Relation of Epstein Barr virus with interleukin-10 Levelamongmen with Prostate Cancer in Ramadi City

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Abstract

The study aimed at evaluating the relation of Epstein Barr virus (EBV)with level of interlukin-10 (IL-10) in men withprostatecancer(PC). The study was carried out in Ramadi city from 12th of January to 12th of September 2018, and included a total of 95 women withbreast cancerwho admitted to oncology clinic ofRamadi Teaching Hospital whose ages were between 40-80 years. Patients were investigated for detection of EBV by using Real Time PCR and interleukin-10 (IL-10) by ELISA technique.The rate of prostate cancer men with EBV was (16.84%). The highest rate of prostate cancer menwas within the age group 50-59 years with no significant relation between prostate cancer and age. The study showed that the highest means of IL-10 level (19.43 pg/ml) were found in prostate cancer men without EBV.There was significant difference (*P value*<0.05) between EBV infection with IL-10. The highest rate of men withprostate cancer was fromurban areas.

Keywords:*Prostate cancer, EBV, IL-10, Ramadi.*

Introduction

Prostate Cancer (PC) is the second most common cancer among men in western populations, and despite its high mortality, its etiology remains unknown. Inflammatory processes are related to the etiology of various types of tumors, andprostate inflammation, in particular, has been associated with prostate cancer carcinogenesis and progression [(1)].

Epstein- Barr Virus (EBV) has been linked to the development of variety of human malignancies including prostate tissues that range from benign prostatic hyperplasia (BPH) to prostatic adenocarcinoma (PAC). Somatic point mutations in *Rb gene* have been detected in prostate cancer and are involved in progression steps of prostate carcinogenesis [(2)].

Interleukin-10 (IL-10) is a key regulator of immune responses described as cytokine synthesis inhibitor, immune suppressive and anti-agiogenic factor produced by Thelper2 (Th2) cells and inhibits Thelper1(Th1) cells by inhibiting pro-inflammatory cytokines. In addition, IL-10 can inhibit monocyte/macrophage functions including monokine synthesis, nitric oxide production, and major histocompatibility complex (MHC) class II and CD80/CD86 co-stimulatory expression. *In vitro* and *in vivo* studies revealed pleotropic activities of IL-10 on B and T cells and , taken together, that a critical function of IL-10 is to suppress multiple immune responses through individual actions on T and B cells, antigen presenting cells and other cell types, and skew the immune response from Th1 to Th2. In malignancy, IL-10 might promote tumor development, by acting to suppress anti- tumor immune responses[(3)].

Material and Method

Acrosssectional study was carried out in Ramadi city from12th of January to 12th of September 2018, andincluded 95men with prostate cancer whose ageswere between 40-80 years old. These patients admitted to oncology clinic ofRamadi Teaching Hospital.

Bloodsamples were taken from prostate cancer men. Samples were examined by immunological methods,enzyme linked immuno sorbent assay (ELISA) for detection interleukin-10(IL-10)and molecular technique (real time PCR)which includedDNA amplification of Epstein Barr virus based on the specific primers.

Seven and half ml of blood was collected by vein puncture using vacutainer tubes from each patient

enrolled in this study. Blood samples were divided into two sterile test tubes, in one of them 2.5 ml of blood was put in test tube containing anticoagulant ethylene diamine tetra acetic acid(EDTA) and used for DNA extraction ofEBV. The second part of sample (5ml) was placed in plain tubes left for 30 minutes at 37 °C then was centrifuged at 3000 round per minute(rpm) for 15 minutes then the clot was removed and the remain re-centrifuged at 3000 rpm for 10 min and the obtained sera were then aspirated using automatic micropipette and transferred into two clean test tubes, for serological tests. Label was fixed on each test tube which then stored in deep freeze at -20°C for late serological testing fordetermination the level of IL-10by usingELISA technique.

For DNA extraction, kit was purchased from Gene Aid(USA) companyfor moleculardetection of EBV by Real Time PCR usingAnatolia Gene Works(Turkey). Detection of IL-10 was done by using ELISA kit Elabscience(China), which depends on the Sandwich-ELISA principle.

Statistical Analysis

Computerized statistically analysis was performed using T-Test probability. The *P value*>0.05 was considered statistically significant, and for result which its P value was less than 0.01 was considered highly significant, while for those which its *P value* greater than 0.05,was consideredstatistically non-significant.

Findings

A total of 95 prostate cancer men, their age ranged between 20-89 years old, were investigated for detection of EBV by using real time PCR and estimation the level of IL-10. The present study revealed that EBV was detected in 16.84% of men with prostate cancer, as shown in Table 1.

Table 1: Frequency of EBV in men with prostate cancer.

EBV	Prostate Cancer Men	
	No.	%
Positive	16	16.84
Negative	79	83.16
Total	95	100

The current study showed that the highest rate of EBV infection (7.36 %) was found in men of prostate cancer within the age group 50-59 years.. Table 2.

Table 2: Distribution of EBV infection according to age of men withprostate cancer.

Age Group (Years)	No. of Men with Prostate Cancer	EBV Infection	
		No.	%
20-29 (No:0)	0	0	0
30-39 (No:3)	3	0	0
40-49 (No:15)	15	1	1.05

Cont.. **Table 2: Distribution of EBV infection according to age of men withprostate cancer.**

50-59 (No:38)	38	7	7.37
60-69 (No:27)	27	5	5.26
70-79 (No:6)	6	3	3.16
80-89 (No:6)	6	0	0
Total (No:95)		16	16.84

The present study revealed that there was anegative correlation betweenEBV infection with ageof men withprostate cancer, but the relation was statistically non-significant. (R value:,-0.09)(Figure 1).

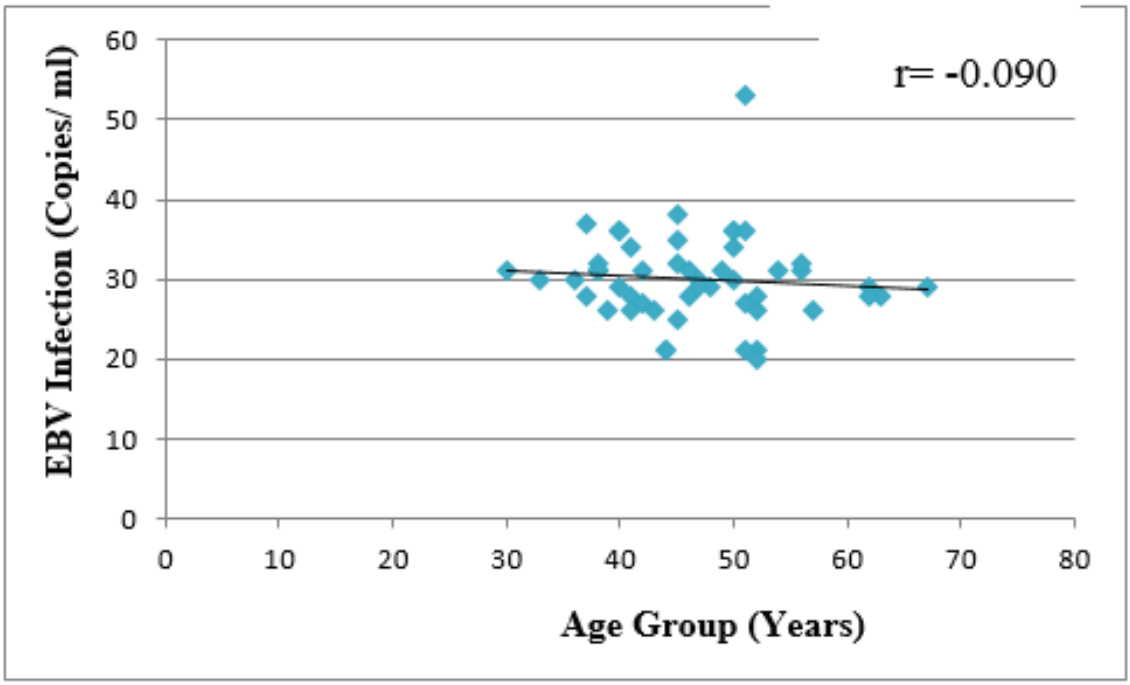

Figure 1: Negative correlation between EBV infection and age in men withprostate cancer.

The current study showed that the highest rate of men with prostate cancer and EBV- infection was from urban areas (68.42%), as shown in Table 3.

Table 3: Distribution of men withprostate canceraccording to residence.

No. of Men with Prostate Cancer	Residence			
	Rural		Urban	
	No.	%	No.	%
95	30	31.58	65	68.42

The highest rate of men with prostate cancer who were infected with EBV were from urban areas (14.73 %), as shown in Table 4.

Table 4: Distribution of EBV infection according to residence of men withprostate cancer.

Residence	No. of Patients	EBV Infection	
		No.	%
Rural	30	2	2.11
Urban	65	14	14.73
Total	95	16	16.84

Relation of EBV infection with IL-10 among prostate cancermen.

Table 5. shows that the means of IL-10 level was higher in prostate cancer men without EBV infection (19.43), highly significant P value <0.05 between EBV and IL-10.

Table 5: Relation of EBV infection with IL-10 amongprostate cancer men.

EBV Infection	No. of Men with Prostate Cancer	IL-10		P value
		Mean	SD	
+ve	16	13.50	±3.39	0.00
-ve	79	19.43	±10.35	

The present study revealed that there was apositive correlation between EBV infection with IL-10 among men withprostate cancer but the difference between them was statistically non- significant (R value:0.198).

Prostate cancer is the second most frequently diagnosed neoplasm in men and the fifth cause of cancer death worldwide [4]. Viruses are etiologic factors in the development of several types of human tumors. At least 15–20% of all human tumors worldwide have a viral cause [5]. The present study revealed that 16.84% of men with prostate cancer had EBV infection. Several studies confirmed the presence of EBV in prostate tissues. In Sweden, EBV was present in 8.8% of benign and malignant prostate tissues [6], and in the United States, EBV was present in 8% of all normal, benign, and malignant prostate tissues [7]. In another study, approximately 37% of prostate cancer patients had EBV infection [8].

The current study showed that the highest rate of EBV infections (7.36%) was found in those within the age group 50-59 years.The reason for EBV to exert its oncogenic influences in a particular patients is unknown but is probably associated with co-factors. The findings in the research by have supported hypothesis that the prostate is a habitat for multiple viral and other infectious agents ,some of which have oncogenic potential[9].

The current study showed that no significant difference was found between EBV infectionand residence in men withprostate cancer*P value*> 0.05. The study of urban–rural differences in prostate cancer in Australia showed that the higher rates of prostate specific antigen (PSA) testing particularly aggressive prostate cancer in regional and rural Australia. Other studies have found urban–rural differences in the management of other cancers [10].

The highest mean of IL-10 level was in men withprostate cancer without EBV infection (19.43%). There was significant difference *P value*<0.05 between EBV with IL-10, and there was apositive correlation between EBV infection and IL-10. Several studies have indicated that IL-10 has both pro- and anti- tumoral effects. IL-10 inhibits NF--B signaling; therefore, it can down regulate pro-inflammatory cytokine expression [11].Interleukin-10 is immunosuppressive and anti-inflammatory.Interleukin-10 inhibits NF-kB activation through ill-defined mechanisms, and consequently inhibits the production of pro inflammatory cytokines,

including TNF-α,IL-6, and IL-12 [12,13,14,15].

Conclusions

The present study revealed that 16.84% of men with prostate cancer had infection with EBV. The relation between EBV infection and age was statistically non-significant. The highest rate of men with prostate cancer who were infected with EBV was from urban areas. The highest mean of IL-10 level was detected in men without EBV infection.

Conflict of Interest: non

Source of Findings: self findings.

Ethical Clearance: non

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