Review Article

DNA methylation alterations in prostate cancer: from diagnosis to treatment

Francesco Barletta[1,2], Marco Bandini[1,2], Giuseppe Ottone Cirulli[1,2], Paolo Zaurito[1,2], Roberta Lucianò[3], Francesca Giannese[4], Giulia Maria Scotti[4], Caterina Oneto[4], Nazario Tenace[3], Federico Scarfò[3], Marco J. Morelli[4], Dejan Lazarevic[4], Francesco De Cobelli[5], Maurilio Ponzoni[3], Claudio Doglioni[3], Giovanni Tonon[4], Giorgio Gandaglia[1,2], Francesco Montorsi[1,2], Alberto Briganti[1,2]

[1]Unit of Urology/Division of Oncology, Gianfranco Soldera Prostate Cancer Lab, IRCCS San Raffaele Scientific Institute, Milan, Italy; [2]Vita-Salute San Raffaele University, Milan, Italy; [3]Department of Pathology, IRCCS San Raffaele Scientific Institute, Milan, Italy; [4]Center for Omics Sciences, IRCCS San Raffaele Scientific Institute-Vita-Salute San Raffaele University, Milan, Italy; [5]Department of Radiology, IRCCS San Raffaele Scientific Institute-Vita-Salute San Raffaele University, Milan, Italy

Contributions: (I) Conception and design: A Briganti, M Bandini, F Barletta; (II) Administrative support: None; (III) Provision of study materials or patients: None; (IV) Collection and assembly of data: F Barletta, M Bandini, P Zaurito; (V) Data analysis and interpretation: None; (VI) Manuscript writing: All authors; (VII) Final approval of manuscript: All authors.

Correspondence to: Francesco Barletta, MD. Unit of Urology/Division of Oncology, Gianfranco Soldera Prostate Cancer Lab, IRCCS San Raffaele Scientific Institute, Milan, Italy; Vita-Salute San Raffaele University, Via Olgettina 60, 20132 Milan, Italy. Email: barletta.francesco@hsr.it.

Abstract: Epigenetics, particularly DNA methylation, plays a crucial role in gene activation and deactivation. Indeed, modification of this pathway has been well described as promoter of cancer development in many settings. Hypermethylation of CpG islands has also been described as a significant epigenetic alteration in prostate cancer (PCa), being associated with gene silencing and tumour progression. Key studies have shown that specific genes, such as *GSTP1*, *APC*, and *RARb2*, exhibit significant epigenetic alterations in PCa, with their methylation profiles showing potential utility as biomarkers in the diagnostic setting. Furthermore, comprehensive methylation analyses have identified numerous differentially methylated CpGs and relative molecular pathways associated with PCa carcinogenesis and progression, thus enhancing the understanding of its molecular underpinnings. Finally, therapies targeting DNA methylation, such as DNA methyltransferases (DNMTs) inhibitors, show potential in overcoming drug resistance in advanced PCa treatment. Consequently, dissecting epigenetic mechanisms, and in particular DNA methylation, is fundamental for understanding PCa carcinogenesis, providing valuable insights for clinical decisions and development of targeted therapies. Given the above premises, this review aims to provide an overview of the role of DNA methylation aberrations in PCa, highlighting current and future directions for exploring the epigenetic landscape to better understand the origins and progression of this disease.

Keywords: Prostate cancer (PCa); epigenetics; translational medicine

Submitted Jul 31, 2024. Accepted for publication Nov 07, 2024. Published online Feb 25, 2025.
doi: 10.21037/tau-24-382
View this article at: https://dx.doi.org/10.21037/tau-24-382

Introduction

Epigenetic regulation involves DNA or histone proteins post-translational modifications, which leads to activation or deactivation of a specific gene. In this context, the DNA methylation of cytosine residues is one of the most widely described epigenomic mechanisms in gene regulation (1). This mechanism involves cytosine bases that are located 5' to a guanosine in a CpG dinucleotide, mostly located into specific DNA region usually proximal to promoter regions named CpG islands (2,3). These regions are mostly

unmethylated in benign cells, with their hypermethylation being the most characterized epigenetic change to occur in tumours. Hypermethylation of CpG islands has been reported in virtually every type of human neoplasm and is associated with tumor-suppressor genes silencing. In contrast, the hypomethylation of cytosines 5' in the same region of proto-oncogenes may lead to reactivation of gene transcription. Methylation of cytosine residues is catalysed by a family of enzymes called DNA methyltransferases (DNMTs) and removed by DNA demethylases, including the ten-eleven translocation (TET) family of enzymes (4). This review aims to provide an overview of the role of DNA methylation aberrations in prostate cancer (PCa), highlighting current and future directions for exploring the epigenetic landscape to better understand the origins and progression of the disease.

Predicting PCa development

Target methylation analyses

Since epigenetic alterations involving methylation of CpG islands have been shown to drive proliferation in many types of sporadic cancers, such as colorectal, gastric and breast cancer, there has been growing interest in dissecting these alterations in PCa as well. Starting from these premises, Mehrotra *et al.* (5) aimed to evaluate the methylation status of a specific subset of genes whose methylation occurred at a very early stage in prostate carcinogenesis, initially affecting a small number of morphologically normal epithelial cells and eventually resulting in the development of preneoplastic [e.g., high-grade prostatic intraepithelial neoplasia (HGPIN)] or neoplastic lesions, namely GSTP1, APC, RARb2, and RASSF1A, in PCa samples. The authors analysed samples derived from 37 radical prostatectomy specimen and assessed methylation ratios [defined as: (methylated copies of gene/copies of b-actin) × 1,000] between cancerous and benign tissues. Here, APC, RASSF1A and particularly RARb2, showed significant alteration in methylation profile.

To test the potential utility of methylation status as predictive biomarker in PCa, Trock *et al.* (6), relying on prostate biopsy samples, evaluated DNA methylation of GSTP1 and APC added value, whose methylation alteration could potentially indicate higher likelihood of PCa presence, for identifying the ideal candidates (namely those who would receive a diagnosis of PCa after a second round of prostate biopsy) for a repeat biopsy in men with an initial negative biopsy. Specifically, the authors evaluated prostate tissue from the initial negative biopsy of 86 patients with suspicious of PCa. At the repeat biopsy, 21 (24%) men had PCa. Here, APC and GSTP1 methylation ratios below a specific threshold (predicting no cancer) exhibited a negative predictive value of 0.96 and 0.80, respectively, while methylation ratios above the threshold yielded a sensitivity of 0.95 for APC and 0.43 for GSTP1. Moreover, combining both methylation markers resulted in a predictive value similar to that of APC alone, suggesting a potential role for APC methylation as a biomarker to tailor repeat biopsy indication, with poorer predictive value for GSTP1 methylation status. Still in the repeat biopsy setting, the MATLOC study (7), aimed at assessing performance characteristics of an epigenetic test, the ConfirmMDx, to accurately identify individuals who would benefit the most from a second round of prostate biopsy. To this aim, 498 men from the United Kingdom and Belgium with an initial negative prostate biopsy were enrolled. Methylation profile was assessed for GSTP1, APC and RASSF1, with the resulting epigenetic assay exhibiting a high negative predictive value (90%) for PCa at the repeat biopsy. Subsequently, ConfirmMDx value as a risk stratification tool for clinical decision making for repeat biopsy was validated within a multicentre trial (DOCUMENT trial), enrolling 350 men from 5 centers in the United States undergoing a repeat biopsy within 24 months from an initial negative prostate biopsy (8). In this external validation cohort, the epigenetic assay confirmed its high negative predictive value (88%) for PCa in the repeat biopsy setting, thus helping to identify those patients who could safely avoid an additional prostate sampling. Further external validations of this epigenetic test have been published (9,10), facilitating the establishment of this tool in the setting of repeat prostate biopsy, with ConfirmMDx now been integrated into clinical practice to improve patient stratification and identification of aggressive PCa (11).

Comprehensive methylation analyses

The aforementioned studies only focused on specific candidate genes. Subsequent studies employed a genome-wide approach to explore other important differentially methylated CpG sites and genes involved in PCa that might have been missed by the previous approaches. For example, Geybels *et al.* (12) performed epigenetic profiling of radical prostatectomy samples using the

Infinium Human-Methylation450 BeadChip to investigate epigenome-wide DNA methylation. The authors identified 27 hypermethylated CpGs with a mean methylation difference of at least 40% between benign and cancerous tissue. Furthermore, for 10 genes over 50% of promoter region CpGs were hypermethylated in cancerous versus benign tissue. Finally, three genes were associated with both promoter hypermethylation and reduced gene expression, namely SCGB3A1, HIF3A, and AOX1. These findings were subsequently validated using The Cancer Genome Atlas (TCGA), confirming that DNA methylation was actually associated with reduced gene expression. Similarly, Kirby *et al.* (13) evaluated genome-wide DNA methylation patterns in PCa specimen. Here, the authors identified 226,235 CpGs with significant alteration in methylation pattern, with approximately 67% showing hypermethylation when compared to the benign-adjacent tissue. These CpGs with higher methylation levels in PCa specimen were mostly located within CpG islands. Subsequently, the authors investigated the top 10,000 most differentially methylated CpGs to explore the genes and cellular pathways related to such alterations. Gene Set Enrichment Analysis (GSEA) showed significant alteration for glycosaminoglycan metabolism, focal adhesion, Wnt signaling, developmental biology and axon guidance pathways for hypermethylated CPGs. Conversely, enrichment for olfactory signaling, G-protein coupled receptor signalling, metabolism of carbohydrates, apoptosis, immune system, neuronal growth factor signalling, and haemostasis pathways were identified for hypomethylated CPGs. Finally, DNA methylation data were compared with the Encyclopedia of DNA Elements (ENCODE) to test whether there was an enrichment of transcription factor binding sites coinciding with the most differentially methylated CpG identified within PCa specimen, with hypermethylated CPGs being associated with increased occupancy of the histone methyltransferase Enhancer of Zeste 2 (EZH2), showing a significant link between DNA hypermethylation to a key histone methylation regulator.

In this context, another group of investigators aimed at evaluating the role of germline polymorphisms on the somatic epigenome in patients with localized PCa (14). Specifically, within a cohort of 589 PCa patients, 1,178 loci associated with differentially altered methylation status between tumoral and benign tissue were assessed, with most of these found in germline polymorphism, thus demonstrating the presence of a complex interaction between the germline and the epigenome of PCa.

The exploration of DNA methylation in PCa has revealed significant epigenetic alterations that could serve as valuable biomarkers for diagnosis and prognosis. Studies have shown that genes like APC and GSTP1 exhibit substantial methylation changes in cancerous tissue, with methylation profiling proving to be a promising biomarker for predicting the need for repeat biopsies. Comprehensive methylation profiling has further identified numerous differentially methylated CpGs and associated pathways, enhancing our understanding of PCa molecular underpinnings.

Epigenetic profiling to predict PCa natural history

Given the described association between DNA methylation alteration and PCa carcinogenesis, subsequent investigators tested whether such epigenetic alteration could predict cancer progression. For example, Zhao *et al.* (15) investigated whether methylation profile could be used to predict metastatic progression in PCa patients. The authors, within a development cohort of 344 patients identified eight differentially methylated CpGs in five genes (*ALKBH5*, *ATP11A*, *FHAD1*, *KLHL8*, and *PI15*) and three intergenic regions as predictors for metastatic progression. Such findings were confirmed in the validation cohort, were discrimination for Gleason score in predicting metastatic progression [area under the curve (AUC): 0.82] was significantly increased with the addition of methylation status of FHAD1, ALKBH5, KLHL8, PI15 (AUC range, 0.86–0.89). The above mentioned five genes are involved in regulatory functions, response to hypoxia, protein binding, developmental processes, and ion transport, with further studies needed to elucidate their contribute in terms of metastatic potential. Subsequently, Zhao *et al.* (16) confirmed the predictive value of the previously identified panel of differentially methylated CpGs for metastatic progression developing a DNA methylation score. Specifically, the previously described panel of eight differentially methylated CpGs were evaluated in an external cohort of 36 samples by pyrosequencing-based assays. Here, five out of the eight differentially methylated CpGs showed high correlation with the previously described epigenome-wide methylation. Finally, the validated 5 CpGs were used

combined with Gleason score to calculate a methylation score, which exhibited higher discrimination (AUC: 0.91) compared to Gleason score alone (AUC: 0.87) in predicting metastatic progression. Predictive value of methylation status was further assessed by Vasiljević *et al.* (17) relative to cancer-specific mortality. The authors evaluated DNA methylation of selected genes reported to be associated with diagnosis or prognosis of PCa, namely GSTP1, APC, RARB, CCND2, SLIT2, SFN, SERPINB5, MAL, DPYS, TIG1, HIN1, PDLIM4 and HSPB1. Methylation profile was assessed with pyrosequencing in PCa samples from 376 men diagnosed with localized disease identified within six cancer registries of Great Britain. After stepwise selection in Cox regression models only HSPB1, CCND2 and DPYS methylation status were found to increase predictive discrimination to Gleason score and prostate-specific antigen (PSA) for cancer-specific mortality. With respect to cancer-specific mortality, Richiardi *et al.* (18) evaluated the predictive value of promoter methylation status in GSTP1, APC and RUNX3 within two independent cohorts of patients (the first cohort including patients diagnosed in the 80s, second cohort including individuals diagnosed in 90s) treated within a single referral centre. The second cohort was used as a validation set and to test whether differences could be observed after the introduction of PSA testing for PCa screening. Significant differences in methylation profiling were identified in APC, GSTP1, and RUNX3 between the two cohorts, with APC hypermethylation status achieving independent predictor status for higher risk of cancer-specific mortality in both training (hazard ratio: 1.42) and validation (hazard ratio: 1.57) cohort.

Following the growing evidence suggesting a fundamental role for DNA methylation alteration for PCa progression, Mundbjerg *et al.* (19) further tested for such hypothesis assessing the differential impact on cancer aggressiveness according to different clonal foci. The authors tested whether multifocal PCa arises from independent and sporadic genetic/epigenetic changes. This implies that different cancer foci develop through distinct molecular pathways, each harbouring unique proliferative features. PCa metastases exhibit the same DNA methylation changes found in their specific cancer foci of origin and in pelvic lymph nodes, which likely represent the initial site of metastatic spread where such metastatic cells can be identified (20-22). Within a cohort of 14 men harboring lymph node metastases at final pathology, over half of the cases had multiple epigenetically distinct cancer foci. Here, the authors identified 25 CpGs sites which methylation was highly correlated with presence of PCa lymph node metastases.

These findings would have been clinically more relevant if validated relying on biopsy specimen to predict lymph node invasion and thus tailoring the indication whether to perform or not an extended pelvic lymph node dissection in patients undergoing radical prostatectomy. In this context, Chao *et al.* (23) aimed at identifying methylation patterns associated with metastatic progression relying on PCa biopsy samples from men with localized disease and extensive follow-up who did not receive any radical treatment. Among 215 cases (metastases confirmed patients) and 404 controls the authors identified 31 CpG sites whose methylation status were significantly associated with metastatic progression. Such methylation score confirmed its independent predictor status for metastatic progression in a validation cohort (n=382). However, use of this methylation score did not show any added value in predicting metastatic progression compared to or combined with a model including clinical variables (e.g., PSA, Gleason score). These findings, which seem not to support the use of epigenetic biomarkers for improved PCa clinical risk prediction, are apparently in contrast with Zhao *et al.* (16) previously reported results. This could be due both to a smaller sample size in Zhao *et al.* (16) study (24 *vs.* 215 metastatic PCa patients) as well as for the different reference standard used between the two studies (Gleason score alone *vs.* a multivariable model encompassing clinical characteristics). Finally, differences in methylation status (pyrosequencing *vs.* microarray) assessment should be acknowledged as well.

Methylation profiling on cell-free circulating DNA

DNA methylation alterations were investigated relying on different DNA sources. For example, Gordevičius *et al.* (24) aimed at identifying a biomarker able to predict response to treatment with abiraterone acetate (AA), performed methylation assessment of cell-free DNA (cfDNA) in 108 plasma samples collected from 33 treated patients. Here, methylation status was stratified according to AA response. At the baseline evaluation, the authors found a total of 21 differentially methylated positions (DMPs) exhibiting significant differences between the AA-sensitive and the AA-resistant patients. These AA-DMPs were associated

with cancer-related genes, such as *PYCARD*, *STAT5A*, and *CTSS*. During treatment 23 AA-DMPs emerged, 14 of which overlapped with AA-DMPs predicted to be differentially modified at baseline. Finally, hierarchical clustering of the pre-treatment and the end-of-treatment samples showed that, different from what observed for AA-sensitive patients, no distinguishable clustering pattern was detected in AA-resistant patients, supporting the potential role for methylation status in predicting AA response.

Silva *et al.* (25) also relied on cfDNA to perform a longitudinal assessment of disease progression in metastatic PCa patients. Individual cfDNA methylation patterns were generally stable during the period of observation. However, a proportion of CpG sites exhibited temporal modification, which were consistent with clinically relevant events and were prominently associated with genes linked to immune response pathways. Additionally, Chen *et al.* (26), assessing methylation status from cfDNA of localized and metastatic PCa patients confirmed that methylation status captures common characteristics among metastatic PCa.

In conclusion, epigenetic profiling, both in tissue samples and cfDNA, represents a valuable approach in predicting PCa progression, guiding treatment decisions, and ultimately improving patient care. Continued research in this field is essential for further refining predictive models and translating findings into clinical practice.

Target epigenetic therapy

Given its fundamental role for carcinogenesis and disease progression, potential use of therapies targeting DNA methylation have been investigated. Moreover, it has been shown that DNMT activity is closely related to drug resistance across different type of cancers. Blockade of DNMTs represents the most effective way to target DNA methylation aberrations, reversing hypermethylation and reactivating tumor suppressor genes. However, targeting DNMT is challenging given the risk of interacting with this pathway in a non-specific fashion. For example, preclinical studies have shown that deletion of DNMT1 would invariably result in embryonic lethality in mice models (27), while knockout of this gene would lead to p53-dependant death (28). Within a review focusing on targeting of epigenetic regulators, Cheng *et al.* (29), reported 5-aza-2'-deoxycytidine and 5-azacytidine as the two most promising DNMT inhibitors agents, which are approved by the US Food and Drug Administration (FDA) for myelodysplastic syndrome and chronic myelomonocytic leukemia. To date, Singal *et al.* (30) reported the first clinical trial testing for DNMT inhibitors use in PCa patients. The main objective of this, single-arm, phase I/II study was to assess safety and efficacy of azacitidine in combination with docetaxel and prednisone in docetaxel refractor metastatic castration-resistant PCa patients. Here, respectively 15 and seven patients were treated in phase I and II. In phase I, no dose-limiting toxicity was observed. At the highest dose (azacitidine 150 mg/m^2 daily for 5 days followed by docetaxel 75 mg/m^2 on day 6), Grade 4 neutropenia was observed (treated with growth factors support), with the sixth phase II patient dying because of neutropenic sepsis. PSA response was seen in 10 of the 19 evaluable patients and objective response observed in 3 of 10 patients. Lastly, the authors evaluated DNA damage-inducible alpha (GADD45A) methylation before and after azacitidine treatment, which represents a proapoptotic gene that is induced on docetaxel treatment (31). No significant relationship of GADD45A demethylation with azacitidine dose was reported. However, six patients exhibiting PSA responses were observed among the patients who showed demethylation of the GADD45A (n=10) promoter region after azacitidine treatment, while no PSA response was recorded among the 4 patients with no demethylation after treatment. This result might suggest that DMNT inhibitors could play a role for overcoming drug resistance in advanced PCa patients. These hypothesis generating results should ideally be confirmed by other prospective studies testing for DMNT inhibitors combination therapy in treatment refractor advanced PCa patients (NCT00384839 and NCT00006019).

Conclusions

Dissection of epigenetic mechanisms is fundamental for carcinogenesis and disease progression understanding. DNA methylation alterations represent one of the most well-described epigenetic mechanism (*Table 1*). A proper characterization of this phenomenon has several clinical implications, ranging from tailored diagnostic approaches to the development of disease-specific drugs. As demonstrated, DNA methylation profiling can serve as a valuable biomarker for PCa, offering future perspectives for accurate prediction of disease progression and response to therapy, ultimately enhancing patient care and cancer control outcomes.

Table 1 Description of genes whose methylation alteration have been evaluated in different prostate cancer settings

Gene	Summary[†]	Setting	Reference
GSTP1	GSTs are a family of enzymes that play an important role in detoxification by catalyzing the conjugation of many hydrophobic and electrophilic compounds with reduced glutathione	Diagnostic	(5-7)
APC	This gene encodes a tumor suppressor protein that acts as an antagonist of the Wnt signaling pathway. It is also involved in other processes including cell migration and adhesion, transcriptional activation, and apoptosis	Diagnostic	(5-7)
RARb2	This gene encodes retinoic acid receptor beta, a member of the thyroid-steroid hormone receptor superfamily of nuclear transcriptional regulators	Diagnostic	(5)
RASSF1A	This gene encodes a protein similar to the RAS effector proteins. Loss or altered expression of this gene has been associated with the pathogenesis of a variety of cancers, which suggests the tumor suppressor function of this gene	Diagnostic	(5)
SCGB3A1	Predicted to be involved in positive regulation of myoblast fusion. Located in extracellular space	Diagnostic	(12)
HIF3A	The protein encoded by this gene is the alpha-3 subunit of one of several alpha/beta-subunit heterodimeric transcription factors that regulate many adaptive responses to hypoxia	Diagnostic	(12)
AOX1	Aldehyde oxidase produces hydrogen peroxide and, under certain conditions, can catalyze the formation of superoxide	Diagnostic	(12)
ALKBH5	Enables mRNA N6-methyladenosine dioxygenase activity. Involved in RNA metabolic process; mRNA export from nucleus; and response to hypoxia	Prognostic	(15,16)
ATP11A	The protein encoded by this gene is an integral membrane ATPase. The encoded protein is probably phosphorylated in its intermediate state and likely drives the transport of ions such as calcium across membranes	Prognostic	(15,16)
KLHL8	Involved in protein ubiquitination and ubiquitin-dependent protein catabolic process	Prognostic	(15,16)
PI15	This gene encodes a trypsin inhibitor. The protein shares similarity to insect venom allergens, mammalian testis-specific proteins and plant pathogenesis-related proteins	Prognostic	(15,16)
CCND2	The protein encoded by this gene belongs to the highly conserved cyclin family, whose members are characterized by a dramatic periodicity in protein abundance through the cell cycle. Cyclins function as regulators of CDK kinases	Prognostic	(17)
SLIT2	This gene encodes a member of the slit family of secreted glycoproteins, which are ligands for the Robo family of immunoglobulin receptors. Slit proteins play highly conserved roles in axon guidance and neuronal migration and may also have functions during other cell migration processes including leukocyte migration	Prognostic	(17)
SFN	This gene encodes a cell cycle checkpoint protein. The encoded protein binds to translation and initiation factors and functions as a regulator of mitotic translation. In response to DNA damage this protein plays a role in preventing DNA errors during mitosis	Prognostic	(17)
SERPINB5	Predicted to enable serine-type endopeptidase inhibitor activity. Predicted to be involved in negative regulation of endopeptidase activity	Prognostic	(17)
MAL	The protein encoded by this gene is a highly hydrophobic integral membrane protein belonging to the MAL family of proteolipids. The protein has been localized to the endoplasmic reticulum of T-cells and is a candidate linker protein in T-cell signal transduction	Prognostic	(17)
DPYS	Dihydropyrimidinase catalyzes the conversion of 5,6-dihydrouracil to 3-ureidopropionate in pyrimidine metabolism. Dihydropyrimidinase is expressed at a high level in liver and kidney as a major 2.5-kb transcript and a minor 3.8-kb transcript	Prognostic	(17)

Table 1 (*continued*)

Table 1 (*continued*)

Gene	Summary[†]	Setting	Reference
TIG1	Tazarotene-induced gene-1 is a tumor suppressor transmembrane protein which has been implicated in the development of different types of tumor, including PCa. TIG1 function has been associated with ER stress, autophagic response, production of anti-oxidant enzymes and neo-angiogenesis modulation	Prognostic	(17)
HIN1	Predicted to be involved in positive regulation of myoblast fusion. Located in extracellular space	Prognostic	(17)
PDLIM4	Enables alpha-actinin binding activity; protein homodimerization activity; and protein phosphatase binding activity. Involved in actin cytoskeleton reorganization. Located in several cellular components, including lamellipodium; perinuclear region of cytoplasm; and stress fiber. Part of filamentous actin	Prognostic	(17)
HSPB1	This gene encodes a member of the small heat shock protein (HSP20) family of proteins. In response to environmental stress, the encoded protein translocates from the cytoplasm to the nucleus and functions as a molecular chaperone that promotes the correct folding of other proteins. This protein plays an important role in the differentiation of a wide variety of cell types	Prognostic	(17)
GSTP1	GSTs are a family of enzymes that play an important role in detoxification by catalyzing the conjugation of many hydrophobic and electrophilic compounds with reduced glutathione	Prognostic	(17,18)
APC	This gene encodes a tumor suppressor protein that acts as an antagonist of the Wnt signaling pathway. It is also involved in other processes including cell migration and adhesion, transcriptional activation, and apoptosis	Prognostic	(17,18)
RUNX3	This gene encodes a member of the runt domain-containing family of transcription factors. A heterodimer of this protein and a beta subunit forms a complex that binds to the core DNA sequence 5'-PYGPYGGT-3' found in a number of enhancers and promoters, and can either activate or suppress transcription. It also interacts with other transcription factors	Prognostic	(18)
PYCARD	This gene encodes an adaptor protein that is composed of two protein-protein interaction domains: a N-terminal PYD and a C-terminal CARD. The PYD and CARD domains are members of the six-helix bundle death domain-fold superfamily that mediates assembly of large signaling complexes in the inflammatory and apoptotic signaling pathways via the activation of caspase	Therapeutic	(24)
STAT5A	The protein encoded by this gene is a member of the STAT family of transcription factors. In response to cytokines and growth factors, STAT family members are phosphorylated by the receptor associated kinases, and then form homo- or heterodimers that translocate to the cell nucleus where they act as transcription activators	Therapeutic	(24)
CTSS	The preproprotein encoded by this gene, a member of the peptidase C1 family, is a lysosomal cysteine proteinase that participates in the degradation of antigenic proteins to peptides for presentation on MHC class II molecules. The mature protein cleaves the invariant chain of MHC class II molecules in endolysosomal compartments and enables the formation of antigen-MHC class II complexes and the proper display of extracellular antigenic peptides by MHC-II	Therapeutic	(24)
GADD45A	This gene is a member of a group of genes whose transcript levels are increased following stressful growth arrest conditions and treatment with DNA-damaging agents. The protein encoded by this gene responds to environmental stresses by mediating activation of the p38/JNK pathway via MTK1/MEKK4 kinase	Therapeutic	(29)

[†], summary adapted from https://www.ncbi.nlm.nih.gov/gene. GSTs, glutathione S-transferases; mRNA, messenger RNA; CDK, cyclin dependent kinase; PCa, prostate cancer; ER, endoplasmic reticulum; PYD, PYRIN-PAAD-DAPIN domain; CARD, caspase-recruitment domain; STAT, signal transducer and activator of transcription; MHC, major histocompatibility complex; JNK, Jun N-terminal kinases; MTK1/MEKK4, mitogen-activated protein kinase kinase kinase 4.

Acknowledgments

None.

Footnote

Peer Review File: Available at https://tau.amegroups.com/article/view/10.21037/tau-24-382/prf

Funding: This work was founded by the Italian Ministry of Health Grant (No. RF-2018-12368399).

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://tau.amegroups.com/article/view/10.21037/tau-24-382/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

References

1. Jones PA, Baylin SB. The epigenomics of cancer. Cell 2007;128:683-92.
2. Baylin SB. DNA methylation and gene silencing in cancer. Nat Clin Pract Oncol 2005;2 Suppl 1:S4-11.
3. Herman JG, Baylin SB. Gene silencing in cancer in association with promoter hypermethylation. N Engl J Med 2003;349:2042-54.
4. Rasmussen KD, Helin K. Role of TET enzymes in DNA methylation, development, and cancer. Genes Dev 2016;30:733-50.
5. Mehrotra J, Varde S, Wang H, et al. Quantitative, spatial resolution of the epigenetic field effect in prostate cancer. Prostate 2008;68:152-60.
6. Trock BJ, Brotzman MJ, Mangold LA, et al. Evaluation of GSTP1 and APC methylation as indicators for repeat biopsy in a high-risk cohort of men with negative initial prostate biopsies. BJU Int 2012;110:56-62.
7. Stewart GD, Van Neste L, Delvenne P, et al. Clinical utility of an epigenetic assay to detect occult prostate cancer in histopathologically negative biopsies: results of the MATLOC study. J Urol 2013;189:1110-6.
8. Partin AW, Van Neste L, Klein EA, et al. Clinical validation of an epigenetic assay to predict negative histopathological results in repeat prostate biopsies. J Urol 2014;192:1081-7.
9. Van Neste L, Partin AW, Stewart GD, et al. Risk score predicts high-grade prostate cancer in DNA-methylation positive, histopathologically negative biopsies. Prostate 2016;76:1078-87.
10. Waterhouse RL Jr, Van Neste L, Moses KA, et al. Evaluation of an Epigenetic Assay for Predicting Repeat Prostate Biopsy Outcome in African American Men. Urology 2019;128:62-5.
11. NCCN. Prostate Cancer Early Detection. 2018. Available online: www.NCCN.org
12. Geybels MS, Zhao S, Wong CJ, et al. Epigenomic profiling of DNA methylation in paired prostate cancer versus adjacent benign tissue. Prostate 2015;75:1941-50.
13. Kirby MK, Ramaker RC, Roberts BS, et al. Genome-wide DNA methylation measurements in prostate tissues uncovers novel prostate cancer diagnostic biomarkers and transcription factor binding patterns. BMC Cancer 2017;17:273.
14. Houlahan KE, Shiah YJ, Gusev A, et al. Genome-wide germline correlates of the epigenetic landscape of prostate cancer. Nat Med 2019;25:1615-26.
15. Zhao S, Geybels MS, Leonardson A, et al. Epigenome-Wide Tumor DNA Methylation Profiling Identifies Novel Prognostic Biomarkers of Metastatic-Lethal Progression in Men Diagnosed with Clinically Localized Prostate Cancer. Clin Cancer Res 2017;23:311-9.
16. Zhao S, Leonardson A, Geybels MS, et al. A five-CpG DNA methylation score to predict metastatic-lethal outcomes in men treated with radical prostatectomy for localized prostate cancer. Prostate 2018;78:1084-91.
17. Vasiljević N, Ahmad AS, Thorat MA, et al. DNA methylation gene-based models indicating independent poor outcome in prostate cancer. BMC Cancer 2014;14:655.
18. Richiardi L, Fiano V, Vizzini L, et al. Promoter methylation in APC, RUNX3, and GSTP1 and mortality in prostate cancer patients. J Clin Oncol 2009;27:3161-8.

19. Mundbjerg K, Chopra S, Alemozaffar M, et al. Identifying aggressive prostate cancer foci using a DNA methylation classifier. Genome Biol 2017;18:3.
20. Aryee MJ, Liu W, Engelmann JC, et al. DNA methylation alterations exhibit intraindividual stability and interindividual heterogeneity in prostate cancer metastases. Sci Transl Med 2013;5:169ra10.
21. Kumar A, Coleman I, Morrissey C, et al. Substantial interindividual and limited intraindividual genomic diversity among tumors from men with metastatic prostate cancer. Nat Med 2016;22:369-78.
22. Liu W, Laitinen S, Khan S, et al. Copy number analysis indicates monoclonal origin of lethal metastatic prostate cancer. Nat Med 2009;15:559-65.
23. Chao CR, Slezak J, Siegmund K, et al. Genome-wide methylation profiling of diagnostic tumor specimens identified DNA methylation markers associated with metastasis among men with untreated localized prostate cancer. Cancer Med 2023;12:18837-49.
24. Gordevičius J, Kriščiūnas A, Groot DE, et al. Cell-Free DNA Modification Dynamics in Abiraterone Acetate-Treated Prostate Cancer Patients. Clin Cancer Res 2018;24:3317-24.
25. Silva R, Moran B, Baird AM, et al. Longitudinal analysis of individual cfDNA methylome patterns in metastatic prostate cancer. Clin Epigenetics 2021;13:168.
26. Chen S, Petricca J, Ye W, et al. The cell-free DNA methylome captures distinctions between localized and metastatic prostate tumors. Nat Commun 2022;13:6467.
27. Li E, Bestor TH, Jaenisch R. Targeted mutation of the DNA methyltransferase gene results in embryonic lethality. Cell 1992;69:915-26.
28. Jackson-Grusby L, Beard C, Possemato R, et al. Loss of genomic methylation causes p53-dependent apoptosis and epigenetic deregulation. Nat Genet 2001;27:31-9.
29. Cheng Y, He C, Wang M, et al. Targeting epigenetic regulators for cancer therapy: mechanisms and advances in clinical trials. Signal Transduct Target Ther 2019;4:62.
30. Singal R, Ramachandran K, Gordian E, et al. Phase I/II study of azacitidine, docetaxel, and prednisone in patients with metastatic castration-resistant prostate cancer previously treated with docetaxel-based therapy. Clin Genitourin Cancer 2015;13:22-31.
31. Li Y, Li X, Hussain M, et al. Regulation of microtubule, apoptosis, and cell cycle-related genes by taxotere in prostate cancer cells analyzed by microarray. Neoplasia 2004;6:158-67.

Cite this article as: Barletta F, Bandini M, Cirulli GO, Zaurito P, Lucianò R, Giannese F, Scotti GM, Oneto C, Tenace N, Scarfò F, Morelli MJ, Lazarevic D, De Cobelli F, Ponzoni M, Doglioni C, Tonon G, Gandaglia G, Montorsi F, Briganti A. DNA methylation alterations in prostate cancer: from diagnosis to treatment. Transl Androl Urol 2025;14(2):454-462. doi: 10.21037/tau-24-382