Relationship between AMACR (Alpha Methyl Acyl CoA Racemase/P504S) Staining Intensity and Histopathological Gleason Grading of Prostatic Adenocarcinoma

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Abstract

Background: Prostate cancer is the fourth most commonly occurring cancer overall and fifth leading cause of cancer related death among male worldwide. Gleason grading system composed of 5 prognostic grade groups based on glandular architecture and microscopic appearance of prostatic adenocarcinoma. Sometimes, for confirmation of prostatic cancer specific single histological feature is not adequate. AMACR is a positive diagnostic marker specific for prostatic adenocarcinoma. This study focuses on AMACR expression in prostatic tumors and evaluate its correlation with histopathological grading.

Methods: Twenty-Four prostatic needle biopsies and 26 transurethral resection of prostate specimens were included in this cross-sectional study conducted in Sir Salimullah Medical College from January, 2019 to December, 2020. Specimens were processed routinely for Haematoxylin and Eosin followed by immunohistochemistry for AMACR. Histopathological and immunohistochemical results were analyzed. Statistical analysis was done using Statistical Package for Social Sciences version 22.

Results: Among the 50 histopathologically diagnosed cases 20 were diagnosed as prostatic adenocarcinoma, 2 were diagnosed as suspicious for malignancy and 28 were diagnosed as nodular hyperplasia of prostate. All 28 cases of NHP showed negative AMACR expression. Among the 20 prostatic adenocarcinoma cases, all showed positive AMACR expression in various intensity. Between the 02 suspicious for malignancy cases, one showed positive expression for AMACR. This case was later confirmed as HGPIN. Another case was negative for AMACR and turned out equivocal later. AMACR showed statistically significant value ($p<0.05$) in prostatic adenocarcinoma. It has been shown that there is no relation between AMACR staining intensity in adenocarcinoma cases with histopathological grading which was also proved by statistically nonsignificant ($p>0.05$) value.

Conclusion: Histopathology is the gold standard for diagnosing prostatic cancer. AMACR helps in confirming the diagnosis of prostatic adenocarcinoma. Gleason score has no correlation with the intensity of AMACR staining.

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Keywords: AMACR- Alpha Methyl Acyl Coenzyme-A Racemase, PAC- Prostatic adenocarcinoma, NHP- Nodular Hyperplasia of Prostate, IHC- Immunohistochemistry.

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Introduction

AMACR (Alpha Methyl Acyl CoA Racemase/P504S) is a mitochondrial and peroxisomal enzyme protein involved in beta-oxidation of dietary branched-chain fatty acids and fatty acid derivatives (including bile acid intermediates), that has been shown to be increased at both the mRNA and protein levels in prostatic adenocarcinoma. It positively stains prostatic adenocarcinoma.[1] AMACR gives negative expression in normal prostatic tissue and nodular hyperplasia of prostate.[2]

Prostate cancer is the second most commonly occurring cancer in male worldwide. In Bangladesh, 1.5% new cases of prostatic malignancy were diagnosed in 2018. Death rate was 1.6%. Five years prevalence rate of all ages of prostate cancer in Bangladesh was 4.63 in 2018.[3]

Prostatic carcinomas can be divided into two major histologic categories: acinar and ductal. The large majority of the tumors are acinar.[4] In 1966, Dr. Donald Gleason[5] devised grades of 1 - 5, based on glandular architecture and microscopic appearance, that were shown to predict outcome in prostate cancer. In 2014, the ISUP and World Health Organization adopted a grading system composed of 5 prognostic grade groups started from grade group 1 comprising (3+3 = 6).[6] Gleason grades 1 and 2 were no longer recommended for use, since those patterns of cancer have an outcome not different from grade 3.[7]

There is no association with AMACR staining intensity and histopathological Gleason grading of prostatic adenocarcinoma.[8]

In this study expression of AMACR in prostatic lesions, evaluation of its diagnostic implication and association with histopathological grading has been done.

Methods

Histopathologically diagnosed cases of prostatic benign, malignant and suspicious for malignancy lesions of (≥50) years male patients were included in this cross-sectional study conducted in Sir Salimullah Medical College from January, 2019 to December, 2020. Twenty-Four prostatic needle biopsies and 26 transurethral resection of prostate specimens were included. Sampling technique was purposive and convenient. Specimens were processed routinely for Haematoxylin and Eosin stain and analyzed under microscope. Carcinoma cases were histologically graded as Gleason scoring system and 2014 WHO/ISUP consensus conference criteria for grading of prostate cancer. Immunohistochemistry for AMACR were performed and results were analyzed in Square Hospital, Dhaka. Interpretation, statistical analysis and evaluation and correlation with histopathological grading were done.

Interpretation of Immunohistochemistry

AMACR: Positive staining for AMACR pertained to dark diffuse or granular, cytoplasmic or luminal, but circumferential. The percentage of positivity were graded from 0+ to 3+ as follows: 0% cells (o+, negative), 1-10% cells (1+, mild), 11-50% cells (2+, moderate), >50% cells (3+, strong) (Rathod et al., 2019).[9] The adjacent benign glands did not show any staining for AMACR. Negative staining pertained to no staining or focal, weak noncircumferential fine granular staining.

Results

This cross-sectional study was conducted on 50 histopathologically diagnosed prostatic tumor patients. Among them 20 (40.0%) cases were diagnosed as prostatic adenocarcinoma, 28 (56.0%) cases nodular hyperplasia of prostate (NHP) and 2 (4.0%) cases were diagnosed as suspicious for

malignancy. According to modified Gleason grade group it was observed that majority cases 9 (45%) belonged to grade group 4, 6 (30.0%) cases belonged to grade group 1 and 5 (25.0%) cases belonged to grade group 3 (Table I).

Table I: Distribution of the cases of histopathologically diagnosed prostatic adenocarcinoma according to 2014 WHO/ISUP modified Gleason grade group (n=20)

Modified Gleason grade Group	Number	%
Grade group 1	6	30.0
Grade group 2	0	0.0
Grade group 3	5	25.0
Grade group 4	9	45.0
Grade group 5	0	0.0

Table II: Association of AMACR expression with different histopathological diagnosis of prostatic lesions (n=50)

AMACR expression	Histopathological diagnosis						*p* value
	NHP (n=28)		Adenocarcinoma (n=20)		Suspicious (n=2)		
	n	%	n	%	n	%	
Negative	28	100.0	0	0.0	1	50.0	0.001[s*]
Mildy positive	0	0.0	6	30.0	0	0.0	
Moderately positive	0	0.0	10	50.0	1	50.0	
Strongly Positive	0	0.0	4	20.0	0	0.0	
Total	28	100	20	100	2	100	

s=significant*
p value reached from Chi-square test

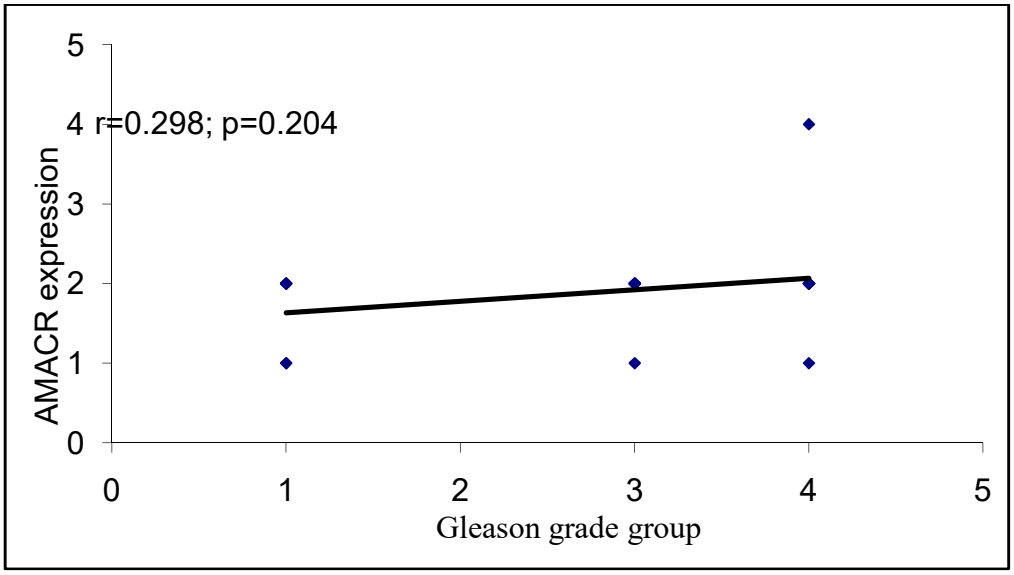

Figure 1. Scatter diagram showing a shaky linear Spearman's rank correlation between Gleason grade group and AMACR expression in adenocarcinoma cases (n=20).

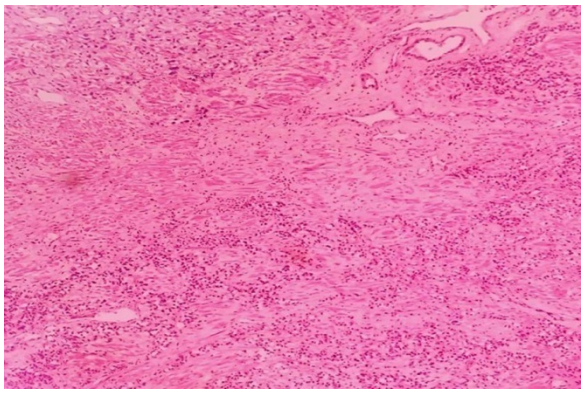

Figure 2. Prostatic adenocarcinoma, Gleason score (4+4) = 8/10, grade group 4 (Case no. 4, H&E, 40x)

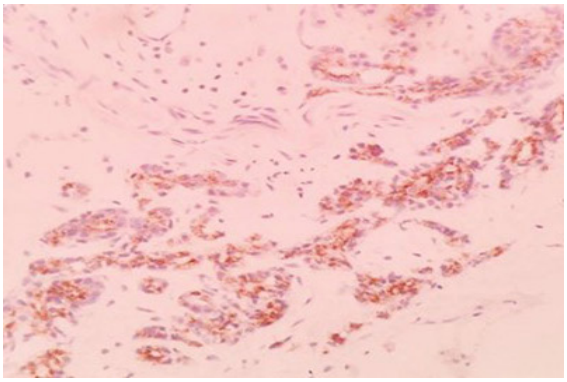

Figure 5. Moderate AMACR expression in PAC, Gleason score (4+3) =7/10, grade group 3 (Case no. 43, 40x)

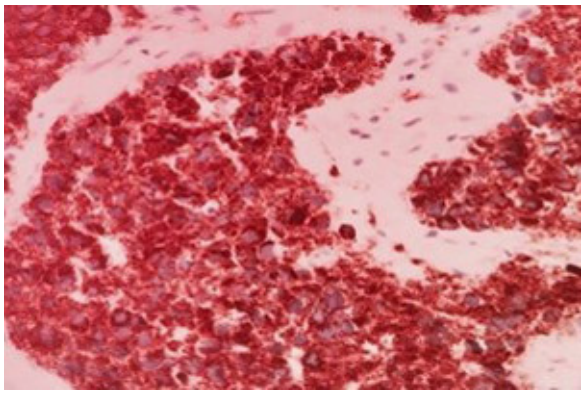

Figure 3. Strong AMACR expression in PAC, Gleason score (4+4) = 8/10, grade group 4 (Case no. 4, 40x)

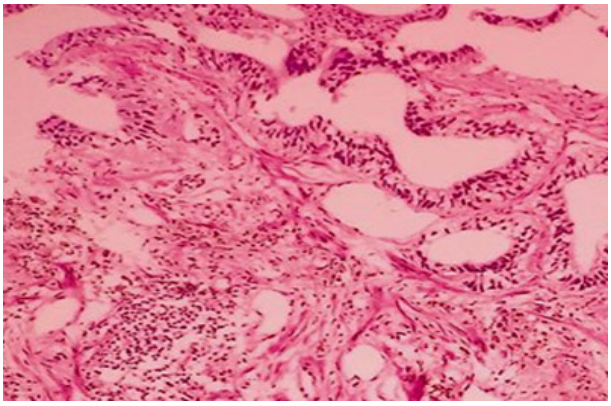

Figure 6. Prostatic adenocarcinoma, Gleason score (4+3) = 7/10, grade group 3 (Case no. 8, H&E, 40x)

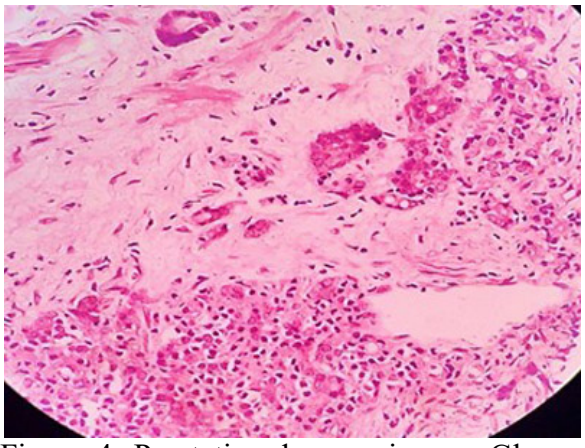

Figure 4. Prostatic adenocarcinoma, Gleason score (4+3) = 7/10, grade group 3 (Case no. 43, H&E, 40x)

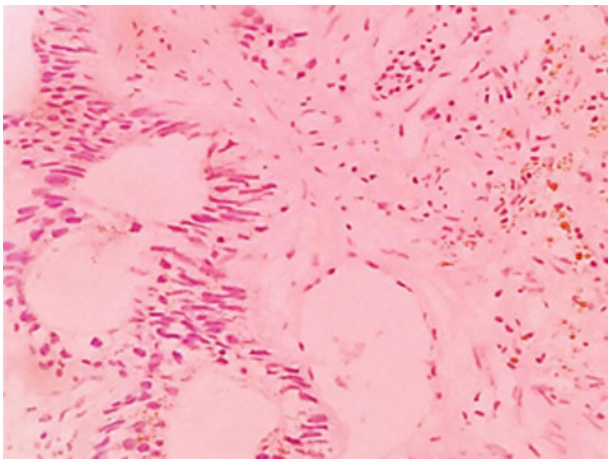

Figure7. Mild AMACR expression in PAC, Gleason score (4+3) = 7/10, grade group 3 (Case no. 8, 40x)

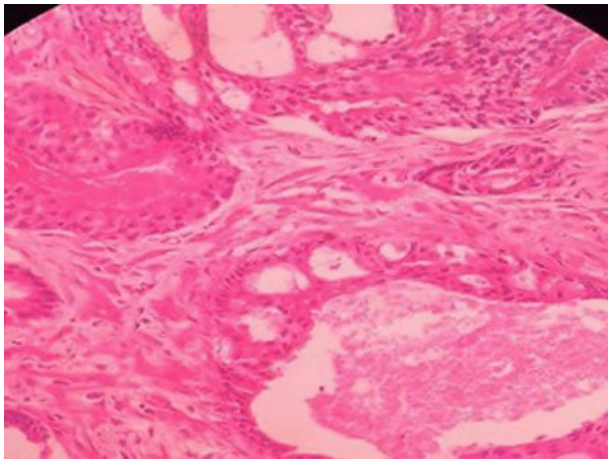

Figure 8. Prostatic adenocarcinoma, Gleason score (3+3) = 6/10, grade group 1 (Case no. 5, H&E, 40x)

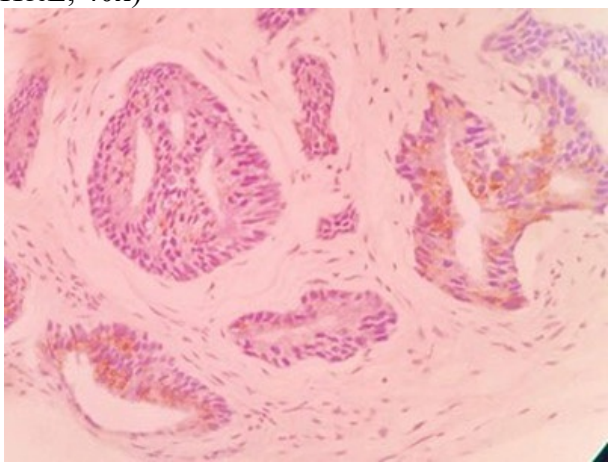

Figure 9. Moderate AMACR expression in PAC, Gleason score (3+3) = 6/10, grade group 1 (Case no. 5, 40x)

Among 50 cases 28(100%) histopathologically diagnosed NHP cases became negative and 20(100%) adenocarcinoma cases became positive on AMACR expression. Among the suspicious for malignancy case 1(50%) gave negative expression and another 1(50%) gave moderately positive expression. The p value reached from Chi-square test and the difference was statistically significant ($p<0.05$) (Table II).

A Scatter diagram showed a shaky linear Spearman's rank correlation between Gleason grade group and AMACR expression in adenocarcinoma cases. Here, $r = 0.298$ indicates a minimal, weak relationship and $p = 0.204$ indicates the difference was not statistically significant ($p>0.05$) (Figure 1).

Discussion

This cross-sectional study was carried out with an aim to determine the expression of AMACR to observe the diagnostic role of this immune marker in patients with prostatic tumors for confirmation of malignancy and to evaluate its correlation with histopathological grading.

A total of 50 male patients of (≥50) years with prostatic tumor diagnosed histopathologically in the pathology departments of Sir Salimullah Medical College and National Institute of Kidney Disease and Urology; Dhaka during 1stJanuary, 2019 to 31st December, 2020, were included in this study. The present study findings were discussed and compared with previously published relevant studies.

Twenty-Four (48.0%) needle biopsies and 26 (52%) TURP specimens were included in this study. Among all the cases 20 (40%) were diagnosed as prostatic adenocarcinoma, 28 (56%) as NHP and 02 (4%) as suspicious for malignancy histopathologically.

The prostatic adenocarcinoma cases were graded according to the modified Gleason scoring system. According to 2014 WHO/ISUP consensus conference criteria for grading of prostatic cancer, grade group 1, grade group 2, grade group 3, grade group 4 and grade group 5 were 06 (30.0%), 00 (0%), 05 (25.0%), 09 (45.0%) and 00 (0%) respectively. In the present study, most common pattern of Gleason grade group in case of adenocarcinoma was Grade group 4 (Gleason score 4+4 = 8) containing 9 (45%) cases (Table I).

Immunohistochemical staining with AMACR were done in all the prostatic adenocarcinoma cases.

In case of AMACR expression it was observed that among 50 cases 28 (100%) NHP cases became negative and 20 (100%) adenocarcinoma cases became positive in various intensity. Among the suspicious for malignancy cases 1 (50%) gave negative expression and another 1 (50%) gave positive expression. The p value reached from Chi-square test and there was a statistical significant difference ($p<0.05$) between AMACR expression and histopathological diagnosis in the current study (Table II). This study is in concordance with Jiang et al., (2001)[10] who found 100% positive staining of prostatic carcinoma cases and 88% negative staining for benign prostates after immunostaining by AMACR. They conclude that AMACR is a highly sensitive and specific positive marker for prostate carcinoma. Rathod et al., (2019)[9] in their study showed AMACR had a sensitivity of 90% and specificity of 100%.

A statistically significant correlation was not observed between histopathological grading of prostatic adenocarcinoma and AMACR expression ($p>0.05$). Here, $r=0.298$ indicates a minimal, weak relationship and $p = 0.204$ indicates the difference was not statistically significant ($p>0.05$) (Figure 1). It indicates that AMACR expression does not depend on tumor differentiation. This finding is similar to the study by Ozgur et al., (2013)[11] who found no significant relationship with tumor grade and AMACR expression pattern ($p>0.5$). Rubin et al., (2002)[8] and Beach et al., (2002)[12] in their separate study made similar observation and concluded that Gleason score has no correlation with the intensity of AMACR staining. Beach et al., (2002)[12] found 82% AMACR expression in prostate cancer whatever may be the degree of differentiation i.e Gleason score or morphological types.

Conclusion

AMACR is a specific and sensitive marker for prostatic adenocarcinoma. However, AMACR expression is not affected by Gleason score or histopathological Gleason grading of prostatic adenocarcinoma.

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