African Journal of Urology (2018) **xxx**, xxx–xxx

African Journal of Urology

Official journal of the Pan African Urological Surgeon's Association
web page of the journal

www.ees.elsevier.com/afju
www.sciencedirect.com

BPH and prostate disease
Original article

Angiogenesis in prostate cancer and benign prostatic hyperplasia assessed by VEGF and CD-34 IHC: A comparative clinico-pathological study

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Received 2 November 2017; received in revised form 17 January 2018; accepted 24 January 2018
Available online xxx

KEYWORDS
Prostate cancer;
Angiogenesis;
Vascular endothelial growth factor;
VEGF;
CD34;
Microvessel density

Abstract

Introduction: Prostate carcinoma is still a dreaded disease wanting more effective treatment and definitive early detection for a better prognosis and cure of life. The present study was planned to investigate the correlation of vascular endothelial growth factor (VEGF) expression level and microvessel density (MVD) between the BPH and prostate cancer subjects to analyze their diagnostic and prognostic value.

Subjects and methods: Freshly diagnosed histopathologically confirmed 50 cases of prostate cancer and 50 cases of BPH were included. Expression level of VEGF was measured using Immunohistochemistry (IHC), while MVD was determined via CD34 endothelium-specific antibodies. In the case group, we have also recorded the Gleason's score of prostate cancer and investigated its correlation with angiogenic factor VEGF and MVD CD34.

Results: The study showed a statistically significant difference value of VEGF expression level between the prostate cancer and BPH group ($p < 0.001$). The mean MVD CD34 in the prostate cancer and BPH groups were 29.66 ± 0.21 and 9.96 ± 0.25, respectively. The difference of MVD CD34 expression between the groups was also found significant ($p < 0.001$). VEGF scoring was significantly correlated with Gleason's scores of prostate cancer ($p = 0.005$).

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Peer review under responsibility of Pan African Urological Surgeons' Association.

https://doi.org/10.1016/j.afju.2018.01.009

Conclusions: The present findings may support the assumption that VEGF and CD34 expression level might have an important role in the prediction of prostate cancer as it was significantly differed with BPH. In addition, VEGF expression level showed intense staining in the tissue samples with higher grading of prostate cancer which reveals its importance as prognostic marker.

Introduction

Prostate cancer is the most prevalent cancer worldwide. In the USA, 233,000 new cases of prostate cancer were diagnosed and 29,490 deaths occurred in 2014 alone [1]. According to Indian Council of Medical Research (ICMR) report, 2009, the estimated age-adjusted incidence rates of prostate cancer in India was 3.7/100,000 person during the year 2008. Expected cases of prostate cancer all over India for the periods 2010, 2015 and 2020 were estimated as 26,120, 28,079 and 30,185, respectively. According to United Nations Development Program, 2013, India has been grouped in the category of medium human development index (HDI). Usually, prostate cancer is the most commonly diagnosed cancer of men in very high HDI countries, but as India is also leading toward westernization with respect to life style, as a result the same pattern of prostate cancer is likely to follow that seen in high HDI countries [2].

Prostate specific antigen (PSA) test is contemporary and preliminary screening test for the early detection of prostate cancer, which is released by normal epithelial cells as well as neoplastic epithelial cells [3]. Since, serum PSA level may also be raised in benign prostatic hyperplasia (BPH), therefore PSA is not specific only in the case of prostate cancer [4,5]. PSA levels between 4.1 and 9.9 ng/ml are more challenging for differentiating BPH and prostate cancer patients. The patients of this group are sent for confirming the abnormal PSA levels with repeat test and if confirmed prostate biopsy is performed for histopathological analysis. In addition, prostate biopsy screening results in pain, fever, bleeding, infection, transient urinary difficulties as well as psychological harm of false-positive test results, and over diagnosis.

The U.S. Preventive Services Task Force (USPSTF, 2012) does not recommend PSA based screening test for prostate cancer in grade D patient [6]. Therefore, due to unequivocal results of PSA in prediction of prostatic neoplasia and its behavior, there is an urgent need of other diagnostic markers for the early screening of progressive prostate cancer. The essential characteristic of any tumor to develop and progress is the formation of new blood vessel from the preexisting vasculature known as angiogenesis. Vascular endothelial growth factor (VEGF) is one of the angiogenic factors that induces neovascularization and allow tumor to grow beyond 2–3 mm [7], whereas MVD is a quantitative parameter of angiogenesis. Endothelium-specific antibodies are used for the IHC staining of vessels.

Differences of VEGF expression level and MVD between prostate cancer group and BPH group have been reported, however results are inconclusive. Recently published research articles have observed that expression level of VEGF and MVD were higher at initial prostate biopsies in patients who were later diagnosed with prostate cancer [8]. Therefore, by observing prostate cancer at initial stage, survival rate can be increased as well as efforts to stop metastasis of cancer through blood stream in other vital organ like brain, lungs, kidney or liver where cancer cells may grow and disrupt tissue organization and destroy normal cells, eventually leading to organ failure and death, can be done.

The purpose of the present study was to evaluate the diagnostic and prognostic value of VEGF expression level and MVD CD34 for prostate cancer by comparing BPH and prostate cancer groups in freshly diagnosed subjects.

Subjects and methods

Study subjects

Initially, we screened 255 patients on the basis of serum PSA level $\geq$2 ng/ml, abnormalities found in digital rectal examination (DRE) investigations and abnormal prostate ultrasound report. These subjects were recommended for the trans-rectal ultrasound (TRUS) guided prostate biopsy as well as sextant biopsy sample were stored for the study of VEGF expression and MVD. On basis of histopathological examination we further categorized these subjects into two groups (Case group and control group) which were age and ethnicity matched. The prostate cancer patients were considered as case group and BPH served as control group. A total of 100 subjects were included on the basis of diagnosis in time interval of June 2013 to July 2016, out of which 50 were in case group and 50 in control group. Demographical and clinical information of all the subjects were recorded in a systematic questionnaire. Subjects having any immunodeficiency disease like AIDS and any other debilitating disease like hepatitis or any patient receiving treatment of cancer were excluded from the study.

The study was approved by the Institutional Ethics Committee of the King George's Medical University, Lucknow, U.P., India (Ref. Code: XLII ECM/B-P31). The informed consent was obtained from all the participants prior to sample collection.

Immunohistochemistry

We used the monoclonal antibodies VEGF (Monoclonal mouse anti-human VEGF clone VG1, Dako, Denmark) and CD34 (RE7290-K Novolink Polymer Detection Systems Novocastra, Leica Biosystems, UK) and the sections were stained using the streptavidin-biotin-peroxidase method as per standardized protocol for the VEGF expression and MVD assessment. Briefly, 4-μm-wide histological sections were retrieved from formalin-fixed paraffin-

Please cite this article in press as: Gautam KA, et al. Angiogenesis in prostate cancer and benign prostatic hyperplasia assessed by VEGF and CD-34 IHC: A comparative clinico-pathological study. Afr J Urol (2018), https://doi.org/10.1016/j.afju.2018.01.009