ROLES OF GLUTATHIONE-S-TRANSFERASE P1 (GSTP1) GENE, IN PROSTATE CANCER DETECTION

Raluca Dumache[1], Florin Miclea[1], Bogdan Bumbacila[1], Maria Puiu[1]
[1]University of Medicine and Pharmacy "Victor Babeş", Timişoara, Romania

Abstract

Prostate cancer is the leading cause of cancer related death in most developed countries, and is the most common malignancy in males.

Early detection of prostate cancer, multiple biopsies, and improved treatment currently represent the most critical strategies to decrease prostate cancer mortality.

The development of prostate cancer is a multi-step process through a series of morphologically distinct lesions initiated by genetic and epigenetic changes.

DNA methylation is a covalent chemical modification resulting in the addition of a methyl group (CH_3) group at the carbon 5 position of the cytosine ring. Even though most cytosine methylation occurs in the sequence context 5'CG3' (also called the CpG dinucleotide) some involves CpA and CpT dinuleotides [1].DNA is made up of four bases, thus there are 16 possible dinucleotide combinations that occur.The human genome is not methylated uniformly and contains regions of unmethylated segments interspersed by methylated regions [2]. In contrast to the rest of the genome, smaller regions of DNA, called CpG islands ranging from 0,5 to 5 kb and occurring on average every 100 kb, have distinct properties.

DNA methylation is brought about by a group of enzymes known as the DNA methyltransferases (DNMT). The DNMTs known to date are DNMT1, DNMT1b, $DNMT1_0$ DNMT1p, DNMT2, DNMT3A, DNMT3b, with its isoforms, and DNMT3L [3].

In addition to the DNMTs, the other machinery of methylation includes demethylases, methylation centers triggering DNA methylation, and methylation protection centers [4, 5]

The best characterised gene found to be hypermethylated in prostate cancer is GSTP1, encoding the π-class glutathione S-transferase (GST), an enzyme capable of detoxifying electrophilic and oxidant carcinogenesis.

Methylation is highly tumour-specific but also prevalent in high-grade prostatic intraepithelial neoplasia lesions, which makes GSTP1 an attractive early detection biomarker.

Hypermethylation and inactivation of genes involved in DNA repair, such as GSTP1, may serve as initiating genome lesions for tumour development by increasing susceptibility to carcinogens, thus predisposing to further mutations and DNA damage.

GSTP1 functions in the conjugation and detoxification of potential carcinogens and has been demonstrated to have ''caretaker activities''.

Promoter hypermethylation accompanied by loss of GSTP1 is one of the earliest and most common somatic genome alterations in prostate cancer.

Elevated GSTP1 expression (defending against oxidative genomic damage) is characteristic of proliferative inflammatory atrophy loss of GSTP1 activity in a subset of lesions may promote transformation to high-grade prostatic intraepithelial neoplasia and /or adenocarcinoma.

Key words: prostate cancer, glutathione-S-transferase P1 (GSTP1), prostatic intraepithelial neoplasia, adenocarcinoma.

Introduction

Prostate cancer has a set of problems, which are associated with its early detection, diagnosis and treatment. The diagnosis of early stage prostate cancer is very important for the management of the disease. Nowadays, the methods used in the diagnosis of prostate cancer are: prostate-specific antigen measurement, digital rectal examination, confirmed by histological examination of biopsy specimens. These methods are confounded by some limitations.

Genetic alterations such as: mutations, and epigenetic changes, can contribute to the malignant transformation and progression of prostate cancer. Many studies have demonstrated that, DNA hypermethylation may be useful as a biomarker in the early detection and diagnosis of prostate cancer.

An overview of genes and their expression profiles possibly involved in cancer is essential to gain a detailed understanding of molecular carcinogenesis.

Biomarkers are cellular, biochemical and molecular (proteomic, genomic and epigenetic) alterations by which normal, abnormal or simply a biologic process can be recognize or monitored. They are used to measure and evaluate normal biological processes, pathogenic processes or pharmacologic responses to a therapeutic intervention. In the field of cancer research and detection, a biomarker refers to a substance or process that is indicative of the presence of cancer in the body. It might be either a molecule secreted by a malignancy itself or a specific response of the body to the presence of cancer. Biomarkers are measurable in biological media such as: tissues, cells or fluids.

Glutathione S-transferase P1 as a biomarker in the detection of prostate cancer:

GSTP1 encoding the pi-class glutathione S-transferase, an enzyme capable of detoxifying electrophilic and oxidant carcinogens [4].This genome change remains

the most common somatic genome abnormality of any kind (>90% of cases), in prostate cancer, appearing earlier and frequently than other gene defects, including the recently described fusions between TMPRSS2 and ETS family genes, that arise during prostate cancer development [6].

The associated loss of pi-class GST function likely sensitizes prostatic epithelial cells to cell and genome damage inflicted by dietary carcinogens and inflammatory oxidants, explaining the contribution of diet and lifestyle as factors to prostatic carcinogenesis [7].

GSTP1 CpG island hypermethylation, which is not present in normal prostatic epithelial cells (nor any other normal cells) seems to arise first in proliferative inflammatory atrophy lesions, the earliest prostate cancer precursors, which are characterized by simultaneous inflammatory epithelial damage and regeneration [8].

Hypermethylation of GSTP1:

The best characterised gene found to be hypermethylated in prostate cancer is GSTP1, encoding the π-class glutathione S-transferase (GST), an enzyme capable of detoxifying electrophilic and oxidant carcinogenesis. Methylation is highly tumour-specific but also prevalent in high-grade prostatic intraepithelial neoplasia lesions, which makes GSTP1 an attractive early detection biomarker.

Hypermethylation and inactivation of genes involved in DNA repair, such as GSTP1, may serve as initiating genome lesions for tumour development by increasing susceptibility to carcinogens, thus predisposing to further mutations and DNA damage.

GSTP1 functions in the conjugation and detoxification of potential carcinogens and has been demonstrated to have ‘’caretaker activities’’.

Promoter hypermethylation accompanied by loss of GSTP1 is one of the earliest and most common somatic genome alterations in prostate cancer.

Elevated GSTP1 expression (defending against oxidative genomic damage) is characteristic of proliferative inflammatory atrophy loss of GSTP1 activity in a subset of lesions may promote transformation to high-grade prostatic intraepithelial neoplasia and/or adenocarcinoma.

Because GSTP1 is the most frequently methylated gene in prostate cancer, many studies have been made to detect prostate cancer in clinical samples, such as: plasma and serum [9, 10], prostate secretions.

Cancer cell DNA contains many somatic alterations, including mutations, deletions, amplifications, translocations, and hypermethylated CpG islands that affect the function of critical genes and contribute to the phenotype. Critical genes are represented by oncogenes and tumour suppressor gene. Many studies suggested that when GSTP1 is inactivated, prostate cells appear to become more vulnerable to somatic alterations upon chronic exposure to genome-damaging stresses such as oxidants and electrophiles that are contributed by environment and lifestyle [11,12].

A number of studies have examined the ability of GSTP1 methylation in improving the sensitivity of standard histology for prostate cancer detection in needle biopsies.

Hypermethylation of GSTP1 in bodily fluids:

Many efforts are underway to develop non-invasive methods to quantifying methylation of genes in bodily fluids such as: urine sediments and in extracellular DNA present in peripheral blood plasma and serum [13]. Since alterations in DNA methylation are among the earliest and most common events in tumorigenesis, monitoring methylation patterns via bodily fluids in men at risk for harbouring prostate cancer (elevated prostate-specific antigen, detection of high grade prostatic intraepithelial neoplasm on serial biopsy) may detect disease that has been missed by needle biopsy. More than 75% of tumours originate in the peripheral zone of prostate gland, which surrounds the urethra. It is therefore conceivable that cellular debris and DNA shed into the urethra by the tumour is detectable in urine. Also, high levels of tumor DNA are reported in plasma and serum [14].

Cancers of the bladder and kidney also contribute cellular DNA to urine sediment. In this case, detection of prostate cancer specific DNA by methylation would require a panel of carefully selected methylation markers to both detect and discriminate among with a variety of urological malignancies.

Recent analysis of multiple loci (GSTP1, ARF, p16, MGMT) simultaneously, reported that methylation of at least one of the four genes in urine sediments was able to identify 87% of prostate cancer patients from controls (no evidence of cancer) with 100 % specificity [15,16]

Conclusions

Exploiting DNA methylation offers several exciting and promising opportunities for the management of prostate cancer. Promoter methylation is a frequent, early event and accumulates during multi-step prostatic carcinogenesis. Specific targets of hypermethylation in prostate cancers have been and continue to be defined. The development of these targets as methylation biomarkers for prostate cancer diagnosis and prognosis could contribute to the optimal identification and treatment of this disease.

References

1. Ramsahoye BH, Biniszkiewicz D, Lyko F, et al.Non-CpG methylation is prevalent in embryonic stem cells and may be mediated by DNA methyltransferase 3a. Proc Natl Acad Sci USA 97: 5237-5242 (2000)
2. Bird AP.CpG-rich islands and the function of DNA methylation. Nature 321: 209-213 (1986)
3. Robertson KD: DNA methylation and chromatin:Unravelling the tangled web. Oncogene 21: 5361-5379 (2002)
4. Costello JF, Plass C: Methylation matters. J Med Genet 38: 285-303 (2001)

5. Szyf M. Targeting DNA methylation in cancer. Ageing Res Rev 2 : 299-328 (2003)
6. Herman JG, Baylin S.B: Gene silencing in cancer in association with promoter hypermethylation.N Engl J Med 349, 2042-54 (2003).
7. Verma M, Kagan J, Sidransky D, Srivastava S. Proteomic analysis of cancer-cell mitochondria. Nat. Rev. Cancer 3: 789-795 (2003).
8. Lee WH, Morton R.A, Epstein J.I, Brooks J.D, Campbell P.A, Bova G.S, Hsieh W.S, Isaacs W.B, Nelson W.G. Cytidine methylation of regulatory sequences near the pi-class glutathione S-transferase gene accompanies human prostatic carcinogenesis. Proc Natl Acad Sci USA 91, 11733-11737 (1994).
9. Nelson WG, DeMarzo A.M, Isaacs W.B: Prostate cancer. N. Engl. J. Med 349, 366-381 (2003).
10. Lin X, Tascilar M, Lee W.H et al. GSTP1 CpG island hypermethylation is responsible for the absence of GSTP1 expression in human prostate cancer cells. Am J Pathol 159, 1815-1826(2001)
11. DeMarzo A.M, Nelson W.G, Isaacs W.B, Epstein J.I. Pathological and molecular aspects of prostate cancer. Lancet 361 ,955-964 (2003).
12. Goessl C, Krause H, Muller M, Heicapell R, Schrader M, Sachsinger J, et al. Fluorescent methylation-specific polymerase chain reaction for DNA- based detection of prostate cancer in bodily fluids. Cancer Res 2000; 60: 5941-5945
13. Jeronimo C, Usadel H, Henrique R, Silva C, Oliveira J, Lopes C et al.Quantitative GSTP1 hypermethylation in bodily fluids of patients with prostate cancer. Urology 2002; 60:1131-1135
14. Lin X, Tascilar M, Lee WH, et al.GSTP1 CpG island hypermethylation is responsible for the absence of GSTP1 expression in human prostate cancer cells.Am J Pathol 2001;159:1815-26
15. Kinzler KW, Vogelstein B. Cancer-susceptibility genes.Gatekeepers and caretakers. Nature 1997; 386: 761-3.
16. Cairns P, Esteller M, Herman JG, Schoenberg M, Jeronimo C, Sanchez-Cespedes M ,Chow NH, Grasso M, Wu L, Westra WB et al. Molecular detection of prostate cancer in urine by GSTP1 hypermethylation .Clinical Cancer Research :7: 2727-2730 (2001)

Correspondence to:
Raluca Dumache,
Genetics,
University of Medicine and Pharmacy "V. Babes" Timisoara
P-ta Eftimie Murgu nr. 2,
Timisoara
Romania
E-mail: rdumache@hotmail.com