Estimation the Role of Epstein-Barr Virus in Prostate Cancer in Kirkuk

Israa Hashim Saadoon[1], , Azzawi Mustafa Hadi[2] Zheeno Niyazi Taha[3]

[1] Ph.D. Medical Microbiology/ Virology/ Department of Miceobiology/College of Medicine/ Tikrit University.. israahs14@yahoo.com

[2] M.B.Ch.B.F.I.C.M.S. Urology/ Department of Surgery/ College of Medicine/ University of Ninevah.. azzawi.hadi@uoninevah.edu.iq

[3] Ph.D. Medical Microbiology/ Kirkuk Health Directorate.. Jeeno_z@yahoo.com

Abstract

The incidence prostate cancer (PCa) in the Arab world is lower than that in Western countries but is rising significantly, with projections showing the highest relative increase globally by 2050. Viral infections are being studied in Iraq for their link to prostate cancer, with research highlighting roles for Epstein-Barr virus (EBV). The aim of this study was to explore the involvement of EBV in prostate cancer and examine its relationship with levels of prostate specific antigen (PSA) shedding light on its possible role in tumor progression.

Keyword: Epstein-Barr Virus, Prostate Cancer, prostate specific antigen (PSA), Kirkuk

Introduction

Cancer is a serious public health problem on a global scale. According to the World Health Organization estimates, the number of newly detected cancer cases shows an increasing tendency and is expected to reach 30.2 million cases in 2040 [1]. Prostate cancer poses a significant worldwide health issue for men, being the fourth most frequently diagnosed cancer globally [2]. According to GLOBOCAN 2020, 1,414,259 new cases of PCa were reported in 2020, which corresponded to 7.3% of all cancer cases. Excluding non-melanoma skin cancer, PCa mortality is the fifth leading cause of cancer deaths in the world [3]. It is caused by an indolent course of disease and symptomless early stage of cancer [4]. It is estimated that prostate cancer account for



3.8% (375,304 men) of all cancer deaths in the male population [3]. Analyzing the data for Poland, a systematic increase in the incidence of PCa has been observed and further growth can be expected. The number of cases is supposed to reach 20,000 per year [5]. This type of cancer has displayed the highest incidence rate with a mortality plateau in the first decade of the 21st century. However, since 2004, there have been indications of its increased prevalence. In 2020, there were nearly 4000 fewer new cases than in 2019, but fatalities from cancer increased by 130 [6].

Materials and Methods

A cross-sectional study was conducted in Kirkuk, Iraq, from November 2017 to February 2019. The study included 180 patients, comprising 73 with histologically confirmed prostate cancer and 107 with benign prostatic hyperplasia (BPH) as a control group. Serum samples were analyzed for EBV-IgG antibodies using enzyme-linked immunosorbent assay (ELISA). Serum PSA level was estimated using ichroma™ PSA which is a fluorescence immunoassay (FIA) for the quantitative determination of PSA in human whole blood/serum/plasma.

The concentration of IL-6, IL-10 and TNF-α in the sera of both groups was quantitatively measured utilizing sandwich ELISA format, the same protocol was done for this purpose since the kits were from the same company following manufacturers instruction. Interleukin-6 standard range 1.56-100 pg/ml, IL-10 standard range 3.1-200 pg/ml and TNF-α standard range 1.56-100 pg/ml .

Results:

The highest rate of EBV infection was in patients with PCa aged between 66-75 year (41.5 %), followed by those aged from 76 to 85 year (37.5 %), the results were statistically nonsignificant, as shown in Table 1.



Table 1: Distribution of EBV infections in patients with PCa according to the age.

Age (years)	No. of PCa patient	EBV +ve		EBV –ve	
		No .	%	No .	%
55-65	14	8	19.5	6	18.8
66-75	30	17	41.5	13	40.6
76-85	27	15	36.6	12	37.5
86-95	2	1	2.4	1	3.1
Total	73	41	100	32	100
X^2= 0.043; *P value*= 0.872, *P* > 0.05, Not Significant (NS)					

EBV-IgG Abs in patients with PCa was detected in 56.16% while it detected in 30.84% of BPH patients. The result was statistically highly significant, as shown in Table 2.

Table 2: Frequency of EBV- IgG antibodies in the study groups.

EBV- IgG antibodies	PCa Patients		BPH Patients	
	No .	%	No .	%
Positive	41	56.16	33	30.84
Negative	32	43.84	74	69.16
Total	73	100	107	100
X^2 = 11.494 *P value* = 0.001 *P* < 0.01, Highly Significant (HS)				

The mean of PSA was 19.7 ng/ml and 21.2 ng/ml in patients with PCa who have EBV and those without EBV infection respectively. The relation was statistically not significant, whereas in BPH patients the mean of PSA was 4.3 ng/ml and 3.3 ng/ml in those with EBV infection and those without EBV respectively. The relation was statistically non-significant, as shown in Table 3.

Table 3: Relation of PSA (ng/ml) levels with EBV infection in the study groups.

Level of PSA (ng/ml)	PCa Patients		BPH Patients	
	EBV+ ve	EBV- ve	EBV+ ve	EBV- ve
Mean	19.7	21.2	4.3	3.3
S.D.	11.7	14.2	3.7	3.8
p value	0.31 Non-significant		0.89 Non-significant	

The mean level of IL-6 was higher in patients with PCa without EBV infection (27.06 pg/ml) than those with EBV infection (24.65 pg/ml). The result was statistically non-significant significant, as shown in Table 4.

Table 4: **Relation of the level of IL-6 with viral infection (EBV) in the study groups.**

Viral infection	IL-6 levels in Patients with PCa pg/ml		IL-6 levels in BPH patients pg/ml Mean (S.D.)	*P value*
	+ve Mean (S.D.)	–ve Mean (S.D.)		
EBV	**24.65 (11.18)**	**27.06 (18.27)**	**18.8 (6.33)**	**0.083 NS**

The mean level of IL-10 was higher in patients with PCa without EBV infection (12.41pg/ml) than those with EBV infection (10.79 pg/ml). The result was statistically non-significant, as shown in Table 5.

Table 5: Relation of the level of IL-10 with viral infection (EBV) in the study groups.

Viral infection	IL-10 level Patients with PCa pg/ml		IL-10 levels in BPH patients pg/ml Mean (S.D.)	*p value*
	+ve Mean (S.D.)	–ve Mean (S.D.)		
EBV	**10.79 (2.34)**	**12.41 (3.22)**	**13.7(2.98)**	**0.091 NS**

Discussion

The current study showed that the highest rate of EBV infection was in patients with PCa who belonged to the age group 66-75 year (41.5 %), followed by those within the age group 76-85 year (37.5 %), the results were statistically non-significant. Prostate cancer is generally considered as a cancer of the elderly, and the median age of presentation is 68 years. However, 10% of new diagnoses in the USA occur in men aged ≤ 55 years (7). Age is often considered an important non-modifiable risk factor for a number of diseases which include, but are not limited to neurodegenerative diseases such as age-specific Alzheimer's disease.; cardiovascular diseases and certain cancers including PCa (8,9). Hadi (10) confirmed that PCa was the third most common tumor affecting men preceded by bladder tumors and bronchogenic carcinoma and more prevalent in men over 80 years old (35/75 cases). No case was reported below the age 40 years.

The findings of the present study provided important insights into the potential association between Epstein-Barr virus (EBV) infection and prostate cancer (PCa). The detection of EBV-IgG antibodies in 56.16% of prostate cancer patients compared to only 30.84% in the benign prostatic hyperplasia (BPH) control group suggests a significant correlation between EBV seropositivity and prostate malignancy. This supports previous research indicating that persistent viral infections, including EBV, may contribute to carcinogenesis in prostate tissues (11,12).

Interestingly, the study found no statistically significant association between PSA levels and EBV infection, suggesting that EBV seropositivity alone may not directly influence PSA production or secretion. PSA remains an important marker for prostate pathology, but its levels can be affected by multiple factors, including tumor burden, inflammation, and hormonal regulation, which may mask any subtle influence of EBV. The present study revealed that the mean of PSA level was lower in patients with PCa with EBV than those without EBV infection (19.7 and 21.2 ng/ml respectively). Prostate-specific antigen has been shown to be the single most significant predictive factor for identifying men at increased risk of PCa (13). However, PSA testing is not specific to PCa; common conditions such as BPH and prostatitis, also increase PSA levels. Approximately 1.5 million men, form age 40 to 69 years, have a



PSA level greater than 4.0 ng/ml, a widely use cutoff value for a positive screening result (14). However, the higher a man's PSA level, the more likely that the PCa is present. In a study done by Palsdottir *et al* (15) found that International Society of Urological Pathology Grade Group (ISUP 1) PCa was not significantly associated with PSA at diagnosis.

Certain cytokines are specific to the pathologically altered prostate, suggests that the cytokine balance differs in pathological and normal prostate (16). One of the most interesting mediators clearly implicated in PCa is IL-6, a multifunctional cytokine, produced by cancer cells. There are multiple lines of clinical and experimental evidence preponderantly showing that IL-6 contributes to PCa progression. Both, patients with PCa and patients with advanced metastatic disease display high expression levels of IL-6 and its soluble receptor in the circulating plasma (17). In PCa patients IL-6 serum levels were found to be strongly elevated and positively correlated to tumor load (18). The present work revealed that the mean level of IL-6 was higher in patients with PCa without EBV infection than those with EBV infection (27.06 and 24.65 pg/ml respectively). The result was statistically non-significant. Interleukin-6 is one of the most common cytokines associated with the progression and metastasis of PCa (19). The serum IL-6 level may be associated with the prognosis of patients with PCa, and high levels of both IL-6 and PSA may indicate patients with a poor prognosis. The expression of IL-6 receptor (IL-6R) may be helpful in predicting the effects of hormonal therapy against PCa (20).

One the other hand, interleukin-10 is known as a cytokine synthesis inhibitory factor that has a censorious effect in the regulation of inflammatory responses which may contribute to angiogenesis and survival of tumor cells (21). The presence of IL-10 in many tumors may influence the native antitumor response, thereby inducing a response against tumor cells. Interlukin-10 down regulates macrophage pro inflammatory cytokines such as IL-6 (22). Epstein–Barr virus produces a homologue of IL-10, a cytokine initially called "cytokine synthesis inhibitory factor"(23).

The present study revealed that mean levels of IL-10 in patients with PCa and viral infection (EBV) were lower than those without viral infection. The results were statistically non-significant. Epstein Barr virus infection is known to induce



endogenous secretion of IL-10 as a mechanism of parasitism because IL-10 seems to be responsible for inhibition synthesis of IFN gamma, the main macrophage–stimulating cytokine involved in the defense against EBV which facilitate the intracellular survival of virus by down-regulating the oxidative and inflammatory response (24). It has been reported that EBV infection was correlated positively with IL-10 elevated level and this increasing will lead to evade of immune response by EBV (25).

Conclusion

This study demonstrates a statistically significant association between EBV seropositivity and prostate cancer, supporting the hypothesis that EBV may play a role in the pathogenesis or progression of the disease.

Although no direct link was found between EBV infection and PSA levels, the overall findings warrant deeper investigation into the molecular mechanisms by which EBV could influence prostate carcinogenesis.

References

1. World Health Organization Cancer Tomorrow. [(accessed on 18 May 2025)]. Available online: https://gco.iarc.fr/tomorrow/en/dataviz/isotype.
2. Prostate Cancer: Statistics Approved by the Cancer.Net Editorial Board. Mar, 2023. [(accessed on 18 May 2025)]. Available online: https://www.cancer.net/cancer-types/prostate-cancer/statistics.
3. World Health Organization The Global Cancer Observatory—All Rights Reserved. Dec, 2020. [(accessed on 28 November 2023)]. Available online: https://gco.iarc.fr/today/data/factsheets/cancers/27-Prostate-fact-sheet.pdf.
4. Rawla P. Epidemiology of Prostate Cancer. World J. Oncol. 2019;10:63–89. doi: 10.14740/wjon1191. [DOI] [PMC free article] [PubMed] [Google Scholar]
5. Prostate Cancer—Is It Worth Getting Tested? [(accessed on 29 November 2023)]; Available online: https://www.gov.pl.

6. National Cancer Registry: Reports. [(accessed on 28 November 2023)]. Available online: https://onkologia.org.pl/pl/raporty.

7. Gupta S, Gupta A, Saini AK, Majumder K, Sinha K, Chahal A. Prostate cancer: how young is too young? Curr Urol 2017;9(4):212-215.

8. Anderson CB, Sternberg IA, Karen-Paz G, *et al*. Age is associated with upgrading at confirmatory biopsy among men with prostate cancer treated with active surveillance. J Urol 2015;194(6):1607-1611.

9. Hall WH, Jani AB, Ryu JK, Narayan S, Vijayakumar S. The impact of age and comorbidity on survival outcomes and treatment patterns in prostate cancer. Prostate Cancer Prostatic Dis 2005;8(1):22-30.

10. Hadi AM. Prostate tumor in Tikrit Teaching Hospital: a review of 75 cases. Tikrit Med J 2008;14(1): 23-27.

11. Kiś, J.; Góralczyk, M.; Sikora, D.; Stępień, E.; Drop, B.; Polz-Dacewicz, M. Can the Epstein–Barr virus plays a role in the development of prostate cancer? *Cancers* **2024**, *16*, 328. https://doi.org/10.3390/cancers16020328

12. Ahmed K., Sheikh A., Fatima S., Haider G., Ghias K., Abbas F., Mughal N., Abidi S.H. Detection and characterization of latency stage of EBV and histopathological analysis of prostatic adenocarcinoma tissues. Sci. Rep. 2022; 12:10399. doi: 10.1038/s41598-022-14511-4. [DOI] [PMC free article] [PubMed] [Google Scholar]

13. Roobol MJ, Schroder FH, Crawford ED *et al*. A framework for the identification of men at increased risk for prostate cancer. J Urol 2009; 182(5): 2112–2120.

14. Welch HG, Schwartz LM, Woloshin S. Prostate-specific antigen levels in the United States: implications of various definitions for abnormal. J Natl Cancer Inst 2005;97(15):1132-1137.

15. Palsdottir T, Nordström T, Aly M, *et al*. Are prostate specific-antigen (PSA) and age associated with the risk of ISUP grade 1 prostate cancer? Results from 72 996 individual biopsy cores in 6 083 men from the Stockholm3 study. PLoS One 2019;14(6):e0218280.

16. Steiner GE, Newman ME, Paikl D, *et al*. Expression and function of pro inflammatory interleukin IL-17 and IL-17 receptor in normal, benign hyperplastic, and malignant prostate. Prostate 2003;56(3):171-182.

17. Nguyen DP, Li J, Tewari AK. Inflammation and prostate cancer: the role of interleukin-6 (IL-6). BJU Int. 2014;113(6):986-992.

18. Shariat SF, Chromecki TF, Hoefer J, *et al*. Soluble gp130 regulates prostate cancer invasion and progression in an interleukin-6 dependent and independent manner. J Urol 2011;186(5):2107-2114.

19. Hong DS, Hui D, Bruera E, *et al*. MABp1, a first-in-class true human antibody targeting interleukin-1α in refractory cancers: an open-label, phase 1 dose-escalation and expansion study. Lancet Oncol 2014;15(6):656-666.

20. Tozawa K, Kawai N, Ando R, Naiki T, Iida K, Yasui T. The role of interleukin-6 and interleukin-6 receptor as prognostic biomarkers in prostate cancer. Clin Oncol 2016; 1:1124.

21. Zadka L, Kram P, Koscinski J, *et al*. Association between interleukin-10 receptors and the CD45-immunophenotype of central nervous system tumors: a preliminary study. Anticancer Res 2017;37: 5777-5783.

22. Zahed A, Chehrazi S. The evaluation of the relationship between serum levels of interleukin-6 and interleukin-10 and metabolic acidosis in hemodialysis patients. Saudi J Kidney Dis Transplant 2017;28(1):23-29.

23. Goering RV, Dockrell HM, Zuckerman M, Chiodini PL. Mims' Medical Microbiology and Immunology. 6th ed. Elsevier: 2019:195.

24. Dukers DF, Meij P, Vervoort MB, *et al*. Direct immunosuppressive effects of EBV encoded latent membrane protein 1. J Immunol 2000; 165(2): 663–670.

25. Liu MY, Shih YY, Li LY, *et al*. Expression of the Epstein-Barr virus BHRF1 gene, a homologue of Bcl-2, in nasopharyngeal carcinoma tissue. J Med Virol 2000;61(2):241-250.