Levels of VEGF and VEGFR-2 in Prostate Cancer Patients

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Abstract

Prostate cancer is the most typical cancer within the world, compared to different cancers, that affects men of previous ages of fifty years and additional, however, it seldom affects men that lesser than this age class. The study included the estimation of the studied chemical parameters by ELISA, which are: VEGF and VEGFR-2. The studied chemical markers were estimated in the serum of patients with prostate tumors (60 with prostate cancer and 60 healthy people), using the ELISA technique. Statistical results of the VEGF protein were highly significant in patients with prostate cancer and the value of P <0.001. Also, the current study showed that VEGFR-2 was highly significant in prostate cancer, it recorded a value of P <0.001.

Keywords: Prostate cancer, VEGF, VEGFR-2

Introduction

Carcinogenesis of prostate is a multistep accumulation of genetic lesions that may result in uncontrolled cellular proliferation, a decrease in cell death or apoptosis, invasion, metastatic spread and blockade of prostatic cell differentiation (Spurgers et al., 2006). In the prostate, the expression of oncogene is a driven malignant conversion and expression of tumor suppressor genes that inhibit this process (Karantanos, Corn and Thompson, 2013). Vasculogenesis is the de novo formation of a vascular network whereas angiogenesis is sprouting of new blood vessels from pre-existing ones. Both processes are highly dependent on regulation by vascular endothelial growth factors (VEGFs) and their interaction with membrane receptors expressed on different cell types (Holmes and Zachary, 2005) (Kipryushina, Yakovlev and Odintsova, 2015). Abnormal angiogenesis is associated with a variety of diseases such as tumor neovascularisation, diabetic retinopathy, rheumatoid arthritis (Risau, 1997). Tumor neovascularisation is triggered by cancer cells to stimulate supply of nutrients and enable metastasis. VEGF is overexpressed in a variety of hematological malignancies (Krejsgaard et al.,
2006) and the vast majority of solid tumors, including prostate cancer (Wegiel et al., 2005), where it is associated with poorer outcomes (Green et al., 2007). In prostate cancer, in addition to its expression in blood and lymphatic endothelial cells, VEGF is also expressed at low levels in prostatic glandular epithelial cells and in nonvascular cells, such as macrophages, fibroblast cells, and mast cells (Hrouda, Nicol and Gardiner, 2003). Binding VEGF to VEGFR-2 stimulates the secretion of von Willebrand factor (vWF) by endothelial cells (Zanetta et al., 2000); the activation of endothelial cells is an essential event for tumor progression (Zanetta et al., 2000). VEGFRs possess a cytoplasmic tyrosine kinase (TK) domain which regulates signal transduction pathways linked to cell proliferation, migration, metabolism, vasodilation, blood vessel formation and remodelling (Cudmore et al., 2012). VEGFR-2 is often involved in tumoral angiogenesis. It is considered that VEGFR-2 has the strongest proangiogenic activity, thus blocking VEGFR-2 may have useful clinical implications (Karadağ et al., 2019).

Materials and Methods

Subject and Study Design

A case-control study recruited 60 prostate cancer patients (50-75 years), who were admitted to a Middle Euphrates Cancer Center in Al-Najaf Al-Ashraf Province, and 60 healthy subjects (aged 50-70 years) who had no history of Prostate diseases as controls. This study was established at the laboratory of the Biochemistry Department in Collage of Medicine / Babylon University. The collection of samples was performed during the period from August 2020 to December 2020. The information has been reported using a questionnaire on every individual by face-to-face interviews, to obtain information on their smoking status and on age, weight, height, exercise, family history, past history of diseases and medications.

Collection of Blood Samples

About 3 ml of blood were obtained from each individual by vein puncture, pushed into a disposable gel tube. The blood was allowed to clot at room temperature for 20 minutes and then centrifuged at 1500 x g for 5 minutes and used for determination of biochemical parameter levels within from collection.

Human VEGF and VEGFR-2 ELISA Kit

This kit is an Enzyme-Linked Immunosorbent Assay (ELISA) according to bioassay technology company. The plate has been pre-coated with human VEGF and VEGFR-2 antibody, Respectively. Certain protein present in the sample is added and binds to antibodies coated on the wells. And then biotinylated human certain protein Antibody is added and binds to Certain protein in the sample. Then Streptavidin-HRP is added and binds to the Biotinylated certain protein antibody. After incubation unbound Streptavidin-HRP is washed away during a washing step. Substrate solution is then added and color develops in proportion to the amount of
human certain protein. The reaction is terminated by addition. All reagents, standard solutions, and samples were prepared as instructed. Brought all reagents to room temperature before used. The assay is performed at room temperature.

**Result and Discussion**

**Vascular endothelial growth factor**

The mean concentration of VEGF in prostate cancer (PCa) to control participants was shown in table (1).

Table 1: P Value, Mean and Standard Division and Mean and Standard Error of VEGF (ng/l)

<table>
<thead>
<tr>
<th>P Value</th>
<th>p&lt;0.001</th>
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<tbody>
<tr>
<td>Mean ± SD</td>
<td>642.5 ± 76.24</td>
</tr>
<tr>
<td>Mean ± SE</td>
<td>642.5 ± 9.84</td>
</tr>
</tbody>
</table>

The results obtained prove that the VEGF concentration is high in patients with prostate cancer compared to its concentration in patients with control participants, figure(1) evident that.

*Figure 1: Level of VEGF (ng/l) in PCa.*

Our results revealed that there is a clear increase in the concentration of VEGF, and this was confirmed by previous studies. The behavior simultaneously of VEGF in benign and malignant prostate environments, both serum and tissue in individuals without comorbidities related to
chronic inflammation (9,29). VEGF is narrowly related to the malignancy grade and metastasis of PCa, suggesting that it has a diagnostic and prognostic value of this illness. The levels of VEGF in the serum of PCa and control participants are significantly different. Probably the inflammatory response in PCa causes an increase in the VEGF expression leading to stromal hypervascularization, endothelial vessel permeability, or it might occur through a decrease in the androgen receptors and inhibition of apoptosis in epithelial cells, the two previous studies that evaluated participants with high risk of prostate cancer also observed no significant associations between cancer and VEGF levels. PCa cells secrete proteic factors such as the vascular endothelial growth factor (VEGF), which is extensively studied and known as the major angiogenic marker. VEGF acts as a direct mediator in endothelial cell proliferation, vascular permeation, tumor growth promotion, and metastasis. Several authors report that there are higher levels of VEGF in biopsies and serum of PCa patients as compared to healthy individuals (Liu et al., 2015) (Gyftopoulos et al., 2011)

**Vascular endothelial growth factor receptor 2**

By using T-Test statistically, the mean concentration of VEGF in prostate cancer (PCa) to control participants was shown in table (2).

<table>
<thead>
<tr>
<th>P Value</th>
<th>Mean ± SD</th>
<th>Mean ± SE</th>
</tr>
</thead>
<tbody>
<tr>
<td>P&lt;0.001</td>
<td>4.51 ± 0.60</td>
<td>4.51 ± 0.08</td>
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The results obtained prove that the VEGFR2 concentration is high in patients with prostate cancer compared to its concentration in patients with control participants, figure(2) evident that.
The results of measuring the VEGFR2 concentration in the serum showed that there is an expected increase in the level in patients with prostate cancer relative to its concentration in the serum of control participants. The results of the thesis agreed with previous studies in terms of increasing the concentration of the VEGFR2 in the serum of patients. Recently, it was reported that the soluble vascular endothelial growth factor receptor-2 (sVEGFR-2) is secreted by microvascular endothelial cells from human prostate tumor (Lian et al., 2019). Further, results of another study show that sVEGFR-2 is able to modify the VEGF effect on the endothelium of BPH (Aweimer et al., 2012). Vascular endothelial growth factor receptor 2 (VEGFR2) is highly levels in tumor associated endothelial cells, where it modulates tumor-promoting angiogenesis, and it is also found on the surface of tumor cells (Lu et al., 2019). Another study found that VEGFR2 is expressed in PC-3 human prostate cancer cell line and associated with malignancy and metastasis of human prostate cancer. The prominent effect of VEGFR-2 in cell proliferation, cell differentiation, migration, survival, angiogenesis, and lymph-angiogenesis support the choice of this receptor as a potential target for the discovery of novel inhibitors (Lu et al., 2019). The tumor vessel was traditionally thought to be an especially attractive target tissue because it is formed from nonmalignant. In addition to the angiogenic actions of VEGFR2 protein in endothelial cells, the receptor is also known to be expressed in various cancer cells, where it is associated with tumor malignancy (Silva et al., 2011) (Chatterjee et al., 2013)

References


Silva, S. R. et al. (2011) ‘VEGFR-2 expression in carcinoid cancer cells and its role in tumor growth and

