International Journal of Clinical and Diagnostic Pathology

ISSN (P): 2617-7226
ISSN (E): 2617-7234
www.patholjournal.com
2024; 7(3): 271-275
Received: 05-07-2024
Accepted: 10-08-2024

Zahraa Tariq Abdulameer
Babylon Health Directorate, Babylon, Iraq

Ali Zaki Al-Asadi
College of Medicine, Babylon University, Babylon, Iraq

Corresponding Author:
Zahraa Tariq Abdulameer
Babylon Health Directorate, Babylon, Iraq

Evaluation of clinicopathological significance of ALDH1a1 expression in prostatic tumors

Zahraa Tariq Abdulameer and Ali Zaki Al-Asadi

DOI: https://doi.org/10.33545/pathol.2024.v7.i3d.612

Abstract

Background: Prostate cancer, the second most common solid organ malignancy among men, is primarily adenocarcinoma. Core needle biopsy is the standard method for non-operative diagnosis. While early-stage prostate carcinoma can be treated with radical prostatectomy or radiotherapy, advanced-stage metastatic prostate carcinoma has high mortality. Prostate cancer stem cells (CSCs), located in the basal cell layer, contribute to cancer progression. Aldehyde Dehydrogenase-1, family member A1 (ALDH1a1), a stem cell marker, plays a central role in tumor biology by protecting CSCs from oxidative injury and promoting proliferation.

Methods: This retrospective cross-sectional study, conducted at the Babylon Training Center for Pathology in Al-Hilla Teaching Hospital from January 2023 to January 2024, analyzed 50 cases of prostatic adenocarcinoma and 10 cases of benign prostatic hyperplasia using ALDH1a1 immunohistochemical staining.

Results: ALDH1a1 expression was negative in benign prostatic hyperplasia but positive in 70% of prostatic adenocarcinoma cases. A significant correlation was found between ALDH1a1 expression and Gleason grades, with higher H-scores in advanced grades ($p = 0.01$). ALDH1a1 H-scores also significantly correlated with pre-operative PSA levels and patient age ($p = 0.03$ and $p = 0.001$, respectively).

Conclusion: ALDH1a1 expression increases with higher Gleason grades, PSA levels, and patient age, indicating tumor aggressiveness. Therefore, ALDH1a1 can be used as a prognostic and predictive marker for prostate cancer.

Keywords: Evaluation, clinicopathological, ALDH1a1, expression, prostatic tumors

Introduction

Prostate cancer (PCa) is the second most common cancer worldwide and the fifth leading cause of death in men [1]. Its incidence increases with age and occurs primarily in African American men; the incidence is higher than in white men [2, 3]. Like many other types of cancer, prostate cancer tends to run in families. 5-10% of prostate cancer cases are described as familial cancers due to inherited genetic risk factors [4]. Most cases of prostate adenocarcinoma are asymptomatic and are diagnosed during PSA screening, but approximately 15% of patients have normal PSA values [5]. Most prostate adenocarcinomas are composed of acini arranged in one or more patterns. The diagnosis depends on a combination of architectural and cytological features. Light microscopic features are usually sufficient for diagnosis, but in rare cases, immunohistochemical studies may be useful. The Gleason score is recommended for routine use in staging and is one of the strongest predictors of the biological behavior of prostate cancer [6]. Benign prostatic hyperplasia is one of the most common urological diseases, characterized by proliferation of prostate stromal and epithelial cells in the periurethral transition zone, and its frequency increases with age. It often leads to lower urinary tract symptoms (LUTS, including urgency, frequency, nocturia, incomplete micturition, and weak urine stream) [7, 8]. This study aimed to assess the expression changes of ALDH1a1 (intensity and percentage) in prostate adenocarcinoma of different grades and benign prostatic hyperplasia, how it can reflect the prognostic and predictive biomarker roles.

Methods: This retrospective cross-sectional study was conducted at the Babylon Training Center for Pathology in Al-Hilla Teaching Hospital from January 2023 to January 2024 to determine ALDH1a1 expression in prostatic adenocarcinoma and benign prostatic hyperplasia (BPH).

The study involved 50 cases of prostatic adenocarcinoma and 10 cases of BPH. Tissue blocks were obtained from the histopathology departments at Al-Hilla Teaching Hospital and the private sector in Babylon, with samples acquired via TURP, transrectal ultrasound-guided core biopsy, and transperineal routes. All samples were formalin-fixed, paraffin-embedded, and re-stained with H&E to verify the morphological diagnosis and grading [9]. Inclusion criteria encompassed cases of prostatic adenocarcinoma with various Gleason grades and cases of benign prostatic hyperplasia. Exclusion criteria included prostate intraepithelial neoplasia, mesenchymal tumors, and blocks with inadequate tissue samples. The immunohistochemical staining procedure utilized ALDH1a1, a rabbit monoclonal antibody. Five µm sections were deparaffinized, rehydrated, and subjected to heat-induced epitope retrieval using Bio SB Immuno DNA Retriever with Citrate solution. Slides were then processed with Polydetector Peroxidase Blocker, incubated with the primary antibody, and stained with PolyDetector DAB Chromogen solution. Positive controls, including normal liver tissue known to express ALDH1a1, and negative controls were included in each staining run [10]. ALDH1a1 immunoreactivity was evaluated based on a Histochemical Score (H-score), calculated by multiplying the intensity of staining by the percentage of stained cells, resulting in a score between 0 and 300. Specimens with H-scores of 33 or less were categorized as low-expressing, while those with scores above 33 were classified as high-expressing [11]. Statistical analysis was conducted using SPSS v26, with chi-square tests applied to evaluate correlations. The results were considered statistically significant if the p-value was ≤ 0.05. This study's methodology and rigorous quality control, including both positive and negative controls, ensured reliable and reproducible results that demonstrated significant associations between ALDH1a1 expression and the histopathological grading of prostatic adenocarcinoma.

Results

In this study, 60 cases were collected, comprising 10 cases of benign prostatic hyperplasia (BPH) and 50 cases of prostatic adenocarcinoma, which were distributed across different Gleason grades. The distribution of adenocarcinoma cases was as follows: 8 cases (16.0%) in grade I, 7 cases (14.0%) in grade II, 8 cases (16.0%) in grade III, 12 cases (24.0%) in grade IV, and 15 cases (30.0%) in grade V. ALDH1a1 immunohistochemical staining was negative in the epithelial cells of all BPH cases. In contrast, ALDH1a1 expression was positive in the epithelial cells (nucleus and cytoplasm) of 35 out of 50 prostatic adenocarcinoma cases (70.0%), while 15 cases (30.0%) were negative. The distribution of ALDH1a1 positivity across Gleason grades was as follows: 3 out of 8 cases (37.5%) in grade I, 3 out of 7 cases (42.9%) in grade II, 5 out of 8 cases (62.5%) in grade III, 10 out of 12 cases (83.3%) in grade IV, and 14 out of 15 cases (93.3%) in grade V. as in Figure 1.

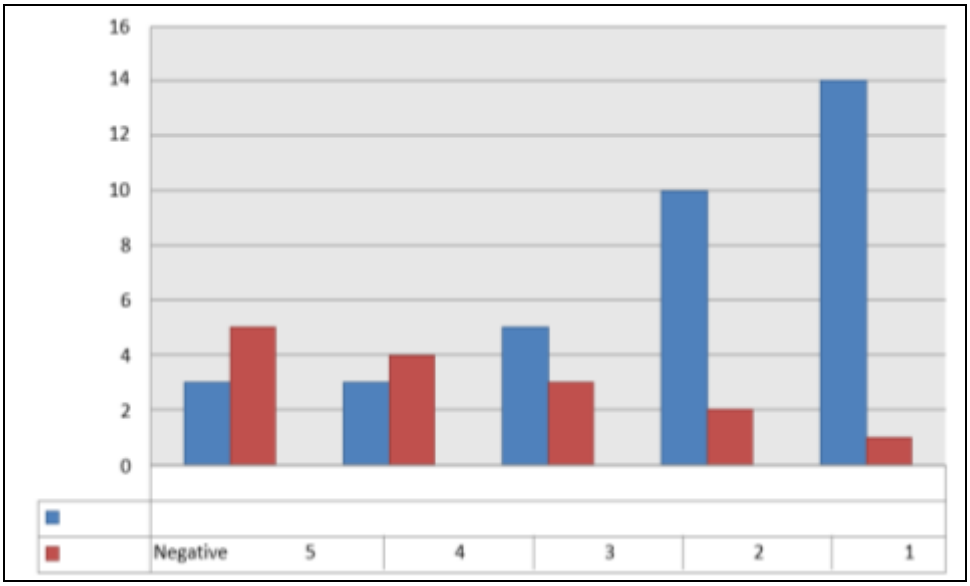

Fig 1: Gleason grades of prostate adenocarcinoma in response to immunostaining of ALDH1a1.

As histochemical (H-score) is the dependent score in our study to determine ALDH1a1 expression which calculated from multiplying the intensity by the percentage of ALDH1a1 immunostaining of cells in order to categorize cases as low H-score and high H-score by using the Cutoff value of 33. In cases of prostate adenocarcinoma, intensity of ALDH1a1 immunostaining was significantly correlated with Gleason grades as shown in table (1). (P Value=0.04).

Table 1: Significant correlation between Gleason grade and intensity of ALDH1a1 staining p-value=0.04 (significant)

Grade	No staining	Faint staining	Moderate staining	Strong staining	Total
Grade I	5(62.50%)	1(12.50%)	1(12.50%)	1(12.50%)	8(16.0%)
Grade II	4(57.15%)	1(14.28%)	1(14.28%)	1(14.29%)	7(14.0%)
Grade III	3(37.50%)	1(12.50%)	3(37.50%)	1(12.50%)	8(16.0%)
Grade IV	2(16.67%)	1(8.33%)	3(25.0%)	6(50.0%)	12(24.0%)
Grade V	1(6.67%)	1(6.67%)	1(6.66%)	12(80.0%)	15(30.0%)
Total	15(30.0%)	5(10.0%)	9(18.0%)	21(42.0%)	50(100%)

Also percentage of ALDH1a1 immunostaining of tumor cells was significantly correlated with Gleason grades of prostate adenocarcinoma as shown in Table 2, (P Value=0.04).

Table 2: Significant correlation between Gleason grades of prostate adenocarcinoma and the percentage of ALDH1a1 staining. (P value 0.04 Significant)

Grade	< 25% percentage	25-50% percentage	50-75% percentage	>75% percentage	Total
Grade 1	5(62.50%)	1(12.50%)	1(12.50%)	1(12.50%)	8(16.0%)
Grade 2	4(57.15%)	1(14.28%)	2(14.28%)	1(14.29%)	7(14.0%)
Grade 3	3(37.5%)	1(12.50%)	3(37.50%)	1(12.50%)	8(16.0%)
Grade 4	2(16.67%)	1(8.33%)	3(25.0%)	6(50.0%)	12(24.0%)
Grade 5	1(6.67%)	1(6.67%)	1(6.66%)	12(80.0%)	15(30.0%)
Total	15(30.0%)	5(10.0%)	9(18.0%)	21(42.0%)	50(100%)

According to results of intensity and percentage of ALDH1a1 immunostaining of tumor cells, significant correlation was present between ALDH1a1 H-score and Gleason grades of prostate adenocarcinoma (P-value=0.01) as shown in Table 3 which illustrate that high H-score present in 31 (62.0%) cases and low H-score present in 19 (38.0%) of cases.

Table 3: Significant correlation between ALDH1a1 H-score and Gleason grades of prostate adenocarcinoma (P-value =0.01significant)

Grades	Low H score < or =33	High H score >33	Total
Grade I	6(75.0%)	2(25.0%)	8(16.0%)
Grade II	5(71.43%)	2(28.57%)	7(14.0%)
Grade III	3(37.5%)	5(62.5%)	8(16.0%)
Grade IV	3(25.0%)	9(75.0%)	12(24.0%)
Grade V	2(13.33%)	13(86.67%)	15(30.0%)
Total	19(38.0%)	31(62.0%)	50(100%)

In benign prostatic hyperplasia ALDH1a1 immunostaining was negative in epithelial cell of all 10 cases (0% intensity and 0% percentage) and staining restricted to basal layer only. Significant statistical correlation in ALDH1a1 H-score were presented between cases of prostatic adenocarcinoma and cases of benign prostatic hyperplasia as shown in Table 4. All cases of BPH present with low H-score, while 31(62.0%) cases of prostatic adenocarcinoma present with high H-score and 19(38.0%) cases of them present with low H-score.

Table 4: Significant correlation in the ALDH1a1 H-score between prostate adenocarcinoma and benign prostate hyperplasia (p-value=0.002) significant

H-score	Prostate adenocarcinoma	Benign prostate hyperplasia	Total
Low H-score < or = 33	19(38.0%)	10(100%)	29(48.33%)
High H-score > 33	31(62.0%)	0 (0.00 %)	31(51.67%)
Total	50(83.33%)	10(16.67%)	60(100%)

The collected results of PSA levels correlate significantly with Gleason grade of prostate adenocarcinoma (p-value 0.02) as shown in table 5.

Table 5: Significant statistical correlation between PSA levels and Gleason grade of prostatic adenocarcinoma p value 0.02

PSA level	Grade I	Grade II	Grade III	Grade IV	Grade V	Total
4-10 ng/ml	6(75.0%)	4(57.10%)	3(37.5%)	2(20.0%)	1(9.10%)	16(36.4%)
>10 ng/ml	2(25.0%)	3(42.90%)	5(62.5%)	8(80.0%)	10(90.9%)	28(63.6%)
Total	8(18.2%)	7(15.90%)	8(18.2%)	10(22.7%)	11(25.0%)	44(100%)

According to our presented study of ALDH1a1 immunostaining, there was significant correlation in the ALDH1a1expression (H-score) and prostate specific antigen levels as illustrated in the Table 6 which explained that cases with higher levels of PSA associated with higher ALDH1a1 expression. < 4 ng/ml PSA level which include 6 cases of BPH, all of them presented with low H-score, while 4-10 ng/ml PSA level, high H-score was equal to 11(68.75%) and low H-score equal to 5(31.25%) of cases. PSA levels >10 ng/ml, high H-score was equal to 20(71.4%) and low H-score was equal to 8(28.6%) of cases.

Table 6: Significant statistical correlation between ALDH1a1 H-score and PSA levels p value 0.03.

PSA levels	Low H-score	High H-score	Total
< 4ng/ml	6(100%)	0	6(12.0%)
4-10 ng/ml	5(31.25%)	11(68.75%)	16(32.0%)
>10 ng/ml	8(28.6%)	20(71.4%)	28(56.0%)
Total	19(38.0%)	31(62.0%)	50(100%)

Age of patients with prostatic adenocarcinoma significantly correlated with Gleason grade p value 0.03 as shown in Table 7.

Table 7: Significant statistical correlation between age groups and Gleason grade of prostatic adenocarcinoma p value 0.03.

Age	Grade I	Grade II	Grade III	Grade IV	Grade V	Total
<65 yr	6(75.0%)	4(57.1%)	3(37.5%)	3(25.0%)	2(13.3%)	18(36.0%)
>or=65yr	2(25.0%)	3(42.9%)	5(62.5%)	9(75.0%)	13(86.7%)	32(64.0%)
Total	8(16.0%)	7(14.0%)	8(16.0%)	12(24.0%)	15(30.0%)	50(100%)

According to our presented study of ALDH1a1 immunostaining, age was significantly correlated with ALDH1a1 expression(H-score) as shown in Table 8 which explained that high H-score in age group of < 65 year equal to 7(28.0%), while in age group of > or = to 65 years was equal to 24(68.6%).

Table 8: Significant statistical correlation between age groups and ALDH1a1 H- score p value 0.001

Age	Low H-score	High H-score	Total
< 65 year	18(72.0%)	7(28.0%)	25(41.67%)
>or =65 year	11(31.4%)	24(68.6%)	35(58.33%)
Total	29(48.33%)	31(51.67%)	60(100%)

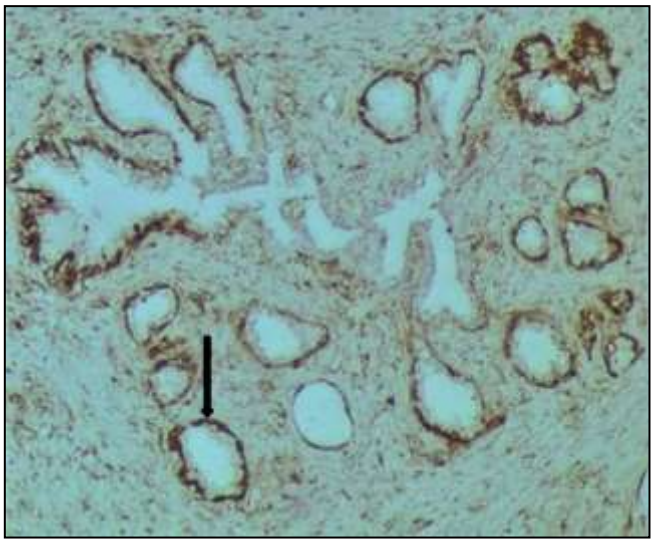

Fig 2: Section of benign prostatic hyperplasia after ALDH1a1immunostaining, show only basal layer staining with no epithelial cells staining, 100.

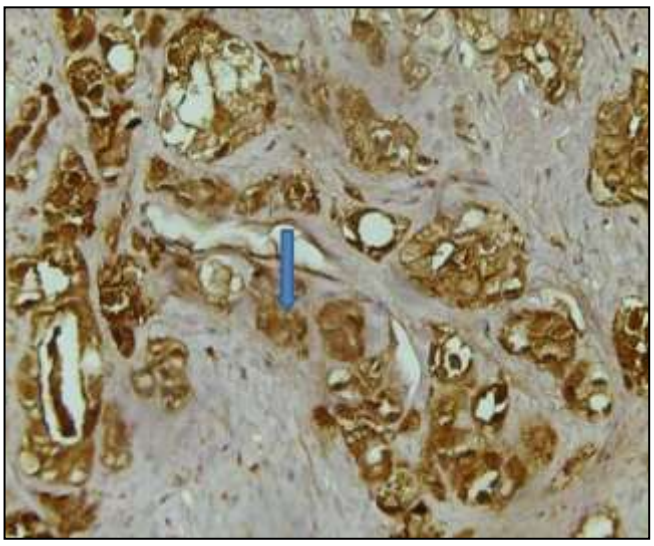

Fig 3: Prostate adenocarcinoma (Gleason score 4+5=9 grade V of infiltrative cribriform glands and solid nest with single file cell infiltration) after staining with ALDH1a1, show strong intensity staining (3+) of epithelial cells (nucleus and cytoplasm), 400.

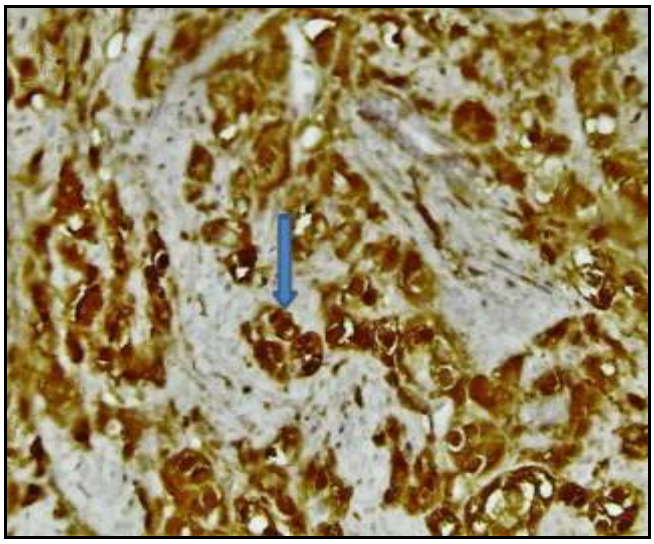

Fig 4: Prostate adenocarcinoma grade V, Gleason pattern 4+5=9 of poorly formed gland and cribriform with single file cell infiltration, after ALDH1a1 staining show strong intensity (3+) of nuclear and cytoplasm of epithelial cells,400

Discussion

This retrospective study analyzed 50 cases of prostatic adenocarcinoma across various Gleason grades and 10 cases of benign prostatic hyperplasia (BPH) to evaluate the clinic histopathological expression of ALDH1a1, focusing on intensity and percentage. ALDH1a1 staining was positive in 35 (70.0%) of the adenocarcinoma cases and negative in 15 (30.0%) cases. Specifically, ALDH1a1 positivity was observed in 3 cases (37.5%) of grade I, 3 cases (42.85%) of grade II, 5 cases (62.5%) of grade III, 10 cases (83.33%) of grade IV, and 14 cases (93.33%) of grade V. The H-score, which reflects both the intensity and percentage of staining, showed a significant correlation with Gleason grades (P-value = 0.01), with higher H-scores more frequently observed in higher grades. The results align with previous studies, including one by Sara G. Masoud *et al.* [10] from Egypt, which reported that 72.7% of adenocarcinoma cases had a high H-score, with a significant correlation between ALDH1a1 expression and Gleason grades (P-value < 0.01). Similarly, Kalantari *et al.* [11] from Iran found that higher Gleason scores were associated with increased ALDH1a1 immunoreactivity (P = 0.01 for intensity, and P = 0.05 for H-score). However, other studies, such as those by Dhafer A. Alghezi *et al.* [12] from Iraq and Matsika *et al.* [13] from Cleveland, did not find a significant correlation between ALDH1a1 expression and Gleason grades, highlighting differences in methodologies, such as the use of Gleason scores rather than the grade group system. In BPH cases, ALDH1a1 expression was negative in the epithelial cells of all cases, with staining restricted to the basal layer. All BPH cases had a low H-score, while 38.0% of adenocarcinoma cases had a low H-score and 62.0% had a high H-score, with a significant correlation (P-value = 0.002). These findings are consistent with previous studies, such as those by Masoud *et al.* [10] and Kalantari *et al.* [11], which also

reported low ALDH1a1 expression in BPH and significant differences in expression between BPH and adenocarcinoma cases. The study also found a significant correlation between prostate-specific antigen (PSA) levels and Gleason grades (P-value = 0.03). Higher PSA levels were associated with higher Gleason grades, consistent with the findings of Masoud *et al*. [10] and Rani *et al*. [14]. However, Gurumurthy *et al*. [15] found no significant correlation between PSA levels and Gleason grades, suggesting that poorly differentiated tumors may release lower amounts of PSA. ALDH1a1 expression was also significantly correlated with PSA levels (P-value = 0.01), with higher expression observed in cases with higher PSA levels. This result aligns with the findings of Masoud *et al*. and Kalantari *et al*., who also reported significant correlations between PSA levels and ALDH1a1 expression [10, 11].

The study further identified a significant correlation between patient age and Gleason grades (P-value = 0.03), with older patients more likely to have higher Gleason grades. This finding is supported by studies from Masoud *et al*. [10] and Pietro Pepe *et al*. [16], which found that Gleason grade progressively increased with age. Additionally, age was significantly correlated with ALDH1a1 expression (P-value = 0.001), with higher H-scores more common in older patients. However, this correlation was not observed in the studies by Masoud *et al*. and Kalantari *et al*. possibly due to differences in sample sizes and the distribution of high-grade adenocarcinoma cases [10, 11]. Overall, this study demonstrates significant associations between ALDH1a1 expression, Gleason grades, PSA levels, and patient age, suggesting that ALDH1a1 could serve as a prognostic marker in prostatic adenocarcinoma.

Conclusion

Immunohistochemical staining for ALDH1a1 in cases of benign prostatic hyperplasia was negative in epithelial cells in all cases and all cases showed a low H score, whereas ALDH1a1 expression (H score) was associated with upgrading to prostate adenocarcinoma cases with higher PSA levels and with upgrading to prostate cancer cases. It increased with increasing patient age. ALDH1a1 expression indicates the degree of tumor aggressiveness (higher expression, more aggressive tumor) and therefore can be used as a prognostic and predictive marker.

Conflict of Interest

Not available

Financial Support

Not available

References

1. Sung H, Ferlay J, Siegel RL, *et al*. Global cancer statistics 2020: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. CA Cancer J Clin. 2021 May;71(3):209-249.
2. Ferlay J, Ervik M, Lam F, *et al*. Global cancer observatory: cancer today. Lyon, France: International Agency for Research on Cancer; 2018 Nov 5. Available from: https://gco.iarc.fr/today
3. Rawla P. Epidemiology of prostate cancer. World J Oncol. 2019 Apr;10(2):63.
4. Al-Badran RA, Al-Badran AI, Azkar M, Al-Mansouri L. Association between some risk factors and prostate cancer progression in Basrah, Iraq. Basrah J Vet Res. 2020;19(2).
5. Hori S, Blanchet JS, McLoughlin J. From prostate-specific antigen (PSA) to precursor PSA (proPSA) isoforms: a review of the emerging role of proPSAs in the detection and management of early prostate cancer. BJU Int. 2013 Oct;112(6):717-728.
6. Epstein JI, Zelefsky MJ, Sjoberg DD, *et al*. A contemporary prostate cancer grading system: a validated alternative to the Gleason score. Eur Urol. 2016 Mar;69(3):428-435.
7. Batai K, Phung M, Bell R, *et al*. Correlation between body mass index and prostate volume in benign prostatic hyperplasia patients undergoing holmium enucleation of the prostate surgery. BMC Urol. 2021 Jun 10;21(1):88.
8. Kim EH, Larson JA, Andriole GL. Management of benign prostatic hyperplasia. Annu Rev Med. 2016 Jan 14;67:137-151.
9. Ram S, Vizcarra P, Whalen P, *et al*. Pixel wise H-score: a novel digital image analysis-based metric to quantify membrane biomarker expression from immunohistochemistry images. PLoS One. 2021 Sep 27;16(9).
10. Masoud SG, Bakr M, Nasif S. The significance of aldehyde dehydrogenase 1A1 expression in benign prostatic hyperplasia, prostatic intraepithelial neoplasia and prostatic adenocarcinoma. Benha Med J. 2020 May 1;37(1):207-219.
11. Kalantari E, Saadi FH, Asgari M, *et al*. Increased expression of ALDH1A1 in prostate cancer is correlated with tumor aggressiveness: a tissue microarray study of Iranian patients. Appl Immunohistochem Mol Morphol. 2017 Sep;25(8):592-598.
12. Alghezi DA, Whitley P, Beresford M, *et al*. ALDH1A1 expression in benign and malignant prostate tissues. Thi-Qar Med J. 2021;21(1).
13. Matsika A, Srinivasan B, Day C, *et al*. Cancer stem cell markers in prostate cancer: an immunohistochemical study of ALDH1, SOX2 and EZH2. Pathology. 2015 Dec;47(7):622-628.
14. Rani E, Nibhoria S, Nagpal N. Outlook of Gleason score in prostate carcinoma and correlation with PSA levels: a study in a tertiary care hospital. J Cancer Res Ther. 2023 Jul 19.
15. Gurumurthy D, Maggad R, Patel S. Prostate carcinoma: correlation of histopathology with serum prostate specific antigen. Sci J Clin Med. 2015 Jul;4(4):1-5.
16. Pepe P, Pennisi M. Gleason score stratification according to age at diagnosis in 1028 men. Contemp Oncol (Pozn). 2015;19(6).

How to Cite This Article

Abdulameer ZT, Al-Asadi AZ. Evaluation of clinicopathological significance of ALDH1a1 expression in prostatic tumors. International Journal of Clinical and Diagnostic Pathology. 2024;7(3):271-275.