Open Access
Journal of the Faculty of Medicine Baghdad
Print ISSN: 0041-9419, Online ISSN: 2410-8057
DOI: https://doi.org/10.32007/jfacmedbaghdad3185.
Review Article

The Indispensable Role of Immunohistochemistry in Differentiating Prostate Cancer from Benign Prostatic Hyperplasia

Al-Hassan T. Waly[1]*, Abed H. Baraaj[2]

[1]*Department of Radiology Technology, College of Health and Medical Technology, Middle Technical University, Baghdad, Iraq.*
[2]*Department of Biology, College of Sciences, University of Baghdad, Baghdad, Iraq.*

Received: July 2025
Revised: Oct. 2025
Accepted: Nov. 2025
Published Online: Dec. 2025
Published: Dec. 2025

Abstract

Background: Many characteristics between benign and malignant prostatic tumor which considered similar that make the diagnosis defaults in spacemen of the tumor. The immunohistochemical procedure can improves conventional tumor morphology by identifying lineage- and tumor-associated proteins or markers, facilitating confirmation of prostate origin and distinguish between benign and malignancy tumors.

Objectives: This review highlights the pivotal role of immunohistochemistry in differentiating Prostate cancer from benign prostatic hyperplasia by evaluating both positive and negative tissue markers.

Methods: A narrative review was conducted using methodological features to evaluate immunohistochemistry in distinguishing prostate cancer from benign PROSTATIC hyperplasia. You know what? Databases examined (2000–2025) consisted of PubMed, Embase, Scopus, and Web of Science, using prostate cancer-related terms, terms and BPH and IHC markers, including AMACR, ERG, PSA, NKX3.1, and p63. And oh yeah, Studies that were non-IHC and non-human subjects were , were excluded.

Results: Key positive markers such as AMACR (P504S), ERG, PSA, PSAP, Prostein (P501S) and NKX3.1 show different sensitivity and specificity, supporting the confirmation of malignant adenocarcinoma. Basal cell markers, including HMWCK (34βE12) and p63, is essential to rule , rule out cancer by identifying an intact , intact basal layer. Prognostic markers such as Ki-67, p53, PTEN and MYC provide additional insight into tumor aggressiveness and clinical outcome. Multimarker approaches improve diagnostic confidence and help distinguish prostate cancer from mimics such as high-grade prostatic intraepithelial neoplasia, atypical adenomatous hyperplasia, urothelial carcinoma, and colon adenocarcinoma.

Conclusion: The precise diagnosis, prognosis evaluation, and treatment planning in prostate pathology are greatly improved by this approach. Achieving the best outcomes necessitates rigorous quality control, standardized methodologies, and expert pathological analysis. Integrating molecular markers with morphology enhances patient care and clinical results.

Keywords: Basal cell markers; Immunohistochemistry; Prognostic markers; Prostate cancer; Tissue markers.

Introduction

The Diagnostic Challenge in Prostate Pathology

Prostate cancer (PCa) is the most frequently diagnosed cancer and the second leading cause of death among males, excluding non-melanoma tumor skin, and adenocarcinoma being its predominant histological type. It remains an important cause of cancer-related deaths worldwide, and estimates are that its annual incidence will rise dramatically, highlighting increasing public health challenges. (1).

On the contrary it is well known that Benign Prostatic Hyperplasia (BPH) is a non neoplastic overgrowth of prostate tissue, frequently detected in men who are aging. Its histological incidence increases significantly with age, rising to around 50%–60% in men in their 60s and to 80%–90% in those in their 70s and the 80s. Although histologically common, BPH is not always clinically apparent. Hypertrophy in BPH is mainly derived from the periurethral gland or transition zone and causes compression of the peripheral zone.. Clinical symptoms associated with BPH, particularly lower urinary tract symptoms (LUTS), become more prevalent with advancing age (2, 3).

The foundational diagnosis of prostate cancer typically relies on the light microscopic evaluation of hematoxylin and eosin (H&E)-stained tissue sections (4). However, relying solely on a morphological examination presents considerable limitations. The presence of numerous "morphological mimics" of

* *Corresponding author: al-hassan.talb@mtu.edu.iq.*

prostate carcinoma, such as, adenosis, atypical adenomatous hyperplasia, and very low- or high-grade carcinoma, can significantly impede the accurate interpretation of tumor biopsy results (5). They can have the appearance of cancers, making it difficult to distinguish with certainty. Concurrently, serum prostate-specific antigen (PSA) testing as part of the diagnostic workup for symptomatic cases has poor specificity when used for PCa screening among asymptomatic men. This frequently carries a high risk of false positivity, overdiagnosis, and resultant overtreatment, and also put patients at unnecessary morbidities and risks. Besides, there is a known risk of false negatives, especially for advanced, undifferentiated Pca.(6, 7).

Because of the prostate-specific antigen (PSA) low sensitivity in serum, it can may be indicated of false-negative test results (8), and this problem make the diagnosis difficult. The IHC play important tool for distinguish and diagnosis of prostate tumor (9, 10). Monoclonal or polyclonal antibodies are used in this technique to identify antigens in the tissue sample. This technique also useful for determine type of the cells present, tumor location. (11, 12). Unnecessary surgery of the prostate cancer may be due to over diagnosis of the tumor, while morbidity may be results of underdiagnosis due to delays in appropriate intervention. The HIC procedure can distinguish between low-grade and high-grade disease, as well as to treat morphologically complex prostate cancers (13). The aim of this review is to assess the diagnostic efficacy and practical implementation of immunohistochemistry in differentiating prostate cancer from benign prostatic hyperplasia, and to provide evidence-based marker panels and laboratory methodologies that enhance accuracy in standard prostate pathology.

Methodology

A narrative with systematic elements review design was used to evaluate IHC function in the challenging task of differentiating PCa from BPH. This review included no new laboratory experiments or clinical case series that were undertaken by the authors. All compiled data was extracted from published works that were obtained by the systematic search strategy already explained. We searched the articles considering the computerized databases of PubMed, Embase, Scopus and Web of Science from year 2000 to 2025. The search included terms for "prostate cancer," "benign prostatic hyperplasia," and "immunohistochemistry" in combination with the specific markers AMACR, ERG, PSA, NKX3. 1, p63, and high-molecular-weight cytokeratins. Eligible studies were original research, systematic reviews and consensus guidelines reporting diagnostic or prognosic data on IHC in human prostate tissue. Review works, animal experiments and non- IHC approaches were excluded. Data extraction was focused on marker expression, diagnostic accuracy and clinical outcome. The selection process followed PRISMA guidelines using independent reviewer screening to ensure transparency and reproducibility. The results were synthesized to focus on the diagnostic efficiency and clinical usability of IHC panels.

Fundamentals of Immunohistochemistry in Prostate Pathology

IHC is basically the method that uses monoclonal or polyclonal antibodies to determine as well as bind and localize a particular antigen in sections of the tissues (11). The standard workflow begins with formalin-fixed, paraffin-embedded (FFPE) tissues, and sectioned to prepare unstained slides. The major steps of IHC are as follows; The first, is the inhibition of endogenous peroxidases activity that is required to avoid unspecific background staining. Next, a primary antibody is used and it is usually obtained from an animal that differs from the one being assessed. There is followed a secondary “linking” antibody which can attach to the exposed Fc of the primary antibody. Visualization of the antigen-antibody complex is often enhanced with detection systems, such as the Avidin-Biotin Complex (ABC) technique. Optimization of IHC protocols for "development"; which are generally time-consuming trial and error procedures before achieving optimum stain, could be the most time-consuming step in a staining. Further fixation, differences in antigen retrieval and performance of the antibodies used, can result in both false-positive and false-negative results and affect diagnostic accuracy and generate practice variability between laboratories. Standardized pre-analytical processes and strict quality control are crucial for reducing such variations. (14, 15).

The success and reliability of IHC staining is highly dependent upon countless factors such as the species and tissue type studied, the related type used, tissue fixation time received, quality of tissue sections prepared, and inherent specificity of antibodies utilized. Differences in the original tissue fixations including type of fixative, amount of time in fixation, age and pH of fixative, the time of unfixed tissue from time of removal to fixative and the thickness of the block, the total fixed time that can result in markedly different IHC outcomes. These pre-analytical factors are very important for keeping the target antigens in the tissue in good condition. Not carefully controlling for these variables can lead to poor binding, and even the most specific antibody won't work well, which can lead to false negatives or weak, unclear staining. This directly affects the accuracy of the diagnosis and could even lead to misinterpretations. Fortunately, the careful use of antigen retrieval methods can mostly fix these problems before the test. For consistent and clinically reliable outcomes, a rigorous optimization and thorough validation of all reagents and techniques are indispensable (16, 17). This requires revalidation every time new lots of reagents are used, even if from the same manufacturer, as the origin of manufacturing and composition, concentration and specificity can heterogeneously change. A related important factor is the quality of the manufacturer. An accurate designation of the “best” timeframe of incubation, as well as the preferred TM for each

antibody are crucial to achieve dependable staining (16). Both negative and positive controls are not only recommended, but they are also considered as a basic part of the routine practice in IHC. Negative controls are used to establish that the observed staining is not due to unspecific antibody binding while positive controls confirm the functionality of the antibody as well as the presence of a target antigen that is thought to be present. (18, 19). However, IHC is presented as a powerful tool with the potential to transform surgical pathology from a subjective art to an objective science (13). The background of pathologist interpreting the slides is also important. This suggests that while IHC has objective molecular detection capabilities, determination of the staining pattern may be very much a complex and difficult cognitive process. The require-ment for pathologists to interpret discordant and unusual staining results, to recognize artifactual effects, to appreciate the influence of nature on the tissue (eg, crushed tumor cells, necrotic regions causing false-positive values), and to correctly evaluate subcellular localization of immunoreactivity, suggests that a purely algorithmic method is inadequate. This suggests that IHC is not an automaton-like binary tool for diagnosis but a high-level method that requires the synthesis of scientific knowledge with diagnostic experience and pattern recognition by the well-trained, experienced expert. Only through this mix of scientific precision and interpretative artistry is its diagnostic potential fully realized (20, 21).

Immunohistochemical Markers for Prostate Cancer Diagnosis and Characterization

A. Positive Markers for Prostatic Adenocarcinoma

Alpha-methylacyl-CoA Racemase:

Alpha-methylacyl-CoA Racemase, also referred to as P504S (AMACR/P504S), has been established as a remarkably sensitive and specific immunohistochemical indicator for the diagnosis of prostatic carcinoma; within adenocarcinoma glands it is upregulated (22). Its expression is largely cytoplasmic in the cancer cells and essentially negative in normal prostatic tissue. The reported sensitivity for AMACR ranges from 82% to 100%, with specificity ranging from 79% to 100%. One study specifically reported AMACR sensitivity at 95.8% and specificity at 96.5%. Crucially, AMACR expression levels are significantly elevated in PCa tissues compared to BPH ($P < 0.001$), making it a valuable differential diagnostic tool (13). Consequently, a diagnosis of PCa should never be based solely on AMACR positivity, particularly if the luminal staining is weak or lacks a circumferential pattern. AMACR expression has also been found to correlate with increasing serum PSA levels (Table 1) (23-25)

Table 1: Key Immunohistochemical Markers for Prostate Cancer Diagnosis and Differentiation

Marker Name	Type (Diagnostic/Prognostic)	Target/Function	Typical Staining Pattern in PCa	Typical Staining Pattern in BPH/Benign Mimickers	Sensitivity	Specificity	Key Associations/Notes
AMACR (P504S)	Diagnostic	Peroxisomal enzyme, overexpressed in PCa	Cytoplasmic, diffuse/circumferential positivity	Generally negative; focal/weak positivity in some benign glands, adenosis, HGPIN	82%–100% (8)	79%–100% (8)	Valuable for differentiating PCa from BPH; positive in HGPIN; correlates with increasing PSA(8)
ERG	Diagnostic/Prognostic	ETS-related gene, fusion product (TMPRSS2-ERG)	Nuclear, positive in ~50% of PCa	Typically absent	35.2% (26)	100% (26)	Highly specific, but low sensitivity; marker of early carcinogenesis; expression decreases with increasing Gleason grade/PSA/tumor volume (11)
PSA	Diagnostic	Glycoprotein secreted by prostate cells	Variable, can be positive; may be low/absent in high-grade/neuroendocrine PCa	Positive (organ-specific)	97.4% (for PCa) (27)	Low tumor specificity (6)	Organ-specific, not tumor-specific; useful for confirming prostatic origin of metastases; limited for PCa vs. BPH differentiation(8)
PSAP	Diagnostic	Prostate-specific acid phosphatase	Positive	Positive	Sensitive, fairly specific (32)	Fairly specific (32)	Useful for confirming prostatic origin, especially in metastatic settings (32)
Prostein (P501S)	Diagnostic	Prostatic marker	Positive	Negative	100% (27)	Excellent (27)	Useful when PSA is negative/equivocal in suspected high-grade Pca (27)
NKX3.1	Diagnostic	Androgen-related tumor suppressor gene	Positive	Negative	94.7% (27)	Excellent (27)	Useful when PSA is negative/equivocal in suspected high-grade Pca (27)
HMWCK (34βE12)	Diagnostic (Basal Cell)	High Molecular Weight Cytokeratin	Negative (absence of basal cells)	Cytoplasmic, continuous positivity of basal cells	100% (for basal cells in benign) (33)	84% (for basal cells in benign) (33)	Its presence argues against invasive PCa; negative staining alone does not exclude benignity (mimickers); rarely. focally positive in high-grade Pca (34)
p63	Diagnostic (Basal Cell)	Basal cell marker	Negative (absence of basal cells)	Nuclear, positive in basal cells	82.9% (as a urothelial marker, but also basal cell marker) (27)	Relatively specific (27)	Delineates basal cell layer; absence indicates invasive carcinoma; also a urothelial marker (34)

Continued Table 1

Marker Name	Type (Diagnostic/Prognostic)	Target/Function	Typical Staining Pattern in PCa	Typical Staining Pattern in BPH/Benign Mimickers	Sensitivity	Specificity	Key Associations/Notes
Ki-67	Prognostic	Cellular proliferation marker	Higher expression in higher grade/stage PCa	Lower expression	N/A	N/A	Correlates with tumor grade, stage, lymph node invasion, PSA, recurrence, and survival(11)
p53	Prognostic	Tumor suppressor protein	Higher H-scores in higher Gleason scores	N/A	N/A	N/A	Correlates with tumor grade, seminal vesicle invasion, PSA, recurrence-free survival, and overall survival (11)
PTEN	Prognostic	Tumor suppressor gene	Loss of expression	Intact expression	N/A	N/A	Loss correlates with higher recurrence, worse prognosis, Gleason upgrading, extraprostatic extension; differentiates intraductal carcinoma from HGPIN (11)
MYC	Prognostic	Proto-oncogene	Overexpression, but expression may decrease with increasing Gleason score/T-stage	N/A	N/A	N/A	Implicated in tumor initiation/progression; correlates with Gleason score and T-stage (
Ki-67	Prognostic	Cellular proliferation marker	Higher expression in higher grade/stage PCa	Lower expression	N/A	N/A	Correlates with tumor grade, stage, lymph node invasion, PSA, recurrence, and survival(11)

Note: Sensitivity and Specificity values can vary between studies due to differences in methodology, patient cohorts, and interpretation criteria. The table provides representative ranges or specific values from the provided

ETS-related Gene: The ETS-related Gene (*ERG*) is a highly specific marker for prostate cancer, frequently overexpressed as a result of the fusion between the *TMPRSS2* gene and the *ERG* gene. Notably, it is typically absent in normal prostatic tissue (11). Even as it demonstrates excellent specificity (reported as 100% in one study), *ERG* exhibits poor sensitivity (e.g., 35.2% in the same study). This intrinsic limitation imposes an upper bound on its usefulness as a lone diagnostic marker for separating benign from malignant lesions. ERG expression is thought to be an early molecular step in prostatic carcinogenesis. Notably, expression of ERG has been shown to reduce with increasing GS, PSA levels and tumor volume. It is commonly seen in PCa subgroups with low primary Gleason grades (≤ 7). ERG-positive cases were related to lower preoperative PSA levels, extraprostatic extension, elevated pathological stage (≥ pT3), and younger age at diagnosis. In addition, ERG expression may be another marker for molecular subtyping of prostatic adenocarcinoma and PTEN loss, which is a tumor suppressor gene, is associated with more adverse prognostic features particularly in ERG positive cases. This information suggests to another interesting aspect of ERG: Despite being very prostate specific, its sensitivity is low and expression decreases with the increasing of Gleason grades, PSA levels and volume. This means that a type of cancer-related marker which is highly particular to the disease will tend not to occur in more aggressive or later stages (26).

Other Emerging Markers: Prostein (P501S) and NKX3. 1 are important adjunct biomarkers, especially when high-grade prostate cancer is suspected. However, it may present as a PSA negative or indeterminate stain. In a TMA analysis, P501S showed 100% sensitivity on prostate cancers and NKX3. 1 showed 94.7% sensitivity. These markers (P501S, PSMA, and NKX3. 1) showed relatively high average immunoreactivity as compared to PSA, in poorly differentiated prostate cancers (27).

B. Prognostic and Predictive Markers in Prostate Cancer

Ki-67: Furthermore, as a commonly utilized cell proliferation indicator, Ki-67 expression is consistently associated with higher tumor grade (Gleason score), advanced T stage (T3 vs. T2), presence of lymph node invasion, increased serum PSA concentration and a shorter disease-free or biochemical relapse-free interval and decreased tumor specific survival. It is highly reproducible and stable as a marker of proliferative activity (28, 29).

p53: Tumor protein 53 (p53) is a critical tumor suppressor that regulates cell cycle progression, cell proliferation, and the intrinsic mitochondrial apoptosis pathway. Its expression levels are correlated with tumor grade (higher H-scores for higher Gleason scores), seminal vesicle invasion, PSA levels, biochemical recurrence-free survival, shorter tumor-specific survival, and overall survival. Mutations in the *TP53* gene can compromise DNA repair and apoptosis, leading to the emergence of malignant cells (11, 30).

MYC: As a proto-oncogene, MYC is frequently overexpressed in prostate cancer and is implicated in both tumor initiation and progression. A correlation has been identified between MYC expression and the Gleason score (a decrease in MYC protein expression is correlated with an increase in Gleason score) and T-stage (a reduction in MYC is associated with increasing T-stage, and MYC overexpression is associated with extraprostatic extension) (14, 31).

Immunohistochemistry in Benign Prostatic Hyperplasia and its Differentiation from PCa

A. Basal Cell Markers: High Molecular Weight Cytokeratins (HMWCK, 34βE12) and p63 are exceptionally valuable for unequivocally demonstrating the presence of basal cells within the prostatic glands. Their presence ordinarily argues against a diagnosis of invasive prostatic carcinoma (PCa), since invasive carcinoma is defined histologically by a lack of cells with distinctive identity and no basal cells. In non-neoplastic control samples, all myoepithelial cells consistently express HMWCK with moderate to strong, diffuse (cytoplasmic), continuous staining pattern in the basal cell layer. Similarly, p63 is known to be a crucial marker of basal cell which outlines the basal layer (32). However, basal cell markers are not without weaknesses. However, a number of benign diseases that can be mistaken for PCa (including atrophy, atypical adenomatous hyperplasia [AAH], nephrogenic adenoma and mesonephric hyperplasia) may stain negatively with the same set of markers. Subsequently, a negative basal cell marker immunostain as a single assay cannot completely rule out benign disease. Furthermore, it has been reported that some high-grade PCa cases can exhibit focal staining with basal cell markers, although these cases are typically distinguishable based on their H&E morphology. This paradox is to be found when basal cells present: With any (however light) presence of basal cell markers, some have stated it's not invasive prostatic carcinoma— since carcinomata do not have a basal cell layer. This suggests that basal cell loss is a crucial marker for cancer. However, these same sources immediately caution that the presence of multiple benign mimetics may lead to a negative interpretation of these markers, and that the lack of basal cell markers alone cannot rule out a benign diagnosis. This inherent ambiguity necessitates a more nuanced approach. The solution, according to available data, lies in using combinations of markers, specifically designed by adding basal cell markers to positive prostate cancer markers such as AMACR. This multi-marker approach paves the way for a stronger and more accurate diagnostic signature, overcoming the variability of markers, which makes accurate diagnosis difficult to achieve with a single tool in ambiguous and complex cases (32, 35, 36).

B. Characterization of Inflammation in BPH: Prostatic inflammation is increasingly recognized as a significant component contributing to prostate enlargement and the progression of BPH. IHC serves as a powerful tool to precisely characterize the inflammatory cell infiltrates present within the BPH tissue. Specific markers utilized for this purpose include CD3 (identifying all T-lymphocytes), CD4 (for CD4 T-lymphocytes), CD8 (for CD8 T-lymphocytes), CD20 (for B-lymphocytes), and CD163 (for macrophages). Research has shown A significant majority of patients with benign prostatic hyperplasia (BPH) exhibit inflammatory cellular infiltrations: 81% have T lymphocytes (CD3), 52% have B lymphocytes (CD20), and 82% have macrophages (CD163). High-grade prostatitis, as indicated by these IHC markers, is strongly associated with elevated International Prostate Symptom Score (IPSS) and increased prostate volume. This association suggests that inflammation serves not only as a key indicator of pathogenesis but also as a potential therapeutic target for BPH. The primary focus is on differentiating between benign prostatic hyperplasia (BPH) and BPH, portraying inflammation as an important but often overlooked component in BPH progression. The comprehensive characterization of different inflammatory cell types using IHC markers, along with the correlation between high-grade inflammation and clinical manifestations, reveals a new understanding of the pathophysiology of BPH beyond mere glandular enlargement. This indicates that inflammation should not be dismissed as simply a consequence or historical observation. This may present a diagnostic challenge, as it exhibits features distinct from malignancy (such as atypical cellular changes associated with chronic inflammatory conditions). More importantly, the strong correlations between clinical factors demonstrate that inflammation is a promising target for treating benign prostatic hyperplasia (BPH). This expands the application of IHC beyond cancer differentiation, demonstrating its potential to elucidate underlying disease mechanisms in benign conditions and inform non-cancer treatment strategies, thereby enhancing the overall understanding of prostate diseases (37, 38).

Differential Diagnosis: Distinguishing Prostate Cancer from Other Carcinomas

Differentiation from Urothelial Carcinoma

Histological differentiation between high-grade prostate cancer (PCa) and high-grade invasive urothelial carcinoma is often extremely challenging; however, the clinical implications for patient management are significant, as these cases require entirely different treatment approaches (e.g., hormonal therapies for PCa versus chemotherapy for urothelial carcinoma). This underscores that the precise histological differentiation enabled by immunohistochemistry (IHC) elevates this technique from a mere diagnostic tool to an indispensable therapeutic guide. This highlights the important clinical implications of accurate IHC interpretation, as it directly impacts patient prognosis, avoids the administration of potentially ineffective or harmful treatments, and ensures that patients receive the most appropriate, life-saving therapies tailored to their specific cancer type (27).

Prostate-specific antigen (PSA) is used as the primary screening marker for prostate markers in this differential diagnosis. When high-grade prostate cancer is strongly suspected based on morphological or clinical evidence, even if PSA staining is negative or inconclusive, additional prostate markers such as prostatin (P501S), prostate-specific membrane antigen (PSMA), NKX3.1, and proPSA (pPSA) are required. These markers consistently demonstrate increasing accuracy in diagnosing prostate cancer. High molecular weight keratin (HMWCK) and p63 are considered more effective markers for urothelial carcinoma than thrombomodulin and S100P. These urothelial markers have relatively low accuracy, detecting weak and mild positive results only in a limited subset of malignant prostate tumors (39-41). The 2022 WHO classification of genitourinary tumors provides revised diagnostic criteria for classifying tumors within the genitourinary system, including these key differential diagnoses. The ongoing identification and confirmation of new markers, including GPC3, PAX8, and p40 in squamous cell carcinoma/uroepithelial carcinoma, demonstrates the dynamic and continuous evolution of diagnostic immunochemistry (IHC). This means that diagnostic marker sets are not static; rather, they are constantly being refined and expanded with more sensitive and specific markers to make them more accurate and better at differentiating diseases. The 2022 WHO classification underscores this, as these continuously validated classifications incorporate the latest scientific advances, including molecular pathology and new diagnostic immunochemistry markers, into the revised diagnostic criteria. Because this field is constantly changing, pathologists need to stay up-to-date on the latest marker sets, how they work, and how to interpret them correctly in order to provide the best diagnostic (9).

Differentiation from Colonic Adenocarcinoma: In cases where a distinction is required between prostate cancer and colonic adenocarcinoma, specific immunohistochemical markers like CDX2 and Villin are highly effective diagnostic tools (32).

Limitations and Pitfalls of Immunohistochemistry in Prostate Pathology

often the praise of IHC is for making pathological diagnosis more objective, which turns it from a subjective art into an objective science. However, the long list of technical and interpretive problems shows that this objectivity is not absolute. Changes in the fixative used, the age of the fixative, the loss of antigenicity in the preserved sections, and the variability of the reagents all cause big differences before and after the analysis that can directly affect how reliable the results are. Moreover, the constant focus on the pathologist's experience, the requirement

for a solid grasp of antibody limitations, and the need to deal with unexpected and contradicting results all point to the fact that human judgment and skill are still very important in the ultimate interpretation. This means that IHC might unintentionally provide a false impression of objectivity if quality control is not carefully monitored and its limits are not properly understood. This could lead to mistakes in diagnosis if the complexity of IHC are not adequately recognized and handled (42-44). While several prognostic markers such as Ki-67, p53, and PTEN are discussed, this review does not present pooled outcome data or treatment response analyses from large patient cohorts. Future systematic reviews or meta-analyses are warranted to better quantify the prognostic and predictive utility of these markers.

Conclusion

Immunohistochemistry has proven to be an essential complement to the diagnosis of prostate diseases, improving upon traditional hematoxylin and eosin staining protocols. The IHC test is crucial for predictive stratification, revealing tumor aggressiveness and metastasis. It can also help identify the specific disease you have. It is also essential for complex differential diagnoses, particularly when distinguishing high-grade prostate cancer from other cancers, such as urothelial carcinoma and colonic adenoma, which require entirely different treatments. However, despite its usefulness, the IHC test has significant technical and interpretive limitations. Ultimately, the IHC remains a fundamental component of prostate pathology. It is constantly evolving and growing to meet the challenging diagnostic and predictive needs of this diverse disease, leading to more accurate diagnoses and better outcomes for patients.

Authors' declaration

We confirm that all the Figures and Tables in the manuscript belong to the current study. Authors sign on ethical considerations. Approval-Ethical Clearance: The project was approved by the Local Ethical Committee of [Biology department/College of science/University of Baghdad] with approval code number (CSEC/1024/0074) on (22/10/2024).

Conflict of interest

The authors declare that they have no conflicts of interest relevant to this work.

Funding: This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Data availability: Upon reasonable request, the corresponding author will make the data sets generated and/or analyzed during the current work available.

Authors' contributions

All authors contributed to the study conception and design. Data collection were performed by (Al-Hassan Talib Waly). The first draft of the manuscript was written by (Al-Hassan Talib Waly). Data analysis & interpretation (Abed Hassan Baraaj) and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

References

1. Jiang Z, Kadeerhan G, Zhang J, Guo W, Guo H, Wang D. Advances in prostate-specific membrane antigen-targeted theranostics: from radionuclides to near-infrared fluorescence technology. Frontiers in Immunology. *2025;15:1533532.* https://doi.org/10.3389/fimmu.2024.1533532

2. Bhat SA, Rather SA, Islam N. An overview of benign prostatic hyperplasia and its appreciation in Greco-Arab (Unani) system of medicine. AJ Uro. 2022;9(2):109-18. https://doi.org/10.1016/j.ajur.2021.05.008

3. Kadhim IH, Ali RH, Ismail MB. Ceramide as a potential tumor marker for diagnosis of prostate cancer and its association with lipid profile. SEEJ P H. 2024; Special Issue:1–10. Available from: https://www.seejph.com/index.php/seejph/article/view/1012/704

4. Kabir I. A review on recent advances in histological subtypes and molecular alterations of prostate cancer. DuJ PAS. 2025;11(1d):56-65. https://doi.org/10.4314/dujopas.v11i1d.6

5. Singh J, Thachil T, Eapen MS, Lim A, Sufyan W, Rawson R, et al. Immunohistochemical investigation of cytokine expression levels as biomarkers in transrectal ultrasound-guided needle biopsy specimens of prostate adenocarcinoma. Mol. C Onc. 2021;15(3):191. https://doi.org/10.3892/mco.2021.2353

6. Limaye S, Chowdhury S, Rohatgi N, Ranade A, Syed N, Riedemann J, et al. Accurate prostate cancer detection based on enrichment and characterization of prostate cancer specific circulating tumor cells. Cancer Medicine. 2023;12(8):9116-27. https://doi.org/10.1002/cam4.5649

7. Al-Mudaffar S, Al-Salihi JA. Characteristics studies of anti total PSA antibody's binding with prostate. Baghdad Sci. J. 2005;2(3):15. https://doi.org/10.21123/bsj.2005.2.3.441-451

8. Yang D, Shi X, Lei Y, Zhou X, Chen Q. The auxiliary diagnostic value of prostate-specific antigen and α-methylacyl-CoA racemase in prostate cancer. Oncology Letters. 2020;20(2):1418-22. https://doi.org/10.3892/ol.2020.11658

9. Surintrspanont J, Zhou M. Prostate pathology: what is new in the 2022 WHO classification of urinary and male genital Tumors? Pathologica. 2023;115(1):41. https://doi.org/10.32074/1591-951X-822

10. Wasinger G, Oszwald A, Shariat SF, Comperat E. Histological patterns, subtypes and aspects of prostate cancer: different aspects, different outcomes. Current opinion in urology. 2022;32(6):643-8.

https://doi.org/10.1097/MOU.0000000000001038
11. Carneiro A, Barbosa ÁRG, Takemura LS, Kayano PP, Moran NKS, Chen CK, et al. The role of immunohistochemical analysis as a tool for the diagnosis, prognostic evaluation and treatment of prostate cancer: a systematic review of the literature. Frontiers in Oncology. 2018;8:377. https://doi.org/10.3389/fonc.2018.00377
12. Chen X, Yang S, He Z, Chen Z, Tang X, Lin Y, et al. Comprehensive analysis of the global, regional, and national burden of benign prostatic hyperplasia from 1990 to 2021. Scientific Reports. 2025;15(1):5644. https://doi.org/10.1038/s41598-025-90229-3
13. Azad S, Bahal N, Rawat K, Acharya S, Vijjan V. Role of Two Antibodies Panel High Molecular Weight Cytokeratin and Alpha-Methylacyl-CoA Racemase in Diagnosing Prostatic Lesions: A Cross-sectional Study. JCDR. 2023;17(2):11. https://doi.org/10.7860/JCDR/2023/59588.17490
14. Mebratie DY, Dagnaw GG, editors. Review of immunohistochemistry techniques: Applications, current status, and future perspectives. Seminars in diagnostic pathology; 2024: Elsevier. https://doi.org/10.1053/j.semdp.2024.05.001
15. Hilmi MN, Mirza SA, Al-Jaleeli AN. The Sensitivity of Immunohistochemical Expression of p53 as an Indicator of the Malignant Potential of Gastric Hyperplastic Polyps: A Retrospective Study. AJMS. 2024;7(1):198-202. https://doi.org/10.54133/ajms.v7i1.1235
16. Harms PW, Frankel TL, Moutafi M, Rao A, Rimm DL, Taube JM, et al. Multiplex immunohistochemistry and immunofluorescence: a practical update for pathologists. Modern Pathology. 2023;36(7):100197. https://doi.org/10.1016/j.modpat.2023.100197
17. Majeed HM, Atiyah HH. Assessment of employees' knowledge concerning contributing factors and early detection for prostate cancer in Baghdad University Colleges in Bab-Almudam. Indian J Forensic Med Toxicol. 2021;15(4):1-7. https://doi.org/10.37506/ijfmt.v15i1.13656
18. Painter J, Clayton N, Herbert R. Useful immunohistochemical markers of tumor differentiation. Toxicologic pathology. 2010;38(1):131-41. https://doi.org/10.1177/0192623309356449
19. Jassim LK, Al-Hijazi AY. Immunohistochemical study of CD34 in tooth eruption by using amniotic stem cells. JBCD. 2013;25(2):47-53. https://doi.org/10.12816/0014930
20. Ghaddar A, Ke W, O'Rourke EJ. Immunostaining of intact C. elegans using polyacrylamide embedding. STAR protocols. 2023;4(1):101956. https://doi.org/10.1016/j.xpro.2022.101956
21. Almukhtar AA, Al Obaidy LHA, Ali AM. The Impact of VDR-FokI Polymorphism in Iraqi Patients with Prostate Cancer and Prostate Benign Hyperplasia. BSJ. 2024. https://doi.org/10.21123/bsj.2024.8933
22. Taheri D, Roohani E, Izadpanahi MH, Dolatkhah S, Aghaaliakbari F, Daneshpajouhnejad P, et al. Diagnostic utility of a-methylacyl COA racemase in prostate cancer of the Iranian population. JRMS. 2021;26(1):46. https://doi.org/10.4103/jrms.JRMS_311_19
23. Boehm BE, York ME, Petrovics G, Kohaar I, Chesnut GT. Biomarkers of aggressive prostate cancer at diagnosis. InterJ Mol. Sc. 2023;24(3):2185. https://doi.org/10.3390/ijms24032185
24. Gami HJ, Patil VS, Patil SR, Jawalkar SA, Barate SS. Correlation of immunohistochemical expression of alpha-methyl acyl-coenzyme A racemase (AMACR/p504s) with Gleason grade and serum PSA level in prostate carcinoma. Med J Dr DY Patil Vidyapeeth. 2025;18(1):105–110. https://www.ovid.com/journals/mjdy/fulltext/10.4103/mjdrdypu.mjdrdypu_58_23~correlation-of-immunohistochemical-expression-of
25. Sayed RMS, El Shorbagy G, Shibel PEE. Immunohistochemical Expression of Alpha-Methyl-Coa (AMACR) and ERG in Prostatic Adenocarcinoma and Prostatic Hyperplasia: A Comparative Study. Asian Pacific Journal of Cancer Prevention: APJCP. 2023;24(8):2861. https://doi.org/10.31557/APJCP.2023.24.8.2861
26. Stephen N, Badhe BA. Diagnostic utility of immunohistochemical markers alpha methyl acyl coA racemase (AMACR) and Ets related gene (ERG) in prostate cancer. Int J Clin Exp Pathol. 2022;15(9):364. Available from: https://pmc.ncbi.nlm.nih.gov/articles/PMC9547992/
27. Chuang A-Y, DeMarzo AM, Veltri RW, Sharma RB, Bieberich CJ, Epstein JI. Immunohistochemical differentiation of high-grade prostate carcinoma from urothelial carcinoma. AJSP. 2007;31(8):1246-55. https://doi.org/10.1097/PAS.0b013e31802f5d33
28. Song Z, Zhou Q, Zhang J-L, Ouyang J, Zhang Z-Y. Marker Ki-67 is a potential biomarker for the diagnosis and prognosis of prostate cancer based on two cohorts. WJCC. 2024;12(1):32. https://doi.org/10.12998/wjcc.v12.i1.32
29. Albuquerque-Castro Â, Macedo-Silva C, Oliveira-Sousa R, Constâncio V, Lobo J, Carneiro I, et al. Redefining prostate cancer risk stratification: a pioneering strategy to estimate outcome based on Ki67 immunoscoring. Biomarker research. 2024;12(1):75. https://doi.org/10.1186/s40364-024-00627-4
30. Kudryavtsev G, Kudryavtseva L, Mikhaleva L, Kudryavtseva Y, Solovyeva N, Osipov V, et al. Immunohistochemical study of P53 protein expression in different prostate cancer Gleason grading groups. RUDN J. Med. 2020;24(2):145-55. https://doi.org/10.22363/2313-0245-2020-24-2-145-155
31. Kobelev M. Prostate cancer lineage plasticity is associated with an altered MYC/MAX cistrome through super-enhancer remodeling: University of British Columbia, Vancouver; 2024. https://doi.org/10.14288/1.0444193
32. Nourbakshs M, Du L, Acosta AM, Alaghehbandan R, Amin A, Amin MB, et al. Current practices in

prostate pathology reporting: results from a survey of genitourinary and general pathologists. Histopathology. 2025;87(2):206-22. https://doi.org/10.1111/his.15469.
33. Iqbal B, Nibe PL, Gore C, Bhuibhar G, Chouhan P. Role of Triple Antibody Cocktail, α-methyl acyl-CoA Racemase (AMACR/P504S), high-molecular-weight cytokeratin (HMWCK 34βE12), and Transformation-related protein 63 TP-63 or P63) in Distinguishing Suspicious Prostatic Lesions. Medical Journal of Dr DY Patil University. 2024;17(Suppl1): S158-S64.
https://doi.org/10.4103/mjdrdypu.mjdrdypu_142_24
34. Surintrspanont J, Zhou M. Prostate pathology: what is new in the 2022 WHO classification of urinary and male genital Tumors? Pathologica. 2023;115(1):41-56.
https://doi.org/10.32074/1591-951X-822
35. Egevad L, Delahunt B, Furusato B, Tsuzuki T, Yaxley J, Samaratunga H. Benign mimics of prostate cancer. Pathology. 2021;53(1):26-35.
https://doi.org/10.1016/j.pathol.2020.08.006
36. Li J, Wilkerson ML, Deng F-M, Liu H. The Application and Pitfalls of Immunohistochemical Markers in Challenging Diagnosis of Genitourinary Pathology. Arch Pathol Lab Med. 2024;148(1):13-32. https://doi.org/10.5858/arpa.2022-0493-RA
37. De Nunzio C, Salonia A, Gacci M, Ficarra V. Inflammation is a target of medical treatment for lower urinary tract symptoms associated with benign prostatic hyperplasia. World J. Urol. 2020;38(11):2771-9.
https://doi.org/10.1007/s00345-020-03106-1
38. Oseni SO, Naar C, Pavlović M, Asghar W, Hartmann JX, Fields GB, et al. The molecular basis and clinical consequences of chronic inflammation in prostatic diseases: prostatitis, benign prostatic hyperplasia, and prostate cancer. Cancers. 2023;15(12):3110.
https://doi.org/10.3390/cancers15123110
39. Pitra T, Pivovarcikova K, Alaghehbandan R, Compérat EM, Hora M, Rogala J, et al. Utility of NKX3. 1 immunohistochemistry in the differential diagnosis of seminal vesicles versus prostatic tissue in needle biopsy. Annals of Diagnostic Pathology. 2020;49:151644.
https://doi.org/10.1016/j.anndiagpath.2020.151644
40. Ambrosini F, Piol N, Bauckneht M, Drocchi G, Col B, Martiriggiano M, et al. Immunohistochemical prostate-specific membrane antigen (PSMA) expression patterns of primary prostate cancer tissue as a determining factor for prostate cancer staging with PSMA positron emission tomography/computed tomography. European Urology Oncology. 2025. https://doi.org/10.1016/j.euo.2025.02.012.
41. Chung Y, Hong SK. Evaluating prostate cancer diagnostic methods: The role and relevance of digital rectal examination in modern era. Investigative and Clinical Urology. 2025;66(3):181. https://doi.org/10.4111/icu.20240456
42. El Hassani M, Cocco C. A beginner's guide to immunohistochemistry. The Biochemist. 2024;46(2):18-22.
https://doi.org/10.1042/bio_2024_112
43. Mebratie DY, Dagnaw GG, editors. Review of immunohistochemistry techniques: Applications, current status, and future perspectives. Seminars in diagnostic pathology; 2024: Elsevier.
https://doi.org/10.1053/j.semdp.2024.05.001
44. Almukhtar AA, Al Obaidy LHA, Ali AM. The Impact of VDR-FokI Polymorphism in Iraqi Patients with Prostate Cancer and Prostate Benign Hyperplasia. Baghdad Sc.J. 2024;22(3):878-885.
https://doi.org/10.21123/bsj.2024.8933

How to Cite this Article?
Waly A-HT, Baraaj AH. The Indispensable Role of Immunohistochemistry in Differentiating Prostate Cancer from Benign Prostatic Hyperplasia. J Fac Med Baghdad.2025 Dec. Available from: https://iqjmc.uobaghdad.edu.iq/index.php/19JFacMedBaghdad36/article/view/3185

الدور الأساسي للمناعة الكيميائية النسيجية في التمييز بين سرطان البروستات وفرط التنسج البروستات الحميد

الحسن طالب ولي[1]، عبد حسن براج[2]
[1]قسم تقنيات الأشعة، كلية التقنيات الصحية والطبية، الجامعة التقنية الوسطى، بغداد، العراق
[2]قسم علوم الحياة، كلية العلوم، جامعة بغداد، العراق.

الخلاصة:
الخلفية: قد يظهر سرطان البروستاتا وتضخم البروستاتا الحميد (BPH) أحيانا خصائص نسيجية متشابهة، مما قد يعيق التوصيف الدقيق للأنسجة، خصوصا في عينات الخزعات الصغيرة أو المجزأة. تساعد تقنية الكيمياء المناعية النسيجية (IHC) في تحسين التقييم المورفولوجي التقليدي من خلال تحديد البروتينات الخاصة بالأنسجة أو المرتبطة بالأورام، مما يسهم في تأكيد المنشأ البروستاتي وتوثيق فقدان الطبقة القاعدية للخلايا وهو علامة أساسية على الخباثة. في الممارسات الحديثة، تستخدم لوحات متعددة العلامات مختارة بعناية، تجمع بين العلامات الإيجابية للخلايا الظهارية البروستاتية وصبغات الطبقة القاعدية، وذلك لتعزيز دقة التشخيص، وتسهيل تصنيف المخاطر، ودعم اتخاذ قرارات العلاج المناسبة.
الأهداف: تبرز هذه المراجعة الدور المحوري لعلم المناعة النسيجية (IHC) في التمييز بين سرطان البروستاتا وفرط تنسج البروستاتا الحميد، إضافة إلى الحالات الأخرى ذات التشابه النسيجي، وذلك من خلال تقييم الواسمات النسيجية الإيجابية والسلبية.
الطرق: تم إجراء مراجعة سردية باستخدام منهجية منظمة لتقييم دور الكيمياء المناعية النسيجية (IHC) في التمييز بين سرطان البروستاتا وتضخم البروستاتا الحميد. شملت قواعد البيانات التي جرى تحليلها خلال الفترة (2000–2025) كلا من PubMed و Embaseو Scopusو Web of Science، باستخدام مصطلحات بحث تتعلق بسرطان البروستاتا و BPHوعلامات IHC بما في ذلك AMACR و ERGو PSAو NKX3.1 و p63.تم استبعاد الدراسات غير المعتمدة على الكيمياء المناعية النسيجية وكذلك الدراسات غير البشرية.
النتائج: أظهرت الواسمات الإيجابية الرئيسة مثل(AMACR (P504S ، وERG، وPSA، وPSAP، و(Prostein (P501S، و NKX3.1 مستويات متباينة من الحساسية والنوعية، مما يدعم تأكيد تكاثر الغدد الخبيث. أما واسمات الخلايا القاعدية مثل HMWCK (34βE12) و p63فهي أساسية لاستبعاد السرطان من خلال إظهار سلامة الطبقة القاعدية. بالإضافة إلى ذلك، توفر الواسمات الإنذارية مثلKi-67 ، وp53، وPTEN، و MYCرؤى إضافية حول شراسة الورم والنتائج السريرية المتوقعة. كما تحسن الاستعانة بالنهج متعدد الواسمات من موثوقية التشخيص وتساعد على التمييز بين سرطان البروستاتا وغيره من الحالات المشابهة، مثل الأورام داخل الظهارية البروستاتية عالية الدرجة، وفرط التنسج الغدي غير النمطي، وسرطان الخلايا البولية، والسرطان الغدي القولوني.
الاستنتاج: يعزز علم المناعة النسيجية بشكل كبير دقة التشخيص، وتقييم الإنذار، واتخاذ القرارات العلاجية في أمراض البروستاتا. ويتطلب الحصول على نتائج مثالية تطبيق ضوابط صارمة للجودة، وتوحيدا منهجيا، وتفسيرا من قبل أخصائي علم الأمراض. إن دمج الواسمات الجزيئية مع التقييم المورفولوجي يسهم في تحسين رعاية المرضى ونتائجهم السريرية.
مفتاح الكلمات :واسمات الخلايا القاعدية؛ علم المناعة النسيجية؛ الواسمات الإنذارية؛ سرطان البروستاتا؛ الواسمات النسيجية.