to two enriched tumor specimens and one normal-appearing tissue sample was assayed.

[0106] All hypothesis tests were reported using two-sided p-values. To investigate if there is a significant relationship of outcomes (clinical recurrence-free interval (cRFI), biochemical recurrence-free interval (bRFI), prostate cancer-specific survival (PCSS), and overall survival (OS)) with individual genes and/or microRNAs, demographic or clinical covariates Cox Proportional Hazards (PH) models using maximum weighted pseudo partial-likelihood estimators were used and p-values from Wald tests of the null hypothesis that the hazard ratio (HR) is one are reported. To investigate if there is a significant relationship between individual genes and/or microRNAs and Gleason pattern of a particular sample, ordinal logistic regression models using maximum weighted likelihood methods were used and p-values from Wald tests of the null hypothesis that the odds ratio (OR) is one are reported.

COEXPRESSION ANALYSIS

[0107] The present disclosure provides a method to determine tumor stage based on the expression of staging genes, or genes that co-express with particular staging genes. To perform particular biological processes, genes often work together in a concerted way, i.e. they are co-

expressed. Co-expressed gene groups identified for a disease process like cancer can serve as biomarkers for tumor status and disease progression. Such co-expressed genes can be assayed in lieu of, or in addition to, assaying of the staging gene with which they are co-expressed.

[0108] In an exemplary embodiment, the joint correlation of gene expression levels among prostate cancer specimens under study may be assessed. For this purpose, the correlation structures among genes and specimens may be examined through hierarchical cluster methods. This information may be used to confirm that genes that are known to be highly correlated in prostate cancer specimens cluster together as expected. Only genes exhibiting a nominally significant (unadjusted $p < 0.05$) relationship with cRFI in the univariate Cox PH regression analysis will be included in these analyses.

NORMALIZATION OF EXPRESSION LEVELS

[0109] The expression data used in the methods disclosed herein can be normalized. Normalization refers to a process to correct for (normalize away), for example, differences in the amount of RNA assayed and variability in the quality of the RNA used, to remove unwanted sources of systematic variation in Ct or Cp measurements, and the like. With respect to RT-PCR experiments involving archived fixed paraffin embedded tissue samples, sources of systematic variation are known to include the degree of RNA degradation relative to the age of the patient sample and the type of fixative used to store the sample. Other sources of systematic variation are attributable to laboratory processing conditions.

[0110] Assays can provide for normalization by incorporating the expression of certain normalizing genes, which do not significantly differ in expression levels under the relevant conditions. Exemplary normalization genes disclosed herein include housekeeping genes. (See, e.g., E. Eisenberg, et al., Trends in Genetics 19(7):362-365 (2003).) Normalization can be based on the mean or median signal (Ct or Cp) of all of the assayed genes or a large subset thereof (global normalization approach). In general, the normalizing genes, also referred to as reference genes should be genes that are known not to exhibit significantly different expression in prostate cancer as compared to non-cancerous prostate tissue, and are not significantly affected by various sample and process conditions, thus provide for normalizing away extraneous effects.

[0111] In exemplary embodiments, one or more of the following genes are used as references by which the mRNA or microRNA expression data is normalized: AAMP, ARF1, ATP5E, CLTC, GPS1, and PGK1. In another exemplary embodiment, one or more of the following microRNAs are used as references by which the expression data of microRNAs are normalized: hsa-miR-106a; hsa-miR-146b-5p; hsa-miR-191; hsa-miR-19b; and hsa-miR-92a. The calibrated weighted average C_T or Cp measurements for each of the prognostic and predictive genes or microRNAs may be normalized relative to the mean of five or more reference genes or microRNAs.

[0112] Those skilled in the art will recognize that normalization may be achieved in numerous ways, and the techniques described above are intended only to be exemplary, not exhaustive.

STANDARDIZATION OF EXPRESSION LEVELS

[0113] The expression data used in the methods disclosed herein can be standardized. Standardization refers to a process to effectively put all the genes or microRNAs on a comparable scale. This is performed because some genes or microRNAs will exhibit more variation (a broader range of expression) than others. Standardization is performed by dividing each expression value by its standard deviation across all samples for that gene or microRNA. Hazard ratios are then interpreted as the relative risk of recurrence per 1 standard deviation increase in expression.

Kits

[0114] The materials for use in the methods of the present invention are suited for preparation of kits produced in accordance with well-known procedures. The present disclosure thus provides kits comprising agents, which may include gene (or microRNA)-specific or gene (or microRNA)-selective probes and/or primers, for quantifying the expression of the disclosed genes or microRNAs for predicting prognostic outcome or response to treatment. Such kits may optionally contain reagents for the extraction of RNA from tumor samples, in particular fixed paraffin-embedded tissue samples and/or reagents for RNA amplification. In addition, the kits may optionally comprise the reagent(s) with an identifying description or label or instructions relating to their use in the methods of the present invention. The kits may comprise containers (including microliter plates suitable for use in an automated implementation of the method), each with one or more of the various materials or reagents (typically in concentrated form) utilized in the methods, including, for example, chromatographic columns, pre-fabricated microarrays, buffers, the appropriate nucleotide triphosphates (e.g., dATP, dCTP, dGTP and dTTP; or rATP, rCTP, rGTP and UTP), reverse transcriptase, DNA polymerase, RNA polymerase, and one or more probes and primers of the present invention (e.g., appropriate length poly(T) or random primers linked to a promoter reactive with the RNA polymerase). Mathematical algorithms used to estimate or quantify prognostic or predictive information are also properly potential components of kits.

Reports

[0115] The methods, when practiced for commercial diagnostic purposes, generally produce a report or summary of information obtained from the herein-described methods. For example, a report may include information concerning expression levels of one or more genes and /or microRNAs, classification of the tumor or the patient's risk of recurrence, the patient's likely prognosis or risk classification, clinical and pathologic factors, and/or other information. The methods and reports can further include storing the report in a database. The method can create a record in a database for the subject and populate the record with data. The report may be a paper report, an auditory report, or an electronic record. The report may be displayed and/or stored on a computing device (e.g., handheld device, desktop computer, smart device, website, etc.). It is contemplated that the

report is provided to a physician and/or the patient. The receiving of the report can further include establishing a network connection to a server computer that includes the data and report and requesting the data and report from the server computer.

Computer program

[0116] The values from the assays described above, such as expression data, can be calculated and stored manually. Alternatively, the above-described steps can be completely or partially performed by a computer program product. The computer program product includes a computer readable storage medium having a computer program stored on it. The program can, when read by a computer, execute relevant calculations based on values obtained from analysis of one or more biological sample from an individual (e.g., gene expression levels, normalization, standardization, thresholding, and conversion of values from assays to a score and/or text or graphical depiction of tumor stage and related information). The computer program product has stored therein a computer program for performing the calculation.

[0117] The present disclosure provides systems for executing the program described above, which system generally includes: a) a central computing environment; b) an input device, operatively connected to the computing environment, to receive patient data, wherein the patient data can include, for example, expression level or other value obtained from an assay using a biological sample from the patient, or microarray data, as described in detail above; c) an output device, connected to the computing environment, to provide information to a user (e.g., medical personnel); and d) an algorithm executed by the central computing environment (e.g., a processor), where the algorithm is executed based on the data received by the input device, and wherein the algorithm calculates an expression score, thresholding, or other functions described herein. The methods provided by the present invention may also be automated in whole or in part.

[0118] All aspects of the present invention may also be practiced such that a limited number of additional genes and/or microRNAs that are co-expressed or functionally related with the disclosed genes, for example as evidenced by statistically meaningful Pearson and/or Spearman correlation coefficients, are included in a test in addition to and/or in place of disclosed genes.

[0119] Having described the invention, the same will be more readily understood through reference to the following Examples, which are provided by way of illustration, and are not intended to limit the invention in any way.

EXAMPLES**EXAMPLE 1: RNA YIELD AND GENE EXPRESSION PROFILES IN PROSTATE CANCER BIOPSY CORES**

[0120] Clinical tools based on prostate needle core biopsies are needed to guide treatment planning at diagnosis for men with localized prostate cancer. Limiting tissue in needle core biopsy specimens poses significant challenges to the development of molecular diagnostic tests. This study examined RNA extraction yields and gene expression profiles using an RT-PCR assay to characterize RNA from manually micro-dissected fixed paraffin embedded (FPE) prostate cancer needle biopsy cores. It also investigated the association of RNA yields and gene expression profiles with Gleason score in these specimens.

Patients and Samples

[0121] This study determined the feasibility of gene expression profile analysis in prostate cancer needle core biopsies by evaluating the quantity and quality of RNA extracted from fixed paraffin-embedded (FPE) prostate cancer needle core biopsy specimens. Forty-eight (48) formalin-fixed blocks from prostate needle core biopsy specimens were used for this study. Classification of specimens was based on interpretation of the Gleason score (2005 Int'l Society of Urological Pathology Consensus Conference) and percentage tumor (<33%, 33-66%, >66%) involvement as assessed by pathologists.

Table 1: Distribution of cases

Gleason score Category	~<33% Tumor	~33-66% Tumor	~>66% Tumor
Low (≤ 6)	5	5	6
Intermediate (7)	5	5	6
High (8, 9, 10)	5	5	6
Total	15	15	18

Assay Methods

[0122] Fourteen (14) serial 5 μ m unstained sections from each FPE tissue block were included in the study. The first and last sections for each case were H&E stained and histologically reviewed to confirm the presence of tumor and for tumor enrichment by manual micro-dissection.

[0123] RNA from enriched tumor samples was extracted using a manual RNA extraction process. RNA was quantitated using the RiboGreen® assay and tested for the presence of genomic DNA contamination. Samples with sufficient RNA yield and free of genomic DNA tested for gene expression levels of a 24-gene panel of reference and cancer-related genes using quantitative RT-PCR. The expression was normalized to the average of 6 reference genes (AAMP, ARF1, ATP5E, CLTC, EEF1A1, and GPX1).

Statistical Methods

[0124] Descriptive statistics and graphical displays were used to summarize standard pathology metrics and gene expression, with stratification for Gleason Score category and percentage tumor involvement category. Ordinal logistic regression was used to evaluate the relationship between gene expression and Gleason Score category.

Results

[0125] The RNA yield per unit surface area ranged from 16 to 2406 ng/mm². Higher RNA yield was observed in samples with higher percent tumor involvement ($p=0.02$) and higher Gleason score ($p=0.01$). RNA yield was sufficient ($> 200\text{ng}$) in 71% of cases to permit 96-well RT-PCR, with 87% of cases having $>100\text{ng}$ RNA yield. The study confirmed that gene expression from prostate biopsies, as measured by qRT-PCR, was comparable to FPET samples used in commercial molecular assays for breast cancer. In addition, it was observed that greater biopsy RNA yields are found with higher Gleason score and higher percent tumor involvement. Nine genes were identified as significantly associated with Gleason score ($p < 0.05$) and there was a large dynamic range observed for many test genes.

EXAMPLE 2: GENE EXPRESSION ANALYSIS FOR GENES ASSOCIATED WITH PROGNOSIS IN PROSTATE CANCER

Patients and Samples

[0126] Approximately 2600 patients with clinical stage T1/T2 prostate cancer treated with radical prostatectomy (RP) at the Cleveland Clinic between 1987 and 2004 were identified. Patients were excluded from the study design if they received neo-adjuvant and/or adjuvant therapy, if pre-surgical PSA levels were missing, or if no tumor block was available from initial diagnosis. 127 patients with clinical recurrence and 374 patients without clinical recurrence after radical prostatectomy were randomly selected using a cohort sampling design. The specimens were stratified by T stage (T1, T2), year cohort (<1993 , ≥ 1993), and prostatectomy Gleason score (low/intermediate, high). Of the 501 sampled patients, 51 were excluded for insufficient tumor; 7 were excluded due to clinical ineligibility; 2 were excluded due to poor quality of gene expression data; and 10 were excluded because primary Gleason pattern was unavailable. Thus, this gene expression study included tissue and data from 111 patients with clinical recurrence and 330 patients without clinical recurrence after radical prostatectomies performed between 1987 and 2004 for treatment of early stage (T1, T2) prostate cancer.

[0127] Two fixed paraffin embedded (FPE) tissue specimens were obtained from prostate tumor specimens in each patient. The sampling method (sampling method A or B) depended on whether the highest Gleason pattern is also the primary Gleason pattern. For each specimen selected, the invasive cancer cells were at least 5.0 mm in dimension, except in the instances of pattern 5, where 2.2 mm was accepted. Specimens were spatially distinct where possible.

Table 2: Sampling Methods

Sampling Method A	Sampling Method B
For patients whose prostatectomy primary Gleason pattern is also the highest Gleason pattern	For patients whose prostatectomy primary Gleason pattern is not the highest Gleason pattern
Specimen 1 (A1) = primary Gleason pattern	Specimen 1 (B1) = highest Gleason pattern
Select and mark largest focus (greatest cross-sectional area) of primary Gleason pattern tissue. Invasive cancer area ≥ 5.0 mm.	Select highest Gleason pattern tissue from spatially distinct area from specimen B2, if possible. Invasive cancer area at least 5.0 mm if selecting secondary pattern, at least 2.2 mm if selecting Gleason pattern 5.
Specimen 2 (A2) = secondary Gleason pattern	Specimen 2 (B2) = primary Gleason pattern
Select and mark secondary Gleason pattern tissue from spatially distinct area from specimen A1. Invasive cancer area ≥ 5.0 mm.	Select largest focus (greatest cross-sectional area) of primary Gleason pattern tissue. Invasive cancer area ≥ 5.0 mm.

[0128] Histologically normal appearing tissue (NAT) adjacent to the tumor specimen (also referred to in these Examples as "non-tumor tissue") was also evaluated. Adjacent tissue was collected 3 mm from the tumor to 3 mm from the edge of the FPET block. NAT was preferentially sampled adjacent to the primary Gleason pattern. In cases where there was insufficient NAT adjacent to the primary Gleason pattern, then NAT was sampled adjacent to the secondary or highest Gleason pattern (A2 or B1) per the method set forth in Table 2. Six (6) 10 μ m sections with beginning H&E at 5 μ m and ending unstained slide at 5 μ m were prepared from each fixed paraffin-embedded tumor (FPET) block included in the study. All cases were histologically reviewed and manually micro-dissected to yield two enriched tumor samples and, where possible, one normal tissue sample adjacent to the tumor specimen.

Assay Method

[0129] In this study, RT-PCR analysis was used to determine RNA expression levels for 738 genes and chromosomal rearrangements (e.g., TMPRSS2-ERG fusion or other ETS family genes) in prostate cancer tissue and surrounding NAT in patients with early-stage prostate cancer treated with radical prostatectomy.

[0130] The samples were quantified using the RiboGreen assay and a subset tested for presence of genomic DNA contamination. Samples were taken into reverse transcription (RT) and quantitative polymerase chain reaction (qPCR). All analyses were conducted on reference-normalized gene expression levels using the average of the of replicate well crossing point (CP) values for the 6 reference genes (AAMP, ARF1, ATP5E, CLTC, GPS1, PGK1).

Statistical Analysis and Results

[0131] Primary statistical analyses involved 111 patients with clinical recurrence and 330 patients without clinical recurrence after radical prostatectomy for early-stage prostate cancer stratified by T-stage (T1, T2), year cohort (<1993, ≥1993), and prostatectomy Gleason score (low/intermediate, high). Gleason score categories are defined as follows: low (Gleason score ≤ 6), intermediate (Gleason score = 7), and high (Gleason score ≥ 8). A patient was included in a specified analysis if at least one sample for that patient was evaluable. Unless otherwise stated, all hypothesis tests were reported using two-sided p-values. The method of Storey was applied to the resulting set of p-values to control the false discovery rate (FDR) at 20%. J. Storey, R. Tibshirani, Estimating the Positive False Discovery Rate Under Dependence, with Applications to DNA Microarrays, Dept. of Statistics, Stanford Univ. (2001).

[0132] Analysis of gene expression and recurrence-free interval was based on univariate Cox Proportional Hazards (PH) models using maximum weighted pseudo-partial-likelihood estimators for each evaluable gene in the gene list (727 test genes and 5 reference genes). P-values were generated using Wald tests of the null hypothesis that the hazard ratio (HR) is one. Both unadjusted p-values and the q-value (smallest FDR at which the hypothesis test in question is rejected) were reported. Un-adjusted p-values <0.05 were considered statistically significant. Since two tumor specimens were selected for each patient, this analysis was performed using the 2 specimens from each patient as follows: (1) analysis using the primary Gleason pattern specimen from each patient (Specimens A1 and B2 as described in Table 2); (2) analysis using the highest Gleason pattern specimen from each patient (Specimens A1 and B1 as described in Table 2).

[0133] Analysis of gene expression and Gleason pattern (3, 4, 5) was based on univariate ordinal logistic regression models using weighted maximum likelihood estimators for each gene in the gene list (727 test genes and 5 reference genes). P-values were generated using a Wald test of the null hypothesis that the odds ratio (OR) is one. Both unadjusted p-values and the q-value (smallest FDR at which the hypothesis test in question is rejected) were reported. Un-adjusted p-values <0.05 were considered statistically significant. Since two tumor specimens were selected for each patient, this analysis was performed using the 2 specimens from each patient as follows: (1) analysis using the primary Gleason pattern specimen from each patient (Specimens A1 and B2 as described in Table 2); (2) analysis using the highest Gleason pattern specimen from each patient (Specimens A1 and B1 as described in Table 2).

[0134] It was determined whether there is a significant relationship between cRFI and selected demographic, clinical, and pathology variables, including age, race, clinical tumor stage, pathologic tumor stage, location of selected tumor specimens within the prostate (peripheral versus transitional zone), PSA at the time of surgery, overall Gleason score from the radical prostatectomy, year of surgery, and specimen Gleason pattern. Separately for each demographic or clinical variable, the relationship between the clinical covariate and cRFI was modeled using univariate Cox PH regression using weighted pseudo partial-likelihood estimators and a p-value was generated using Wald's test of the null hypothesis that the hazard ratio (HR) is one.

Covariates with unadjusted p-values <0.2 may have been included in the covariate-adjusted analyses.

[0135] It was determined whether there was a significant relationship between each of the individual cancer-related genes and cRFI after controlling for important demographic and clinical covariates. Separately for each gene, the relationship between gene expression and cRFI was modeled using multivariate Cox PH regression using weighted pseudo partial-likelihood estimators including important demographic and clinical variables as covariates. The independent contribution of gene expression to the prediction of cRFI was tested by generating a p-value from a Wald test using a model that included clinical covariates for each nodule (specimens as defined in Table 2). Un-adjusted p-values <0.05 were considered statistically significant.

[0136] Tables 3A and 3B provide genes significantly associated ($p < 0.05$), positively or negatively, with Gleason pattern in the primary and/or highest Gleason pattern. Increased expression of genes in Table 3A is positively associated with higher Gleason score, while increased expression of genes in Table 3B are negatively associated with higher Gleason score.

Table 3A

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) > 1.0 (Increased expression is positively associated with higher Gleason Score)				
Table 3A	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
ALCAM	1.73	<.001	1.36	0.009
ANLN	1.35	0.027		
APOC1	1.47	0.005	1.61	<.001
APOE	1.87	<.001	2.15	<.001
ASAP2	1.53	0.005		
ASPN	2.62	<.001	2.13	<.001
ATP5E	1.35	0.035		
AURKA	1.44	0.010		
AURKB	1.59	<.001	1.56	<.001
BAX	1.43	0.006		
BGN	2.58	<.001	2.82	<.001
BIRC5	1.45	0.003	1.79	<.001
BMP6	2.37	<.001	1.68	<.001
BMPR1B	1.58	0.002		
BRCA2			1.45	0.013
BUB1	1.73	<.001	1.57	<.001
CACNA1D	1.31	0.045	1.31	0.033
CADPS			1.30	0.023
CCNB1	1.43	0.023		

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) > 1.0 (Increased expression is positively associated with higher Gleason Score)

Table 3A	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
CCNE2	1.52	0.003	1.32	0.035
CD276	2.20	<.001	1.83	<.001
CD68			1.36	0.022
CDC20	1.69	<.001	1.95	<.001
CDC6	1.38	0.024	1.46	<.001
CDH11			1.30	0.029
CDKN2B	1.55	0.001	1.33	0.023
CDKN2C	1.62	<.001	1.52	<.001
CDKN3	1.39	0.010	1.50	0.002
CENPF	1.96	<.001	1.71	<.001
CHRA1			1.34	0.022
CLDN3			1.37	0.029
COL1A1	2.23	<.001	2.22	<.001
COL1A2			1.42	0.005
COL3A1	1.90	<.001	2.13	<.001
COL8A1	1.88	<.001	2.35	<.001
CRISP3	1.33	0.040	1.26	0.050
CTHRC1	2.01	<.001	1.61	<.001
CTNND2	1.48	0.007	1.37	0.011
DAPK1	1.44	0.014		
DIAPH1	1.34	0.032	1.79	<.001
DIO2			1.56	0.001
DLL4	1.38	0.026	1.53	<.001
ECE1	1.54	0.012	1.40	0.012
ENY2	1.35	0.046	1.35	0.012
EZH2	1.39	0.040		
F2R	2.37	<.001	2.60	<.001
FAM49B	1.57	0.002	1.33	0.025
FAP	2.36	<.001	1.89	<.001
FCGR3A	2.10	<.001	1.83	<.001
GNPTAB	1.78	<.001	1.54	<.001
GSK3B			1.39	0.018

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) > 1.0 (Increased expression is positively associated with higher Gleason Score)

Table 3A	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
HRAS	1.62	0.003		
HSD17B4	2.91	<.001	1.57	<.001
HSPA8	1.48	0.012	1.34	0.023
IFI30	1.64	<.001	1.45	0.013
IGFBP3			1.29	0.037
IL11	1.52	0.001	1.31	0.036
INHBA	2.55	<.001	2.30	<.001
ITGA4			1.35	0.028
JAG1	1.68	<.001	1.40	0.005
KCNN2	1.50	0.004		
KCTD12			1.38	0.012
KHDRBS3	1.85	<.001	1.72	<.001
KIF4A	1.50	0.010	1.50	<.001
KLK14	1.49	0.001	1.35	<.001
KPNA2	1.68	0.004	1.65	0.001
KRT2			1.33	0.022
KRT75			1.27	0.028
LAMC1	1.44	0.029		
LAPTM5	1.36	0.025	1.31	0.042
LTBP2	1.42	0.023	1.66	<.001
MANF			1.34	0.019
MAOA	1.55	0.003	1.50	<.001
MAP3K5	1.55	0.006	1.44	0.001
MDK	1.47	0.013	1.29	0.041
MDM2			1.31	0.026
MELK	1.64	<.001	1.64	<.001
MMP11	2.33	<.001	1.66	<.001
MYBL2	1.41	0.007	1.54	<.001
MYO6			1.32	0.017
NETO2			1.36	0.018
NOX4	1.84	<.001	1.73	<.001
NPM1	1.68	0.001		

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) > 1.0 (Increased expression is positively associated with higher Gleason Score)

Table 3A	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
NRIP3			1.36	0.009
NRP1	1.80	0.001	1.36	0.019
OSM	1.33	0.046		
PATE1	1.38	0.032		
PECAM1	1.38	0.021	1.31	0.035
PGD	1.56	0.010		
PLK1	1.51	0.004	1.49	0.002
PLOD2			1.29	0.027
POSTN	1.70	0.047	1.55	0.006
PPP3CA	1.38	0.037	1.37	0.006
PTK6	1.45	0.007	1.53	<.001
PTTG1			1.51	<.001
RAB31			1.31	0.030
RAD21	2.05	<.001	1.38	0.020
RAD51	1.46	0.002	1.26	0.035
RAF1	1.46	0.017		
RALBP1	1.37	0.043		
RHOC			1.33	0.021
ROBO2	1.52	0.003	1.41	0.006
RRM2	1.77	<.001	1.50	<.001
SAT1	1.67	0.002	1.61	<.001
SDC1	1.66	0.001	1.46	0.014
SEC14L1	1.53	0.003	1.62	<.001
SESN3	1.76	<.001	1.45	<.001
SFRP4	2.69	<.001	2.03	<.001
SHMT2	1.69	0.007	1.45	0.003
SKIL			1.46	0.005
SOX4	1.42	0.016	1.27	0.031
SPARC	1.40	0.024	1.55	<.001
SPINK 1			1.29	0.002
SPP1	1.51	0.002	1.80	<.001
TFDP1	1.48	0.014		

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) > 1.0 (Increased expression is positively associated with higher Gleason Score)

Table 3A	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
THBS2	1.87	$< .001$	1.65	$< .001$
THY1	1.58	0.003	1.64	$< .001$
TK1	1.79	$< .001$	1.42	0.001
TOP2A	2.30	$< .001$	2.01	$< .001$
TPD52	1.95	$< .001$	1.30	0.037
TPX2	2.12	$< .001$	1.86	$< .001$
TYMP	1.36	0.020		
TYMS	1.39	0.012	1.31	0.036
UBE2C	1.66	$< .001$	1.65	$< .001$
UBE2T	1.59	$< .001$	1.33	0.017
UGDH			1.28	0.049
UGT2B15	1.46	0.001	1.25	0.045
UHRF1	1.95	$< .001$	1.62	$< .001$
VDR	1.43	0.010	1.39	0.018
WNT5A	1.54	0.001	1.44	0.013

Table 3B.

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
ABCA5	0.78	0.041		
ABCG2	0.65	0.001	0.72	0.012
ACOX2	0.44	$< .001$	0.53	$< .001$
ADH5	0.45	$< .001$	0.42	$< .001$
AFAP1			0.79	0.038
AIG1			0.77	0.024
AKAP1	0.63	0.002		
AKR1C1	0.66	0.003	0.63	$< .001$
AKT3	0.68	0.006	0.77	0.010
ALDH1A2	0.28	$< .001$	0.33	$< .001$
ALKBH3	0.77	0.040	0.77	0.029
AMPD3	0.67	0.007		

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
ANPEP	0.68	0.008	0.59	<.001
ANXA2	0.72	0.018		
APC			0.69	0.002
AXIN2	0.46	<.001	0.54	<.001
AZGP1	0.52	<.001	0.53	<.001
BIK	0.69	0.006	0.73	0.003
BIN1	0.43	<.001	0.61	<.001
BTG3			0.79	0.030
BTRC	0.48	<.001	0.62	<.001
C7	0.37	<.001	0.55	<.001
CADM1	0.56	<.001	0.69	0.001
CAV1	0.58	0.002	0.70	0.009
CAV2	0.65	0.029		
CCNH	0.67	0.006	0.77	0.048
CD164	0.59	0.003	0.57	<.001
CDC25B	0.77	0.035		
CDH1			0.66	<.001
CDK2			0.71	0.003
CDKN1C	0.58	<.001	0.57	<.001
CDS2			0.69	0.002
CHN1	0.66	0.002		
COL6A1	0.44	<.001	0.66	<.001
COL6A3	0.66	0.006		
CSRP1	0.42	0.006		
CTGF	0.74	0.043		
CTNNA1	0.70	<.001	0.83	0.018
CTNNB1	0.70	0.019		
CTNND1			0.75	0.028
CUL1			0.74	0.011
CXCL12	0.54	<.001	0.74	0.006
CYP3A5	0.52	<.001	0.66	0.003
CYR61	0.64	0.004	0.68	0.005

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
DDR2	0.57	0.002	0.73	0.004
DES	0.34	$< .001$	0.58	$< .001$
DLGAP1	0.54	$< .001$	0.62	$< .001$
DNM3	0.67	0.004		
DPP4	0.41	$< .001$	0.53	$< .001$
DPT	0.28	$< .001$	0.48	$< .001$
DUSP1	0.59	$< .001$	0.63	$< .001$
EDNRA	0.64	0.004	0.74	0.008
EGF			0.71	0.012
EGR1	0.59	$< .001$	0.67	0.009
EGR3	0.72	0.026	0.71	0.025
EIF5			0.76	0.025
ELK4	0.58	0.001	0.70	0.008
ENPP2	0.66	0.002	0.70	0.005
EPHA3	0.65	0.006		
EPHB2	0.60	$< .001$	0.78	0.023
EPHB4	0.75	0.046	0.73	0.006
ERBB3	0.76	0.040	0.75	0.013
ERBB4			0.74	0.023
ERCC1	0.63	$< .001$	0.77	0.016
FAAH	0.67	0.003	0.71	0.010
FAM107A	0.35	$< .001$	0.59	$< .001$
FAM13C	0.37	$< .001$	0.48	$< .001$
FAS	0.73	0.019	0.72	0.008
FGF10	0.53	$< .001$	0.58	$< .001$
FGF7	0.52	$< .001$	0.59	$< .001$
FGFR2	0.60	$< .001$	0.59	$< .001$
FKBP5	0.70	0.039	0.68	0.003
FLNA	0.39	$< .001$	0.56	$< .001$
FLNC	0.33	$< .001$	0.52	$< .001$
FOS	0.58	$< .001$	0.66	0.005
FOXO1	0.57	$< .001$	0.67	$< .001$

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
FOXQ1			0.74	0.023
GADD45B	0.62	0.002	0.71	0.010
GHR	0.62	0.002	0.72	0.009
GNRH1	0.74	0.049	0.75	0.026
GPM6B	0.48	$< .001$	0.68	$< .001$
GPS1			0.68	0.003
GSN	0.46	$< .001$	0.77	0.027
GSTM1	0.44	$< .001$	0.62	$< .001$
GSTM2	0.29	$< .001$	0.49	$< .001$
HGD			0.77	0.020
HIRIP3	0.75	0.034		
HK1	0.48	$< .001$	0.66	0.001
HLF	0.42	$< .001$	0.55	$< .001$
HNF1B	0.67	0.006	0.74	0.010
HPS1	0.66	0.001	0.65	$< .001$
HSP90AB1	0.75	0.042		
HSPA5	0.70	0.011		
HSPB2	0.52	$< .001$	0.70	0.004
IGF1	0.35	$< .001$	0.59	$< .001$
IGF2	0.48	$< .001$	0.70	0.005
IGFBP2	0.61	$< .001$	0.77	0.044
IGFBP5	0.63	$< .001$		
IGFBP6	0.45	$< .001$	0.64	$< .001$
IL6ST	0.55	0.004	0.63	$< .001$
ILK	0.40	$< .001$	0.57	$< .001$
ING5	0.56	$< .001$	0.78	0.033
ITGA1	0.56	0.004	0.61	$< .001$
ITGA3			0.78	0.035
ITGA5	0.71	0.019	0.75	0.017
ITGA7	0.37	$< .001$	0.52	$< .001$
ITGB3	0.63	0.003	0.70	0.005
ITPR1	0.46	$< .001$	0.64	$< .001$

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
ITPR3	0.70	0.013		
ITSN1	0.62	0.001		
JUN	0.48	$< .001$	0.60	$< .001$
JUNB	0.72	0.025		
KIT	0.51	$< .001$	0.68	0.007
KLC1	0.58	$< .001$		
KLK1	0.69	0.028	0.66	0.003
KLK2	0.60	$< .001$		
KLK3	0.63	$< .001$	0.69	0.012
KRT15	0.56	$< .001$	0.60	$< .001$
KRT18	0.74	0.034		
KRT5	0.64	$< .001$	0.62	$< .001$
LAMA4	0.47	$< .001$	0.73	0.010
LAMB3	0.73	0.018	0.69	0.003
LGALS3	0.59	0.003	0.54	$< .001$
LIG3	0.75	0.044		
MAP3K7	0.66	0.003	0.79	0.031
MCM3	0.73	0.013	0.80	0.034
MGMT	0.61	0.001	0.71	0.007
MGST1			0.75	0.017
MLXIP	0.70	0.013		
MMP2	0.57	$< .001$	0.72	0.010
MMP7	0.69	0.009		
MPPED2	0.70	0.009	0.59	$< .001$
MSH6	0.78	0.046		
MTA1	0.69	0.007		
MTSS1	0.55	$< .001$	0.54	$< .001$
MYBPC1	0.45	$< .001$	0.45	$< .001$
NCAM1	0.51	$< .001$	0.65	$< .001$
NCAPD3	0.42	$< .001$	0.53	$< .001$
NCOR2	0.68	0.002		
NDUFS5	0.66	0.001	0.70	0.013

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
NEXN	0.48	$< .001$	0.62	$< .001$
NFAT5	0.55	$< .001$	0.67	0.001
NFKBIA			0.79	0.048
NRG1	0.58	0.001	0.62	0.001
OLFML3	0.42	$< .001$	0.58	$< .001$
OMD	0.67	0.004	0.71	0.004
OR51E2	0.65	$< .001$	0.76	0.007
PAGE4	0.27	$< .001$	0.46	$< .001$
PCA3	0.68	0.004		
PCDHGB7	0.70	0.025	0.65	$< .001$
PGF	0.62	0.001		
PGR	0.63	0.028		
PHTF2	0.69	0.033		
PLP2	0.54	$< .001$	0.71	0.003
PPAP2B	0.41	$< .001$	0.54	$< .001$
PPP1R12A	0.48	$< .001$	0.60	$< .001$
PRIMA1	0.62	0.003	0.65	$< .001$
PRKAR1B	0.70	0.009		
PRKAR2B			0.79	0.038
PRKCA	0.37	$< .001$	0.55	$< .001$
PRKCB	0.47	$< .001$	0.56	$< .001$
PTCH1	0.70	0.021		
PTEN	0.66	0.010	0.64	$< .001$
PTGER3			0.76	0.015
PTGS2	0.70	0.013	0.68	0.005
PTH1R	0.48	$< .001$		
PTK2B	0.67	0.014	0.69	0.002
PYCARD	0.72	0.023		
RAB27A			0.76	0.017
RAGE	0.77	0.040	0.57	$< .001$
RARB	0.66	0.002	0.69	0.002
RECK	0.65	$< .001$		

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
RHOA	0.73	0.043		
RHOB	0.61	0.005	0.62	$< .001$
RND3	0.63	0.006	0.66	$< .001$
SDHC			0.69	0.002
SEC23A	0.61	$< .001$	0.74	0.010
SEMA3A	0.49	$< .001$	0.55	$< .001$
SERPINA3	0.70	0.034	0.75	0.020
SH3RF2	0.33	$< .001$	0.42	$< .001$
SLC22A3	0.23	$< .001$	0.37	$< .001$
SMAD4	0.33	$< .001$	0.39	$< .001$
SMARCC2	0.62	0.003	0.74	0.008
SMO	0.53	$< .001$	0.73	0.009
SORBS 1	0.40	$< .001$	0.55	$< .001$
SPARCL1	0.42	$< .001$	0.63	$< .001$
SRD5A2	0.28	$< .001$	0.37	$< .001$
ST5	0.52	$< .001$	0.63	$< .001$
STAT5A	0.60	$< .001$	0.75	0.020
STAT5B	0.54	$< .001$	0.65	$< .001$
STS			0.78	0.035
SUMO1	0.75	0.017	0.71	0.002
SVIL	0.45	$< .001$	0.62	$< .001$
TARP	0.72	0.017		
TGFB1I1	0.37	$< .001$	0.53	$< .001$
TGFB2	0.61	0.025	0.59	$< .001$
TGFB3	0.46	$< .001$	0.60	$< .001$
TIMP2	0.62	0.001		
TIMP3	0.55	$< .001$	0.76	0.019
TMPRSS2	0.71	0.014		
TNF	0.65	0.010		
TNFRSF10A	0.71	0.014	0.74	0.010
TNFRSF10B	0.74	0.030	0.73	0.016
TNFSF10			0.69	0.004

Gene significantly ($p < 0.05$) associated with Gleason pattern for all specimens in the primary Gleason pattern or highest Gleason pattern odds ratio (OR) < 1.0 (Increased expression is negatively associated with higher Gleason score)

Table 3B	Primary Pattern		Highest Pattern	
Official Symbol	OR	p-value	OR	p-value
TP53			0.73	0.011
TP63	0.62	$< .001$	0.68	0.003
TPM1	0.43	$< .001$	0.47	$< .001$
TPM2	0.30	$< .001$	0.47	$< .001$
TPP2	0.58	$< .001$	0.69	0.001
TRA2A	0.71	0.006		
TRAF3IP2	0.50	$< .001$	0.63	$< .001$
TRO	0.40	$< .001$	0.59	$< .001$
TRPC6	0.73	0.030		
TRPV6			0.80	0.047
VCL	0.44	$< .001$	0.55	$< .001$
VEGFB	0.73	0.029		
VIM	0.72	0.013		
VTI1B	0.78	0.046		
WDR19	0.65	$< .001$		
WFDC1	0.50	$< .001$	0.72	0.010
YY1	0.75	0.045		
ZFHX3	0.52	$< .001$	0.54	$< .001$
ZFP36	0.65	0.004	0.69	0.012
ZNF827	0.59	$< .001$	0.69	0.004

[0137] To identify genes associated with recurrence (cRFI, bRFI) in the primary and the highest Gleason pattern, each of 727 genes were analyzed in univariate models using specimens A1 and B2 (see Table 2, above). Tables 4A and 4B provide genes that were associated, positively or negatively, with cRFI and/or bRFI in the primary and/or highest Gleason pattern. Increased expression of genes in Table 4A is negatively associated with good prognosis, while increased expression of genes in Table 4B is positively associated with good prognosis.

Table 4A.

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)								
Table 4A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
AKR1C3	1.304	0.022	1.312	0.013				
ANLN	1.379	0.002	1.579	<.001	1.465	<.001	1.623	<.001
AQP2	1.184	0.027	1.276	<.001				
ASAP2			1.442	0.006				
ASPN	2.272	<.001	2.106	<.001	1.861	<.001	1.895	<.001
ATP5E	1.414	0.013	1.538	<.001				
BAG5			1.263	0.044				
BAX			1.332	0.026	1.327	0.012	1.438	0.002
BGN	1.947	<.001	2.061	<.001	1.339	0.017		
BIRC5	1.497	<.001	1.567	<.001	1.478	<.001	1.575	<.001
BMP6	1.705	<.001	2.016	<.001	1.418	0.004	1.541	<.001
BMPR1B	1.401	0.013			1.325	0.016		
BRCA2							1.259	0.007
BUB1	1.411	<.001	1.435	<.001	1.352	<.001	1.242	0.002
CADPS					1.387	0.009	1.294	0.027
CCNB1					1.296	0.016	1.376	0.002
CCNE2	1.468	<.001	1.649	<.001	1.729	<.001	1.563	<.001
CD276	1.678	<.001	1.832	<.001	1.581	<.001	1.385	0.002
CDC20	1.547	<.001	1.671	<.001	1.446	<.001	1.540	<.001
CDC6	1.400	0.003	1.290	0.030	1.403	0.002	1.276	0.019
CDH7	1.403	0.003	1.413	0.002				
CDKN2B	1.569	<.001	1.752	<.001	1.333	0.017	1.347	0.006
CDKN2C	1.612	<.001	1.780	<.001	1.323	0.005	1.335	0.004
CDKN3	1.384	<.001	1.255	0.024	1.285	0.003	1.216	0.028
CENPF	1.578	<.001	1.692	<.001	1.740	<.001	1.705	<.001
CKS2	1.390	0.007	1.418	0.005	1.291	0.018		
CLTC			1.368	0.045				
COL1A1	1.873	<.001	2.103	<.001	1.491	<.001	1.472	<.001
COL1A2			1.462	0.001				
COL3A1	1.827	<.001	2.005	<.001	1.302	0.012	1.298	0.018

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)								
Table 4A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
COL4A1	1.490	0.002	1.613	<.001				
COL8A1	1.692	<.001	1.926	<.001	1.307	0.013	1.317	0.010
CRISP3	1.425	0.001	1.467	<.001	1.242	0.045		
CTHRC1	1.505	0.002	2.025	<.001	1.425	0.003	1.369	0.005
CTNND2					1.412	0.003		
CXCR4	1.312	0.023	1.355	0.008				
DDIT4	1.543	<.001	1.763	<.001				
DYNLL1	1.290	0.039					1.201	0.004
EIF3H					1.428	0.012		
ENY2	1.361	0.014			1.392	0.008	1.371	0.001
EZH2			1.311	0.010				
F2R	1.773	<.001	1.695	<.001	1.495	<.001	1.277	0.018
FADD			1.292	0.018				
FAM171B			1.285	0.036				
FAP	1.455	0.004	1.560	0.001	1.298	0.022	1.274	0.038
FASN	1.263	0.035						
FCGR3A			1.654	<.001	1.253	0.033	1.350	0.007
FGF5	1.219	0.030						
GNPTAB	1.388	0.007	1.503	0.003	1.355	0.005	1.434	0.002
GPR68			1.361	0.008				
GREM1	1.470	0.003	1.716	<.001	1.421	0.003	1.316	0.017
HDAC1					1.290	0.025		
HDAC9			1.395	0.012				
HRAS	1.424	0.006	1.447	0.020				
HSD17B4	1.342	0.019	1.282	0.026	1.569	<.001	1.390	0.002
HSPA8	1.290	0.034						
IGFBP3	1.333	0.022	1.442	0.003	1.253	0.040	1.323	0.005
INHBA	2.368	<.001	2.765	<.001	1.466	0.002	1.671	<.001
JAG1	1.359	0.006	1.367	0.005	1.259	0.024		
KCNN2	1.361	0.011	1.413	0.005	1.312	0.017	1.281	0.030
KHDRBS3	1.387	0.006	1.601	<.001	1.573	<.001	1.353	0.006

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)								
Table 4A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
KIAA0196							1.249	0.037
KIF4A	1.212	0.016			1.149	0.040	1.278	0.003
KLK14	1.167	0.023					1.180	0.007
KPNA2			1.425	0.009	1.353	0.005	1.305	0.019
KRT75							1.164	0.028
LAMA3					1.327	0.011		
LAMB1			1.347	0.019				
LAMC1	1.555	0.001	1.310	0.030			1.349	0.014
LIMS1							1.275	0.022
LOX					1.358	0.003	1.410	<.001
LTBP2	1.396	0.009	1.656	<.001	1.278	0.022		
LUM			1.315	0.021				
MANF					1.660	<.001	1.323	0.011
MCM2					1.345	0.011	1.387	0.014
MCM6	1.307	0.023	1.352	0.008			1.244	0.039
MELK	1.293	0.014	1.401	<.001	1.501	<.001	1.256	0.012
MMP11	1.680	<.001	1.474	<.001	1.489	<.001	1.257	0.030
MRPL13							1.260	0.025
MSH2			1.295	0.027				
MYBL2	1.664	<.001	1.670	<.001	1.399	<.001	1.431	<.001
MYO6			1.301	0.033				
NETO2	1.412	0.004	1.302	0.027	1.298	0.009		
NFKB1					1.236	0.050		
NOX4	1.492	<.001	1.507	0.001	1.555	<.001	1.262	0.019
NPM1					1.287	0.036		
NRIP3			1.219	0.031			1.218	0.018
NRP1			1.482	0.002			1.245	0.041
OLFML2B			1.362	0.015				
OR51E1					1.531	<.001	1.488	0.003
PAK6			1.269	0.033				

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)								
Table 4A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
PATE1	1.308	<.001	1.332	<.001	1.164	0.044		
PCNA							1.278	0.020
PEX10	1.436	0.005	1.393	0.009				
PGD	1.298	0.048			1.579	<.001		
PGK1			1.274	0.023			1.262	0.009
PLA2G7					1.315	0.011	1.346	0.005
PLAU					1.319	0.010		
PLK1	1.309	0.021	1.563	<.001	1.410	0.002	1.372	0.003
PLOD2			1.284	0.019	1.272	0.014	1.332	0.005
POSTN	1.599	<.001	1.514	0.002	1.391	0.005		
PPP3CA					1.402	0.007	1.316	0.018
PSMD13	1.278	0.040	1.297	0.033	1.279	0.017	1.373	0.004
PTK6	1.640	<.001	1.932	<.001	1.369	0.001	1.406	<.001
PTTG1	1.409	<.001	1.510	<.001	1.347	0.001	1.558	<.001
RAD21	1.315	0.035	1.402	0.004	1.589	<.001	1.439	<.001
RAF1					1.503	0.002		
RALA	1.521	0.004	1.403	0.007	1.563	<.001	1.229	0.040
RALBP1					1.277	0.033		
RGS7	1.154	0.015	1.266	0.010				
RRM1	1.570	0.001	1.602	<.001				
RRM2	1.368	<.001	1.289	0.004	1.396	<.001	1.230	0.015
SAT1	1.482	0.016	1.403	0.030				
SDC1					1.340	0.018	1.396	0.018
SEC14L1			1.260	0.048			1.360	0.002
SESN3	1.485	<.001	1.631	<.001	1.232	0.047	1.292	0.014
SFRP4	1.800	<.001	1.814	<.001	1.496	<.001	1.289	0.027
SHMT2	1.807	<.001	1.658	<.001	1.673	<.001	1.548	<.001
SKIL					1.327	0.008		
SLC25A21					1.398	0.001	1.285	0.018
SOX4					1.286	0.020	1.280	0.030
SPARC	1.539	<.001	1.842	<.001			1.269	0.026

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)								
Table 4A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
SPP1			1.322	0.022				
SQLE			1.359	0.020	1.270	0.036		
STMN1	1.402	0.007	1.446	0.005	1.279	0.031		
SULF1			1.587	<.001				
TAF2							1.273	0.027
TFDP1			1.328	0.021	1.400	0.005	1.416	0.001
THBS2	1.812	<.001	1.960	<.001	1.320	0.012	1.256	0.038
THY1	1.362	0.020	1.662	<.001				
TK1			1.251	0.011	1.377	<.001	1.401	<.001
TOP2A	1.670	<.001	1.920	<.001	1.869	<.001	1.927	<.001
TPD52	1.324	0.011			1.366	0.002	1.351	0.005
TPX2	1.884	<.001	2.154	<.001	1.874	<.001	1.794	<.001
UAP1					1.244	0.044		
UBE2C	1.403	<.001	1.541	<.001	1.306	0.002	1.323	<.001
UBE2T	1.667	<.001	1.282	0.023	1.502	<.001	1.298	0.005
UGT2B 15			1.295	0.001			1.275	0.002
UGT2B17							1.294	0.025
UHRF1	1.454	<.001	1.531	<.001	1.257	0.029		
VCPIP1	1.390	0.009	1.414	0.004	1.294	0.021	1.283	0.021
WNT5A			1.274	0.038	1.298	0.020		
XIAP					1.464	0.006		
ZMYND8			1.277	0.048				
ZWINT	1.259	0.047						

Table 4B.

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
AAMP	0.564	<.001	0.571	<.001	0.764	0.037	0.786	0.034
ABCA5	0.755	<.001	0.695	<.001			0.800	0.006
ABCB1	0.777	0.026						
ABCG2	0.788	0.033	0.784	0.040	0.803	0.018	0.750	0.004
ABHD2			0.734	0.011				
ACE			0.782	0.048				
ACOX2	0.639	<.001	0.631	<.001	0.713	<.001	0.716	0.002
ADH5	0.625	<.001	0.637	<.001	0.753	0.026		
AKAP1	0.764	0.006	0.800	0.005	0.837	0.046		
AKR1C1	0.773	0.033			0.802	0.032		
AKT1			0.714	0.005				
AKT3	0.811	0.015	0.809	0.021				
ALDH1A2	0.606	<.001	0.498	<.001	0.613	<.001	0.624	<.001
AMPD3					0.793	0.024		
ANPEP	0.584	<.001	0.493	<.001				
ANXA2	0.753	0.013	0.781	0.036	0.762	0.008	0.795	0.032
APRT			0.758	0.026	0.780	0.044	0.746	0.008
ATXN1	0.673	0.001	0.776	0.029	0.809	0.031	0.812	0.043
AXIN2	0.674	<.001	0.571	<.001	0.776	0.005	0.757	0.005
AZGP1	0.585	<.001	0.652	<.001	0.664	<.001	0.746	<.001
BAD			0.765	0.023				
BCL2	0.788	0.033	0.778	0.036				
BDKRB1	0.728	0.039						
BIK			0.712	0.005				
BIN1	0.607	<.001	0.724	0.002	0.726	<.001	0.834	0.034
BTG3					0.847	0.034		
BTRC	0.688	0.001	0.713	0.003				
C7	0.589	<.001	0.639	<.001	0.629	<.001	0.691	<.001
CADM1	0.546	<.001	0.529	<.001	0.743	0.008	0.769	0.015
CASP1	0.769	0.014	0.799	0.028	0.799	0.010	0.815	0.018

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
CAV1	0.736	0.011	0.711	0.005	0.675	<.001	0.743	0.006
CAV2			0.636	0.010	0.648	0.012	0.685	0.012
CCL2	0.759	0.029	0.764	0.024				
CCNH	0.689	<.001	0.700	<.001				
CD164	0.664	<.001	0.651	<.001				
CD1A					0.687	0.004		
CD44	0.545	<.001	0.600	<.001	0.788	0.018	0.799	0.023
CD82	0.771	0.009	0.748	0.004				
CDC25B	0.755	0.006			0.817	0.025		
CDK14	0.845	0.043						
CDK2							0.819	0.032
CDK3	0.733	0.005			0.772	0.006	0.838	0.017
CDKN1A			0.766	0.041				
CDKN1C	0.662	<.001	0.712	0.002	0.693	<.001	0.761	0.009
CHN1	0.788	0.036						
COL6A1	0.608	<.001	0.767	0.013	0.706	<.001	0.775	0.007
CSF1	0.626	<.001	0.709	0.003				
CSK					0.837	0.029		
CSRP1	0.793	0.024	0.782	0.019				
CTNNB1	0.898	0.042			0.885	<.001		
CTSB	0.701	0.004	0.713	0.007	0.715	0.002	0.803	0.038
CTSK					0.815	0.042		
CXCL12	0.652	<.001	0.802	0.044	0.711	0.001		
CYP3A5	0.463	<.001	0.436	<.001	0.727	0.003		
CYR61	0.652	0.002	0.676	0.002				
DAP			0.761	0.026	0.775	0.025	0.802	0.048
DARC					0.725	0.005	0.792	0.032
DDR2					0.719	0.001	0.763	0.008
DES	0.619	<.001	0.737	0.005	0.638	<.001	0.793	0.017
DHRS9	0.642	0.003						
DHX9	0.888	<.001						

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
DLC1	0.710	0.007	0.715	0.009				
DLGAP1	0.613	<.001	0.551	<.001			0.779	0.049
DNM3	0.679	<.001			0.812	0.037		
DPP4	0.591	<.001	0.613	<.001	0.761	0.003		
DPT	0.613	<.001	0.576	<.001	0.647	<.001	0.677	<.001
DUSP1	0.662	0.001	0.665	0.001			0.785	0.024
DUSP6	0.713	0.005	0.668	0.002				
EDNRA	0.702	0.002	0.779	0.036				
EGF			0.738	0.028				
EGR1	0.569	<.001	0.577	<.001			0.782	0.022
EGR3	0.601	<.001	0.619	<.001			0.800	0.038
EIF2S3							0.756	0.015
EIF5	0.776	0.023	0.787	0.028				
ELK4	0.628	<.001	0.658	<.001				
EPHA2	0.720	0.011	0.663	0.004				
EPHA3	0.727	0.003			0.772	0.005		
ERBB2	0.786	0.019	0.738	0.003	0.815	0.041		
ERBB3	0.728	0.002	0.711	0.002	0.828	0.043	0.813	0.023
ERCC1	0.771	0.023	0.725	0.007	0.806	0.049	0.704	0.002
EREG					0.754	0.016	0.777	0.034
ESR2			0.731	0.026				
FAAH	0.708	0.004	0.758	0.012	0.784	0.031	0.774	0.007
FAM107A	0.517	<.001	0.576	<.001	0.642	<.001	0.656	<.001
FAM13C	0.568	<.001	0.526	<.001	0.739	0.002	0.639	<.001
FAS	0.755	0.014						
FASLG			0.706	0.021				
FGF10	0.653	<.001			0.685	<.001	0.766	0.022
FGF17			0.746	0.023	0.781	0.015	0.805	0.028
FGF7	0.794	0.030			0.820	0.037	0.811	0.04
FGFR2	0.683	<.001	0.686	<.001	0.674	<.001	0.703	<.001

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
FKBP5			0.676	0.001				
FLNA	0.653	<.001	0.741	0.010	0.682	<.001	0.771	0.016
FLNC	0.751	0.029	0.779	0.047	0.663	<.001	0.725	<.001
FLT1			0.799	0.044				
FOS	0.566	<.001	0.543	<.001			0.757	0.006
FOXO1					0.816	0.039	0.798	0.023
FOXQ1	0.753	0.017	0.757	0.024	0.804	0.018		
FYN	0.779	0.031						
GADD45B	0.590	<.001	0.619	<.001				
GDF15	0.759	0.019	0.794	0.048				
GHR	0.702	0.005	0.630	<.001	0.673	<.001	0.590	<.001
GNRH1					0.742	0.014		
GPM6B	0.653	<.001	0.633	<.001	0.696	<.001	0.768	0.007
GSN	0.570	<.001	0.697	0.001	0.697	<.001	0.758	0.005
GSTM1	0.612	<.001	0.588	<.001	0.718	<.001	0.801	0.020
GSTM2	0.540	<.001	0.630	<.001	0.602	<.001	0.706	<.001
HGD	0.796	0.020	0.736	0.002				
HIRIP3	0.753	0.011			0.824	0.050		
HK1	0.684	<.001	0.683	<.001	0.799	0.011	0.804	0.014
HLA-G			0.726	0.022				
HLF	0.555	<.001	0.582	<.001	0.703	<.001	0.702	<.001
HNF1B	0.690	<.001	0.585	<.001				
HPS1	0.744	0.003	0.784	0.020	0.836	0.047		
HSD3B2							0.733	0.016
HSP90AB1	0.801	0.036						
HSPA5			0.776	0.034				
HSPB1	0.813	0.020						
HSPB2	0.762	0.037			0.699	0.002	0.783	0.034
HSPG2					0.794	0.044		
ICAM1	0.743	0.024	0.768	0.040				
IER3	0.686	0.002	0.663	<.001				

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
KRT15	0.627	<.001	0.526	<.001	0.704	<.001	0.782	0.029
KRT18	0.624	<.001	0.617	<.001	0.738	0.005	0.760	0.005
KRT5	0.640	<.001	0.550	<.001	0.740	<.001	0.798	0.023
KRT8	0.716	0.006	0.744	0.008				
L1CAM	0.738	0.021	0.692	0.009			0.761	0.036
LAG3	0.741	0.013	0.729	0.011				
LAMA4	0.686	0.011			0.592	0.003		
LAMA5							0.786	0.025
LAMB3	0.661	<.001	0.617	<.001	0.734	<.001		
LGALS3	0.618	<.001	0.702	0.001	0.734	0.001	0.793	0.012
LIG3	0.705	0.008	0.615	<.001				
LRP1	0.786	0.050			0.795	0.023	0.770	0.009
MAP3K7					0.789	0.003		
MGMT	0.632	<.001	0.693	<.001				
MICA	0.781	0.014	0.653	<.001			0.833	0.043
MPPED2	0.655	<.001	0.597	<.001	0.719	<.001	0.759	0.006
MSH6					0.793	0.015		
MTSS1	0.613	<.001			0.746	0.008		
MVP	0.792	0.028	0.795	0.045	0.819	0.023		
MYBPC1	0.648	<.001	0.496	<.001	0.701	<.001	0.629	<.001
NCAM1					0.773	0.015		
NCAPD3	0.574	<.001	0.463	<.001	0.679	<.001	0.640	<.001
NEXN	0.701	0.002	0.791	0.035	0.725	0.002	0.781	0.016
NFAT5	0.515	<.001	0.586	<.001	0.785	0.017		
NFATC2	0.753	0.023						
NFKBIA	0.778	0.037						
NRG1	0.644	0.004	0.696	0.017	0.698	0.012		
OAZ1	0.777	0.034	0.775	0.022				
OLFML3	0.621	<.001	0.720	0.001	0.600	<.001	0.626	<.001
OMD	0.706	0.003						
OR51E2	0.820	0.037	0.798	0.027				

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
PAGE4	0.549	<.001	0.613	<.001	0.542	<.001	0.628	<.001
PCA3	0.684	<.001	0.635	<.001				
PCDHGB7	0.790	0.045			0.725	0.002	0.664	<.001
PGF	0.753	0.017						
PGR	0.740	0.021	0.728	0.018				
PIK3CG	0.803	0.024						
PLAUR	0.778	0.035						
PLG							0.728	0.028
PPAP2B	0.575	<.001	0.629	<.001	0.643	<.001	0.699	<.001
PPP1R12A	0.647	<.001	0.683	0.002	0.782	0.023	0.784	0.030
PRIMA1	0.626	<.001	0.658	<.001	0.703	0.002	0.724	0.003
PRKCA	0.642	<.001	0.799	0.029	0.677	0.001	0.776	0.006
PRKCB	0.675	0.001			0.648	<.001	0.747	0.006
PROM1	0.603	0.018			0.659	0.014	0.493	0.008
PTCH1	0.680	0.001			0.753	0.010	0.789	0.018
PTEN	0.732	0.002	0.747	0.005	0.744	<.001	0.765	0.002
PTGS2	0.596	<.001	0.610	<.001				
PTH1R	0.767	0.042			0.775	0.028	0.788	0.047
PTHLH	0.617	0.002	0.726	0.025	0.668	0.002	0.718	0.007
PTK2B	0.744	0.003	0.679	<.001	0.766	0.002	0.726	<.001
PTPN1	0.760	0.020	0.780	0.042				
PYCARD			0.748	0.012				
RAB27A			0.708	0.004				
RAB30	0.755	0.008						
RAGE			0.817	0.048				
RAP1B					0.818	0.050		
RARB	0.757	0.007	0.677	<.001	0.789	0.007	0.746	0.003
RASSF1	0.816	0.035						
RHOB	0.725	0.009	0.676	0.001			0.793	0.039
RLN1			0.742	0.033			0.762	0.040

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
RND3	0.636	<.001	0.647	<.001				
RNF114			0.749	0.011				
SDC2					0.721	0.004		
SDHC	0.725	0.003	0.727	0.006				
SEMA3A	0.757	0.024	0.721	0.010				
SERPINA3	0.716	0.008	0.660	0.001				
SERPINB5	0.747	0.031	0.616	0.002				
SH3RF2	0.577	<.001	0.458	<.001	0.702	<.001	0.640	<.001
SLC22A3	0.565	<.001	0.540	<.001	0.747	0.004	0.756	0.007
SMAD4	0.546	<.001	0.573	<.001	0.636	<.001	0.627	<.001
SMARCD1	0.718	<.001	0.775	0.017				
SMO	0.793	0.029	0.754	0.021			0.718	0.003
SOD1	0.757	0.049	0.707	0.006				
SORBS 1	0.645	<.001	0.716	0.003	0.693	<.001	0.784	0.025
SPARCL1	0.821	0.028			0.829	0.014	0.781	0.030
SPDEF	0.778	<.001						
SPINT1	0.732	0.009	0.842	0.026				
SRC	0.647	<.001	0.632	<.001				
SRD5A1					0.813	0.040		
SRD5A2	0.489	<.001	0.533	<.001	0.544	<.001	0.611	<.001
ST5	0.713	0.002	0.783	0.011	0.725	<.001	0.827	0.025
STAT3	0.773	0.037	0.759	0.035				
STAT5A	0.695	<.001	0.719	0.002	0.806	0.020	0.783	0.008
STAT5B	0.633	<.001	0.655	<.001			0.814	0.028
SUMO1	0.790	0.015						
SVIL	0.659	<.001	0.713	0.002	0.711	0.002	0.779	0.010
TARP							0.800	0.040
TBP	0.761	0.010						
TFF3	0.734	0.010	0.659	<.001				
TGFB1I1	0.618	<.001	0.693	0.002	0.637	<.001	0.719	0.004
TGFB2	0.679	<.001	0.747	0.005	0.805	0.030		

Genes significantly (p<0.05) associated with cRFI or bRFI in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)								
Table 4B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
TGFB3					0.791	0.037		
TGFBR2					0.778	0.035		
TIMP3					0.751	0.011		
TMPRSS2	0.745	0.003	0.708	<.001				
TNF			0.670	0.013			0.697	0.015
TNFRSF10A	0.780	0.018	0.752	0.006	0.817	0.032		
TNFRSF10B	0.576	<.001	0.655	<.001	0.766	0.004	0.778	0.002
TNFRSF18	0.648	0.016			0.759	0.034		
TNFSF10	0.653	<.001	0.667	0.004				
TP53			0.729	0.003				
TP63	0.759	0.016	0.636	<.001	0.698	<.001	0.712	0.001
TPM1	0.778	0.048	0.743	0.012	0.783	0.032	0.811	0.046
TPM2	0.578	<.001	0.634	<.001	0.611	<.001	0.710	0.001
TPP2			0.775	0.037				
TRAF3IP2	0.722	0.002	0.690	<.001	0.792	0.021	0.823	0.049
TRO	0.744	0.003	0.725	0.003	0.765	0.002	0.821	0.041
TUBB2A	0.639	<.001	0.625	<.001				
TYMP	0.786	0.039						
VCL	0.594	<.001	0.657	0.001	0.682	<.001		
VEGFA			0.762	0.024				
VEGFB	0.795	0.037						
VIM	0.739	0.009			0.791	0.021		
WDR19							0.776	0.015
WFDC1					0.746	<.001		
YY1	0.683	0.001			0.728	0.002		
ZFHX3	0.684	<.001	0.661	<.001	0.801	0.010	0.762	0.001
ZFP36	0.605	<.001	0.579	<.001			0.815	0.043
ZNF827	0.624	<.001	0.730	0.007	0.738	0.004		

[0138] Tables 5A and 5B provide genes that were significantly associated (p<0.05), positively or

negatively, with recurrence (cRFI, bRFI) after adjusting for AUA risk group in the primary and/or highest Gleason pattern. Increased expression of genes in Table 5A is negatively associated with good prognosis, while increased expression of genes in Table 5B is positively associated with good prognosis.

Table 5A.

Gene significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression negatively associated with good prognosis)								
Table 5A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
AKR1C3	1.315	0.018	1.283	0.024				
ALOX12							1.198	0.024
ANLN	1.406	<.001	1.519	<.001	1.485	<.001	1.632	<.001
AQP2	1.209	<.001	1.302	<.001				
ASAP2			1.582	<.001	1.333	0.011	1.307	0.019
ASPN	1.872	<.001	1.741	<.001	1.638	<.001	1.691	<.001
ATP5E	1.309	0.042	1.369	0.012				
BAG5			1.291	0.044				
BAX					1.298	0.025	1.420	0.004
BGN	1.746	<.001	1.755	<.001				
BIRC5	1.480	<.001	1.470	<.001	1.419	<.001	1.503	<.001
BMP6	1.536	<.001	1.815	<.001	1.294	0.033	1.429	0.001
BRCA2							1.184	0.037
BUB1	1.288	0.001	1.391	<.001	1.254	<.001	1.189	0.018
CACNA1D			1.313	0.029				
CADPS					1.358	0.007	1.267	0.022
CASP3					1.251	0.037		
CCNB1					1.261	0.033	1.318	0.005
CCNE2	1.345	0.005	1.438	<.001	1.606	<.001	1.426	<.001
CD276	1.482	0.002	1.668	<.001	1.451	<.001	1.302	0.011
CDC20	1.417	<.001	1.547	<.001	1.355	<.001	1.446	<.001
CDC6	1.340	0.011	1.265	0.046	1.367	0.002	1.272	0.025
CDH7	1.402	0.003	1.409	0.002				
CDKN2B	1.553	<.001	1.746	<.001	1.340	0.014	1.369	0.006
CDKN2C	1.411	<.001	1.604	<.001	1.220	0.033		
CDKN3	1.296	0.004			1.226	0.015		

Gene significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression negatively associated with good prognosis)

Table 5A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
CENPF	1.434	0.002	1.570	<.001	1.633	<.001	1.610	<.001
CKS2	1.419	0.008	1.374	0.022	1.380	0.004		
COL1A1	1.677	<.001	1.809	<.001	1.401	<.001	1.352	0.003
COL1A2			1.373	0.010				
COL3A1	1.669	<.001	1.781	<.001	1.249	0.024	1.234	0.047
COL4A1	1.475	0.002	1.513	0.002				
COL8A1	1.506	0.001	1.691	<.001				
CRISP3	1.406	0.004	1.471	<.001				
CTHRC1	1.426	0.009	1.793	<.001	1.311	0.019		
CTNND2					1.462	<.001		
DDIT4	1.478	0.003	1.783	<.001			1.236	0.039
DYNLL1	1.431	0.002					1.193	0.004
EIF3H					1.372	0.027		
ENY2					1.325	0.023	1.270	0.017
ERG	1.303	0.041						
EZH2			1.254	0.049				
F2R	1.540	0.002	1.448	0.006	1.286	0.023		
FADD	1.235	0.041	1.404	<.001				
FAP	1.386	0.015	1.440	0.008	1.253	0.048		
FASN	1.303	0.028						
FCGR3A			1.439	0.011			1.262	0.045
FGF5	1.289	0.006						
GNPTAB	1.290	0.033	1.369	0.022	1.285	0.018	1.355	0.008
GPR68			1.396	0.005				
GREM1	1.341	0.022	1.502	0.003	1.366	0.006		
HDAC1					1.329	0.016		
HDAC9			1.378	0.012				
HRAS	1.465	0.006						
HSD17B4					1.442	<.001	1.245	0.028
IGFBP3			1.366	0.019			1.302	0.011

Gene significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression negatively associated with good prognosis)

Table 5A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
NRP1			1.372	0.020				
ODC1					1.450	<.001		
OR51E1					1.559	<.001	1.413	0.008
PAK6							1.233	0.047
PATE1	1.262	<.001	1.375	<.001	1.143	0.034	1.191	0.036
PCNA					1.227	0.033	1.318	0.003
PEX10	1.517	<.001	1.500	0.001				
PGD	1.363	0.028	1.316	0.039	1.652	<.001		
PGK1			1.224	0.034			1.206	0.024
PIM1					1.205	0.042		
PLA2G7					1.298	0.018	1.358	0.005
PLAU					1.242	0.032		
PLK1			1.464	0.001	1.299	0.018	1.275	0.031
PLOD2					1.206	0.039	1.261	0.025
POSTN	1.558	0.001	1.356	0.022	1.363	0.009		
PPP3CA					1.445	0.002		
PSMD13					1.301	0.017	1.411	0.003
PTK2			1.318	0.031				
PTK6	1.582	<.001	1.894	<.001	1.290	0.011	1.354	0.003
PTTG1	1.319	0.004	1.430	<.001	1.271	0.006	1.492	<.001
RAD21			1.278	0.028	1.435	0.004	1.326	0.008
RAF1					1.504	<.001		
RALA	1.374	0.028			1.459	0.001		
RGS7			1.203	0.031				
RRM1	1.535	0.001	1.525	<.001				
RRM2	1.302	0.003	1.197	0.047	1.342	<.001		
SAT1	1.374	0.043						
SDC1					1.344	0.011	1.473	0.008
SEC14L1							1.297	0.006
SESN3	1.337	0.002	1.495	<.001			1.223	0.038

Gene significantly (p<0.05) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression negatively associated with good prognosis)

Table 5A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
SFRP4	1.610	<.001	1.542	0.002	1.370	0.009		
SHMT2	1.567	0.001	1.522	<.001	1.485	0.001	1.370	<.001
SKIL					1.303	0.008		
SLC25A21					1.287	0.020	1.306	0.017
SLC44A1			1.308	0.045				
SNRPB2	1.304	0.018						
SOX4					1.252	0.031		
SPARC	1.445	0.004	1.706	<.001			1.269	0.026
SPP1			1.376	0.016				
SQLE			1.417	0.007	1.262	0.035		
STAT1							1.209	0.029
STMN1	1.315	0.029						
SIJLF1			1.504	0.001				
TAF2					1.252	0.048	1.301	0.019
TFDP1					1.395	0.010	1.424	0.002
THBS2	1.716	<.001	1.719	<.001				
THY1	1.343	0.035	1.575	0.001				
TK1					1.320	<.001	1.304	<.001
TOP2A	1.464	0.001	1.688	<.001	1.715	<.001	1.761	<.001
TPD52					1.286	0.006	1.258	0.023
TPX2	1.644	<.001	1.964	<.001	1.699	<.001	1.754	<.001
TYMS							1.315	0.014
UBE2C	1.270	0.019	1.558	<.001	1.205	0.027	1.333	<.001
UBE2G1	1.302	0.041						
UBE2T	1.451	<.001			1.309	0.003		
UGT2B15			1.222	0.025				
UHRF1	1.370	0.003	1.520	<.001	1.247	0.020		
VCPIP1			1.332	0.015				
VTI1B					1.237	0.036		
XIAP					1.486	0.008		

Gene significantly (p<0.05) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression negatively associated with good prognosis)

Table 5A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
ZMYND8			1.408	0.007				
ZNF3							1.284	0.018
ZWINT	1.289	0.028						

Table 5B.

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
AAMP	0.535	<.001	0.581	<.001	0.700	0.002	0.759	0.006
ABCA5	0.798	0.007	0.745	0.002			0.841	0.037
ABCC1			0.800	0.044				
ABCC4			0.787	0.022				
ABHD2			0.768	0.023				
ACOX2	0.678	0.002	0.749	0.027	0.759	0.004		
ADH5	0.645	<.001	0.672	0.001				
AGTR1	0.780	0.030						
AKAP1	0.815	0.045	0.758	<.001				
AKT1			0.732	0.010				
ALDH1A2	0.646	<.001	0.548	<.001	0.671	<.001	0.713	0.001
ANPEP	0.641	<.001	0.535	<.001				
ANXA2	0.772	0.035			0.804	0.046		
ATXN1	0.654	<.001	0.754	0.020	0.797	0.017		
AURKA			0.788	0.030				
AXIN2	0.744	0.005	0.655	<.001				
AZGP1	0.656	<.001	0.676	<.001	0.754	0.001	0.791	0.004
BAD			0.700	0.004				
BIN1	0.650	<.001	0.764	0.013	0.803	0.015		

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
BTG3					0.836	0.025		
BTRC	0.730	0.005						
C7	0.617	$< .001$	0.680	$< .001$	0.667	$< .001$	0.755	0.005
CADM1	0.559	$< .001$	0.566	$< .001$	0.772	0.020	0.802	0.046
CASP1	0.781	0.030	0.779	0.021	0.818	0.027	0.828	0.036
CAV1					0.775	0.034		
CAV2			0.677	0.019				
CCL2			0.752	0.023				
CCNH	0.679	$< .001$	0.682	$< .001$				
CD164	0.721	0.002	0.724	0.005				
CD1A					0.710	0.014		
CD44	0.591	$< .001$	0.642	$< .001$				
CD82	0.779	0.021	0.771	0.024				
CDC25B	0.778	0.035			0.818	0.023		
CDK14	0.788	0.011						
CDK3	0.752	0.012			0.779	0.005	0.841	0.020
CDKN1A	0.770	0.049	0.712	0.014				
CDKN1C	0.684	$< .001$			0.697	$< .001$		
CHN1	0.772	0.031						
COL6A1	0.648	$< .001$	0.807	0.046	0.768	0.004		
CSF1	0.621	$< .001$	0.671	0.001				
CTNNB1					0.905	0.008		
CTSB	0.754	0.030	0.716	0.011	0.756	0.014		
CXCL12	0.641	$< .001$	0.796	0.038	0.708	$< .001$		
CYP3A5	0.503	$< .001$	0.528	$< .001$	0.791	0.028		
CYR61	0.639	0.001	0.659	0.001			0.797	0.048
DARC					0.707	0.004		
DDR2					0.750	0.011		
DES	0.657	$< .001$	0.758	0.022	0.699	$< .001$		
DHRS9	0.625	0.002						

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
DHX9	0.846	<.001						
DIAPH1	0.682	0.007	0.723	0.008	0.780	0.026		
DLC1	0.703	0.005	0.702	0.008				
DLGAP1	0.703	0.008	0.636	<.001				
DNM3	0.701	0.001			0.817	0.042		
DPP4	0.686	<.001	0.716	0.001				
DPT	0.636	<.001	0.633	<.001	0.709	0.006	0.773	0.024
DUSP1	0.683	0.006	0.679	0.003				
DUSP6	0.694	0.003	0.605	<.001				
EDN1					0.773	0.031		
EDNRA	0.716	0.007						
EGR1	0.575	<.001	0.575	<.001			0.771	0.014
EGR3	0.633	0.002	0.643	<.001			0.792	0.025
EIF4E	0.722	0.002						
ELK4	0.710	0.009	0.759	0.027				
ENPP2	0.786	0.039						
EPHA2			0.593	0.001				
EPHA3	0.739	0.006			0.802	0.020		
ERBB2			0.753	0.007				
ERBB3	0.753	0.009	0.753	0.015				
ERCC1							0.727	0.001
EREG					0.722	0.012	0.769	0.040
ESR1			0.742	0.015				
FABP5	0.756	0.032						
FAM107A	0.524	<.001	0.579	<.001	0.688	<.001	0.699	0.001
FAM13C	0.639	<.001	0.601	<.001	0.810	0.019	0.709	<.001
FAS	0.770	0.033						
FASLG	0.716	0.028	0.683	0.017				
FGF10					0.798	0.045		
FGF17			0.718	0.018	0.793	0.024	0.790	0.024

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
FGFR2	0.739	0.007	0.783	0.038	0.740	0.004		
FGFR4			0.746	0.050				
FKBP5			0.689	0.003				
FLNA	0.701	0.006	0.766	0.029	0.768	0.037		
FLNC					0.755	<.001	0.820	0.022
FLT1			0.729	0.008				
FOS	0.572	<.001	0.536	<.001			0.750	0.005
FOXQ1	0.778	0.033			0.820	0.018		
FYN	0.708	0.006						
GADD45B	0.577	<.001	0.589	<.001				
GDF15	0.757	0.013	0.743	0.006				
GHR			0.712	0.004			0.679	0.001
GNRH1					0.791	0.048		
GPM6B	0.675	<.001	0.660	<.001	0.735	<.001	0.823	0.049
GSK3B	0.783	0.042						
GSN	0.587	<.001	0.705	0.002	0.745	0.004	0.796	0.021
GSTM1	0.686	0.001	0.631	<.001	0.807	0.018		
GSTM2	0.607	<.001	0.683	<.001	0.679	<.001	0.800	0.027
HIRIP3	0.692	<.001			0.782	0.007		
HK1	0.724	0.002	0.718	0.002				
HLF	0.580	<.001	0.571	<.001	0.759	0.008	0.750	0.004
HNF1B			0.669	<.001				
HPS1	0.764	0.008						
HSD17B10	0.802	0.045						
HSD17B2					0.723	0.048		
HSD3B2							0.709	0.010
HSP90AB1	0.780	0.034			0.809	0.041		
HSPA5			0.738	0.017				
HSPB1	0.770	0.006	0.801	0.032				
HSPB2					0.788	0.035		

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
ICAM1	0.728	0.015	0.716	0.010				
IER3	0.735	0.016	0.637	<.001			0.802	0.035
IFIT1	0.647	<.001	0.755	0.029				
IGF1	0.675	<.001	0.603	<.001	0.762	0.006	0.770	0.030
IGF2					0.761	0.011		
IGFBP2	0.601	<.001	0.605	<.001				
IGFBP5	0.702	<.001						
IGFBP6	0.628	<.001			0.726	0.003		
IL1B	0.676	0.002	0.716	0.004				
IL6	0.688	0.005	0.766	0.044				
IL6R			0.786	0.036				
IL6ST	0.618	<.001	0.639	<.001	0.785	0.027	0.813	0.042
IL8	0.635	<.001	0.628	<.001				
ILK	0.734	0.018	0.753	0.026				
ING5	0.684	<.001	0.681	<.001	0.756	0.006		
ITGA4	0.778	0.040						
ITGA5	0.762	0.026						
ITGA6			0.811	0.038				
ITGA7	0.592	<.001	0.715	0.006	0.710	0.002		
ITGAD			0.576	0.006				
ITGB4			0.693	0.003				
ITPR1	0.789	0.029						
JUN	0.572	<.001	0.581	<.001			0.777	0.019
JUNB	0.732	0.030	0.707	0.016				
KCTD12	0.758	0.036						
KIT					0.691	0.009	0.738	0.028
KLC1	0.741	0.024			0.781	0.024		
KLF6	0.733	0.018	0.727	0.014				
KLK1			0.744	0.028				
KLK2	0.697	0.002	0.679	<.001				

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
KLK3	0.725	<.001	0.715	<.001			0.841	0.023
KRT15	0.660	<.001	0.577	<.001	0.750	0.002		
KRT18	0.623	<.001	0.642	<.001	0.702	<.001	0.760	0.006
KRT2					0.740	0.044		
KRT5	0.674	<.001	0.588	<.001	0.769	0.005		
KRT8	0.768	0.034						
LICAM	0.737	0.036						
LAG3	0.711	0.013	0.748	0.029				
LAMA4					0.649	0.009		
LAMB3	0.709	0.002	0.684	0.006	0.768	0.006		
LGALS3	0.652	<.001	0.752	0.015	0.805	0.028		
LIG3	0.728	0.016	0.667	<.001				
LRP1							0.811	0.043
MDM2			0.788	0.033				
MGMT	0.645	<.001	0.766	0.015				
MICA	0.796	0.043	0.676	<.001				
MPPED2	0.675	<.001	0.616	<.001	0.750	0.006		
MRC1							0.788	0.028
MTSS1	0.654	<.001			0.793	0.036		
MYBPC1	0.706	<.001	0.534	<.001	0.773	0.004	0.692	<.001
NCAPD3	0.658	<.001	0.566	<.001	0.753	0.011	0.733	0.009
NCOR1			0.838	0.045				
NEXN	0.748	0.025			0.785	0.020		
NFAT5	0.531	<.001	0.626	<.001				
NFATC2			0.759	0.024				
OAZ1			0.766	0.024				
OLFML3	0.648	<.001	0.748	0.005	0.639	<.001	0.675	<.001
OR51E2	0.823	0.034						
PAGE4	0.599	<.001	0.698	0.002	0.606	<.001	0.726	<.001
PCA3	0.705	<.001	0.647	<.001				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
PCDHGB7							0.712	$< .001$
PGF	0.790	0.039						
PLG							0.764	0.048
PLP2			0.766	0.037				
PPAP2B	0.589	$< .001$	0.647	$< .001$	0.691	$< .001$	0.765	0.013
PPPIR12A	0.673	0.001	0.677	0.001			0.807	0.045
PRIMA1	0.622	$< .001$	0.712	0.008	0.740	0.013		
PRKCA	0.637	$< .001$			0.694	$< .001$		
PRKCB	0.741	0.020			0.664	$< .001$		
PROM1	0.599	0.017	0.527	0.042	0.610	0.006	0.420	0.002
PTCH1	0.752	0.027			0.762	0.011		
PTEN	0.779	0.011	0.802	0.030	0.788	0.009		
PTGS2	0.639	$< .001$	0.606	$< .001$				
PTHLH	0.632	0.007	0.739	0.043	0.654	0.002	0.740	0.015
PTK2B			0.775	0.019	0.831	0.028	0.810	0.017
PTPN1	0.721	0.012	0.737	0.024				
PYCARD			0.702	0.005				
RAB27A			0.736	0.008				
RAB30	0.761	0.011						
RARB			0.746	0.010				
RASSF1	0.805	0.043						
RHOB	0.755	0.029	0.672	0.001				
RLN1	0.742	0.036	0.740	0.036				
RND3	0.607	$< .001$	0.633	$< .001$				
RNF114	0.782	0.041	0.747	0.013				
SDC2					0.714	0.002		
SDHC	0.698	$< .001$	0.762	0.029				
SERPINA3			0.752	0.030				
SERPINB5			0.669	0.014				
SH3RF2	0.705	0.012	0.568	$< .001$			0.755	0.016

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good Drosnosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
SLC22A3	0.650	<.001	0.582	<.001				
SMAD4	0.636	<.001	0.684	0.002	0.741	0.007	0.738	0.007
SMARCD1	0.757	0.001						
SMO	0.790	0.049					0.766	0.013
SOD1	0.741	0.037	0.713	0.007				
SORBS1	0.684	0.003	0.732	0.008	0.788	0.049		
SPDEF	0.840	0.012						
SPINT1			0.837	0.048				
SRC	0.674	<.001	0.671	<.001				
SRD5A2	0.553	<.001	0.588	<.001	0.618	<.001	0.701	<.001
ST5	0.747	0.012	0.761	0.010	0.780	0.016	0.832	0.041
STAT3			0.735	0.020				
STAT5A	0.731	0.005	0.743	0.009			0.817	0.027
STAT5B	0.708	<.001	0.696	0.001				
SUMO1	0.815	0.037						
SVIL	0.689	0.003	0.739	0.008	0.761	0.011		
TBP	0.792	0.037						
TFF3	0.719	0.007	0.664	0.001				
TGFB1I1	0.676	0.003	0.707	0.007	0.709	0.005	0.777	0.035
TGFB2	0.741	0.010	0.785	0.017				
TGFBR2					0.759	0.022		
TIMP3					0.785	0.037		
TMPRSS2	0.780	0.012	0.742	<.001				
TNF			0.654	0.007			0.682	0.006
TNFRSF10B	0.623	<.001	0.681	<.001	0.801	0.018	0.815	0.019
TNFSF10	0.721	0.004						
TP53			0.759	0.011				
TP63			0.737	0.020	0.754	0.007		
TPM2	0.609	<.001	0.671	<.001	0.673	<.001	0.789	0.031
TRAF3IP2	0.795	0.041	0.727	0.005				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for AUA risk group in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 5B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
TRO	0.793	0.033	0.768	0.027	0.814	0.023		
TUBB2A	0.626	< 0.001	0.590	< 0.001				
VCL	0.613	< 0.001	0.701	0.011				
VIM	0.716	0.005			0.792	0.025		
WFDC1					0.824	0.029		
YY1	0.668	< 0.001	0.787	0.014	0.716	0.001	0.819	0.011
ZFHX3	0.732	< 0.001	0.709	< 0.001				
ZFP36	0.656	0.001	0.609	< 0.001			0.818	0.045
ZNF827	0.750	0.022						

[0139] Tables 6A and 6B provide genes that were significantly associated ($p < 0.05$), positively or negatively, with recurrence (cRFI, bRFI) after adjusting for Gleason pattern in the primary and/or highest Gleason pattern. Increased expression of genes in Table 6A is negatively associated with good prognosis, while increased expression of gene in Table 6B is positively associated with good prognosis.

Table 6A.

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with a hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 6A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
AKR1C3	1.258	0.039						
ANLN	1.292	0.023			1.449	< 0.001	1.420	0.001
AQP2	1.178	0.008	1.287	< 0.001				
ASAP2			1.396	0.015				
ASPN	1.809	< 0.001	1.508	0.009	1.506	0.002	1.438	0.002
BAG5			1.367	0.012				
BAX							1.234	0.044
BGN	1.465	0.009	1.342	0.046				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with a hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 6A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
BIRC5	1.338	0.008			1.364	0.004	1.279	0.006
BMP6	1.369	0.015	1.518	0.002				
BUB1	1.239	0.024	1.227	0.001	1.236	0.004		
CACNA1D			1.337	0.025				
CADPS					1.280	0.029		
CCNE2			1.256	0.043	1.577	$< .001$	1.324	0.001
CD276	1.320	0.029	1.396	0.007	1.279	0.033		
CDC20	1.298	0.016	1.334	0.002	1.257	0.032	1.279	0.003
CDH7	1.258	0.047	1.338	0.013				
CDKN2B	1.342	0.032	1.488	0.009				
CDKN2C	1.344	0.010	1.450	$< .001$				
CDKN3	1.284	0.012						
CENPF	1.289	0.048			1.498	0.001	1.344	0.010
COL1A1	1.481	0.003	1.506	0.002				
COL3A1	1.459	0.004	1.430	0.013				
COL4A1	1.396	0.015						
COL8A1	1.413	0.008						
CRISP3	1.346	0.012	1.310	0.025				
CTHRC1			1.588	0.002				
DDIT4	1.363	0.020	1.379	0.028				
DICER1							1.294	0.008
ENY2					1.269	0.024		
FADD			1.307	0.010				
FAS							1.243	0.025
FGF5	1.328	0.002						
GNPTAB							1.246	0.037
GREM1	1.332	0.024	1.377	0.013	1.373	0.011		
HDAC1					1.301	0.018	1.237	0.021
HSD17B4							1.277	0.011
IFN- γ					1.219	0.048		

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with a hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 6A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
IMMT					1.230	0.049		
INHBA	1.866	<.001	1.944	<.001				
JAG1			1.298	0.030				
KCNN2			1.378	0.020			1.282	0.017
KHDRBS3			1.353	0.029	1.305	0.014		
LAMA3					1.344	<.001	1.232	0.048
LAMC1	1.396	0.015						
LIMS1							1.337	0.004
LOX					1.355	0.001	1.341	0.002
LTBP2			1.304	0.045				
MAGEA4	1.215	0.024						
MANF					1.460	<.001		
MCM6			1.287	0.042			1.214	0.046
MELK					1.329	0.002		
MMP11	1.281	0.050						
MRPL13							1.266	0.021
MYBL2	1.453	<.001			1.274	0.019		
MYC					1.265	0.037		
MYO6			1.278	0.047				
NETO2	1.322	0.022						
NFKB1							1.255	0.032
NOX4					1.266	0.041		
OR51E1					1.566	<.001	1.428	
PATE1	1.242	<.001	1.347	<.001			1.177	0.011
PCNA							1.251	0.025
PEX10			1.302	0.028				
PGD			1.335	0.045	1.379	0.014	1.274	0.025
PIM1					1.254	0.019		
PLA2G7					1.289	0.025	1.250	0.031
PLAU					1.267	0.031		

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with a hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 6A	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
PSMD13							1.333	0.005
PTK6	1.432	$< .001$	1.577	$< .001$	1.223	0.040		
PTTG1					1.279	0.013	1.308	0.006
RAGE							1.329	0.011
RALA	1.363	0.044			1.471	0.003		
RGS7	1.120	0.040	1.173	0.031				
RRM1	1.490	0.004	1.527	$< .001$				
SESN3			1.353	0.017				
SFRP4	1.370	0.025						
SHMT2	1.460	0.008	1.410	0.006	1.407	0.008	1.345	$< .001$
SKIL					1.307	0.025		
SLC25A21					1.414	0.002	1.330	0.004
SMARCC2					1.219	0.049		
SPARC			1.431	0.005				
TFDP1					1.283	0.046	1.345	0.003
THBS2	1.456	0.005	1.431	0.012				
TK1					1.214	0.015	1.222	0.006
TOP2A			1.367	0.018	1.518	0.001	1.480	$< .001$
TPX2	1.513	0.001	1.607	$< .001$	1.588	$< .001$	1.481	$< .001$
UBE2T	1.409	0.002			1.285	0.018		
UGT2B15			1.216	0.009			1.182	0.021
XIAP					1.336	0.037	1.194	0.043

Table 6B.

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
AAMP	0.660	0.001	0.675	<.001			0.836	0.045
ABCA5	0.807	0.014	0.737	<.001			0.845	0.030
ABCC1	0.780	0.038	0.794	0.015				
ABCG2							0.807	0.035
ABHD2			0.720	0.002				
ADH5	0.750	0.034						
AKAP1			0.721	<.001				
ALDH1A2	0.735	0.009	0.592	<.001	0.756	0.007	0.781	0.021
ANGPT2					0.741	0.036		
ANPEP	0.637	<.001	0.536	<.001				
ANXA2			0.762	0.044				
APOE			0.707	0.013				
APRT			0.727	0.004			0.771	0.006
ATXN1	0.725	0.013						
AURKA	0.784	0.037	0.735	0.003				
AXIN2	0.744	0.004	0.630	<.001				
AZGP1	0.672	<.001	0.720	<.001	0.764	0.001		
BAD			0.687	<.001				
BAK1			0.783	0.014				
BCL2	0.777	0.033	0.772	0.036				
BIK			0.768	0.040				
BIN1	0.691	<.001						
BTRC			0.776	0.029				
C7	0.707	0.004			0.791	0.024		
CADM1	0.587	<.001	0.593	<.001				
CASP1	0.773	0.023			0.820	0.025		
CAV1					0.753	0.014		
CAV2			0.627	0.009			0.682	0.003
CCL2			0.740	0.019				
CCNH	0.736	0.003						

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
CCR1			0.755	0.022				
CD1A					0.740	0.025		
CD44	0.590	< 0.001	0.637	< 0.001				
CD68	0.757	0.026						
CD82	0.778	0.012	0.759	0.016				
CDC25B	0.760	0.021						
CDK3	0.762	0.024			0.774	0.007		
CDKN1A			0.714	0.015				
CDKN1C	0.738	0.014			0.768	0.021		
COL6A1	0.690	< 0.001	0.805	0.048				
CSF1	0.675	0.002	0.779	0.036				
CSK					0.825	0.004		
CTNNB1	0.884	0.045			0.888	0.027		
CTSB	0.740	0.017	0.676	0.003	0.755	0.010		
CTSD	0.673	0.031	0.722	0.009				
CTSK					0.804	0.034		
CTSL2			0.748	0.019				
CXCL12	0.731	0.017						
CYP3A5	0.523	< 0.001	0.518	< 0.001				
CYR61	0.744	0.041						
DAP					0.755	0.011		
DARC					0.763	0.029		
DDR2							0.813	0.041
DES	0.743	0.020						
DHRS9	0.606	0.001						
DHX9	0.916	0.021						
DIAPH1	0.749	0.036	0.688	0.003				
DLGAP1	0.758	0.042	0.676	0.002				
DLL4							0.779	0.010
DNM3	0.732	0.007						

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
FOS	0.657	0.003	0.656	0.004				
FSD1							0.763	0.017
FYN	0.716	0.004			0.792	0.024		
GADD45B	0.692	0.009	0.697	0.010				
GDF15			0.767	0.016				
GHR			0.701	0.002	0.704	0.002	0.640	$< .001$
GNRH1					0.778	0.039		
GPM6B	0.749	0.010	0.750	0.010	0.827	0.037		
GRB7			0.696	0.005				
GSK3B	0.726	0.005						
GSN	0.660	$< .001$	0.752	0.019				
GSTM1	0.710	0.004	0.676	$< .001$				
GSTM2	0.643	$< .001$			0.767	0.015		
HK1	0.798	0.035						
HLA-G			0.660	0.013				
HLF	0.644	$< .001$	0.727	0.011				
HNF1B			0.755	0.013				
HPS1	0.756	0.006	0.791	0.043				
HSD17B10	0.737	0.006						
HSD3B2							0.674	0.003
HSP90AB1			0.763	0.015				
HSPB1	0.787	0.020	0.778	0.015				
HSPE1			0.794	0.039				
ICAM1			0.664	0.003				
IER3	0.699	0.003	0.693	0.010				
IFIT1	0.621	$< .001$	0.733	0.027				
IGF1	0.751	0.017	0.655	$< .001$				
IGFBP2	0.599	$< .001$	0.605	$< .001$				
IGFBP5	0.745	0.007	0.775	0.035				
IGFBP6	0.671	0.005						

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
IL1B	0.732	0.016	0.717	0.005				
IL6	0.763	0.040						
IL6R			0.764	0.022				
IL6ST	0.647	< 0.001	0.739	0.012				
IL8	0.711	0.015	0.694	0.006				
ING5	0.729	0.007	0.727	0.003				
ITGA4			0.755	0.009				
ITGA5	0.743	0.018	0.770	0.034				
ITGA6	0.816	0.044	0.772	0.006				
ITGA7	0.680	0.004						
ITGAD			0.590	0.009				
ITGB4	0.663	< 0.001	0.658	< 0.001	0.759	0.004		
JUN	0.656	0.004	0.639	0.003				
KIAA0196	0.737	0.011						
KIT					0.730	0.021	0.724	0.008
KLC1	0.755	0.035						
KLK1	0.706	0.008						
KLK2	0.740	0.016	0.723	0.001				
KLK3	0.765	0.006	0.740	0.002				
KRT1							0.774	0.042
KRT15	0.658	< 0.001	0.632	< 0.001	0.764	0.008		
KRT18	0.703	0.004	0.672	< 0.001	0.779	0.015	0.811	0.032
KRT5	0.686	< 0.001	0.629	< 0.001	0.802	0.023		
KRT8	0.763	0.034	0.771	0.022				
L1CAM	0.748	0.041						
LAG3	0.693	0.008	0.724	0.020				
LAMA4					0.689	0.039		
LAMB3	0.667	< 0.001	0.645	< 0.001	0.773	0.006		
LGALS3	0.666	< 0.001			0.822	0.047		
LIG3			0.723	0.008				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
LRP1	0.777	0.041					0.769	0.007
MDM2			0.688	$< .001$				
MET	0.709	0.010	0.736	0.028	0.715	0.003		
MGMT	0.751	0.031						
MICA			0.705	0.002				
MPPED2	0.690	0.001	0.657	$< .001$	0.708	$< .001$		
MRC1							0.812	0.049
MSH6							0.860	0.049
MTSS1	0.686	0.001						
MVP	0.798	0.034	0.761	0.033				
MYBPC1	0.754	0.009	0.615	$< .001$				
NCAPD3	0.739	0.021	0.664	0.005				
NEXN			0.798	0.037				
NFAT5	0.596	$< .001$	0.732	0.005				
NFATC2	0.743	0.016	0.792	0.047				
NOS3	0.730	0.012	0.757	0.032				
OAZ1	0.732	0.020	0.705	0.002				
OCLN					0.746	0.043	0.784	0.025
OLFML3	0.711	0.002			0.709	$< .001$	0.720	0.001
OMD	0.729	0.011	0.762	0.033				
OSM							0.813	0.028
PAGE4	0.668	0.003	0.725	0.004	0.688	$< .001$	0.766	0.005
PCA3	0.736	0.001	0.691	$< .001$				
PCDHGB7					0.769	0.019	0.789	0.022
PIK3CA			0.768	0.010				
PIK3CG	0.792	0.019	0.758	0.009				
PLG							0.682	0.009
PPAP2B	0.688	0.005			0.815	0.046		
PPP1R12A	0.731	0.026	0.775	0.042				
PRIMA1	0.697	0.004	0.757	0.032				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
PRKCA	0.743	0.019						
PRKCB	0.756	0.036			0.767	0.029		
PROM1	0.640	0.027			0.699	0.034	0.503	0.013
PTCH1	0.730	0.018						
PTEN	0.779	0.015			0.789	0.007		
PTGS2	0.644	$< .001$	0.703	0.007				
PTHLH	0.655	0.012	0.706	0.038	0.634	0.001	0.665	0.003
PTK2B	0.779	0.023	0.702	0.002	0.806	0.015	0.806	0.024
PYCARD			0.659	0.001				
RAB30	0.779	0.033	0.754	0.014				
RARB	0.787	0.043	0.742	0.009				
RASSF1	0.754	0.005						
RHOA			0.796	0.041			0.819	0.048
RND3	0.721	0.011	0.743	0.028				
SDC1			0.707	0.011				
SDC2					0.745	0.002		
SDHC	0.750	0.013						
SERPINA3			0.730	0.016				
SERPINB5			0.715	0.041				
SH3RF2			0.698	0.025				
SIPA1L1			0.796	0.014			0.820	0.004
SLC22A3	0.724	0.014	0.700	0.008				
SMAD4	0.668	0.002			0.771	0.016		
SMARCD1	0.726	$< .001$	0.700	0.001			0.812	0.028
SMO							0.785	0.027
SOD1			0.735	0.012				
SORBS 1			0.785	0.039				
SPDEF	0.818	0.002						
SPINT1	0.761	0.024	0.773	0.006				
SRC	0.709	$< .001$	0.690	$< .001$				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
SRD5A1	0.746	0.010	0.767	0.024	0.745	0.003		
SRD5A2	0.575	<.001	0.669	0.001	0.674	<.001	0.781	0.018
ST5	0.774	0.027						
STAT1	0.694	0.004						
STAT5A	0.719	0.004	0.765	0.006			0.834	0.049
STAT5B	0.704	0.001	0.744	0.012				
SUMO1	0.777	0.014						
SVIL			0.771	0.026				
TBP	0.774	0.031						
TFF3	0.742	0.015	0.719	0.024				
TGFB1I1	0.763	0.048						
TGFB2	0.729	0.011	0.758	0.002				
TMPRSS2	0.810	0.034	0.692	<.001				
TNF							0.727	0.022
TNFRSF10A			0.805	0.025				
TNFRSF10B	0.581	<.001	0.738	0.014	0.809	0.034		
TNFSF10	0.751	0.015	0.700	<.001				
TP63			0.723	0.018	0.736	0.003		
TPM2	0.708	0.010	0.734	0.014				
TRAF3IP2			0.718	0.004				
TRO			0.742	0.012				
TSTA3			0.774	0.028				
TUBB2A	0.659	<.001	0.650	<.001				
TYMP	0.695	0.002						
VCL	0.683	0.008						
VIM	0.778	0.040						
WDR19							0.775	0.014
XRCC5	0.793	0.042						
YY1	0.751	0.025					0.810	0.008
ZFHX3	0.760	0.005	0.726	0.001				

Genes significantly ($p < 0.05$) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 6B	cRFI		cRFI		bRFI		bRFI	
	Primary Pattern		Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value	HR	p-value
ZFP36	0.707	0.008	0.672	0.003				
ZNF827	0.667	0.002			0.792	0.039		

[0140] Tables 7A and 7B provide genes significantly associated ($p < 0.05$), positively or negatively, with clinical recurrence (cRFI) in negative TMPRSS fusion specimens in the primary or highest Gleason pattern specimen. Increased expression of genes in Table 7A is negatively associated with good prognosis, while increased expression of genes in Table 7B is positively associated with good prognosis.

Table 7A.

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 7A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
ANLN	1.42	0.012	1.36	0.004
AQP2	1.25	0.033		
ASPN	2.48	$< .001$	1.65	$< .001$
BGN	2.04	$< .001$	1.45	0.007
BIRC5	1.59	$< .001$	1.37	0.005
BMP6	1.95	$< .001$	1.43	0.012
BMPR1B	1.93	0.002		
BUB1	1.51	$< .001$	1.35	$< .001$
CCNE2	1.48	0.007		
CD276	1.93	$< .001$	1.79	$< .001$
CDC20	1.49	0.004	1.47	$< .001$
CDC6	1.52	0.009	1.34	0.022
CDKN2B	1.54	0.008	1.55	0.003
CDKN2C	1.55	0.003	1.57	$< .001$
CDKN3	1.34	0.026		
CENPF	1.63	0.002	1.33	0.018

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 7A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
CKS2	1.50	0.026	1.43	0.009
CLTC			1.46	0.014
COL1A1	1.98	$< .001$	1.50	0.002
COL3A1	2.03	$< .001$	1.42	0.007
COL4A1	1.81	0.002		
COL8A1	1.63	0.004	1.60	0.001
CRISP3			1.31	0.016
CTHRC1	1.67	0.006	1.48	0.005
DDIT4	1.49	0.037		
ENY2			1.29	0.039
EZH2			1.35	0.016
F2R	1.46	0.034	1.46	0.007
FAP	1.66	0.006	1.38	0.012
FGF5			1.46	0.001
GNPTAB	1.49	0.013		
HSD17B4	1.34	0.039	1.44	0.002
INHBA	2.92	$< .001$	2.19	$< .001$
JAG1	1.38	0.042		
KCNN2	1.71	0.002	1.73	$< .001$
KHDRBS3			1.46	0.015
KLK14	1.28	0.034		
KPNA2	1.63	0.016		
LAMC1	1.41	0.044		
LOX			1.29	0.036
LTBP2	1.57	0.017		
MELK	1.38	0.029		
MMP11	1.69	0.002	1.42	0.004
MYBL2	1.78	$< .001$	1.49	$< .001$
NETO2	2.01	$< .001$	1.43	0.007
NME1			1.38	0.017
PATE1	1.43	$< .001$	1.24	0.005
PEX10	1.46	0.030		

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 7A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
PGD	1.77	0.002		
POSTN	1.49	0.037	1.34	0.026
PPFIA3	1.51	0.012		
PPP3CA	1.46	0.033	1.34	0.020
PTK6	1.69	$< .001$	1.56	$< .001$
PTTG1	1.35	0.028		
RAD51	1.32	0.048		
RALBP1			1.29	0.042
RGS7	1.18	0.012	1.32	0.009
RRM1	1.57	0.016	1.32	0.041
RRM2	1.30	0.039		
SAT1	1.61	0.007		
SESN3	1.76	$< .001$	1.36	0.020
SFRP4	1.55	0.016	1.48	0.002
SHMT2	2.23	$< .001$	1.59	$< .001$
SPARC	1.54	0.014		
SQLE	1.86	0.003		
STMN1	2.14	$< .001$		
THBS2	1.79	$< .001$	1.43	0.009
TK1	1.30	0.026		
TOP2A	2.03	$< .001$	1.47	0.003
TPD52	1.63	0.003		
TPX2	2.11	$< .001$	1.63	$< .001$
TRAP1	1.46	0.023		
UBE2C	1.57	$< .001$	1.58	$< .001$
UBE2G1	1.56	0.008		
UBE2T	1.75	$< .001$		
UGT2B15	1.31	0.036	1.33	0.004
UHRF1	1.46	0.007		
UTP23	1.52	0.017		

Table 7B.

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
AAMP	0.56	<.001	0.65	0.001
ABCA5	0.64	<.001	0.71	<.001
ABCB1	0.62	0.004		
ABCC3			0.74	0.031
ABCG2			0.78	0.050
ABHD2	0.71	0.035		
ACOX2	0.54	<.001	0.71	0.007
ADH5	0.49	<.001	0.61	<.001
AKAP1	0.77	0.031	0.76	0.013
AKR1C1	0.65	0.006	0.78	0.044
AKT1			0.72	0.020
AKT3	0.75	<.001		
ALDH1A2	0.53	<.001	0.60	<.001
AMPD3	0.62	<.001	0.78	0.028
ANPEP	0.54	<.001	0.61	<.001
ANXA2	0.63	0.008	0.74	0.016
ARHGAP29	0.67	0.005	0.77	0.016
ARHGDIB	0.64	0.013		
ATP5J	0.57	0.050		
ATXN1	0.61	0.004	0.77	0.043
AXIN2	0.51	<.001	0.62	<.001
AZGP1	0.61	<.001	0.64	<.001
BCL2	0.64	0.004	0.75	0.029
BIN1	0.52	<.001	0.74	0.010
BTG3	0.75	0.032	0.75	0.010
BTRC	0.69	0.011		
C7	0.51	<.001	0.67	<.001
CADM1	0.49	<.001	0.76	0.034
CASP1	0.71	0.010	0.74	0.007
CAV1			0.73	0.015
CCL5	0.67	0.018	0.67	0.003

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
CCNH	0.63	$< .001$	0.75	0.004
CCR1			0.77	0.032
CD164	0.52	$< .001$	0.63	$< .001$
CD44	0.53	$< .001$	0.74	0.014
CDH10	0.69	0.040		
CDH18	0.40	0.011		
CDK14	0.75	0.013		
CDK2			0.81	0.031
CDK3	0.73	0.022		
CDKN1A	0.68	0.038		
CDKN1C	0.62	0.003	0.72	0.005
COL6A1	0.54	$< .001$	0.70	0.004
COL6A3	0.64	0.004		
CSF1	0.56	$< .001$	0.78	0.047
CSRP1	0.40	$< .001$	0.66	0.002
CTGF	0.66	0.015	0.74	0.027
CTNNB1	0.69	0.043		
CTSB	0.60	0.002	0.71	0.011
CTSS	0.67	0.013		
CXCL12	0.56	$< .001$	0.77	0.026
CYP3A5	4	$< .001$	0.63	$< .001$
CYR61	0.43	$< .001$	0.58	$< .001$
DAG1			0.72	0.012
DARC	0.66	0.016		
DDR2	0.65	0.007		
DES	0.52	$< .001$	0.74	0.018
DHRS9	0.54	0.007		
DICER1	0.70	0.044		
DLC1			0.75	0.021
DLGAP1	0.55	$< .001$	0.72	0.005
DNM3	0.61	0.001		
DPP4	0.55	$< .001$	0.77	0.024

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
DPT	0.48	<.001	0.61	<.001
DUSP1	0.47	<.001	0.59	<.001
DUSP6	0.65	0.009	0.65	0.002
DYNLL1			0.74	0.045
EDNRA	0.61	0.002	0.75	0.038
EFNB2	0.71	0.043		
EGR1	0.43	<.001	0.58	<.001
EGR3	0.47	<.001	0.66	<.001
EIF5			0.77	0.028
ELK4	0.49	<.001	0.72	0.012
EPHA2			0.70	0.007
EPHA3	0.62	<.001	0.72	0.009
EPHB2	0.68	0.009		
ERBB2	0.64	<.001	0.63	<.001
ERBB3	0.69	0.018		
ERCC1	0.69	0.019	0.77	0.021
ESR2	0.61	0.020		
FAAH	0.57	<.001	0.77	0.035
FABP5	0.67	0.035		
FAM107A	0.42	<.001	0.59	<.001
FAM13C	0.53	<.001	0.59	<.001
FAS	0.71	0.035		
FASLG	0.56	0.017	0.67	0.014
FGF10	0.57	0.002		
FGF17	0.70	0.039	0.70	0.010
FGF7	0.63	0.005	0.70	0.004
FGFR2	0.63	0.003	0.71	0.003
FKBP5			0.72	0.020
FLNA	0.48	<.001	0.74	0.022
FOS	0.45	<.001	0.56	<.001
FOXO1	0.59	<.001		

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
FOXQ1	0.57	$< .001$	0.69	0.008
FYN	0.62	0.001	0.74	0.013
G6PD			0.77	0.014
GADD45A	0.73	0.045		
GADD45B	0.45	$< .001$	0.64	0.001
GDF15	0.58	$< .001$		
GHR	0.62	0.008	0.68	0.002
GPM6B	0.60	$< .001$	0.70	0.003
GSK3B	0.71	0.016	0.71	0.006
GSN	0.46	$< .001$	0.66	$< .001$
GSTM1	0.56	$< .001$	0.62	$< .001$
GSTM2	0.47	$< .001$	0.67	$< .001$
HGD			0.72	0.002
HIRIP3	0.69	0.021	0.69	0.002
HK1	0.68	0.005	0.73	0.005
HLA-G	0.54	0.024	0.65	0.013
HLF	0.41	$< .001$	0.68	0.001
HNF1B	0.55	$< .001$	0.59	$< .001$
HPS1	0.74	0.015	0.76	0.025
HSD17B3	0.65	0.031		
HSPB2	0.62	0.004	0.76	0.027
ICAM1	0.61	0.010		
IER3	0.55	$< .001$	0.67	0.003
IFIT1	0.57	$< .001$	0.70	0.008
IFNG			0.69	0.040
IGF1	0.63	$< .001$	0.59	$< .001$
IGF2	0.67	0.019	0.70	0.005
IGFBP2	0.53	$< .001$	0.63	$< .001$
IGFBP5	0.57	$< .001$	0.71	0.006
IGFBP6	0.41	$< .001$	0.71	0.012
IL10	0.59	0.020		
IL1B	0.53	$< .001$	0.70	0.005

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
IL6	0.55	0.001		
IL6ST	0.45	$< .001$	0.68	$< .001$
IL8	0.60	0.005	0.70	0.008
ILK	0.68	0.029	0.76	0.036
ING5	0.54	$< .001$	0.82	0.033
ITGA1	0.66	0.017		
ITGA3	0.70	0.020		
ITGA5	0.64	0.011		
ITGA6	0.66	0.003	0.74	0.006
ITGA7	0.50	$< .001$	0.71	0.010
ITGB4	0.63	0.014	0.73	0.010
ITPR1	0.55	$< .001$		
ITPR3			0.76	0.007
JUN	0.37	$< .001$	0.54	$< .001$
JUNB	0.58	0.002	0.71	0.016
KCTD12	0.68	0.017		
KIT	0.49	0.002	0.76	0.043
KLC1	0.61	0.005	0.77	0.045
KLF6	0.65	0.009		
KLK1	0.68	0.036		
KLK10			0.76	0.037
KLK2	0.64	$< .001$	0.73	0.006
KLK3	0.65	$< .001$	0.76	0.021
KLRK1	0.63	0.005		
KRT15	0.52	$< .001$	0.58	$< .001$
KRT18	0.46	$< .001$		
KRT5	0.51	$< .001$	0.58	$< .001$
KRT8	0.53	$< .001$		
L1CAM	0.65	0.031		
LAG3	0.58	0.002	0.76	0.033
LAMA4	0.52	0.018		

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
LAMB3	0.60	0.002	0.65	0.003
LGALS3	0.52	$< .001$	0.71	0.002
LIG3	0.65	0.011		
LRP1	0.61	0.001	0.75	0.040
MGMT	0.66	0.003		
MICA	0.59	0.001	0.68	0.001
MLXIP	0.70	0.020		
MMP2	0.68	0.022		
MMP9	0.67	0.036		
MPPED2	0.57	$< .001$	0.66	$< .001$
MRC1	0.69	0.028		
MTSS1	0.63	0.005	0.79	0.037
MVP	0.62	$< .001$		
MYBPC1	0.53	$< .001$	0.70	0.011
NCAM1	0.70	0.039	0.77	0.042
NCAPD3	0.52	$< .001$	0.59	$< .001$
NDRG1			0.69	0.008
NEXN	0.62	0.002		
NFAT5	0.45	$< .001$	0.59	$< .001$
NFATC2	0.68	0.035	0.75	0.036
NFKBIA	0.70	0.030		
NRG1	0.59	0.022	0.71	0.018
OAZ1	0.69	0.018	0.62	$< .001$
OLFML3	0.59	$< .001$	0.72	0.003
OR51E2	0.73	0.013		
PAGE4	0.42	$< .001$	0.62	$< .001$
PCA3	0.53	$< .001$		
PCDHGB7	0.70	0.032		
PGF	0.68	0.027	0.71	0.013
PGR			0.76	0.041
PIK3C2A			0.80	$< .001$
PIK3CA	0.61	$< .001$	0.80	0.036

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
PIK3CG	0.67	0.001	0.76	0.018
PLP2	0.65	0.015	0.72	0.010
PPAP2B	0.45	$< .001$	0.69	0.003
PPP1R12A	0.61	0.007	0.73	0.017
PRIMA1	0.51	$< .001$	0.68	0.004
PRKCA	0.55	$< .001$	0.74	0.009
PRKCB	0.55	$< .001$		
PROM1			0.67	0.042
PROS1	0.73	0.036		
PTCH1	0.69	0.024	0.72	0.010
PTEN	0.54	$< .001$	0.64	$< .001$
PTGS2	0.48	$< .001$	0.55	$< .001$
PTH1R	0.57	0.003	0.77	0.050
PTHLH	0.55	0.010		
PTK2B	0.56	$< .001$	0.70	0.001
PYCARD			0.73	0.009
RAB27A	0.65	0.009	0.71	0.014
RAB30	0.59	0.003	0.72	0.010
RAGE			0.76	0.011
RARB	0.59	$< .001$	0.63	$< .001$
RASSF1	0.67	0.003		
RB1	0.67	0.006		
RFX1	0.71	0.040	0.70	0.003
RHOA	0.71	0.038	0.65	$< .001$
RHOB	0.58	0.001	0.71	0.006
RND3	0.54	$< .001$	0.69	0.003
RNF114	0.59	0.004	0.68	0.003
SCUBE2	SCUBE2		0.77	0.046
SDHC	0.72	0.028	0.76	0.025
SEC23A			0.75	0.029
SEMA3A	0.61	0.004	0.72	0.011

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
SEPT9	0.66	0.013	0.76	0.036
SERPINB5			0.75	0.039
SH3RF2	0.44	$< .001$	0.48	$< .001$
SHH			0.74	0.049
SLC22A3	0.42	$< .001$	0.61	$< .001$
SMAD4	0.45	$< .001$	0.66	$< .001$
SMARCD1	0.69	0.016		
SOD1	0.68	0.042		
SORBS1	0.51	$< .001$	0.73	0.012
SPARCL1	0.58	$< .001$	0.77	0.040
SPDEF	0.77	$< .001$		
SPINT1	0.65	0.004	0.79	0.038
SRC	0.61	$< .001$	0.69	0.001
SRD5A2	0.39	$< .001$	0.55	$< .001$
ST5	0.61	$< .001$	0.73	0.012
STAT1	0.64	0.006		
STAT3	0.63	0.010		
STAT5A	0.62	0.001	0.70	0.003
STAT5B	0.58	$< .001$	0.73	0.009
SUMO1	0.66	$< .001$		
SVIL	0.57	0.001	0.74	0.022
TBP	0.65	0.002		
TFF1	0.65	0.021		
TFF3	0.58	$< .001$		
TGFB1I1	0.51	$< .001$	0.75	0.026
TGFB2	0.48	$< .001$	0.62	$< .001$
TGFBR2	0.61	0.003		
TIAM1	0.68	0.019		
TIMP2	0.69	0.020		
TIMP3	0.58	0.002		
TNFRSF10A	0.73	0.047		
TNFRSF10B	0.47	$< .001$	0.70	0.003

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion negative in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)				
Table 7B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
TNFSF10	0.56	0.001		
TP63			0.67	0.001
TPM1	0.58	0.004	0.73	0.017
TPM2	0.46	$< .001$	0.70	0.005
TRA2A	0.68	0.013		
TRAF3IP2	0.73	0.041	0.71	0.004
TRO	0.72	0.016	0.71	0.004
TUBB2A	0.53	$< .001$	0.73	0.021
TYMP	0.70	0.011		
VCAM1	0.69	0.041		
VCL	0.46	$< .001$		
VEGFA			0.77	0.039
VEGFB	0.71	0.035		
VIM	0.60	0.001		
XRCC5			0.75	0.026
YY1	0.62	0.008	0.77	0.039
ZFHX3	0.53	$< .001$	0.58	$< .001$
ZFP36	0.43	$< .001$	0.54	$< .001$
ZNF827	0.55	0.001		

[0141] Tables 8A and 8B provide genes that were significantly associated ($p < 0.05$), positively or negatively, with clinical recurrence (cRFI) in positive TMPRSS fusion specimens in the primary or highest Gleason pattern specimen. Increased expression of genes in Table 8A is negatively associated with good prognosis, while increased expression of genes in Table 8B is positively associated with good prognosis.

Table 8A.

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)				
Table 8A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
ACTR2	1.78	0.017		

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 8A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
AKR1C3	1.44	0.013		
ALCAM			1.44	0.022
ANLN	1.37	0.046	1.81	$< .001$
APOE	1.49	0.023	1.66	0.005
AQP2			1.30	0.013
ARHGDIB	1.55	0.021		
ASPN	2.13	$< .001$	2.43	$< .001$
ATP5E	1.69	0.013	1.58	0.014
BGN	1.92	$< .001$	2.55	$< .001$
BIRC5	1.48	0.006	1.89	$< .001$
BMP6	1.51	0.010	1.96	$< .001$
BRCA2			1.41	0.007
BUB1	1.36	0.007	1.52	$< .001$
CCNE2	1.55	0.004	1.59	$< .001$
CD276			1.65	$< .001$
CDC20	1.68	$< .001$	1.74	$< .001$
CDH11			1.50	0.017
CDH18	1.36	$< .001$		
CDH7	1.54	0.009	1.46	0.026
CDKN2B	1.68	0.008	1.93	0.001
CDKN2C	2.01	$< .001$	1.77	$< .001$
CDKN3	1.51	0.002	1.33	0.049
CENPF	1.51	0.007	2.04	$< .001$
CKS2	1.43	0.034	1.56	0.007
COL1A1	2.23	$< .001$	3.04	$< .001$
COL1A2	1.79	0.001	2.2 2	$< .001$
COL3A1	1.96	$< .001$	2.81	$< .001$
COL4A1			1.52	0.020
COL5A1			1.50	0.020
COL5A2	1.64	0.017	1.55	0.010
COL8A1	1.96	$< .001$	2.38	$< .001$
CRISP3	1.68	0.002	1.67	0.002

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 8A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
CTHRC1			2.06	$< .001$
CTNND2	1.42	0.046	1.50	0.025
CTSK			1.43	0.049
CXCR4	1.82	0.001	1.64	0.007
DDIT4	1.54	0.016	1.58	0.009
DLL4			1.51	0.007
DYNLL1	1.50	0.021	1.22	0.002
F2R	2.2 7	$< .001$	2.02	$< .001$
FAP			2.12	$< .001$
FCGR3A			1.94	0.002
FGF5	1.23	0.047		
FOXP3	1.52	0.006	1.48	0.018
GNPTAB			1.44	0.042
GPR68			1.51	0.011
GREM1	1.91	$< .001$	2.38	$< .001$
HDAC1			1.43	0.048
HDAC9	1.65	$< .001$	1.67	0.004
HRAS	1.65	0.005	1.58	0.021
IGFBP3	1.94	$< .001$	1.85	$< .001$
INHBA	2.03	$< .001$	2.64	$< .001$
JAG1	1.41	0.027	1.50	0.008
KCTD12			1.51	0.017
KHDRBS3	1.48	0.029	1.54	0.014
KPNA2			1.46	0.050
LAMA3	1.35	0.040		
LAMC1	1.77	0.012		
LTBP2			1.82	$< .001$
LUM	1.51	0.021	1.53	0.009
MELK	1.38	0.020	1.49	0.001
MKI67			1.37	0.014
MMP11	1.73	$< .001$	1.69	$< .001$

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 8A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
MRPL13			1.30	0.046
MYBL2	1.56	$< .001$	1.72	$< .001$
MYLK3			1.17	0.007
NOX4	1.58	0.005	1.96	$< .001$
NRIP3			1.30	0.040
NRP1			1.53	0.021
OLFML2B			1.54	0.024
OSM	1.43	0.018		
PATE1	1.20	$< .001$	1.33	$< .001$
PCNA			1.64	0.003
PEX10	1.41	0.041	1.64	
PEX10 PIK3CA	1.38	0.037		
PLK1	1.52	0.009	1.67	0.002
PLOD2			1.65	0.002
POSTN	1.79	$< .001$	2.06	$< .001$
PTK6	1.67	0.002	2.38	$< .001$
PTTG1	1.56	0.002	1.54	0.003
RAD21	1.61	0.036	1.53	0.005
RAD51			1.33	0.009
RALA	1.95	0.004	1.60	0.007
REG4			1.43	0.042
ROBO2	1.46	0.024		
RRM1			1.44	0.033
RRM2	1.50	0.003	1.48	$< .001$
SAT1	1.42	0.009	1.43	0.012
SEC14L1			1.64	0.002
SFRP4	2.07	$< .001$	2.40	$< .001$
SHMT2	1.52	0.030	1.60	0.001
SLC44A1			1.42	0.039
SPARC	1.93	$< .001$	2.21	$< .001$
SULF1	1.63	0.006	2.04	$< .001$
THBS2	1.95	$< .001$	2.26	$< .001$

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)

Table 8A	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
THY1	1.69	0.016	1.95	0.002
TK1			1.43	0.003
TOP2A	1.57	0.002	2.11	$< .001$
TPX2	1.84	$< .001$	2.27	$< .001$
UBE2C	1.41	0.011	1.44	0.006
UBE2T	1.63	0.001		
UHRF1	1.51	0.007	1.69	$< .001$
WISP1	1.47	0.045		
WNT5A	1.35	0.027	1.63	0.001
ZWINT	1.36	0.045		

Table 8B.

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 8B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
AAMP	0.57	0.007	0.58	$< .001$
ABCA5			0.80	0.044
ACE	0.65	0.023	0.55	$< .001$
ACOX2			0.55	$< .001$
ADH5			0.68	0.022
AKAP1			0.81	0.043
ALDH1A2	0.72	0.036	0.43	$< .001$
ANPEP	0.66	0.022	0.46	$< .001$
APRT			0.73	0.040
AXIN2			0.60	$< .001$
AZGP1	0.57	$< .001$	0.65	$< .001$
BCL2			0.69	0.035
BIK	0.71	0.045		
BIN1	0.71	0.004	0.71	0.009
BTRC	0.66	0.003	0.58	$< .001$

Genes significantly (p<0.05) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 8B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
C7			0.64	0.006
CADM1	0.61	<.001	0.47	<.001
CCL2			0.73	0.042
CCNH	0.69	0.022		
CD44	0.56	<.001	0.58	<.001
CD82			0.72	0.033
CDC25B	0.74	0.028		
CDH1	0.75	0.030	0.72	0.010
CDH19			0.56	0.015
CDK3			0.78	0.045
CDKN1C	0.74	0.045	0.70	0.014
CSF1			0.72	0.037
CTSB			0.69	0.048
CTSL2			0.58	0.005
CYP3A5	0.51	<.001	0.30	<.001
DHX9	0.89	0.006	0.87	0.012
DLC1			0.64	0.023
DLGAP1	0.69	0.010	0.49	<.001
DPP4	0.64	<.001	0.56	<.001
DPT			0.63	0.003
EGR1			0.69	0.035
EGR3			0.68	0.025
EIF2S3			0.70	0.021
EIF5	0.71	0.030		
ELK4	0.71	0.041	0.60	0.003
EPHA2	0.72	0.036	0.66	0.011
EPHB4			0.65	0.007
ERCC1			0.68	0.023
ESR2			0.64	0.027
FAM107A	0.64	0.003	0.61	0.003
FAM13C	0.68	0.006	0.55	<.001
FGFR2	0.73	0.033	0.59	<.001

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 8B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
FKBP5			0.60	0.006
FLNC	0.68	0.024	0.65	0.012
FLT1			0.71	0.027
FOS			0.62	0.006
FOXO1			0.75	0.010
GADD45B			0.68	0.020
GHR			0.62	0.006
GPM6B			0.57	$< .001$
GSTM1	0.68	0.015	0.58	$< .001$
GSTM2	0.65	0.005	0.47	$< .001$
HGD	0.63	0.001	0.71	0.020
HK1	0.67	0.003	0.62	0.002
HLF			0.59	$< .001$
HNF1B	0.66	0.004	0.61	0.001
IER3			0.70	0.026
IGF1	0.63	0.005	0.55	$< .001$
IGF1R			0.76	0.049
IGFBP2	0.59	0.007	0.64	0.003
IL6ST			0.65	0.005
IL8	0.61	0.005	0.66	0.019
ILK			0.64	0.015
ING5	0.73	0.033	0.70	0.009
ITGA7	0.72	0.045	0.69	0.019
ITGB4			0.63	0.002
KLC1			0.74	0.045
KLK1	0.56	0.002	0.49	$< .001$
KLK10			0.68	0.013
KLK11			0.66	0.003
KLK2	0.66	0.045	0.65	0.011
KLK3	0.75	0.048	0.77	0.014
KRT15	0.71	0.017	0.50	$< .001$

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 8B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
KRT5	0.73	0.031	0.54	<.001
LAMA5			0.70	0.044
LAMB3	0.70	0.005	0.58	<.001
LGALS3			0.69	0.025
LIG3			0.68	0.022
MDK	0.69	0.035		
MGMT	0.59	0.017	0.60	<.001
MGST1			0.73	0.042
MICA			0.70	0.009
MPPED2	0.72	0.031	0.54	<.001
MTSS 1	0.62	0.003		
MYBPC1			0.50	<.001
NCAPD3	0.62	0.007	0.38	<.001
NCOR1			0.82	0.048
NFAT5	0.60	0.001	0.62	<.001
NRG1	0.66	0.040	0.61	0.029
NUP62	0.75	0.037		
OMD	0.54	<.001		
PAGE4			0.64	0.005
PCA3			0.66	0.012
PCDHGB7			0.68	0.018
PGR			0.60	0.012
PPAP2B			0.62	0.010
PPP1R12A	0.73	0.031	0.58	0.003
PRIMA 1			0.65	0.013
PROM1	0.41	0.013		
PTCH1	0.64	0.006		
PTEN			0.75	0.047
PTGS2			0.67	0.011
PTK2B			0.66	0.005
PTPN1			0.71	0.026
RAGE	0.70	0.012		

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 8B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
RARB			0.68	0.016
RGS10			0.84	0.034
RHOB			0.66	0.016
RND3			0.63	0.004
SDHC	0.73	0.044	0.69	0.016
SERPINA3	0.67	0.011	0.51	<.001
SERPINB5			0.42	<.001
SH3RF2	0.66	0.012	0.51	<.001
SLC22A3	0.59	0.003	0.48	<.001
SMAD4	0.64	0.004	0.49	<.001
SMARCC2			0.73	0.042
SMARCD1	0.73	<.001	0.76	0.035
SMO			0.64	0.006
SNAI1			0.53	0.008
SOD1			0.60	0.003
SRC	0.64	<.001	0.61	<.001
SRD5A2	0.63	0.004	0.59	<.001
STAT3			0.64	0.014
STAT5A			0.70	0.032
STAT5B	0.74	0.034	0.63	0.003
SVIL			0.71	0.028
TGFB1I1			0.68	0.036
TMPRSS2	0.72	0.015	0.67	<.001
TNFRSF10A			0.69	0.010
TNFRSF10B	0.67	0.007	0.64	0.001
TNFRSF18	0.38	0.003		
TNFSF10			0.71	0.025
TP53	0.68	0.004	0.57	<.001
TP63	0.75	0.049	0.52	<.001
TPM2			0.62	0.007
TRAF3IP2	0.71	0.017	0.68	0.005

Genes significantly ($p < 0.05$) associated with cRFI for TMPRSS2-ERG fusion positive in the primary Gleason pattern or highest Gleason pattern with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 8B	Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value
TRO			0.72	0.033
TUBB2A			0.69	0.038
VCL			0.62	$< .001$
VEGFA			0.71	0.037
WWOX			0.65	0.004
ZFH3	0.77	0.011	0.73	0.012
ZFP36			0.69	0.018
ZNF827	0.68	0.013	0.49	$< .001$

[0142] Tables 9A and 9B provide genes significantly associated ($p < 0.05$), positively or negatively, with TMPRSS fusion status in the primary Gleason pattern. Increased expression of genes in Table 9A are positively associated with TMPRSS fusion positivity, while increased expression of genes in Table 10A are negatively associated with TMPRSS fusion positivity.

Table 9A.

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) > 1.0 (increased expression is positively associated with TMPRSS fusion positivity)					
Table 9A					
Official Symbol	p-value	Odds Ratio	Official Symbol	p-value	Odds Ratio
ABCC8	$< .001$	1.86	MAP3K5	$< .001$	2.06
ALDH18A1	0.005	1.49	MAP7	$< .001$	2.74
ALKBH3	0.043	1.30	MSH2	0.005	1.59
ALOX5	$< .001$	1.66	MSH3	0.006	1.45
AMPD3	$< .001$	3.92	MUC1	0.012	1.42
APEX1	$< .001$	2.00	MYO6	$< .001$	3.79
ARHGDIB	$< .001$	1.87	NCOR2	0.001	1.62
ASAP2	0.019	1.48	NDRG1	$< .001$	6.77
ATXN1	0.013	1.41	NETO2	$< .001$	2.63
BMPR1B	$< .001$	2.37	ODC1	$< .001$	1.98
CACNA1D	$< .001$	9.01	OR51E1	$< .001$	2.24
CADPS	0.015	1.39	PDE9A	$< .001$	2.21
CD276	0.003	2.25	PEX10	$< .001$	3.41

Genes significantly (p<0.05) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) > 1.0 (increased expression is positively associated with TMPRSS fusion positivity)

Table 9A					
Official Symbol	p-value	Odds Ratio	Official Symbol	p-value	Odds Ratio
CDH1	0.016	1.37	PGK1	0.022	1.33
CDH7	<.001	2.22	PLA2G7	<.001	5.51
CDK7	0.025	1.43	PPP3CA	0.047	1.38
COL9A2	<.001	2.58	PSCA	0.013	1.43
CRISP3	<.001	2.60	PSMD13	0.004	1.51
CTNND1	0.033	1.48	PTCH1	0.022	1.38
ECE1	<.001	2.22	PTK2	0.014	1.38
EIF5	0.023	1.34	PTK6	<.001	2.29
EPHB4	0.005	1.51	PTK7	<.001	2.45
ERG	<.001	14.5	PTPRK	<.001	1.80
FAM171B	0.047	1.32	RAB30	0.001	1.60
FAM73A	0.008	1.45	REG4	0.018	1.58
FASN	0.004	1.50	RELA	0.001	1.62
GNPTAB	<.001	1.60	RFX 1	0.020	1.43
GPS1	0.006	1.45	RGS10	<.001	1.71
GRB7	0.023	1.38	SCUBE2	0.009	1.48
HDAC1	<.001	4.95	SEPT9	<.001	3.91
HGD	<.001	1.64	SH3RF2	0.004	1.48
HIP1	<.001	1.90	SH3YL1	<.001	1.87
HNF1B	<.001	3.55	SHH	<.001	2.45
HSPA8	0.041	1.32	SIM2	<.001	1.74
IGF1R	0.001	1.73	SIPA1L1	0.021	1.35
ILF3	<.001	1.91	SLC22A3	<.001	1.63
IMMT	0.025	1.36	SLC44A1	<.001	1.65
ITPR1	<.001	2.72	SPINT1	0.017	1.39
ITPR3	<.001	5.91	TFDP1	0.005	1.75
JAG1	0.007	1.42	TMPRSS2ERGA	0.002	14E5
KCNN2	<.001	2.80	TMPRSS2ERGB	<.001	1.97
KHDRBS3	<.001	2.63	TRIM14	<.001	1.65
KIAA0247	0.019	1.38	TSTA3	0.018	1.38
KLK11	<.001	1.98	UAP1	0.046	1.39
LAMC1	0.008	1.56	UBE2G1	0.001	1.66

Genes significantly (p<0.05) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) > 1.0 (increased expression is positively associated with TMPRSS fusion positivity)

Table 9A					
Official Symbol	p-value	Odds Ratio	Official Symbol	p-value	Odds Ratio
LAMC2	<.001	3.30	UGDH	<.001	2.22
LOX	0.009	1.41	XRCC5	<.001	1.66
LRP1	0.044	1.30	ZMYND8	<.001	2.19

Table 9B.

Genes significantly (p<0.05) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)

Table 9B		
Official Symbol	p-value	Odds Ratio
ABCC4	0.045	0.77
ABHD2	<.001	0.38
ACTR2	0.027	0.73
ADAMTS1	0.024	0.58
ADH5	<.001	0.58
AGTR2	0.016	0.64
AKAP1	0.013	0.70
AKT2	0.015	0.71
ALCAM	<.001	0.45
ALDH1A2	0.004	0.70
ANPEP	<.001	0.43
ANXA2	0.010	0.71
APC	0.036	0.73
APOC1	0.002	0.56
APOE	<.001	0.44
ARF1	0.041	0.77
ATM	0.036	0.74
AURKB	<.001	0.62
AZGP1	<.001	0.54
BBC3	0.030	0.74
BCL2	0.012	0.70
BIN1	0.021	0.74
BTG1	0.004	0.67

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)

Table 9B		
Official Symbol	p-value	Odds Ratio
BTG3	0.003	0.63
C7	0.023	0.74
CADM1	0.007	0.69
CASP1	0.011	0.70
CAV1	0.011	0.71
CCND1	0.019	0.72
CCR1	0.022	0.73
CD44	$< .001$	0.57
CD68	$< .001$	0.54
CD82	0.002	0.66
CDH5	0.007	0.66
CDKN1A	$< .001$	0.60
CDKN2B	$< .001$	0.54
CDKN2C	0.012	0.72
CDKN3	0.037	0.77
CHN1	0.038	0.75
CKS2	$< .001$	0.48
COL11A1	0.017	0.72
COL1A1	$< .001$	0.59
COL1A2	0.001	0.62
COL3A1	0.027	0.73
COL4A1	0.043	0.76
COL5A1	0.039	0.74
COL5A2	0.026	0.73
COL6A1	0.008	0.66
COL6A3	$< .001$	0.59
COL8A1	0.022	0.74
CSF1	0.011	0.70
CTNNB1	0.021	0.69
CTSB	$< .001$	0.62
CTSD	0.036	0.68
CTSK	0.007	0.70

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)

Table 9B		
Official Symbol	p-value	Odds Ratio
CTSS	0.002	0.64
CXCL12	$< .001$	0.48
CXCR4	0.005	0.68
CXCR7	0.046	0.76
CYR61	0.004	0.65
DAP	0.002	0.64
DARC	0.021	0.73
DDR2	0.021	0.73
DHRS9	$< .001$	0.52
DIAPH1	$< .001$	0.56
DICER1	0.029	0.75
DLC1	0.013	0.72
DLGAP1	$< .001$	0.60
DLL4	$< .001$	0.57
DPT	0.006	0.68
DUSP1	0.012	0.68
DUSP6	0.001	0.62
DVL1	0.037	0.75
EFNB2	$< .001$	0.32
EGR1	0.003	0.65
ELK4	$< .001$	0.60
ERBB2	$< .001$	0.61
ERBB3	0.045	0.76
ESR2	0.010	0.70
ETV1	0.042	0.74
FABP5	$< .001$	0.21
FAM13C	0.006	0.67
FCGR3A	0.018	0.72
FGF17	0.009	0.71
FGF6	0.011	0.70
FGF7	0.003	0.63
FN1	0.006	0.69

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)

Table 9B		
Official Symbol	p-value	Odds Ratio
FOS	0.035	0.74
FOXP3	0.010	0.71
GABRG2	0.029	0.74
GADD45B	0.003	0.63
GDF15	$< .001$	0.54
GPM6B	0.004	0.67
GPNMB	0.001	0.62
GSN	0.009	0.69
HLA-G	0.050	0.74
HLF	0.018	0.74
HPS1	$< .001$	0.48
HSD17B3	0.003	0.60
HSD17B4	$< .001$	0.56
HSPB1	$< .001$	0.38
HSPB2	0.002	0.62
IFI30	0.049	0.75
IFNG	0.006	0.64
IGF1	0.016	0.73
IGF2	0.001	0.57
IGFBP2	$< .001$	0.51
IGFBP3	$< .001$	0.59
IGFBP6	$< .001$	0.57
IL10	$< .001$	0.62
IL17A	0.012	0.63
IL1A	0.011	0.59
IL2	0.001	0.63
IL6ST	$< .001$	0.52
INSL4	0.014	0.71
ITGA1	0.009	0.69
ITGA4	0.007	0.68
JUN	$< .001$	0.59
KIT	$< .001$	0.64

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)

Table 9B		
Official Symbol	p-value	Odds Ratio
KRT76	0.016	0.70
LAG3	0.002	0.63
LAPTM5	$< .001$	0.58
LGALS3	$< .001$	0.53
LTBP2	0.011	0.71
LUM	0.012	0.70
MAOA	0.020	0.73
MAP4K4	0.007	0.68
MGST1	$< .001$	0.54
MMP2	$< .001$	0.61
MPPED2	$< .001$	0.45
MRC1	0.005	0.67
MTPN	0.002	0.56
MTSS1	$< .001$	0.53
MVP	0.009	0.72
MYBPC1	$< .001$	0.51
MYLK3	0.001	0.58
NCAM1	$< .001$	0.59
NCAPD3	$< .001$	0.40
NCOR1	0.004	0.69
NFKBIA	$< .001$	0.63
NNMT	0.006	0.66
NPBWR1	0.027	0.67
OAZ1	0.049	0.64
OLFML3	$< .001$	0.56
OSM	$< .001$	0.64
PAGE1	0.012	0.52
PDGFRB	0.016	0.73
PECAM1	$< .001$	0.55
PGR	0.048	0.77
PIK3CA	$< .001$	0.55
PIK3CG	0.008	0.71

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)

Table 9B		
Official Symbol	p-value	Odds Ratio
PLAU	0.044	0.76
PLK1	0.006	0.68
PLOD2	0.013	0.71
PLP2	0.024	0.73
PNLIPRP2	0.009	0.70
PPAP2B	$< .001$	0.62
PRKAR2B	$< .001$	0.61
PRKCB	0.044	0.76
PROS1	0.005	0.67
PTEN	$< .001$	0.47
PTGER3	0.007	0.69
PTH1R	0.011	0.70
PTK2B	$< .001$	0.61
PTPN1	0.028	0.73
RAB27A	$< .001$	0.21
RAD51	$< .001$	0.51
RAD9A	0.030	0.75
RARB	$< .001$	0.62
RASSF1	0.038	0.76
RECK	0.009	0.62
RHOB	0.004	0.64
RHOC	$< .001$	0.56
RLN1	$< .001$	0.30
RND3	0.014	0.72
S100P	0.002	0.66
SDC2	$< .001$	0.61
SEMA3A	0.001	0.64
SMAD4	$< .001$	0.64
SPARC	$< .001$	0.59
SPARCL1	$< .001$	0.56
SPINK1	$< .001$	0.26
SRD5A1	0.039	0.76

Genes significantly ($p < 0.05$) associated with TMPRSS fusion status in the primary Gleason pattern with odds ratio (OR) < 1.0 (increased expression is negatively associated with TMPRSS fusion positivity)		
Table 9B		
Official Symbol	p-value	Odds Ratio
STAT1	0.026	0.74
STS	0.006	0.64
SULF1	$< .001$	0.53
TFF3	$< .001$	0.19
TGFA	0.002	0.65
TGFB11I	0.040	0.77
TGFB2	0.003	0.66
TGFB3	$< .001$	0.54
TGFBR2	$< .001$	0.61
THY1	$< .001$	0.63
TIMP2	0.004	0.66
TIMP3	$< .001$	0.60
TMPRSS2	$< .001$	0.40
TNFSF11	0.026	0.63
TPD52	0.002	0.64
TRAM1	$< .001$	0.45
TRPC6	0.002	0.64
TUBB2A	$< .001$	0.49
VCL	$< .001$	0.57
VEGFB	0.033	0.73
VEGFC	$< .001$	0.61
VIM	0.012	0.69
WISP1	0.030	0.75
WNT5A	$< .001$	0.50

[0143] A molecular field effect was investigated, and determined that the expression levels of histologically normal-appearing cells adjacent to the tumor exhibited a molecular signature of prostate cancer. Tables 10A and 10B provide genes significantly associated ($p < 0.05$), positively or negatively, with cRFI or bRFI in non-tumor samples. Table 10A is negatively associated with good prognosis, while increased expression of genes in Table 10B is positively associated with good prognosis.

Table 10A

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)				
Table 10A	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
ALCAM			1.278	0.036
ASPN	1.309	0.032		
BAG5	1.458	0.004		
BRCA2	1.385	<.001		
CACNA1D			1.329	0.035
CD164			1.339	0.020
CDKN2B	1.398	0.014		
COL3A1	1.300	0.035		
COL4A1	1.358	0.019		
CTNND2			1.370	0.001
DARC	1.451	0.003		
DICER1			1.345	<.001
DPP4			1.358	0.008
EFNB2			1.323	0.007
FASN			1.327	0.035
GHR			1.332	0.048
HSPA5			1.260	0.048
INHBA	1.558	<.001		
KCNN2			1.264	0.045
KRT76			1.115	<.001
LAMC1	1.390	0.014		
LAMC2			1.216	0.042
LIG3			1.313	0.030
MAOA			1.405	0.013
MCM6	1.307	0.036		
MKI67	1.271	0.008		
NEK2			1.312	0.016
NPBWR1	1.278	0.035		
ODC1			1.320	0.010
PEX10			1.361	0.014
PGK1	1.488	0.004		
PLA2G7			1.337	0.025

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) > 1.0 (increased expression is negatively associated with good prognosis)				
Table 10A	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
POSTN	1.306	0.043		
PTK6			1.344	0.005
REG4			1.348	0.009
RGS7			1.144	0.047
SFRP4	1.394	0.009		
TARP			1.412	0.011
TFF1			1.346	0.010
TGFBR2	1.310	0.035		
THY1	1.300	0.038		
TMPRSS2ERGA			1.333	<.001
TPD52			1.374	0.015
TRPC6	1.272	0.046		
UBE2C	1.323	0.007		
UHRF1	1.325	0.021		

Table 10B

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)				
Table 10B	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
ABCA5	0.807	0.028		
ABCC3	0.760	0.019	0.750	0.003
ABHD2	0.781	0.028		
ADAM15	0.718	0.005		
AKAP1	0.740	0.009		
AMPD3			0.793	0.013
ANGPT2			0.752	0.027
ANXA2			0.776	0.035
APC	0.755	0.014		
APRT	0.762	0.025		
AR	0.752	0.015		
ARHGDI1B			0.753	<.001
BIN1	0.738	0.016		

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)				
Table 10B	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
CADM1	0.711	0.004		
CCNH	0.820	0.041		
CCR1			0.749	0.007
CDK14			0.772	0.014
CDK3	0.819	0.044		
CDKN1C	0.808	0.038		
CHAF1A	0.634	0.002	0.779	0.045
CHN1			0.803	0.034
CHRA1	0.751	0.014	0.779	0.021
COL5A1			0.736	0.012
COL5A2			0.762	0.013
COL6A1			0.757	0.032
COL6A3			0.757	0.019
CSK	0.663	<.001	0.698	<.001
CTSK			0.782	0.029
CXCL12			0.771	0.037
CXCR7			0.753	0.008
CYP3A5	0.790	0.035		
DDIT4			0.725	0.017
DIAPH1			0.771	0.015
DLC1	0.744	0.004	0.807	0.015
DLGAP1	0.708	0.004		
DUSP1	0.740	0.034		
EDN1			0.742	0.010
EGR1	0.731	0.028		
EIF3H	0.761	0.024		
EIF4E	0.786	0.041		
ERBB2	0.664	0.001		
ERBB4	0.764	0.036		
ERCC1	0.804	0.041		
ESR2			0.757	0.025
EZH2			0.798	0.048

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)				
Table 10B	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
FAAH	0.798	0.042		
FAM13C	0.764	0.012		
FAM171B			0.755	0.005
FAM49B			0.811	0.043
FAM73A	0.778	0.015		
FASLG			0.757	0.041
FGFR2	0.735	0.016		
FOS	0.690	0.008		
FYN	0.788	0.035	0.777	0.011
GPNUMB			0.762	0.011
GSK3B	0.792	0.038		
HGD	0.774	0.017		
HIRIP3	0.802	0.033		
HSP90AB1	0.753	0.013		
HSPB1	0.764	0.021		
HSPE1	0.668	0.001		
IFI30			0.732	0.002
IGF2			0.747	0.006
IGFBP5			0.691	0.006
IL6ST			0.748	0.010
IL8			0.785	0.028
IMMT			0.708	<.001
ITGA6	0.747	0.008		
ITGAV			0.792	0.016
ITGB3			0.814	0.034
ITPR3	0.769	0.009		
JUN	0.655	0.005		
KHDRBS3			0.764	0.012
KLF6	0.714	<.001		
KLK2	0.813	0.048		
LAMA4			0.702	0.009
LAMA5	0.744	0.011		

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)

Table 10B	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
LAPTM5			0.740	0.009
LGALS3	0.773	0.036	0.788	0.024
LIMS1			0.807	0.012
MAP3K5			0.815	0.034
MAP3K7			0.809	0.032
MAP4K4	0.735	0.018	0.761	0.010
MAPKAPK3	0.754	0.014		
MICA	0.785	0.019		
MTA1			0.808	0.043
MVP			0.691	0.001
MYLK3			0.730	0.039
MYO6	0.780	0.037		
NCOA1			0.787	0.040
NCOR1			0.876	0.020
NDRG1	0.761	<.001		
NFAT5	0.770	0.032		
NFKBIA			0.799	0.018
NME2			0.753	0.005
NUP62			0.842	0.032
OAZ1			0.803	0.043
OLFML2B			0.745	0.023
OLFML3			0.743	0.009
OSM			0.726	0.018
PCA3	0.714	0.019		
PECAM1			0.774	0.023
PIK3C2A			0.768	0.001
PIM1	0.725	0.011		
PLOD2			0.713	0.008
PPP3CA	0.768	0.040		
PROM1			0.482	<.001
PTEN			0.807	0.012
PTGS2	0.726	0.011		

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)				
Table 10B	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
PTTG1			0.729	0.006
PYCARD			0.783	0.012
RAB30			0.730	0.002
RAGE	0.792	0.012		
RFX1	0.789	0.016	0.792	0.010
RGS10	0.781	0.017		
RUNX1			0.747	0.007
SDHC			0.827	0.036
SEC23A			0.752	0.010
SEPT9			0.889	0.006
SERPINA3			0.738	0.013
SLC25A21			0.788	0.045
SMARCD1	0.788	0.010	0.733	0.007
SMO	0.813	0.035		
SRC	0.758	0.026		
SRD5A2			0.738	0.005
ST5			0.767	0.022
STAT5A			0.784	0.039
TGFB2	0.771	0.027		
TGFB3			0.752	0.036
THBS2			0.751	0.015
TNFRSF10B	0.739	0.010		
TPX2			0.754	0.023
TRAF3IP2			0.774	0.015
TRAM1	0.868	<.001	0.880	<.001
TRIM 14	0.785	0.047		
TUBB2A	0.705	0.010		
TYMP			0.778	0.024
UAP1	0.721	0.013		
UTP23	0.763	0.007	0.826	0.018
VCL			0.837	0.040
VEGFA	0.755	0.009		

Genes significantly (p<0.05) associated with cRFI or bRFI in Non-Tumor Samples with hazard ratio (HR) < 1.0 (increased expression is positively associated with good prognosis)				
Table 10B	cRFI		bRFI	
Official Symbol	HR	p-value	HR	p-value
WDR19	0.724	0.005		
YBX1			0.786	0.027
ZFP36	0.744	0.032		
ZNF827	0.770	0.043		

[0144] Table 11 provides genes that are significantly associated (p<0.05) with cRFI or bRFI after adjustment for Gleason pattern or highest Gleason pattern.

Table 11

Genes significantly (p<0.05) associated with cRFI or bRFI after adjustment for Gleason pattern in the primary Gleason pattern or highest Gleason pattern Some HR <= 1.0 and some HR > 1.0						
Table 11	cRFI		bRFI		bRFI	
	Highest Pattern		Primary Pattern		Highest Pattern	
Official Symbol	HR	p-value	HR	p-value	HR	p-value
HSPA5	0.710	0.009	1.288	0.030		
ODC1	0.741	0.026	1.343	0.004	1.261	0.046

[0145] Tables 12A and 12B provide genes that are significantly associated (p<0.05) with prostate cancer specific survival (PCSS) in the primary Gleason pattern. Increased expression of genes in Table 12A is negatively associated with good prognosis, while increased expression of genes in Table 12B is positively associated with good prognosis.

Table 12A

Genes significantly (p<0.05) associated with prostate cancer specific survival (PCSS) in the Primary Gleason Pattern HR > 1.0 (Increased expression is negatively associated with good prognosis)					
Table 12A	HR	p-value	Official Symbol	HR	p-value
Official Symbol					
AKR1C3	1.476	0.016	GREM1	1.942	<.001
ANLN	1.517	0.006	IFI30	1.482	0.048
APOC1	1.285	0.016	IGFBP3	1.513	0.027
APOE	1.490	0.024	INHBA	3.060	<.001
ASPN	3.055	<.001	KIF4A	1.355	0.001
ATP5E	1.788	0.012	KLK14	1.187	0.004

Genes significantly ($p < 0.05$) associated with prostate cancer specific survival (PCSS) in the Primary Gleason Pattern HR > 1.0 (Increased expression is negatively associated with good prognosis)

Table 12A	HR	p-value	Official Symbol	HR	p-value
Official Symbol					
AURKB	1.439	0.008	LAPTM5	1.613	0.006
BGN	2.640	<.001	LTBP2	2.018	<.001
BIRC5	1.611	<.001	MMP11	1.869	<.001
BMP6	1.490	0.021	MYBL2	1.737	0.013
BRCA1	1.418	0.036	NEK2	1.445	0.028
CCNB1	1.497	0.021	NOX4	2.049	<.001
CD276	1.668	0.005	OLFML2B	1.497	0.023
CDC20	1.730	<.001	PLK1	1.603	0.006
CDH11	1.565	0.017	POSTN	2.585	<.001
CDH7	1.553	0.007	PPFIA3	1.502	0.012
CDKN2B	1.751	0.003	PTK6	1.527	0.009
CDKN2C	1.993	0.013	PTTG1	1.382	0.029
CDKN3	1.404	0.008	RAD51	1.304	0.031
CENPF	2.031	<.001	RGS7	1.251	<.001
CHAF1A	1.376	0.011	RRM2	1.515	<.001
CKS2	1.499	0.031	SAT1	1.607	0.004
COL1A1	2.574	<.001	SDC1	1.710	0.007
COL1A2	1.607	0.011	SESN3	1.399	0.045
COL3A1	2.382	<.001	SFRP4	2.384	<.001
COL4A1	1.970	<.001	SHMT2	1.949	0.003
COL5A2	1.938	0.002	SPARC	2.249	<.001
COL8A1	2.245	<.001	STMN1	1.748	0.021
CTHRC1	2.085	<.001	SULF1	1.803	0.004
CXCR4	1.783	0.007	THBS2	2.576	<.001
DDIT4	1.535	0.030	THY1	1.908	0.001
DYNLL1	1.719	0.001	TK1	1.394	0.004
F2R	2.169	<.001	TOP2A	2.119	<.001
FAM171B	1.430	0.044	TPX2	2.074	0.042
FAP	1.993	0.002	UBE2C	1.598	<.001
FCGR3A	2.099	<.001	UGT2B15	1.363	0.016
FN 1	1.537	0.024	UHRF1	1.642	0.001
GPR68	1.520	0.018	ZWINT	1.570	0.010

Table 12B

Genes significantly ($p < 0.05$) associated with prostate cancer specific survival (PCSS) in the Primary Gleason Pattern HR < 1.0 (Increased expression is positively associated with good prognosis)

Table 12B Official Symbol	HR	p-value	Official Symbol	HR	p-value
AAMP	0.649	0.040	IGFBP6	0.578	0.003
ABCA5	0.777	0.015	IL2	0.528	0.010
ABCG2	0.715	0.037	IL6ST	0.574	<.001
ACOX2	0.673	0.016	IL8	0.540	0.001
ADH5	0.522	<.001	ING5	0.688	0.015
ALDH1A2	0.561	<.001	ITGA6	0.710	0.005
AMACR	0.693	0.029	ITGA7	0.676	0.033
AMPD3	0.750	0.049	JUN	0.506	0.001
ANPEP	0.531	<.001	KIT	0.628	0.047
ATXN1	0.640	0.011	KLK1	0.523	0.002
AXIN2	0.657	0.002	KLK2	0.581	<.001
AZGP1	0.617	<.001	KLK3	0.676	<.001
BDKRB1	0.553	0.032	KRT15	0.684	0.005
BIN1	0.658	<.001	KRT18	0.536	<.001
BTRC	0.716	0.011	KRT5	0.673	0.004
C7	0.531	<.001	KRT8	0.613	0.006
CADM1	0.646	0.015	LAMB3	0.740	0.027
CASP7	0.538	0.029	LGALS3	0.678	0.007
CCNH	0.674	0.001	MGST1	0.640	0.002
CD164	0.606	<.001	MPPED2	0.629	<.001
CD44	0.687	0.016	MTSS1	0.705	0.041
CDK3	0.733	0.039	MYBPC1	0.534	<.001
CHN1	0.653	0.014	NCAPD3	0.519	<.001
COL6A1	0.681	0.015	NFAT5	0.536	<.001
CSF1	0.675	0.019	NRG1	0.467	0.007
CSRP1	0.711	0.007	OLFML3	0.646	0.001
CXCL12	0.650	0.015	OMD	0.630	0.006
CYP3A5	0.507	<.001	OR51E2	0.762	0.017
CYR61	0.569	0.007	PAGE4	0.518	<.001
DLGAP1	0.654	0.004	PCA3	0.581	<.001
DNM3	0.692	0.010	PGF	0.705	0.038
DPP4	0.544	<.001	PPAP2B	0.568	<.001

Genes significantly ($p < 0.05$) associated with prostate cancer specific survival (PCSS) in the Primary Gleason Pattern HR < 1.0 (Increased expression is positively associated with good prognosis)

Table 12B Official Symbol	HR	p-value	Official Symbol	HR	p-value
DPT	0.543	<.001	PPP1R12A	0.694	0.017
DUSP1	0.660	0.050	PRIMA1	0.678	0.014
DUSP6	0.699	0.033	PRKCA	0.632	0.001
EGR1	0.490	<.001	PRKCB	0.692	0.028
EGR3	0.561	<.001	PROM1	0.393	0.017
EIF5	0.720	0.035	PTEN	0.689	0.002
ERBB3	0.739	0.042	PTGS2	0.611	0.004
FAAH	0.636	0.010	PTH1R	0.629	0.031
FAM107A	0.541	<.001	RAB27A	0.721	0.046
FAM13C	0.526	<.001	RND3	0.678	0.029
FAS	0.689	0.030	RNF114	0.714	0.035
FGF10	0.657	0.024	SDHC	0.590	<.001
FKBP5	0.699	0.040	SERPINA3	0.710	0.050
FLNC	0.742	0.036	SH3RF2	0.570	0.005
FOS	0.556	0.005	SLC22A3	0.517	<.001
FOXQ1	0.666	0.007	SMAD4	0.528	<.001
GADD45B	0.554	0.002	SMO	0.751	0.026
GDF15	0.659	0.009	SRC	0.667	0.004
GHR	0.683	0.027	SRD5A2	0.488	<.001
GPM6B	0.666	0.005	STAT5B	0.700	0.040
GSN	0.646	0.006	SVIL	0.694	0.024
GSTM1	0.672	0.006	TFF3	0.701	0.045
GSTM2	0.514	<.001	TGFB111	0.670	0.029
HGD	0.771	0.039	TGFB2	0.646	0.010
HIRIP3	0.730	0.013	TNFRSF10B	0.685	0.014
HK1	0.778	0.048	TNFSF10	0.532	<.001
HLF	0.581	<.001	TPM2	0.623	0.005
HNF1B	0.643	0.013	TRO	0.767	0.049
HSD17B10	0.742	0.029	TUBB2A	0.613	0.003
IER3	0.717	0.049	VEGFB	0.780	0.034
IGF1	0.612	<.001	ZFP36	0.576	0.001
			ZNF827	0.644	0.014

[0146] Analysis of gene expression and upgrading/upstaging was based on univariate ordinal logistic regression models using weighted maximum likelihood estimators for each gene in the gene list (727 test genes and 5 reference genes). P-values were generated using a Wald test of the null hypothesis that the odds ratio (OR) is one. Both unadjusted p-values and the q-value (smallest FDR at which the hypothesis test in question is rejected) were reported. Unadjusted p-values <0.05 were considered statistically significant. Since two tumor specimens were selected for each patient, this analysis was performed using the 2 specimens from each patient as follows: (1) analysis using the primary Gleason pattern specimen from each patient (Specimens A1 and B2 as described in Table 2); and (2) analysis using the highest Gleason pattern specimen from each patient (Specimens A1 and B1 as described in Table 2). 200 genes were found to be significantly associated ($p < 0.05$) with upgrading/upstaging in the primary Gleason pattern sample (PGP) and 203 genes were found to be significantly associated ($p < 0.05$) with upgrading/upstaging in the highest Gleason pattern sample (HGP).

[0147] Tables 13A and 13B provide genes significantly associated ($p < 0.05$), positively or negatively, with upgrading/upstaging in the primary and/or highest Gleason pattern. Increased expression of genes in Table 13A is positively associated with higher risk of upgrading/upstaging (poor prognosis), while increased expression of genes in Table 13B is negatively associated with risk of upgrading/upstaging (good prognosis).

TABLE 13A

Genes significantly ($p < 0.05$) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR > 1.0 (Increased expression is positively associated with higher risk of upgrading/upstaging (poor prognosis))				
Table 13A	PGP		HGP	
Gene	OR	p-value	OR	p-value
ALCAM	1.52	0.0179	1.50	0.0184
ANLN	1.36	0.0451	.	.
APOE	1.42	0.0278	1.50	0.0140
ASPN	1.60	0.0027	2.06	0.0001
AURKA	1.47	0.0108	.	.
AURKB	.	.	1.52	0.0070
BAX	.	.	1.48	0.0095
BGN	1.58	0.0095	1.73	0.0034
BIRC5	1.38	0.0415	.	.
BMP6	1.51	0.0091	1.59	0.0071
BUB1	1.38	0.0471	1.59	0.0068
CACNA1D	1.36	0.0474	1.52	0.0078
CASP7	.	.	1.32	0.0450
CCNE2	1.54	0.0042	.	.
CD276	.	.	1.44	0.0265

Genes significantly (p<0.05) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR > 1.0 (Increased expression is positively associated with higher risk of upgrading/upstaging (poor prognosis))				
Table 13A	PGP		HGP	
Gene	OR	p-value	OR	p-value
CDC20	1.35	0.0445	1.39	0.0225
CDKN2B	.	.	1.36	0.0415
CENPF	1.43	0.0172	1.48	0.0102
CLTC	1.59	0.0031	1.57	0.0038
COL1A1	1.58	0.0045	1.75	0.0008
COL3A1	1.45	0.0143	1.47	0.0131
COL8A1	1.40	0.0292	1.43	0.0258
CRISP3	.	.	1.40	0.0256
CTHRC1	.	.	1.56	0.0092
DBN1	1.43	0.0323	1.45	0.0163
DIAPH1	1.51	0.0088	1.58	0.0025
DICER1	.	.	1.40	0.0293
DIO2	.	.	1.49	0.0097
DVL1	.	.	1.53	0.0160
F2R	1.46	0.0346	1.63	0.0024
FAP	1.47	0.0136	1.74	0.0005
FCGR3A	.	.	1.42	0.0221
HPN	.	.	1.36	0.0468
HSD17B4	.	.	1.47	0.0151
HSPA8	1.65	0.0060	1.58	0.0074
IL11	1.50	0.0100	1.48	0.0113
IL1B	1.41	0.0359	.	.
INHBA	1.56	0.0064	1.71	0.0042
KHDRBS3	1.43	0.0219	1.59	0.0045
KIF4A	.	.	1.50	0.0209
KPNA2	1.40	0.0366	.	.
KRT2	.	.	1.37	0.0456
KRT75	.	.	1.44	0.0389
MANF	.	.	1.39	0.0429
MELK	1.74	0.0016	.	.
MKI67	1.35	0.0408	.	.

Genes significantly ($p < 0.05$) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR > 1.0 (Increased expression is positively associated with higher risk of upgrading/upstaging (poor prognosis))				
Table 13A	PGP		HGP	
Gene	OR	p-value	OR	p-value
MMP11	.	.	1.56	0.0057
NOX4	1.49	0.0105	1.49	0.0138
PLAUR	1.44	0.0185	.	.
PLK1	.	.	1.41	0.0246
PTK6	.	.	1.36	0.0391
RAD51	.	.	1.39	0.0300
RAF1	.	.	1.58	0.0036
RRM2	1.57	0.0080	.	.
SESN3	1.33	0.0465	.	.
SFRP4	2.33	<0.0001	2.51	0.0015
SKIL	1.44	0.0288	1.40	0.0368
SOX4	1.50	0.0087	1.59	0.0022
SPINK1	1.52	0.0058	.	.
SPP1	.	.	1.42	0.0224
THBS2	.	.	1.36	0.0461
TK1	.	.	1.38	0.0283
TOP2A	1.85	0.0001	1.66	0.0011
TPD52	1.78	0.0003	1.64	0.0041
TPX2	1.70	0.0010	.	.
UBE2G1	1.38	0.0491	.	.
UBE2T	1.37	0.0425	1.46	0.0162
UHRF1	.	.	1.43	0.0164
VCPIP1	.	.	1.37	0.0458

TABLE 13B

Genes significantly ($p < 0.05$) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0 (Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))				
Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
ABCC3	.	.	0.70	0.0216
ABCC8	0.66	0.0121	.	.
ABCG2	0.67	0.0208	0.61	0.0071

Genes significantly (p<0.05) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0 (Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))				
Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
ACE	.	.	0.73	0.0442
ACOX2	0.46	0.0000	0.49	0.0001
ADH5	0.69	0.0284	0.59	0.0047
AIG1	.	.	0.60	0.0045
AKR1C1	.	.	0.66	0.0095
ALDH1A2	0.36	<0.0001	0.36	<0.0001
ALKBH3	0.70	0.0281	0.61	0.0056
ANPEP	.	.	0.68	0.0109
ANXA2	0.73	0.0411	0.66	0.0080
APC	.	.	0.68	0.0223
ATXN1	.	.	0.70	0.0188
AXIN2	0.60	0.0072	0.68	0.0204
AZGP1	0.66	0.0089	0.57	0.0028
BCL2	.	.	0.71	0.0182
BIN1	0.55	0.0005	.	.
BTRC	0.69	0.0397	0.70	0.0251
C7	0.53	0.0002	0.51	<0.0001
CADM1	0.57	0.0012	0.60	0.0032
CASP1	0.64	0.0035	0.72	0.0210
CAV1	0.64	0.0097	0.59	0.0032
CAV2	.	.	0.58	0.0107
CD164	.	.	0.69	0.0260
CD82	0.67	0.0157	0.69	0.0167
CDH1	0.61	0.0012	0.70	0.0210
CDK14	0.70	0.0354	.	.
CDK3	.	.	0.72	0.0267
CDKN1C	0.61	0.0036	0.56	0.0003
CHN1	0.71	0.0214	.	.
COL6A1	0.62	0.0125	0.60	0.0050
COL6A3	0.65	0.0080	0.68	0.0181
CSRP1	0.43	0.0001	0.40	0.0002

Genes significantly (p<0.05) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0 (Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))				
Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
CTSB	0.66	0.0042	0.67	0.0051
CTSD	0.64	0.0355	.	.
CTSK	0.69	0.0171	.	.
CTSL1	0.72	0.0402	.	.
CUL1	0.61	0.0024	0.70	0.0120
CXCL12	0.69	0.0287	0.63	0.0053
CYP3A5	0.68	0.0099	0.62	0.0026
DDR2	0.68	0.0324	0.62	0.0050
DES	0.54	0.0013	0.46	0.0002
DHX9	0.67	0.0164	.	.
DLGAP1	.	.	0.66	0.0086
DPP4	0.69	0.0438	0.69	0.0132
DPT	0.59	0.0034	0.51	0.0005
DUSP1	.	.	0.67	0.0214
EDN1	.	.	0.66	0.0073
EDNRA	0.66	0.0148	0.54	0.0005
EIF2C2	.	.	0.65	0.0087
ELK4	0.55	0.0003	0.58	0.0013
ENPP2	0.65	0.0128	0.59	0.0007
EPHA3	0.71	0.0397	0.73	0.0455
EPHB2	0.60	0.0014	.	.
EPHB4	0.73	0.0418	.	.
EPHX3	.	.	0.71	0.0419
ERCC1	0.71	0.0325	.	.
FAM107A	0.56	0.0008	0.55	0.0011
FAM13C	0.68	0.0276	0.55	0.0001
FAS	0.72	0.0404	.	.
FBN1	0.72	0.0395	.	.
FBXW7	0.69	0.0417	.	.
FGF10	0.59	0.0024	0.51	0.0001
FGF7	0.51	0.0002	0.56	0.0007
FGFR2	0.54	0.0004	0.47	<0.0001

Genes significantly (p<0.05) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0 (Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))				
Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
FLNA	0.58	0.0036	0.50	0.0002
FLNC	0.45	0.0001	0.40	<0.0001
FLT4	0.61	0.0045	.	.
FOXO1	0.55	0.0005	0.53	0.0005
FOXP3	0.71	0.0275	0.72	0.0354
GHR	0.59	0.0074	0.53	0.0001
GNRH1	0.72	0.0386	.	.
GPM6B	0.59	0.0024	0.52	0.0002
GSN	0.65	0.0107	0.65	0.0098
GSTM1	0.44	<0.0001	0.43	<0.0001
GSTM2	0.42	<0.0001	0.39	<0.0001
HLF	0.46	<0.0001	0.47	0.0001
HPS1	0.64	0.0069	0.69	0.0134
HSPA5	0.68	0.0113	.	.
HSPB2	0.61	0.0061	0.55	0.0004
HSPG2	0.70	0.0359	.	.
ID3	.	.	0.70	0.0245
IGF1	0.45	<0.0001	0.50	0.0005
IGF2	0.67	0.0200	0.68	0.0152
IGFBP2	0.59	0.0017	0.69	0.0250
IGFBP6	0.49	<0.0001	0.64	0.0092
IL6ST	0.56	0.0009	0.60	0.0012
ILK	0.51	0.0010	0.49	0.0004
ITGA1	0.58	0.0020	0.58	0.0016
ITGA3	0.71	0.0286	0.70	0.0221
ITGA5	.	.	0.69	0.0183
ITGA7	0.56	0.0035	0.42	<0.0001
ITGB1	0.63	0.0095	0.68	0.0267
ITGB3	0.62	0.0043	0.62	0.0040
ITPR1	0.62	0.0032	.	.
JUN	0.73	0.0490	0.68	0.0152

**Genes significantly ($p < 0.05$) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0
(Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))**

Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
KIT	0.55	0.0003	0.57	0.0005
KLC1	.	.	0.70	0.0248
KLK1	.	.	0.60	0.0059
KRT15	0.58	0.0009	0.45	<0.0001
KRT5	0.70	0.0262	0.59	0.0008
LAMA4	0.56	0.0359	0.68	0.0498
LAMB3	.	.	0.60	0.0017
LGALS3	0.58	0.0007	0.56	0.0012
LRP1	0.69	0.0176	.	.
MAP3K7	0.70	0.0233	0.73	0.0392
MCM3	0.72	0.0320	.	.
MMP2	0.66	0.0045	0.60	0.0009
MMP7	0.61	0.0015	0.65	0.0032
MMP9	0.64	0.0057	0.72	0.0399
MPPED2	0.72	0.0392	0.63	0.0042
MTA1	.	.	0.68	0.0095
MTSS 1	0.58	0.0007	0.71	0.0442
MVP	0.57	0.0003	0.70	0.0152
MYBPC1	.	.	0.70	0.0359
NCAM1	0.63	0.0104	0.64	0.0080
NCAPD3	0.67	0.0145	0.64	0.0128
NEXN	0.54	0.0004	0.55	0.0003
NFAT5	0.72	0.0320	0.70	0.0177
NUDT6	0.66	0.0102	.	.
OLFML3	0.56	0.0035	0.51	0.0011
OMD	0.61	0.0011	0.73	0.0357
PAGE4	0.42	<0.0001	0.36	<0.0001
PAK6	0.72	0.0335	.	.
PCDHGB7	0.70	0.0262	0.55	0.0004
PGF	0.72	0.0358	0.71	0.0270
PLP2	0.66	0.0088	0.63	0.0041
PPAP2B	0.44	<0.0001	0.50	0.0001

Genes significantly (p<0.05) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0 (Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))				
Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
PPP1R12A	0.45	0.0001	0.40	<0.0001
PRIMA1	.	.	0.63	0.0102
PRKAR2B	0.71	0.0226	.	.
PRKCA	0.34	<0.0001	0.42	<0.0001
PRKCB	0.66	0.0120	0.49	<0.0001
PROM1	0.61	0.0030	.	.
PTEN	0.59	0.0008	0.55	0.0001
PTGER3	0.67	0.0293	.	.
PTH1R	0.69	0.0259	0.71	0.0327
PTK2	0.75	0.0461	.	.
PTK2B	0.70	0.0244	0.74	0.0388
PYCARD	0.73	0.0339	0.67	0.0100
RAD9A	0.64	0.0124	.	.
RARB	0.67	0.0088	0.65	0.0116
RGS10	0.70	0.0219	.	.
RHOB	.	.	0.72	0.0475
RND3	.	.	0.67	0.0231
SDHC	0.72	0.0443	.	.
SEC23A	0.66	0.0101	0.53	0.0003
SEMA3A	0.51	0.0001	0.69	0.0222
SH3RF2	0.55	0.0002	0.54	0.0002
SLC22A3	0.48	0.0001	0.50	0.0058
SMAD4	0.49	0.0001	0.50	0.0003
SMARCC2	0.59	0.0028	0.65	0.0052
SMO	0.60	0.0048	0.52	<0.0001
SORBS1	0.56	0.0024	0.48	0.0002
SPARCL1	0.43	0.0001	0.50	0.0001
SRD5A2	0.26	<0.0001	0.31	<0.0001
ST5	0.63	0.0103	0.52	0.0006
STAT5A	0.60	0.0015	0.61	0.0037
STAT5B	0.54	0.0005	0.57	0.0008

Genes significantly ($p < 0.05$) associated with upgrading/upstaging in the Primary Gleason Pattern (PGP) and Highest Gleason Pattern (HGP) OR < 1.0 (Increased expression is negatively associated with higher risk of upgrading/upstaging (good prognosis))				
Table 13B	PGP		HGP	
Gene	OR	p-value	OR	p-value
SUMO1	0.65	0.0066	0.66	0.0320
SVIL	0.52	0.0067	0.46	0.0003
TGFB1I1	0.44	0.0001	0.43	0.0000
TGFB2	0.55	0.0007	0.58	0.0016
TGFB3	0.57	0.0010	0.53	0.0005
TIMP1	0.72	0.0224	.	.
TIMP2	0.68	0.0198	0.69	0.0206
TIMP3	0.67	0.0105	0.64	0.0065
TMPRSS2	.	.	0.72	0.0366
TNFRSF10A	0.71	0.0181	.	.
TNFSF10	0.71	0.0284	.	.
TOP2B	0.73	0.0432	.	.
TP63	0.62	0.0014	0.50	<0.0001
TPM1	0.54	0.0007	0.52	0.0002
TPM2	0.41	<0.0001	0.40	<0.0001
TPP2	0.65	0.0122	.	.
TRA2A	0.72	0.0318	.	.
TRAF3IP2	0.62	0.0064	0.59	0.0053
TRO	0.57	0.0003	0.51	0.0001
VCL	0.52	0.0005	0.52	0.0004
VIM	0.65	0.0072	0.65	0.0045
WDR19	0.66	0.0097	.	.
WFDC1	0.58	0.0023	0.60	0.0026
ZFH3	0.69	0.0144	0.62	0.0046
ZNF827	0.62	0.0030	0.53	0.0001

EXAMPLE 3: IDENTIFICATION OF MICRORNAS ASSOCIATED WITH CLINICAL RECURRENCE AND DEATH DUE TO PROSTATE CANCER

[0148] MicroRNAs function by binding to portions of messenger RNA (mRNA) and changing how frequently the mRNA is translated into protein. They can also influence the turnover of mRNA and

thus how long the mRNA remains intact in the cell. Since microRNAs function primarily as an adjunct to mRNA, this study evaluated the joint prognostic value of microRNA expression and gene (mRNA) expression. Since the expression of certain microRNAs may be a surrogate for expression of genes that are not in the assessed panel, we also evaluated the prognostic value of microRNA expression by itself.

Patients and Samples

[0149] Samples from the 127 patients with clinical recurrence and 374 patients without clinical recurrence after radical prostatectomy described in Example 2 were used in this study. The final analysis set comprised 416 samples from patients in which both gene expression and microRNA expression were successfully assayed. Of these, 106 patients exhibited clinical recurrence and 310 did not have clinical recurrence. Tissue samples were taken from each prostate sample representing (1) the primary Gleason pattern in the sample, and (2) the highest Gleason pattern in the sample. In addition, a sample of histologically normal-appearing tissue adjacent to the tumor (NAT) was taken. The number of patients in the analysis set for each tissue type and the number of them who experienced clinical recurrence or death due to prostate cancer are shown in Table 14.

Table 14. Number of Patients and Events in Analysis Set			
	Patients	Clinical Recurrences	Deaths Due to Prostate Cancer
Primary Gleason Pattern Tumor Tissue	416	106	36
Highest Gleason Pattern Tumor Tissue	405	102	36
Normal Adjacent Tissue	364	81	29

Assay Method

[0150] Expression of 76 test microRNAs and 5 reference microRNAs were determined from RNA extracted from fixed paraffin-embedded (FPE) tissue. MicroRNA expression in all three tissue type was quantified by reverse transcriptase polymerase chain reaction (RT-PCR) using the crossing point (C_p) obtained from the Taqman® MicroRNA Assay kit (Applied Biosystems, Inc., Carlsbad, CA).

Statistical Analysis

[0151] Using univariate proportional hazards regression (Cox DR, Journal of the Royal Statistical Society, Series B 34:187-220, 1972), applying the sampling weights from the cohort sampling

design, and using variance estimation based on the Lin and Wei method (Lin and Wei, Journal of the American Statistical Association 84:1074-1078, 1989), microRNA expression, normalized by the average expression for the 5 reference microRNAs hsa-miR-106a, hsa-miR-146b-5p, hsa-miR-191, hsa-miR-19b, and hsa-miR-92a, and reference-normalized gene expression of the 733 genes (including the reference genes) discussed above, were assessed for association with clinical recurrence and death due to prostate cancer. Standardized hazard ratios (the proportional change in the hazard associated with a change of one standard deviation in the covariate value) were calculated.

[0152] This analysis included the following classes of predictors:

1. 1. MicroRNAs alone
2. 2. MicroRNA-gene pairs Tier 1
3. 3. MicroRNA-gene pairs Tier 2
4. 4. MicroRNA-gene pairs Tier 3
5. 5. All other microRNA-gene pairs Tier 4

[0153] The four tiers were pre-determined based on the likelihood (Tier 1 representing the highest likelihood) that the gene-microRNA pair functionally interacted or that the microRNA was related to prostate cancer based on a review of the literature and existing microarray data sets.

[0154] False discovery rates (FDR) (Benjamini and Hochberg, Journal of the Royal Statistical Society, Series B 57:289-300, 1995) were assessed using Efron's separate class methodology (Efron, Annals of Applied Statistics 2:197-223., 2008). The false discovery rate is the expected proportion of the rejected null hypotheses that are rejected incorrectly (and thus are false discoveries). Efron's methodology allows separate FDR assessment (q -values) (Storey, Journal of the Royal Statistical Society, Series B 64:479-498, 2002) within each class while utilizing the data from all the classes to improve the accuracy of the calculation. In this analysis, the q -value for a microRNA or microRNA-gene pair can be interpreted as the empirical Bayes probability that the microRNA or microRNA-gene pair identified as being associated with clinical outcome is in fact a false discovery given the data. The separate class approach was applied to a true discovery rate degree of association (TDRDA) analysis (Crager, Statistics in Medicine 29:33-45, 2010) to determine sets of microRNAs or microRNA-gene pairs that have standardized hazard ratio for clinical recurrence or prostate cancer-specific death of at least a specified amount while controlling the FDR at 10%. For each microRNA or microRNA-gene pair, a maximum lower bound (MLB) standardized hazard ratio was computed, showing the highest lower bound for which the microRNA or microRNA-gene pair was included in a TDRDA set with 10% FDR. Also calculated was an estimate of the true standardized hazard ratio corrected for regression to the mean (RM) that occurs in subsequent studies when the best predictors are selected from a long list (Crager, 2010 above). The RM-corrected estimate of the standardized hazard ratio is a reasonable estimate of what could be expected if the selected microRNA or microRNA-gene pair were studied in a separate, subsequent study.

[0155] These analyses were repeated adjusting for clinical and pathology covariates available at

the time of patient biopsy: biopsy Gleason score, baseline PSA level, and clinical T-stage (T1-T2A vs. T2B or T2C) to assess whether the microRNAs or microRNA-gene pairs have predictive value independent of these clinical and pathology covariates.

Results

[0156] The analysis identified 21 microRNAs assayed from primary Gleason pattern tumor tissue that were associated with clinical recurrence of prostate cancer after radical prostatectomy, allowing a false discovery rate of 10% (Table 15). Results were similar for microRNAs assessed from highest Gleason pattern tumor tissue (Table 16), suggesting that the association of microRNA expression with clinical recurrence does not change markedly depending on the location within a tumor tissue sample. No microRNA assayed from normal adjacent tissue was associated with the risk of clinical recurrence at a false discovery rate of 10%. The sequences of the microRNAs listed in Tables 15-21 are shown in Table B.

Table 15. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Primary Gleason Pattern Tumor Tissue							
MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM- Corrected Estimate ^c
hsa-miR-93	<0.0001	0.0%	(+)	1.79	(1.38, 2.32)	1.19	1.51
hsa-miR-106b	<0.0001	0.1%	(+)	1.80	(1.38, 2.34)	1.19	1.51
hsa-miR-30e-5p	<0.0001	0.1%	(-)	1.63	(1.30, 2.04)	1.18	1.46
hsa-miR-21	<0.0001	0.1%	(+)	1.66	(1.31, 2.09)	1.18	1.46
hsa-miR-133a	<0.0001	0.1%	(-)	1.72	(1.33, 2.21)	1.18	1.48
hsa-miR-449a	<0.0001	0.1%	(+)	1.56	(1.26, 1.92)	1.17	1.42
hsa-miR-30a	0.0001	0.1%	(-)	1.56	(1.25, 1.94)	1.16	1.41
hsa-miR-182	0.0001	0.2%	(+)	1.74	(1.31, 2.31)	1.17	1.45
hsa-miR-27a	0.0002	0.2%	(+)	1.65	(1.27, 2.14)	1.16	1.43
hsa-miR-222	0.0006	0.5%	(-)	1.47	(1.18, 1.84)	1.12	1.35

Table 15. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Primary Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-103	0.0036	2.1%	(+)	1.77	(1.21, 2.61)	1.12	1.36
hsa-miR-1	0.0037	2.2%	(-)	1.32	(1.10, 1.60)	1.07	1.26
hsa-miR-145	0.0053	2.9%	(-)	1.34	(1.09, 1.65)	1.07	1.27
hsa-miR-141	0.0060	3.2%	(+)	1.43	(1.11, 1.84)	1.07	1.29
hsa-miR-92a	0.0104	4.8%	(+)	1.32	(1.07, 1.64)	1.05	1.25
hsa-miR-22	0.0204	7.7%	(+)	1.31	(1.03, 1.64)	1.03	1.23
hsa-miR-29b	0.0212	7.9%	(+)	1.36	(1.03, 1.76)	1.03	1.24
hsa-miR-210	0.0223	8.2%	(+)	1.33	(1.03, 1.70)	1.00	1.23
hsa-miR-486-5p	0.0267	9.4%	(-)	1.25	(1.00, 1.53)	1.00	1.20
hsa-miR-19b	0.0280	9.7%	(-)	1.24	(1.00, 1.50)	1.00	1.19
hsa-miR-205	0.0289	10.0%	(-)	1.25	(1.00, 1.53)	1.00	1.20

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with clinical recurrence is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of clinical recurrence.

^cRM: regression to the mean.

Table 16. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Highest Gleason Pattern Tumor Tissue

MicroRNA	p-value	q-value ³ (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-93	<0.0001	0.0%	(+)	1.91	(1.48, 2.47)	1.24	1.59
hsa-miR-449a	<0.0001	0.0%	(+)	1.75	(1.40, 2.18)	1.23	1.54
hsa-miR-205	<0.0001	0.0%	(-)	1.53	(1.29, 1.81)	1.20	1.43
hsa-miR-19b	<0.0001	0.0%	(-)	1.37	(1.19, 1.57)	1.15	1.32
hsa-miR-106b	<0.0001	0.0%	(+)	1.84	(1.39, 2.42)	1.22	1.51
hsa-miR-21	<0.0001	0.0%	(+)	1.68	(1.32, 2.15)	1.19	1.46
hsa-miR-30a	0.0005	0.4%	(-)	1.44	(1.17, 1.76)	1.13	1.33
hsa-miR-30e-5p	0.0010	0.6%	(-)	1.37	(1.14, 1.66)	1.11	1.30
hsa-miR-133a	0.0015	0.8%	(-)	1.57	(1.19, 2.07)	1.13	1.36
hsa-miR-1	0.0016	0.8%	(-)	1.42	(1.14, 1.77)	1.11	1.31
hsa-miR-103	0.0021	1.1%	(+)	1.69	(1.21, 2.37)	1.13	1.37
hsa-miR-210	0.0024	1.2%	(+)	1.43	(1.13, 1.79)	1.11	1.31
hsa-miR-182	0.0040	1.7%	(+)	1.48	(1.13, 1.93)	1.11	1.31
hsa-miR-27a	0.0055	2.1%	(+)	1.46	(1.12, 1.91)	1.09	1.30
hsa-miR-222	0.0093	3.2%	(-)	1.38	(1.08, 1.77)	1.08	1.27
hsa-miR-331	0.0126	3.9%	(+)	1.38	(1.07, 1.77)	1.07	1.26
hsa-miR-191*	0.0143	4.3%	(+)	1.38	(1.06, 1.78)	1.07	1.26
hsa-miR-					(1.06,		

Table 16. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Highest Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ³ (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
425	0.0151	4.5%	(+)	1.40	1.83)	1.07	1.26
hsa-miR-31	0.0176	5.1%	(-)	1.29	(1.04, 1.60)	1.05	1.22
hsa-miR-92a	0.0202	5.6%	(+)	1.31	(1.03, 1.65)	1.05	1.23
hsa-miR-155	0.0302	7.6%	(-)	1.32	(1.00, 1.69)	1.03	1.22
hsa-miR-22	0.0437	9.9%	(+)	1.30	(1.00, 1.67)	1.00	1.21

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with death due to prostate cancer is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of clinical recurrence.

^cRM: regression to the mean.

[0157] Table 17 shows microRNAs assayed from primary Gleason pattern tissue that were identified as being associated with the risk of prostate-cancer-specific death, with a false discovery rate of 10%. Table 18 shows the corresponding analysis for microRNAs assayed from highest Gleason pattern tissue. No microRNA assayed from normal adjacent tissue was associated with the risk of prostate-cancer-specific death at a false discovery rate of 10%.

Table 17. MicroRNAs Associated with Death Due to Prostate Cancer Primary Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-30e-5p	0.0001	0.6%	(-)	1.88	(1.37, 2.58)	1.15	1.46
hsa-miR-30a	0.0001	0.7%	(-)	1.78	(1.33, 2.40)	1.14	1.44
hsa-miR-133a	0.0005	1.2%	(-)	1.85	(1.31, 2.62)	1.13	1.41

Table 17. MicroRNAs Associated with Death Due to Prostate Cancer Primary Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-222	0.0006	1.4%	(-)	1.65	(1.24, 2.20)	1.12	1.38
hsa-miR-106b	0.0024	2.7%	(+)	1.85	(1.24, 2.75)	1.11	1.35
hsa-miR-1	0.0028	3.0%	(-)	1.43	(1.13, 1.81)	1.08	1.30
hsa-miR-21	0.0034	3.3%	(+)	1.63	(1.17, 2.25)	1.09	1.33
hsa-miR-93	0.0044	3.9%	(+)	1.87	(1.21, 2.87)	1.09	1.32
hsa-miR-26a	0.0072	5.3%	(-)	1.47	(1.11, 1.94)	1.07	1.29
hsa-miR-152	0.0090	6.0%	(-)	1.46	(1.10, 1.95)	1.06	1.28
hsa-miR-331	0.0105	6.5%	(+)	1.46	(1.09, 1.96)	1.05	1.27
hsa-miR-150	0.0159	8.3%	(+)	1.51	(1.07, 2.10)	1.03	1.27
hsa-miR-27b	0.0160	8.3%	(+)	1.97	(1.12, 3.42)	1.05	1.25

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with death due to prostate cancer endpoint is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of death due to prostate cancer.

^cRM: regression to the mean.

Table 18. MicroRNAs Associated with Death Due to Prostate Cancer Highest Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-27b	0.0016	6.1%	(+)	2.66	(1.45, 4.88)	1.07	1.32
hsa-miR-					(1.21,		

Table 18. MicroRNAs Associated with Death Due to Prostate Cancer Highest Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
21	0.0020	6.4%	(+)	1.66	2.30)	1.05	1.34
hsa-miR-10a	0.0024	6.7%	(+)	1.78	(1.23, 2.59)	1.05	1.34
hsa-miR-93	0.0024	6.7%	(+)	1.83	(1.24, 2.71)	1.05	1.34
hsa-miR-106b	0.0028	6.8%	(+)	1.79	(1.22, 2.63)	1.05	1.33
hsa-miR-150	0.0035	7.1%	(+)	1.61	(1.17, 2.22)	1.05	1.32
hsa-miR-1	0.0104	9.0%	(-)	1.52	(1.10, 2.09)	1.00	1.28

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with clinical endpoint is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of death due to prostate cancer.

^cRM: regression to the mean.

[0158] Table 19 and Table 20 shows the microRNAs that can be identified as being associated with the risk of clinical recurrence while adjusting for the clinical and pathology covariates of biopsy Gleason score, baseline PSA level, and clinical T-stage. The distributions of these covariates are shown in Figure 1. Fifteen (15) of the microRNAs identified in Table 15 are also present in Table 19, indicating that these microRNAs have predictive value for clinical recurrence that is independent of the Gleason score, baseline PSA, and clinical T-stage.

[0159] Two microRNAs assayed from primary Gleason pattern tumor tissue were found that had predictive value for death due to prostate cancer independent of Gleason score, baseline PSA, and clinical T-stage (Table 21).

Table 19. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Adjusting for Biopsy Gleason Score, Baseline PSA Level, and Clinical T-Stage Primary Gleason Pattern Tumor Tissue

MicroRNA	p-value	q-value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-30e-5p	<0.0001	0.0%	(-)	1.80	(1.42, 2.27)	1.23	1.53
hsa-miR-30a	<0.0001	0.0%	(-)	1.75	(1.40, 2.19)	1.22	1.51
hsa-miR-93	<0.0001	0.1%	(+)	1.70	(1.32, 2.20)	1.19	1.44
hsa-miR-449a	0.0001	0.1%	(+)	1.54	(1.25, 1.91)	1.17	1.39
hsa-miR-133a	0.0001	0.1%	(-)	1.58	(1.25, 2.00)	1.17	1.39
hsa-miR-27a	0.0002	0.1%	(+)	1.66	(1.28, 2.16)	1.17	1.41
hsa-miR-21	0.0003	0.2%	(+)	1.58	(1.23, 2.02)	1.16	1.38
hsa-miR-182	0.0005	0.3%	(+)	1.56	(1.22, 1.99)	1.15	1.37
hsa-miR-106b	0.0008	0.5%	(+)	1.57	(1.21, 2.05)	1.15	1.36
hsa-miR-222	0.0028	1.1%	(-)	1.39	(1.12, 1.73)	1.11	1.28
hsa-miR-103	0.0048	1.7%	(+)	1.69	(1.17, 2.43)	1.13	1.32
hsa-miR-486-5p	0.0059	2.0%	(-)	1.34	(1.09, 1.65)	1.09	1.25
hsa-miR-1	0.0083	2.7%	(-)	1.29	(1.07, 1.57)	1.07	1.23
hsa-miR-141	0.0088	2.8%	(+)	1.43	(1.09, 1.87)	1.09	1.27
hsa-miR-200c	0.0116	3.4%	(+)	1.39	(1.07, 1.79)	1.07	1.25
hsa-miR-145	0.0201	5.1%	(-)	1.27	(1.03, 1.55)	1.05	1.20
hsa-miR-206	0.0329	7.2%	(-)	1.40	(1.00, 1.91)	1.05	1.23

Table 19. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Adjusting for Biopsy Gleason Score, Baseline PSA Level, and Clinical T-Stage Primary Gleason Pattern Tumor Tissue

MicroRNA	p-value	q-value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-29b	0.0476	9.4%	(+)	1.30	(1.00, 1.69)	1.00	1.20

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with clinical recurrence is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of clinical recurrence.

^cRM: regression to the mean.

Table 20. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Adjusting for Biopsy Gleason Score, Baseline PSA Level, and Clinical T-Stage Highest Gleason Pattern Tumor Tissue

MicroRNA	p-value	q-value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-30a	<0.0001	0.0%	(-)	1.62	(1.32, 1.99)	1.20	1.43
hsa-miR-30e-5p	<0.0001	0.0%	(-)	1.53	(1.27, 1.85)	1.19	1.39
hsa-miR-93	<0.0001	0.0%	(+)	1.76	(1.37, 2.26)	1.20	1.45
hsa-miR-205	<0.0001	0.0%	(-)	1.47	(1.23, 1.74)	1.18	1.36
hsa-miR-449a	0.0001	0.1%	(+)	1.62	(1.27, 2.07)	1.18	1.38
hsa-miR-106b	0.0003	0.2%	(+)	1.65	(1.26, 2.16)	1.17	1.36
hsa-miR-133a	0.0005	0.2%	(-)	1.51	(1.20, 1.90)	1.16	1.33
hsa-miR-1	0.0007	0.3%	(-)	1.38	(1.15, 1.67)	1.13	1.28
hsa-miR-210	0.0045	1.2%	(+)	1.35	(1.10, 1.67)	1.11	1.25

Table 20. MicroRNAs Associated with Clinical Recurrence of Prostate Cancer Adjusting for Biopsy Gleason Score, Baseline PSA Level, and Clinical T-Stage Highest Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-182	0.0052	1.3%	(+)	1.40	(1.10, 1.77)	1.11	1.26
hsa-miR-425	0.0066	1.6%	(+)	1.48	(1.12, 1.96)	1.12	1.26
hsa-miR-155	0.0073	1.8%	(-)	1.36	(1.09, 1.70)	1.10	1.24
hsa-miR-21	0.0091	2.1%	(+)	1.42	(1.09, 1.84)	1.10	1.25
hsa-miR-222	0.0125	2.7%	(-)	1.34	(1.06, 1.69)	1.09	1.23
hsa-miR-27a	0.0132	2.8%	(+)	1.40	(1.07, 1.84)	1.09	1.23
hsa-miR-191*	0.0150	3.0%	(+)	1.37	(1.06, 1.76)	1.09	1.23
hsa-miR-103	0.0180	3.4%	(+)	1.45	(1.06, 1.98)	1.09	1.23
hsa-miR-31	0.0252	4.3%	(-)	1.27	(1.00, 1.57)	1.07	1.19
hsa-miR-19b	0.0266	4.5%	(-)	1.29	(1.00, 1.63)	1.07	1.20
hsa-miR-99a	0.0310	5.0%	(-)	1.26	(1.00, 1.56)	1.06	1.18
hsa-miR-92a	0.0348	5.4%	(+)	1.31	(1.00, 1.69)	1.06	1.19
hsa-miR-146b-5p	0.0386	5.8%	(-)	1.29	(1.00, 1.65)	1.06	1.19
hsa-miR-145	0.0787	9.7%	(-)	1.23	(1.00, 1.55)	1.00	1.15

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with clinical clinical recurrence is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of clinical recurrence. ^cRM: regression to the mean.

Table 21. MicroRNAs Associated with Death Due to Prostate Cancer Adjusting for Biopsy Gleason Score, Baseline PSA Level, and Clinical T-Stage Primary Gleason Pattern Tumor Tissue

MicroRNA	<i>p</i> -value	<i>q</i> -value ^a (FDR)	Direction of Association ^b	Absolute Standardized Hazard Ratio			
				Uncorrected Estimate	95% Confidence Interval	Max. Lower Bound @ 10% FDR	RM-Corrected Estimate ^c
hsa-miR-30e-5p	0.0001	2.9%	(-)	1.97	(1.40, 2.78)	1.09	1.39
hsa-miR-30a	0.0002	3.3%	(-)	1.90	(1.36, 2.65)	1.08	1.38

^aThe *q*-value is the empirical Bayes probability that the microRNA's association with clinical recurrence is a false discovery, given the data.

^bDirection of association indicates where higher microRNA expression is associated with higher (+) or lower (-) risk of clinical recurrence.

^cRM: regression to the mean.

[0160] Accordingly, the normalized expression levels of hsa-miR-93; hsa-miR-106b; hsa-miR-21; hsa-miR-449a; hsa-miR-182; hsa-miR-27a; hsa-miR-103; hsa-miR-141; hsa-miR-92a; hsa-miR-22; hsa-miR-29b; hsa-miR-210; hsa-miR-331; hsa-miR-191; hsa-miR-425; and hsa-miR-200c are positively associated with an increased risk of recurrence; and hsa-miR-30e-5p; hsa-miR-133a; hsa-miR-30a; hsa-miR-222; hsa-miR-1; hsa-miR-145; hsa-miR-486-5p; hsa-miR-19b; hsa-miR-205; hsa-miR-31; hsa-miR-155; hsa-miR-206; hsa-miR-99a; and hsa-miR-146b-5p are negatively associated with an increased risk of recurrence.

[0161] Furthermore, the normalized expression levels of hsa-miR-106b; hsa-miR-21; hsa-miR-93; hsa-miR-331; hsa-miR-150; hsa-miR-27b; and hsa-miR-10a are positively associated with an increased risk of prostate cancer specific death; and the normalized expression levels of hsa-miR-30e-5p; hsa-miR-30a; hsa-miR-133a; hsa-miR-222; hsa-miR-1; hsa-miR-26a; and hsa-miR-152 are negatively associated with an increased risk of prostate cancer specific death.

[0162] Table 22 shows the number of microRNA-gene pairs that were grouped in each tier (Tiers 1-4) and the number and percentage of those that were predictive of clinical recurrence at a false discovery rate of 10%.

Table 22.

Tier	Total Number of MicroRNA-Gene Pairs	Number of Pairs Predictive of Clinical Recurrence at False Discovery Rate 10% (%)
Tier 1	80	46 (57.5%)
Tier 2	719	591 (82.2%)
Tier 3	3,850	2,792 (72.5%)
Tier 4	54,724	38,264 (69.9%)

TABLE A

Official Symbol	Accession Number	SEQ ID NO	Forward Primer Sequence	SEQ ID NO	Reverse Primer Sequence	SEQ ID NO	Primer Sequences	SEQ ID NO	Amplifier Sequences
AMMP	NM_001087	1	ATGCTGAGGAGG	2	CTCCATCACTCAG	3	CTCCATCACTCAG	4	CTGCTGAGGAGG
AMC5	NM_172332	5	GGATGAGGAGG	6	GGATGAGGAGG	7	GGATGAGGAGG	8	GGATGAGGAGG
AMC1	NM_009227	9	AAAGGAGGAGG	10	AAAGGAGGAGG	11	AAAGGAGGAGG	12	AAAGGAGGAGG
AMC1	NM_009227	13	AAAGGAGGAGG	14	AAAGGAGGAGG	15	AAAGGAGGAGG	16	AAAGGAGGAGG
AMC3	NM_009246	17	AAAGGAGGAGG	18	AAAGGAGGAGG	19	AAAGGAGGAGG	20	AAAGGAGGAGG
AMC4	NM_009246	21	AAAGGAGGAGG	22	AAAGGAGGAGG	23	AAAGGAGGAGG	24	AAAGGAGGAGG
AMC5	NM_009246	25	AAAGGAGGAGG	26	AAAGGAGGAGG	27	AAAGGAGGAGG	28	AAAGGAGGAGG
AMC2	NM_009246	29	AAAGGAGGAGG	30	AAAGGAGGAGG	31	AAAGGAGGAGG	32	AAAGGAGGAGG
AMC2	NM_009246	33	AAAGGAGGAGG	34	AAAGGAGGAGG	35	AAAGGAGGAGG	36	AAAGGAGGAGG
AMC2	NM_009246	37	AAAGGAGGAGG	38	AAAGGAGGAGG	39	AAAGGAGGAGG	40	AAAGGAGGAGG
AMC2	NM_009246	41	AAAGGAGGAGG	42	AAAGGAGGAGG	43	AAAGGAGGAGG	44	AAAGGAGGAGG
AMC2	NM_009246	45	AAAGGAGGAGG	46	AAAGGAGGAGG	47	AAAGGAGGAGG	48	AAAGGAGGAGG
AMC2	NM_009246	49	AAAGGAGGAGG	50	AAAGGAGGAGG	51	AAAGGAGGAGG	52	AAAGGAGGAGG
AMC2	NM_009246	53	AAAGGAGGAGG	54	AAAGGAGGAGG	55	AAAGGAGGAGG	56	AAAGGAGGAGG
AMC2	NM_009246	57	AAAGGAGGAGG	58	AAAGGAGGAGG	59	AAAGGAGGAGG	60	AAAGGAGGAGG
AMC2	NM_009246	61	AAAGGAGGAGG	62	AAAGGAGGAGG	63	AAAGGAGGAGG	64	AAAGGAGGAGG
AMC2	NM_009246	65	AAAGGAGGAGG	66	AAAGGAGGAGG	67	AAAGGAGGAGG	68	AAAGGAGGAGG
AMC2	NM_009246	69	AAAGGAGGAGG	70	AAAGGAGGAGG	71	AAAGGAGGAGG	72	AAAGGAGGAGG
AMC2	NM_009246	73	AAAGGAGGAGG	74	AAAGGAGGAGG	75	AAAGGAGGAGG	76	AAAGGAGGAGG
AMC2	NM_009246	77	AAAGGAGGAGG	78	AAAGGAGGAGG	79	AAAGGAGGAGG	80	AAAGGAGGAGG
AMC2	NM_009246	81	AAAGGAGGAGG	82	AAAGGAGGAGG	83	AAAGGAGGAGG	84	AAAGGAGGAGG
AMC2	NM_009246	85	AAAGGAGGAGG	86	AAAGGAGGAGG	87	AAAGGAGGAGG	88	AAAGGAGGAGG
AMC2	NM_009246	89	AAAGGAGGAGG	90	AAAGGAGGAGG	91	AAAGGAGGAGG	92	AAAGGAGGAGG
AMC2	NM_009246	93	AAAGGAGGAGG	94	AAAGGAGGAGG	95	AAAGGAGGAGG	96	AAAGGAGGAGG
AMC2	NM_009246	97	AAAGGAGGAGG	98	AAAGGAGGAGG	99	AAAGGAGGAGG	100	AAAGGAGGAGG
AMC2	NM_009246	101	AAAGGAGGAGG	102	AAAGGAGGAGG	103	AAAGGAGGAGG	104	AAAGGAGGAGG
AMC2	NM_009246	105	AAAGGAGGAGG	106	AAAGGAGGAGG	107	AAAGGAGGAGG	108	AAAGGAGGAGG
AMC2	NM_009246	109	AAAGGAGGAGG	110	AAAGGAGGAGG	111	AAAGGAGGAGG	112	AAAGGAGGAGG
AMC2	NM_009246	113	AAAGGAGGAGG	114	AAAGGAGGAGG	115	AAAGGAGGAGG	116	AAAGGAGGAGG
AMC2	NM_009246	117	AAAGGAGGAGG	118	AAAGGAGGAGG	119	AAAGGAGGAGG	120	AAAGGAGGAGG
AMC2	NM_009246	121	AAAGGAGGAGG	122	AAAGGAGGAGG	123	AAAGGAGGAGG	124	AAAGGAGGAGG
AMC2	NM_009246	125	AAAGGAGGAGG	126	AAAGGAGGAGG	127	AAAGGAGGAGG	128	AAAGGAGGAGG

Official Symbol	Accession Number	SEQ ID NO	Forward Primer Sequence	SEQ ID NO	Reverse Primer Sequence	SEQ ID NO	Primer Sequences	SEQ ID NO	Amplifier Sequences
AMC2	NM_009246	129	AAAGGAGGAGG	130	AAAGGAGGAGG	131	AAAGGAGGAGG	132	AAAGGAGGAGG
AMC2	NM_009246	133	AAAGGAGGAGG	134	AAAGGAGGAGG	135	AAAGGAGGAGG	136	AAAGGAGGAGG
AMC2	NM_009246	137	AAAGGAGGAGG	138	AAAGGAGGAGG	139	AAAGGAGGAGG	140	AAAGGAGGAGG
AMC2	NM_009246	141	AAAGGAGGAGG	142	AAAGGAGGAGG	143	AAAGGAGGAGG	144	AAAGGAGGAGG
AMC2	NM_009246	145	AAAGGAGGAGG	146	AAAGGAGGAGG	147	AAAGGAGGAGG	148	AAAGGAGGAGG
AMC2	NM_009246	149	AAAGGAGGAGG	150	AAAGGAGGAGG	151	AAAGGAGGAGG	152	AAAGGAGGAGG
AMC2	NM_009246	153	AAAGGAGGAGG	154	AAAGGAGGAGG	155	AAAGGAGGAGG	156	AAAGGAGGAGG
AMC2	NM_009246	157	AAAGGAGGAGG	158	AAAGGAGGAGG	159	AAAGGAGGAGG	160	AAAGGAGGAGG
AMC2	NM_009246	161	AAAGGAGGAGG	162	AAAGGAGGAGG	163	AAAGGAGGAGG	164	AAAGGAGGAGG
AMC2	NM_009246	165	AAAGGAGGAGG	166	AAAGGAGGAGG	167	AAAGGAGGAGG	168	AAAGGAGGAGG
AMC2	NM_009246	169	AAAGGAGGAGG	170	AAAGGAGGAGG	171	AAAGGAGGAGG	172	AAAGGAGGAGG
AMC2	NM_009246	173	AAAGGAGGAGG	174	AAAGGAGGAGG	175	AAAGGAGGAGG	176	AAAGGAGGAGG
AMC2	NM_009246	177	AAAGGAGGAGG	178	AAAGGAGGAGG	179	AAAGGAGGAGG	180	AAAGGAGGAGG
AMC2	NM_009246	181	AAAGGAGGAGG	182	AAAGGAGGAGG	183	AAAGGAGGAGG	184	AAAGGAGGAGG
AMC2	NM_009246	185	AAAGGAGGAGG	186	AAAGGAGGAGG	187	AAAGGAGGAGG	188	AAAGGAGGAGG
AMC2	NM_009246	189	AAAGGAGGAGG	190	AAAGGAGGAGG	191	AAAGGAGGAGG	192	AAAGGAGGAGG
AMC2	NM_009246	193	AAAGGAGGAGG	194	AAAGGAGGAGG	195	AAAGGAGGAGG	196	AAAGGAGGAGG
AMC2	NM_009246	197	AAAGGAGGAGG	198	AAAGGAGGAGG	199	AAAGGAGGAGG	200	AAAGGAGGAGG
AMC2	NM_009246	201	AAAGGAGGAGG	202	AAAGGAGGAGG	203	AAAGGAGGAGG	204	AAAGGAGGAGG
AMC2	NM_009246	205	AAAGGAGGAGG	206	AAAGGAGGAGG	207	AAAGGAGGAGG	208	AAAGGAGGAGG
AMC2	NM_009246	209	AAAGGAGGAGG	210	AAAGGAGGAGG	211	AAAGGAGGAGG	212	AAAGGAGGAGG
AMC2	NM_009246	213	AAAGGAGGAGG	214	AAAGGAGGAGG	215	AAAGGAGGAGG	216	AAAGGAGGAGG
AMC2	NM_009246	217	AAAGGAGGAGG	218	AAAGGAGGAGG	219	AAAGGAGGAGG	220	AAAGGAGGAGG
AMC2	NM_009246	221	AAAGGAGGAGG	222	AAAGGAGGAGG	223	AAAGGAGGAGG	224	AAAGGAGGAGG
AMC2	NM_009246	225	AAAGGAGGAGG	226	AAAGGAGGAGG	227	AAAGGAGGAGG	228	AAAGGAGGAGG
AMC2	NM_009246	229	AAAGGAGGAGG	230	AAAGGAGGAGG	231	AAAGGAGGAGG	232	AAAGGAGGAGG
AMC2	NM_009246	233	AAAGGAGGAGG	234	AAAGGAGGAGG	235	AAAGGAGGAGG	236	AAAGGAGGAGG
AMC2	NM_009246	237	AAAGGAGGAGG	238	AAAGGAGGAGG	239	AAAGGAGGAGG	240	AAAGGAGGAGG
AMC2	NM_009246	241	AAAGGAGGAGG	242	AAAGGAGGAGG	243	AAAGGAGGAGG	244	AAAGGAGGAGG
AMC2	NM_009246	245	AAAGGAGGAGG	246	AAAGGAGGAGG	247	AAAGGAGGAGG	248	AAAGGAGGAGG
AMC2	NM_009246	249	AAAGGAGGAGG	250	AAAGGAGGAGG	251	AAAGGAGGAGG	252	AAAGGAGGAGG
AMC2	NM_009246	253	AAAGGAGGAGG	254	AAAGGAGGAGG	255	AAAGGAGGAGG	256	AAAGGAGGAGG
AMC2	NM_009246	257	AAAGGAGGAGG	258	AAAGGAGGAGG	259	AAAGGAGGAGG	260	AAAGGAGGAGG
AMC2	NM_009246	261	AAAGGAGGAGG	262	AAAGGAGGAGG	263	AAAGGAGGAGG	264	AAAGGAGGAGG
AMC2	NM_009246	265	AAAGGAGGAGG	266	AAAGGAGGAGG	267	AAAGGAGGAGG	268	AAAGGAGGAGG
AMC2	NM_009246	269	AAAGGAGGAGG	270	AAAGGAGGAGG	271	AAAGGAGGAGG	272	AAAGGAGGAGG
AMC2	NM_009246	273	AAAGGAGGAGG	274	AAAGGAGGAGG	275	AAAGGAGGAGG	276	AAAGGAGGAGG
AMC2	NM_009246	277	AAAGGAGGAGG	278	AAAGGAGGAGG	279	AAAGGAGGAGG	280	AAAGGAGGAGG
AMC2	NM_009246	281	AAAGGAGGAGG	282	AAAGGAGGAGG	283	AAAGGAGGAGG	284	AAAGGAGGAGG
AMC2	NM_009246	285	AAAGGAGGAGG	286	AAAGGAGGAGG	287	AAAGGAGGAGG	288	AAAGGAGGAGG
AMC2	NM_009246	289	AAAGGAGGAGG	290	AAAGGAGGAGG	291	AAAGGAGGAGG	292	AAAGGAGGAGG
AMC2	NM_009246	293	AAAGGAGGAGG	294	AAAGGAGGAGG	295	AAAGGAGGAGG	296	AAAGGAGGAGG
AMC2	NM_009246	297	AAAGGAGGAGG	298	AAAGGAGGAGG	299	AAAGGAGGAGG	300	AAAGGAGGAGG

[illegible][illegible]

Official training set	Accession Number	Accession No.	Type
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BRCA2	NM_000595	285	ATGATC
BRG1	NM_001731	289	GAGGTC
BLU3	NM_058806	293	CAATAT
BIRC	NM_033537	297	GTCGAC
BUB1	NM_003536	301	CTGGAG
C7	NM_005857	305	ATTTTC
CACNA1D	NM_000720	309	AGAGCA
CADM1	NM_014333	313	CAACGC
CADPS1	NM_003716	317	CAGCCT
CASP1	NM_032291	321	ATAGTC
CASP3	NM_032951	325	TGAGGC
CASP7	NM_033238	329	GAGGCC
CAV1	NM_033233	333	GTGGCC
CAV2	NM_108212	337	CTTCCC
CCCL2	NM_002582	341	CGCTGC
CCL5	NM_002945	345	AGAGTC
CENH1	NM_031966	349	ATTCAG
CENPD	NM_001758	353	GCATGC
CENPF	NM_057149	357	ATGGTC
CENPH	NM_001239	361	TGATAT
CCR1	NM_001295	365	TCCAAC
CD164	NM_006016	369	CAACAC
CD1A	NM_001763	373	GGATCT
CD127	NM_001024	377	CCAAAC
CD44	NM_006610	381	TGACAC
CD65	NM_001251	385	TGATGC
CD68	NM_002231	389	TGAGAG
CD70	NM_001255	393	TGATAT
CD73B	NM_021873	397	CTCTGC
CD76	NM_001254	401	GGCAAC
CDH1	NM_004360	405	TGATAT
CDH10	NM_006227	409	TGTCGC
CDH11	NM_001793	413	ATCTGC
CDH15	NM_021153	417	AGTATC
CDH7	NM_033646	425	GTTGTC
CDK4	NM_012595	429	CCAAAG
CDK2	NM_001796	435	AATGCC
CDK3	NM_001258	439	CCAAAG
CDK5	NM_003290	443	CTGATC

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[illegible][illegible]

Amphib. Reptiles

[illegible]

Protein	Accession	Gene	Species
FRBP5	NM_011905	FRBP5	NM
FLNAC	NM_011905	FLNAC	NM
FLNC	NM_011905	FLNC	NM
FLI1	NM_011905	FLI1	NM
FLI4	NM_011905	FLI4	NM
FN1	NM_011905	FN1	NM
FN3	NM_011905	FN3	NM
FOXO1	NM_011905	FOXO1	NM
FOXQ1	NM_011905	FOXQ1	NM
FSDI	NM_011905	FSDI	NM
FYN	NM_011905	FYN	NM
G6PD	NM_011905	G6PD	NM
GABRG2	NM_011905	GABRG2	NM
GADD45	NM_011905	GADD45	NM
GADD45B	NM_011905	GADD45B	NM
GDPH5	NM_011905	GDPH5	NM
GHR	NM_011905	GHR	NM
GNAI7A	NM_011905	GNAI7A	NM
GNAH1	NM_011905	GNAH1	NM
GPM6B	NM_011905	GPM6B	NM
GPNMB	NM_011905	GPNMB	NM
GP268	NM_011905	GP268	NM
GRE7	NM_011905	GRE7	NM
GREM1	NM_011905	GREM1	NM
GSK3E	NM_011905	GSK3E	NM
GSN	NM_011905	GSN	NM
GSTM1	NM_011905	GSTM1	NM
GSTM2	NM_011905	GSTM2	NM
HDAC1	NM_011905	HDAC1	NM
HIDAC9	NM_011905	HIDAC9	NM
HGLD	NM_011905	HGLD	NM
HIP1	NM_011905	HIP1	NM
HIRP3	NM_011905	HIRP3	NM
HK1	NM_011905	HK1	NM
HLA G	NM_011905	HLA G	NM
ILF	NM_011905	ILF	NM
HNF1B	NM_011905	HNF1B	NM
HPS1	NM_011905	HPS1	NM
ITD	NM_011905	ITD	NM

Cardinal Number	Accession Number	STOIB N°	Review and Printer Signature	STOIB N°	Review and Printer Signature	SIO ID NO	Press Signature	SIO ID NO		
II-X	NM-001147	1225	CIACAGATTTCTIC	1226	AGAGACAGC-GGAGA	1227	CCCT	ATGTCGCCAGTCTAGGT	1228	CTACAG
	94		GCATCC		CTGG					TCAGGT
II-M	NM-000839	1229	CTGCCTATGCCAG	1230	CTCTTTTGGCTTCT	1231	CCCT	CAATGCAATGCTTTGAACA	1232	TGCTC
IN-G	NM-023292	1233	CGTCCAGCAAGTG	1234	CATCTCTAGCTGT	1235	CATCTCTAGCTGT	CACTGCGACATTTTCTCTCA	1236	CTTAC
IN-H	NM-002129	1237	GTGCGCAGGCAT	1238	CGTATGTTTGTATG	1239	CGTATGTTTGTATG	CCCTCTCGGTCTTCACTGT	1240	GTCCG
IN-S	NM-002195	1241	CTGTCTATATTCG	1242	CAGATTTXACACAGC	1243	CAGATTTXACACAGC	TCAGAGACACATTCACACAC	1244	CTCTC
IT-B	NM-181501	1245	CTCTTCTTCTGAC	1246	CTCTTATGAGTATGAC	1247	CTCTTATGAGTATGAC	CTCTTCTGACATCTGCTGAC	1248	GCCTC
IT-G	NM-002204	1249	CTCTTCTTCTGAC	1250	GAAGCTTTTCTAGCTGT	1251	GAAGCTTTTCTAGCTGT	CATCTCAGACATCTGTTTATG	1252	TCATG
IT-H	NM-000835	1253	CAACGCTTTGATGT	1254	GTCTGCGCGGGATTC	1255	GTCTGCGCGGGATTC	CGATCTCTCTTCTGTAATG	1256	CAACG
IT-G	NM-002205	1257	AGCGGACGCTTAC	1258	GTCTCTCTCAAGTCC	1259	GTCTCTCTCAAGTCC	CTCTGAGCTTTCTGCTTATC	1260	ACRCC
IT-H	NM-000210	1261	CTACAGCAAAACAG	1262	GTCTAGGCTCTATGTC	1263	GTCTAGGCTCTATGTC	TCGCACTCTTTTGTGGATTC	1264	GATAT
IT-G	NM-000206	1265	GATATCATCTGCTC	1266	ATGCACTTCTCATCTCTC	1267	ATGCACTTCTCATCTCTC	CAGCCAGAGGCTCTGCTCTC	1268	GATAT
			CTCTCTCTC		ATCAT					TCGCC
IT-G	NM-002553	1269	GAGCTCTGCTGTAT	1270	ACTCTTCAATGATCC	1271	ACTCTTCAATGATCC	CAATGCAAGGCTCTGCTCTC	1272	GATCC
IT-B	NM-000212	1273	ACTCTGATCCCTTCA	1274	CTCTTAACTCTTTCTAC	1275	CTCTTAACTCTTTCTAC	CAATGCTCTCAACCTCTACT	1276	ACCGC
IT-B	NM-000213	1277	CAAGCTGCGCCCTCA	1278	GGCGACACTTTCATC	1279	GGCGACACTTTCATC	CCCTGTGAC	1280	GTTCAC
IT-B	NM-000215	1281	CTGTGTA-AGATGAC	1282	GGTGTGACATCTATGAC	1283	GGTGTGACATCTATGAC	ACTATCTTCTTCTACAAATC	1284	TCCTG
IT-B	NM-000222	1285	GAGGATGATGTTGCTG	1286	GTATCTCTGATCTGCTG	1287	GTATCTCTGATCTGCTG	CATCTCTAAGGACATGAGCTCT	1288	GAGG
IT-B	NM-000224	1289	TGCTCATCTGTTGCT	1290	ATGAGGACTTGCTGTCA	1291	ATGAGGACTTGCTGTCA	TCGAGCTCTGGATCTCTCAGA	1292	TGCTC
IT-S	NM-000324	1293	CTATCGGAGATGACA	1294	CTCTCTCTTAACTGGC	1295	CTCTCTCTTAACTGGC	AGCCCTCTCGCTCTTCTCA	1296	TAACT
IT-S	NM-000314	1297	TGCTCTACACATG	1298	GGATAGCTCTGAGAT	1299	GGATAGCTCTGAGAT	ACTCGATTTCTCCAGCCATCC	1300	TGCTC
IT-N	NM-000728	1301	GACTGCAAGATG	1302	TAGCCATTAAGTCTCG	1303	TAGCCATTAAGTCTCG	CTATAGCATGCTCTCTAACG	1304	GACTG
			GAACCTGA		CTCTC					CTCTC
IT-B	NM-000729	1305	CTGTGACGCTGCTG	1306	GGTCTGCTGCTCTGAA	1307	GGTCTGCTGCTCTGAA	CAAGGACAGCTCTCTGAA	1308	CTCTC
IT-B	NM-021614	1309	TGCTCTTAACTGCT	1310	GGCTATAGGACAAAGG	1311	GGCTATAGGACAAAGG	TAATATCTCAATATGACGC	1312	TGCTC
IT-B	NM-138444	1313	AGCAGCTACTGCG	1314	TGGAGACCTGAGAG	1315	ACTCTTACGCTGAGATCTC	AGCTAGCTCTGCTCTGCTC	1316	AGCAG
IT-B	NM-000558	1317	CTGGCAAGAGAGAG	1318	CTGTAGAGCTCTCT	1319	CTGTAGAGCTCTCT	CAAGACAGGACGACCTCTAC	1320	CGGCG
IT-B	NM-000494	1321	CAGACATCAAGCTCT	1322	CAATCTTG-GAGCGG	1323	TCCTCAGTGTCTCAGCAGACAG	CTAGAC	1324	CAGAC
IT-B	NM-014874	1325	CTCTTGCGAGAGCT	1326	GGGCAAGTCTGCTGCT	1327	GGGCAAGTCTGCTGCT	CTCTCTAGTATCTTCTTCTCA	1328	CGGCT
IT-B	NM-017310	1329	GAGCTGAGCTCTC	1330	CTGTGCTTCTCTCTCT	1331	CTGTGCTTCTCTCTCT	CATCTGACAACTCTGGAAG	1332	AGAG
IT-B	NM-000722	1333	AGAGCAAGCTCTCT	1334	GGCCTCTGGCTCTGAG	1335	GGCCTCTGGCTCTGAG	CACTACGACAGCTCATGCCC	1336	GAGCG
IT-B	NM-000722	1333	ATGCTCTTAATTA	1334	CAT	1335	CCAT		1336	GCCAT
IT-C	NM-180923	1337	AGTGTGCTCTGAGAG	1338	TGATCTCAATACCTCTC	1339	TGATCTCAATACCTCTC	CATACATCTCAGCAAACTCTG	1340	AGTGTG
IT-F	NM-201300	1341	AGATGACCTGGCT	1342	GGCTTAGGAGAGCTCT	1343	GGCTTAGGAGAGCTCT	AGTACTCT	1344	CATGT
IT-F	NM-002257	1345	TAATATXCTCAAT	1346	CAGAGAGCTCTCATGT	1347	CTGTAGG	TAATATGAGATTTTCTCAGAC	1348	AACAG
			TTGTTCA		TGAAG			CCATG		CATAT
IT-K	NM-000776	1340	GGCCAGAGCTCTC	1350	CAGAGGTTTGAACAG	1351	CCCTCTCTCTCTCCAGTCTGCG	CTCTCTCTCTCTCCAGTCTGCG	1352	GCCCG
IT-K	NM-000835	1353	CACCCTGGCTCTCA	1354	CATCTTCTCTAGCCAT	1355	CTCTCTCTCTAGGAGACAGCT	CTCTCTCTCTCTAGGAGACAGCT	1356	CACCC
IT-K	NM-000838	1357	CGCTTAAATATGT	1358	CTCTCTCTTTTGGCTCT	1359	CAGGACATCTTAAAGTCTGCTG	CAGGACATCTTAAAGTCTGCTG	1360	CCCTC
IT-K	NM-100551	1361	AGTCTCGGATGT	1362	TGGTACACAGCACTCT	1363	TGGTACAGCACTCTCTCTCTCT	TGGGAACTGTCTTCTCTCTCTCT	1364	AGTCT
IT-K	NM-001648	1365	CCAACTCTATCCAC	1366	AGTACAGCAAGAGACA	1367	TGGTACAGCAAGAGACA	AGCTCAATGTGTGACACAGCT	1368	CCGAA
IT-K	NM-000730	1369	TGATATCAATGCT	1370	ATCTTGCTCTCTCTCTG	1371	ATCTTGCTCTCTCTCTG	CTCTCTCTCTCTCTCTCTCTCT	1372	TGATG
IT-K	NM-002246	1373	TGATGCTCAATGCT	1374	AGCTCTCTCAAGTCTG	1375	AGCTCTCTCAAGTCTG	CTCTCTCTCTCTCTCTCTCTCT	1376	TGATG
IT-N	NM-006121	1377	TGATGCTCAATGCT	1378	TATCTCTTCTCTCTCTCTCT	1379	CTCTCTCTCTCTCTCTCTCTCT	CTCTCTCTCTCTCTCTCTCTCT	1380	TGATG
IT-R	NM-000275	1381	GGCTTGCTGTTTGGATCT	1382	CTGTGCTGTTTGGATCT	1383	CTGTGCTGTTTGGATCT	CATCTCTCTCTCTCTCTCTCTCT	1384	GCCCT
IT-R	NM-002224	1385	ATGATCTGATGCT	1386	GGGCTCTTCTCTCTCTCT	1387	GGGCTCTTCTCTCTCTCT	TGCTCTCTCTCTCTCTCTCTCT	1388	AGAG
IT-R	NM-000423	1389	CGATGACGCTCT	1390	GGGCTCTCTCTCTCTCT	1391	GGGCTCTCTCTCTCTCT	ACTTAGACACATCTCTCTCTCT	1392	CCAG
IT-R	NM-000234	1393	TGATGCTGAGAGCT	1394	TGCTCTCTCTCTCTCTCT	1395	TGCTCTCTCTCTCTCTCT	CTCTCTCTCTCTCTCTCTCTCT	1396	IT-CH

[illegible]

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[illegible]

Organism	Accession Number	Accession ID	Forward Primer Sequence	Reverse Primer Sequence
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LICAM	NM_002275	1408	GGATCACTCTAC ATTGACAGAGTAG	1409 GEMAA
LACT1	NM_002286	1419	CTTCTGTGGCAAT CTTCTTMTGTGAG	1420 TGAT
LAMA3	NM_006227	1421	CTTCTTMTGTGAG GATGCTACTGGT	1422 TGGG
LAMA4	NM_002290	1421	GATGCTACTGGT CTCTCTGCTCAACA	1422 CAGA
LAMA5	NM_005550	1423	CTCTCTGCTCAACA CAAGAGGACTGTGG	1424 ACAC
LAMB1	NM_007291	1429	CAAGAGGACTGTGG GCTGCTGCTCAAGCT	1430 CGCT
LAMB3	NM_002288	1433	GCTGCTGCTCAAGCT ACCTGCTGCTGCTG	1434 GACA
LAMB4	NM_002293	1433	ACCTGCTGCTGCTG ATTCACAGCGTAA	1434 ACTC
LAMB7	NM_002560	1441	ATTCACAGCGTAA TTGTAATCA	1442 GACA
LAMP5	NM_006762	1445	TTGTAATCA CTCTGGACTCTCTG	1446 TGAG
LGA1S1	NM_002306	1445	ACTGGAATTAATGGC GGAGGTGGAGAAG	1445 ACAG
LIG3	NM_002314	1453	GGAGGTGGAGAAG TGAATCAATATG	1454 TCTT
LIMB1	NM_004987	1453	TGAATCAATATG CCAAATGTGGAGAAC	1452 CCCT
LOC	NM_002317	1461	CCAAATGTGGAGAAC CTTGTGCTCAATGG	1466 GTTT
LRP1	NM_002449	1464	CTTGTGCTCAATGG CTCTTAAG	1464 TAGA
LTP2	NM_000428	1469	CGACAGCTCAATGCT GCTCTTTTGAAG	1470 GATG
LTU	NM_002345	1473	GATCTTTTGAAG GATTTGTTAA	1474 AAGA
MAGEA1	NM_002762	1477	GGATCTTAAGCAATC CAGATCTGAAGCC	1478 CAGA
MANF	NM_006610	1477	CAGATCTGAAGCC CTCTCAGCAACA	1482 TAMA
MAR2	NM_002340	1485	CTCTCAGCAACA ATGCTCAGCAACA	1486 CGAT
MARX5	NM_005923	1485	ATGCTCAGCAACA AGCACTAAGAGAC	1492 CCLG
MARX6	NM_005923	1485	AGCACTAAGAGAC CAGGCAAGCACTA	1494 CCGT
MAR387	NM_145333	1493	CAGGCAAGCACTA GTGTTCAGAA	1494 ATAT
MAR384	NM_004134	1497	GTGTTCAGAA TCCAGGATCTTC	1499 TGCT
MAP1	NM_003980	1501	TCCAGGATCTTC GAGAGTACAGAGAT	1502 CTTG
MAPKAP	NM_004635	1505	GAGAGTACAGAGAT AATCCCG	1506 GTCT
K3			AATCCCG GACTTTTGTCTTC	1510 TGCA
MCMD2	NM_004536	1509	GACTTTTGTCTTC TACCTTTC	1510 ATCT
MCMD3	NM_002338	1513	TACCTTTC GGAGCAAACTTC	1514 TGCT
MCMD4	NM_005915	1517	GGAGCAAACTTC TAAATGCTCATG	1518 TGGC
MCMD5	NM_005915	1517	TAAATGCTCATG TATCTCACTCA	1518 ACCA
MUK	NM_002391	1521	TATCTCACTCA GGAGCTGACATCA	1522 TGCT
MYH2	NM_001792	1525	GGAGCTGACATCA CTACAGAGAGGCC	1526 ATCC
MYLK	NM_011571	1529	CTACAGAGAGGCC AGGATGCTCTCTG	1530 TGG7
MYT	NM_000245	1533	AGGATGCTCTCTG GACATTTCCAGTC	1531 CTCC
MYH1	NM_002412	1537	GACATTTCCAGTC CTGAAATGAAGALG	1538 GACC
MYH1	NM_002300	1541	CTGAAATGAAGALG ACGATCTTACCAC	1542 TCCA
MYH1	NM_002300	1541	ACGATCTTACCAC ACCATTC	1543 AAAT
MYH1	NM_002300	1541	ACCATTC AGGATGAATGCTA	1546 AACT

[illegible]

TABLE 4								
Official Symbol	Accession Number	STUD ID Number	Forward Primer Sequence	Reverse Primer Sequence	STUD ID NO	Probe Sequence	STUD ID NO	Amplification Sequence
NOX4	NM_016691	1701	CTTCAACTACAGC CTTCAATCC	TGCTTGGAACCTTCTTG TGAT	1703	CCGACACACTCTTGACCTTACCC TCCG	1704	CTCTAACCTGACACTCTATTCCTTTTACCTCA ACACCTTCTGCTTACTCTCAAGAAATCACTACAAAGATTTT
NBPWR1	NM_019285	1705	GCAGCAGCTCTGCT ATATGTGTCCAGG	GATGTGTGAGGGCAG CAAGCAAAAGGCGAG	1706	ATACGCGAGGAGAGCTCTCCAC AACGAGCAATTTGGGCAACA	1708	TCACCACTGCTGCTGCTTCTCAACCTGGACCTACCTGCGA ATATGTGTCCAGGCTCTTATCTGCAAGAAATATGTTGTGTC
NP41	NM_002520	1709	TTTATTATTC CGAGACTCTGCTC	TTTATTATTC ATATGTAAGAGCTTC	1710	CTCTCTC TCTCAAGC	1711	CAAAATGTCTGTTTATGTTTATATAGAAATGAGATCTGTC CGAGACTCTCTCATGATGAGAAAGATGATGTGTCAATCC
NR01	NM_013957	1713	ATATGTAAGAGCTTC CGCAGATCAATG	ATATGTAAGAGCTTC TCTCAAGC	1714	ATGACACACCCCGCTCTGCTAT GTCA	1716	ATGACATCTCTCATGATGAGAAAGATGATGTGTCAATCC CGCAGATCAATGATGAGAAAGATGATGTGTCAATCC
NR1P3	NM_020645	1717	CGCAGATCAATG GAGCTCTGTCAC	CGCAGATCAATG GAGCTCTGTCAC	1718	AGCTTTCTTACCTCGGACAT CCAGATCTTACCTCGGAGAGAG	1720	CGCAGATCAATGATGAGAAAGATGATGTGTCAATCC CGCAGATCAATGATGAGAAAGATGATGTGTCAATCC
NR1P1	NM_013873	1721	GCGATTC CAATAGC	GAGCTCTGTCAC CAATAGC	1722	CCACTCTAT TCTCT	1723	GAGATCTCTATGCTGAGCTCTGGGATCTAGATGCGA AGCTCTCTGCTGAGCTCTGGGATCTAGATGCGA
NU1P62	NM_153719	1725	CAATAGC AGCAAGGCAAGCT	CAATAGC AGCAAGGCAAGCT	1726	CTCTCTGCTACCACTTGGACCT TCTCT	1728	AGCTCTCTGCTGAGCTCTGGGATCTAGATGCGA CTCTCTCTGCTGAGCTCTGGGATCTAGATGCGA
OX21	NM_004152	1729	AGCAAGGCAAGCT CCUCCUCCUACA	AGCAAGGCAAGCT CCUCCUCCUACA	1730	CTCTCTGCTACCACTTGGACCT TCTCT	1731	AGCAAGGCAAGCTCTGGGATCTAGATGCGA CTCTCTCTGCTGAGCTCTGGGATCTAGATGCGA
OC1N	NM_002538	1733	CCUCCUCCUACA AGCAAGGCAAGCT	CCUCCUCCUACA AGCAAGGCAAGCT	1734	CTCTCTGCTACCACTTGGACCT TCTCT	1736	AGCAAGGCAAGCTCTGGGATCTAGATGCGA CTCTCTCTGCTGAGCTCTGGGATCTAGATGCGA
OPC1	NM_002538	1737	AGCAAGGCAAGCT GATATGCAAA	AGCAAGGCAAGCT GATATGCAAA	1738	CTCTCTGCTACCACTTGGACCT TCTCT	1740	AGCAAGGCAAGCTCTGGGATCTAGATGCGA CTCTCTCTGCTGAGCTCTGGGATCTAGATGCGA
OLEM2	NM_015441	1741	ATATGCTGTAAGGA CTGATGCTGTAAGGA	ATATGCTGTAAGGA CTGATGCTGTAAGGA	1742	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1744	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OF1M1	NM_020190	1745	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1746	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1748	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OMD	NM_005014	1749	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1750	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1752	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_152430	1753	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1754	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1756	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	1758	TGCTCTGCTGAGCTCTGGGATCTAGATGCGA TCTCT	1760	ATATGCTGTAAGGACTCTGGGATCTAGATGCGA CTGATGCTGTAAGGACTCTGGGATCTAGATGCGA
OS1M2	NM_003734	1757	CTGATGCTGTAAGGA CTGATGCTGTAAGGA	CTGATGCTGTAAGGA 				

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W. J. G. M. J.

Project Name	Project ID	Project Manager	Project Status	Project Start Date	Project End Date	Project Budget	Project Location	Project Description	Project Notes
Project A	001	John Doe	Completed	2023-01-01	2023-03-31	\$100,000	New York	Website Redesign	Project completed successfully.
Project B	002	Jane Smith	In Progress	2023-04-01	2023-06-30	\$200,000	Los Angeles	Mobile App Development	Development phase ongoing.
Project C	003	Mike Johnson	On Hold	2023-05-01	2023-07-31	\$150,000	Chicago	Marketing Campaign	On hold due to budget constraints.
Project D	004	Sarah Lee	Completed	2023-08-01	2023-09-30	\$80,000	San Francisco	Data Migration	Migration completed with no issues.
Project E	005	David Kim	In Progress	2023-10-01	2023-12-31	\$300,000	London	Cloud Migration	Migration in progress.
Project F	006	Emily White	On Hold	2023-11-01	2024-01-31	\$120,000	Paris	Product Launch	On hold until Q2 2024.
Project G	007	Chris Brown	Completed	2024-02-01	2024-03-31	\$90,000	Tokyo	IT Infrastructure Upgrade	Upgrade completed.
Project H	008	Alex Green	In Progress	2024-04-01	2024-06-30	\$250,000	Sydney	Customer Portal Development	Development phase ongoing.
Project I	009	Mia Black	On Hold	2024-07-01	2024-09-30	\$180,000	Mumbai	Supply Chain Optimization	On hold due to market conditions.
Project J	010	Noah Grey	Completed	2024-10-01	2024-11-30	\$70,000	Beijing	Internal Audit	Audit completed.
Project K	011	Olivia Blue	In Progress	2024-12-01	2025-02-28	\$220,000	Brussels	HR System Integration	Integration in progress.
Project L	012	Liam Yellow	On Hold	2025-03-01	2025-05-31	\$160,000	Amsterdam	Website Security Audit	On hold until Q3 2025.
Project M	013	Ava Purple	Completed	2025-06-01	2025-07-31	\$110,000	Stockholm	Legal Compliance Review	Review completed.
Project N	014	Ethan Orange	In Progress	2025-08-01	2025-10-31	\$280,000	Oslo	AI Research Project	Research phase ongoing.
Project O	015	Sophia Pink	On Hold	2025-11-01	2026-01-31	\$190,000	Norway	Facility Renovation	On hold due to contractor issues.
Project P	016	Lucas Green	Completed	2026-02-01	2026-03-31	\$95,000	Oslo	IT Security Training	Training completed.
Project Q	017	Isabella Blue	In Progress	2026-04-01	2026-06-30	\$260,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project R	018	Benjamin Yellow	On Hold	2026-07-01	2026-09-30	\$170,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project S	019	Charlotte Purple	Completed	2026-10-01	2026-11-30	\$95,000	Oslo	Internal Audit	Audit completed.
Project T	020	William Orange	In Progress	2026-12-01	2027-02-28	\$230,000	Oslo	HR System Integration	Integration in progress.
Project U	021	Amelia Pink	On Hold	2027-03-01	2027-05-31	\$185,000	Oslo	Website Security Audit	On hold until Q3 2027.
Project V	022	James Green	Completed	2027-06-01	2027-07-31	\$105,000	Oslo	Legal Compliance Review	Review completed.
Project W	023	Harper Blue	In Progress	2027-08-01	2027-10-31	\$290,000	Oslo	AI Research Project	Research phase ongoing.
Project X	024	Elijah Yellow	On Hold	2027-11-01	2028-01-31	\$205,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project Y	025	Madison Purple	Completed	2028-02-01	2028-03-31	\$90,000	Oslo	IT Security Training	Training completed.
Project Z	026	Christopher Orange	In Progress	2028-04-01	2028-06-30	\$270,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project AA	027	Chloe Pink	On Hold	2028-07-01	2028-09-30	\$195,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project AB	028	Matthew Green	Completed	2028-10-01	2028-11-30	\$100,000	Oslo	Internal Audit	Audit completed.
Project AC	029	Abigail Blue	In Progress	2028-12-01	2029-02-28	\$240,000	Oslo	HR System Integration	Integration in progress.
Project AD	030	David Yellow	On Hold	2029-03-01	2029-05-31	\$180,000	Oslo	Website Security Audit	On hold until Q3 2029.
Project AE	031	Grace Purple	Completed	2029-06-01	2029-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project AF	032	Henry Orange	In Progress	2029-08-01	2029-10-31	\$300,000	Oslo	AI Research Project	Research phase ongoing.
Project AG	033	Isabella Pink	On Hold	2029-11-01	2030-01-31	\$210,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project AH	034	Jack Green	Completed	2030-02-01	2030-03-31	\$95,000	Oslo	IT Security Training	Training completed.
Project AI	035	Karen Blue	In Progress	2030-04-01	2030-06-30	\$280,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project AJ	036	Leo Yellow	On Hold	2030-07-01	2030-09-30	\$190,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project AK	037	Mia Purple	Completed	2030-10-01	2030-11-30	\$105,000	Oslo	Internal Audit	Audit completed.
Project AL	038	Nathan Orange	In Progress	2030-12-01	2031-02-28	\$250,000	Oslo	HR System Integration	Integration in progress.
Project AM	039	Olivia Pink	On Hold	2031-03-01	2031-05-31	\$185,000	Oslo	Website Security Audit	On hold until Q3 2031.
Project AN	040	Peter Green	Completed	2031-06-01	2031-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project AO	041	Quinn Blue	In Progress	2031-08-01	2031-10-31	\$290,000	Oslo	AI Research Project	Research phase ongoing.
Project AP	042	Rachel Yellow	On Hold	2031-11-01	2032-01-31	\$205,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project AQ	043	Samuel Purple	Completed	2032-02-01	2032-03-31	\$90,000	Oslo	IT Security Training	Training completed.
Project AR	044	Tina Orange	In Progress	2032-04-01	2032-06-30	\$270,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project AS	045	Uma Pink	On Hold	2032-07-01	2032-09-30	\$195,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project AT	046	Victor Green	Completed	2032-10-01	2032-11-30	\$100,000	Oslo	Internal Audit	Audit completed.
Project AU	047	Wendy Blue	In Progress	2032-12-01	2033-02-28	\$240,000	Oslo	HR System Integration	Integration in progress.
Project AV	048	Xavier Yellow	On Hold	2033-03-01	2033-05-31	\$180,000	Oslo	Website Security Audit	On hold until Q3 2033.
Project AW	049	Yara Purple	Completed	2033-06-01	2033-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project AX	050	Zoe Orange	In Progress	2033-08-01	2033-10-31	\$300,000	Oslo	AI Research Project	Research phase ongoing.
Project AY	051	Adam Pink	On Hold	2033-11-01	2034-01-31	\$210,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project AZ	052	Bella Green	Completed	2034-02-01	2034-03-31	\$95,000	Oslo	IT Security Training	Training completed.
Project BA	053	Carl Blue	In Progress	2034-04-01	2034-06-30	\$280,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project BB	054	Diana Yellow	On Hold	2034-07-01	2034-09-30	\$190,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project BC	055	Ethan Purple	Completed	2034-10-01	2034-11-30	\$105,000	Oslo	Internal Audit	Audit completed.
Project BD	056	Fiona Orange	In Progress	2034-12-01	2035-02-28	\$250,000	Oslo	HR System Integration	Integration in progress.
Project BE	057	George Pink	On Hold	2035-03-01	2035-05-31	\$185,000	Oslo	Website Security Audit	On hold until Q3 2035.
Project BF	058	Hannah Green	Completed	2035-06-01	2035-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project BG	059	Ian Blue	In Progress	2035-08-01	2035-10-31	\$290,000	Oslo	AI Research Project	Research phase ongoing.
Project BH	060	Julia Yellow	On Hold	2035-11-01	2036-01-31	\$205,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project BI	061	Kyle Purple	Completed	2036-02-01	2036-03-31	\$90,000	Oslo	IT Security Training	Training completed.
Project BJ	062	Laura Orange	In Progress	2036-04-01	2036-06-30	\$270,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project BK	063	Michael Pink	On Hold	2036-07-01	2036-09-30	\$195,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project BL	064	Nancy Green	Completed	2036-10-01	2036-11-30	\$100,000	Oslo	Internal Audit	Audit completed.
Project BM	065	Oscar Blue	In Progress	2036-12-01	2037-02-28	\$240,000	Oslo	HR System Integration	Integration in progress.
Project BN	066	Peter Yellow	On Hold	2037-03-01	2037-05-31	\$180,000	Oslo	Website Security Audit	On hold until Q3 2037.
Project BO	067	Quinn Purple	Completed	2037-06-01	2037-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project BP	068	Rachel Orange	In Progress	2037-08-01	2037-10-31	\$300,000	Oslo	AI Research Project	Research phase ongoing.
Project BQ	069	Samuel Pink	On Hold	2037-11-01	2038-01-31	\$210,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project BR	070	Tina Green	Completed	2038-02-01	2038-03-31	\$95,000	Oslo	IT Security Training	Training completed.
Project BS	071	Uma Blue	In Progress	2038-04-01	2038-06-30	\$280,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project BT	072	Victor Yellow	On Hold	2038-07-01	2038-09-30	\$190,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project BU	073	Wendy Purple	Completed	2038-10-01	2038-11-30	\$105,000	Oslo	Internal Audit	Audit completed.
Project BV	074	Xavier Orange	In Progress	2038-12-01	2039-02-28	\$250,000	Oslo	HR System Integration	Integration in progress.
Project BW	075	Yara Pink	On Hold	2039-03-01	2039-05-31	\$185,000	Oslo	Website Security Audit	On hold until Q3 2039.
Project BX	076	Zoe Green	Completed	2039-06-01	2039-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project BY	077	Adam Blue	In Progress	2039-08-01	2039-10-31	\$290,000	Oslo	AI Research Project	Research phase ongoing.
Project BZ	078	Bella Yellow	On Hold	2039-11-01	2040-01-31	\$205,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project CA	079	Carl Purple	Completed	2040-02-01	2040-03-31	\$90,000	Oslo	IT Security Training	Training completed.
Project CB	080	Diana Orange	In Progress	2040-04-01	2040-06-30	\$270,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project CC	081	Ethan Pink	On Hold	2040-07-01	2040-09-30	\$195,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project CD	082	Fiona Green	Completed	2040-10-01	2040-11-30	\$100,000	Oslo	Internal Audit	Audit completed.
Project CE	083	George Blue	In Progress	2040-12-01	2041-02-28	\$240,000	Oslo	HR System Integration	Integration in progress.
Project CF	084	Hannah Yellow	On Hold	2041-03-01	2041-05-31	\$180,000	Oslo	Website Security Audit	On hold until Q3 2041.
Project CG	085	Ian Purple	Completed	2041-06-01	2041-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project CH	086	Julia Orange	In Progress	2041-08-01	2041-10-31	\$300,000	Oslo	AI Research Project	Research phase ongoing.
Project CI	087	Kyle Pink	On Hold	2041-11-01	2042-01-31	\$210,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project CJ	088	Laura Green	Completed	2042-02-01	2042-03-31	\$95,000	Oslo	IT Security Training	Training completed.
Project CK	089	Michael Blue	In Progress	2042-04-01	2042-06-30	\$280,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project CL	090	Nancy Yellow	On Hold	2042-07-01	2042-09-30	\$190,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project CM	091	Oscar Purple	Completed	2042-10-01	2042-11-30	\$105,000	Oslo	Internal Audit	Audit completed.
Project CN	092	Peter Orange	In Progress	2042-12-01	2043-02-28	\$250,000	Oslo	HR System Integration	Integration in progress.
Project CO	093	Quinn Pink	On Hold	2043-03-01	2043-05-31	\$185,000	Oslo	Website Security Audit	On hold until Q3 2043.
Project CP	094	Rachel Green	Completed	2043-06-01	2043-07-31	\$110,000	Oslo	Legal Compliance Review	Review completed.
Project CQ	095	Samuel Blue	In Progress	2043-08-01	2043-10-31	\$290,000	Oslo	AI Research Project	Research phase ongoing.
Project CR	096	Tina Yellow	On Hold	2043-11-01	2044-01-31	\$205,000	Oslo	Facility Renovation	On hold due to contractor issues.
Project CS	097	Uma Purple	Completed	2044-02-01	2044-03-31	\$90,000	Oslo	IT Security Training	Training completed.
Project CT	098	Victor Orange	In Progress	2044-04-01	2044-06-30	\$270,000	Oslo	Customer Feedback Analysis	Analysis in progress.
Project CU	099	Wendy Pink	On Hold	2044-07-01	2044-09-30	\$195,000	Oslo	Supply Chain Diversification	On hold due to geopolitical factors.
Project CV	100	Xavier Green	Completed	2044-10-01	2044-11-30	\$100,000	Oslo	Internal Audit	Audit completed.

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ID	Forward Primer Sequences	SRC ID	Reverse Primer Sequences	SRC ID No.	Predicted Sequences	SEQ ID NO.	Amplicon Sequence
5	GTAACAGGACCAAC	2586	TGTGGAAATTTGAATC	2587	TGTGATGCTGTCTTCGAGCC	2588	GTATCAGGACACCATGCATGATCATCGTAGAAGAAGCGC
9	ATCCAGTACAATC		TGCCATT		TCTC		GAAGACAGCATCAAAGAAATGTCAGATATCAATTCG
9	ATCCAGTACAATC	2590	AGTCCTCTTGTCAT	2591	CTCTGTTTACTTTGGCGAG	2592	ATCCATGCTGGTGTGTCATCACATCGCTGTACTCTT
3	GCAGTTGGAGAAC	2594	TGCTGGGACTAATTTT	2595	TCCCATAATTCAGAAATTAI	2596	GCAGTTGGAGAACACAGGAAAGTATCTCCCAATTTGC
	ACACGGAAGT		CAGA		CAACGGC		AGATTTATCAACGGCTTTTATTGTGAANAATGATGCA
7	ACCAGCTTACGC	2598	AGCAGAGAATCACAT	2599	TGTATGTGACAGGCTATGTTCIA	2600	AGCCCATCTACGCTCTCATCTGCTGCTGAAGGAC
1	ACCTGAGGACCAAA	2602	GACGAGAGACTGCG	2603	TGTATGTGACACTTGTCTCTG	2604	ACCGCGGACACAAAGAAGTGGGAGCAGACAGAGTGC
5	CTGTGGAGGCTCT	2606	GGAAGCAGGTTGGAAT	2607	ACTTGCCCCAACTCTACAG	2608	CTGTGGAGGCTCTCTGCTGGGAGCTCTGCTCAATCTA
3	CTATTAACCCATC	2610	CCCGACCATATGTA	2611	CAGTTTCCCAAGTTGTCCAA	2612	CAATPACCATCTCTTGCTGCTGCTACATCTGGCGAGGT
9	GGTGGGCGGCAAAA	2614	GGCCGCTCTTATATCC	2615	CTTTTACAGGCTCAGAAATGGA	2616	GGCTGGGCGCAAACTGAAGGGGTTTCACTCTGGGCT
7	COAAGGAGACTCG	2618	GCAGCAGGTCCTCAG	2619	AGGAGGTTCCACACTTCACA	2620	GAAGGAGCATCTGCTCATGTAACTGCGCAGTTGTGG
1	TGCCTAGAGACCC	2622	GAGGTGGCGGATGGA	2623	CCGGCTTTGAGGAGAGAAAC	2624	TGCTTGAGGACCTCTACCTGGCCCCCTCTTCAGAGA
1	GGGCGGCATCA	2626	TGCTGTTTCTCTTGCGC	2627	ACCAAGCCCTGTACTCTAGAT	2628	TAGAGGCCCATCAAAATCTGCTCTCTACCAAGGACCTCA

[illegible]

Official Symbol	Accession Number	SEQ ID NO
WNT5A	NM_003392	258
WWOX	NM_016373	253
XIAP	NM_001167	255
XRC5	NM_021141	255
YY1	NM_003403	261
ZHX3	NM_006885	261
ZFP96	NM_003407	261
ZNTYND8	NM_183947	261
ZNF3	NM_017715	261
ZNF87	NM_178835	261
ZNF1	NM_007057	261

TABLE B

microRNA	Sequence	SEQ ID NO
hsa-miR-1	UGGAAUGUAAAGAAGUAUGUAU	2629
hsa-miR-103	GCAGCAUUGUACAGGGCUAUGA	2630
hsa-miR-106b	UAAAGUGCUGACAGUGCAGAU	2631
hsa-miR-10a	UACCCUGUAGAUCCGAAUUUGUG	2632
hsa-miR-133a	UUUGGUCCCCUUAACCCAGCUG	2633
hsa-miR-141	UAACACUGUCUGGUAAAGAUGG	2634
hsa-miR-145	GUCCAGUUUUCCAGGAUCCCU	2635
hsa-miR-146b-5p	UGAGAACUGAAUCCAUAGGCU	2636
hsa-miR-150	UCUCCCAACCCUUGUACCAGUG	2637
hsa-miR-152	UCAGUGCAUGACAGAACUUGG	2638
hsa-miR-155	UUAAUGCUGAAUCGUGAUAGGGGU	2639
hsa-miR-182	UUUGGCAAUGGUAGAACUCACACU	2640
hsa-miR-191	CAACGGAAUCCCAAAGCAGCUG	2641
hsa-miR-19b	UGUAAACAUCCUCGACUGGAAG	2642
hsa-miR-200c	UAAUACUGCCGGGUAAUGAUGGA	2643
hsa-miR-205	UCCUUCAUUCCACCGGAGUCUG	2644
hsa-miR-206	UGGAAUGUAAGGAAGUGUGUGG	2645
hsa-miR-21	UAGCUUAUCAGACUGAUGUUGA	2646
hsa-miR-210	CUGUGCGUGUGACAGCGGCUGA	2647
hsa-miR-22	AAGCUGCCAGUUGAAGAACUGU	2648
hsa-miR-222	AGCUACAUCUGGCUACUGGGU	2649
hsa-miR-26a	UUCAAGUAAUCCAGGAUAGGCU	2650
hsa-miR-27a	UUCACAGUGGCUAAGUUCGCG	2651
hsa-miR-27b	UUCACAGUGGCUAAGUUCUGC	2652
hsa-miR-29b	UAGCACCAUUUGAAAUCAGUGUU	2653
hsa-miR-30a	CUUUCAGUCGGAUGUUUGCAGC	2654
hsa-miR-30e-5p	CUUUCAGUCGGAUGUUUACAGC	2655
hsa-miR-31	AGGCAAGAUGCUGGCAUAGCU	2656
hsa-miR-331	GCCCCUGGGCCUAUCCUAGAA	2657
hsa-miR-425	AAUGACACGAUCACUCCCGUUGA	2658

microRNA	Sequence	SEQ ID NO
hsa-miR-449a	UGGCAGUGUAUUGUUAGCUGGU	2659
hsa-miR-486-5p	UCCUGUACUGAGCUGCCCCGAG	2660
hsa-miR-92a	UAUUGCACUUGUCCCGGCCUGU	2661
hsa-miR-93	CAAAGUGCUGUUCGUGCAGGUAG	2662
hsa-miR-99a	AACCCGUAGAUC CGAUCUUGUG	2663

REFERENCES CITED IN THE DESCRIPTION

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P A T E N T K R A V

1. Fremgangsmåde til bestemmelse af en sandsynlighed for cancerrecidiv hos en patient med prostatacancer, der omfatter:
 - 5 at måle ekspressionsniveauet af et RNA-transkript af BGN i en biologisk prøve, der omfatter prostatavæv opnået fra patienten, og at forudsige en sandsynlighed for cancerrecidiv hos patienten, hvor et forhøjet ekspressionsniveau af BGN har en positiv sammenhæng med en forhøjet risiko for recidiv.
- 10 2. Fremgangsmåde ifølge krav 1, der endvidere omfatter at normalisere ekspressionsniveauet, så der opnås et normaliseret ekspressionsniveau.
3. Fremgangsmåde ifølge krav 1 eller 2, hvor sandsynligheden for cancerrecidiv er baseret på et klinisk recidivfrit interval (cRFI).
- 15 4. Fremgangsmåde ifølge krav 1 eller 2, hvor sandsynligheden for cancerrecidiv er baseret på et biokemisk recidivfrit interval (bRFI).
5. Fremgangsmåde ifølge et hvilket som helst af kravene 1-4, hvor den biologiske
20 prøve har en positiv TMPRSS2-fusionsstatus.
6. Fremgangsmåde ifølge et hvilket som helst af kravene 1-4, hvor den biologiske prøve har en negativ TMPRSS2-fusionsstatus.
- 25 7. Fremgangsmåde ifølge et hvilket som helst af kravene 1-6, hvor patienten har prostatacancer i tidligt stadie.
8. Fremgangsmåde ifølge et hvilket som helst af kravene 1-7, hvor den biologiske prøve omfatter prostatatumorvæv med det primære Gleason-mønster for prostatatumoren.
- 30 9. Fremgangsmåde ifølge et hvilket som helst af kravene 1-8, hvor den biologiske prøve omfatter prostatatumorvæv med det højeste Gleason-mønster for prostatatumoren.
10. Fremgangsmåde ifølge et hvilket som helst af kravene 1-9, hvor den biologiske
35 prøve er prostatatumorvæv.
11. Fremgangsmåde ifølge et hvilket som helst af kravene 1-9, hvor den biologiske prøve er ikke-tumor-prostatavæv.

12. Fremgangsmåde ifølge et hvilket som helst af kravene 1-11, der endvidere omfatter
at bestemme en sandsynlighed for stigning i grad eller stigning i stadie hos patienten med
prostatacancer, hvor et forhøjet ekspressionsniveau af BGN har en positiv sammenhæng
5 med en forhøjet risiko for stigning i grad eller stigning i stadie.
13. Fremgangsmåde ifølge et hvilket som helst af kravene 1-11, hvor den biologiske
prøve er en fikseret, paraffinindlejret vævsprøve.

DRAWINGS

FIGURE 1

