EXPRESSION OF VEGF IN PATIENTS OF URINARY BLADDER CARCINOMA

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ABSTRACT

BACKGROUND
Urinary bladder cancer is the 9th most common cancer worldwide and 13th most numerous cause of death from cancer. VEGF expression has been shown to be a prognostic marker and predicts the risk of recurrence and its high expression is related to lymph node metastasis. The study was undertaken to determine the expression of VEGF in urinary bladder tumours and its possible benefits in determining the prognosis of patients.

MATERIALS AND METHODS
50 cases of transitional cell carcinoma of urinary bladder diagnosed by histopathology were taken and expression of VEGF was observed on immunohistochemistry the results of staining intensity and percentage of positive cells were added and final scoring was done.

RESULTS
Expression of VEGF correlated with grade of tumor and was found to be statistically significant ($p < 0.05$). Maximum number of patients with high grade TCC showed a score of 3 for VEGF whereas in maximum patients with low grade TCC score of 1 for VEGF was obtained.

CONCLUSION
Expression of VEGF was found to be higher in patients with high grade TCC as compared to low grade TCC. Therefore, VEGF can be used as an important IHC marker to assess the utilization of antiangiogenic therapy in patients with urinary bladder malignancy.

KEYWORDS
VEGF: Vascular Endothelial Growth Factor, IHC: Immunohistochemistry, TCC: Transitional Cell Carcinoma. VEGF, Urinary Bladder.

BACKGROUND

Urinary Bladder malignancy is a common malignancy being the 9th most common cancer worldwide and 13th most numerous cause of death from cancer.[1] It is the 5th most common malignancy in Europe.[2] and 4th most common malignancy in USA.[3] As per the Indian cancer registry data in men it is the ninth most common cancer accounting for 3.9% of all cancer cases.[4] No literature has shown the incidence of urinary bladder cancer in Uttarakhand. The main presenting symptom of all bladder cancers is painless haematuria.[5-6]

Among urinary bladder neoplasms, transitional cell carcinomas are the most common tumours accounting for 90% of all urinary bladder carcinomas. Other tumours include squamous cell carcinoma, mixed urothelial carcinomas with areas of squamous carcinoma, adenocarcinomas, signet ring cell carcinoma and mixed adenocarcinomas.[7]

Cigarette smoking is the well-established risk factor and contributes to more than 40% cancer of urinary bladder.[8] Angiogenesis, as quantified by vascular density, is an independent prognostic tumor marker in several tumor types and VEGF is a major factor in this process.[9]

The process of angiogenesis is regulated within a complex homeostasis by the balance between proangiogenic and antiangiogenetic signals. Some proangiogenetic factors including bFGF, VEGF and IL-8 are produced by tumours growing in their relevant microenvironment. Overexpression of VEGF has been identified in tissue, serum, and urine of patients with bladder cancer and correlates with disease recurrence.[10-14]

In many studies VEGF expression has been shown to be a prognostic marker and also predicts the risk of recurrence and high expression is related to lymph node metastasis. This could help in choosing the most convenient therapy for patients.

Aims and Objectives

Since HIHT tertiary care center the incidence of patients with urinary bladder tumours, majority of which are transitional cell carcinoma is very high here. As per many previous studies VEGF is a good predictor of tumor grade as well as prognosis. Therefore this study is being undertaken to determine the expression of this marker in bladder tumours and its possible benefits in determining prognosis of patients.

MATERIALS AND METHODS

The study was conducted in the Department of Pathology, Swami Rama Himalayan University (SRHU) over a period of 12 months. 50 cases of transitional cell carcinoma of urinary bladder were included in the study.

Study Design- Type of the study: Observational and Cross sectional.

Selection of Subject- 50 cases of transitional cell carcinoma urinary bladder diagnosed by histopathology were included in study.

Study Tools

- Histomorphological diagnosis was done using Haematoxylin and Eosin (H & E) stain.
- Immunohistochemistry was done by using Anti VEGF antibody. (BIOGENEX Company kit).
- Structured study instruments (data reporting form) was developed and used to generate data.

Scoring- The immunostaining was graded and this grading was based on staining intensity and relative abundance of VEGF and Factor 8 immunoreactive cells.

The staining intensities for VEGF were graded as follows.[4,3]

- 0- No staining (colourless)
- 1- Slight staining (yellowish)
- 2- Moderate staining (brown-yellow)
- 3- Maximal staining (dark brown)

The scoring was done by using the X40 objective lens and counting the immunopositive cells in at least 10 fields and was recorded as-

- 0 no staining
- 1 positive cells<25%
- 2 positive cells 25-50%
- 3 positive cells>50%

The results of staining intensity and the percentage of positive cells were added and the final scoring of immunohistochemistry results for VEGF was-

- 0(-)- taken as negative
- 1-2 points(+)- taken as positive
- 3-4 points(++) - taken as positive
- 5-6 points(+++)- taken as positive

(-) was regarded as negative and (+, ++,+++) was positive. [15-21]

RESULTS

1. Maximum number of cases (n=31; 62%) in the study group were more than 60 years of age, followed by 40-50 years (n=10;20%) as shown in table 1. The youngest patient was 45 years old. Oldest patient was 85 years old. Mean age of the patient was 64.42 ± 11.78 years.

2. 88% of the patients in the study group were males and 12% were females making male to female ratio of 7:1.

3. Out of total 50 cases, 41 (82%) cases were cigarette smokers, 38 (76%) were tobacco chewers and 24 (48%) cases were alcoholics. Cigarette smoking was found to be the most common addiction.

4. A correlation between cigarette smoking and grade of tumor was found. Out of 41 patients who smoked 32 had high grade lesion which was statistically significant ($p<0.05$). Amongst 38 tobacco chewers, 28 had high grade lesion which was also statistically significant ($p<0.05$). Out of 24 alcoholics only 18 had high grade lesion which was statistically insignificant.

5. The commonest presenting symptom was painless haematuria accounting for total 40 (80%) cases. This was followed by increased frequency of micturition accounting for 30 (60%) cases. Other symptoms were dysuria, urgency and weight loss.

6. 74% patients had high grade malignancy whereas 26% patients had low grade malignancy.

7. Out of 50 total cases, only 15 patients with high grade lesion had deeper muscle involvement which accounted for 30% of total cases.

8. Co-relation of VEGF score and sex of the patients was seen. No significant association was found between the two ($p<0.711$).

9. Increasing VEGF score was associated with increasing grade of tumor. Amongst high grade cases, 22 had VEGF score of 3+ which accounted for 55% of cases. In low grade lesions maximum number of patients had VEGF score of 1+. A significant association was seen between VEGF score and grade of tumor ($p<0.05$).

10. VEGF score was compared between smokers and non-smokers. No significant association was found between cigarette smoking and VEGF score ($p>0.05$).

11. Association between VEGF score and tobacco chewing was seen in the patients. Tobacco chewing was not associated with high VEGF score ($p=0.364$).

12. In total 15 patients with deep muscle invasion, VEGF score was seen. No significant association was seen between high VEGF score and deep muscle invasion ($p=0.616$).

DISCUSSION

Urinary Bladder cancer is a worldwide health problem. It is a common malignancy being the 9th most common cancer worldwide. Transitional cell carcinoma accounts for the most common histological type. Tobacco smoking is the most well established risk factor of bladder cancer followed by occupational exposure to certain chemicals, aniline dyes and petroleum. Men are affected more than females.

This study was conducted on 50 patients in Department of Pathology, Swami Rama Himalayan University (SRHU), Dehradun over a period of 12 months in whom transitional cell carcinoma of urinary bladder cancer was diagnosed histomorphologically.

The incidence of bladder carcinoma is higher in older males and transitional cell carcinoma is the most common variety of urinary bladder malignancy.[22]

Age is now widely accepted as the greatest single risk factor for developing UCB. While UCB can occur at any age, it is generally a disease of middle-aged and elderly people. In fact, with the median age at diagnosis being 70 years.[23]

UCB is primarily considered a disease of the elderly. Because of the close link between age and incidence of UCB, UCB can be expected to become an enormous challenge with the growth in the ageing population in the years ahead.[24]

A study was conducted on 125 (104 males and 21 females) by Rehmani et al. The peak incidence of bladder cancer was observed in the age group of 50-70 years with the mean age being 55 years.[9]

In the present study the ages of patients ranged from 40-85 years. Maximum number of patients (n=31:62%) were more than 60 years of age, followed by 40-50 years (n=10:20%). The youngest patient was 45 year old and oldest patient was 85 years old. The mean age was 64.2 ± 11.78 years. This was slightly higher as seen in the previous studies.

In the present study 88% of the patients were males and females comprised of 12% of the cases making a male to female ratio of 7:1. However the ratio was different from other studies in which the ratio was higher. In our study the peak incidence of bladder cancer was observed in the age group of more than 60 years.

In the present study cigarette smoking was seen to be associated with urinary bladder cancer. 82% (n=41) of the patients were cigarette smokers. Out of 41 people who smoked 33 had high grade lesion and 8 had low grade lesion. A significant association was found between cigarette smoking and bladder cancer (p=0.003).

In this study the commonest presenting symptom was painless haematuria accounting for total 40(80%) cases. This finding was in consistent with the results seen in previous studies.

In addition, patients with high VEGF-C expression had a worse prognosis compared with patients with low VEGF-C expression. In a separate study of 126 patients treated with transurethral resection for UCB, higher expression of VEGF-A, VEGF-C, and VEGF-D were all associated with increasing tumor stage and grade.[25]

The expression of VEGF in high stage bladder cancer is significantly higher than in low stage cancers. Superficial bladder cancer with high VEGF expression shows higher progression (muscle invasion or metastasis) than that with low VEGF expression.[24]

VEGF expression in tumor cells was also suggested as an important predictive factor of pelvic lymph node metastasis in bladder cancer patients.[8]

Another study done in china suggested that the positive rate of VEGF expression significantly increased with the progression of tumor grade and therefore abnormal expression of VEGF genes could be used as a prognostic marker in bladder cancer patients.[26]

In the present study we found 15 patients of high grade lesions having deep muscle involvement by the tumor cells. We studied the VEGF score in these patients separately. Chi square was used to compare the two. No significant association was found. Thereby deep muscle invasion cannot be identified as a criteria for high VEGF expression.

CONCLUSION

In the present study transitional cell carcinoma of bladder was studied. The patients were divided into groups of low grade and high grade urothelial carcinoma. Smoking status was evaluated in all patients and a statistically significant association was seen between smoking and grade of tumor. The expression of VEGF was studied in all patients. And was found to higher in patients with high grade lesion as compared to patients with low grade lesion. This expression was statistically significant (p<0.05). This suggests that VEGF, is a major angiogenic factor in patients with malignancy and contributes to the pathogenesis of this disease. Therefore it can also can be helpful in assessing the utilization of antiangiogenic therapy in patients with urinary bladder malignancy.

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