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Analyzing the Possible Impact of Herpes Simplex Virus-1 in Relation to Interleukin-6 Levels in Patients with Oral Squamous Cell Carcinoma

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Abstract

Background: Oral squamous cell carcinoma (OSCC) is the most common malignant tumor of the oral cavity, accounting for more than 90% of all cases. Oral herpes is caused by herpes simplex virus type 1 (HSV-1), a common, benign virus that affects the skin and mucous membranes of individuals with weakened immune systems, causing ulcers. There are several cellular processes that rely on the pleiotropic cytokine interleukin-6 (IL-6), including migration, invasion, differentiation, proliferation, and survival. By modifying tumor angiogenesis and tumor lymphangiogenesis, IL-6 controls tumor growth and its effects on tumor cells.

Methods: The study involved 60 patients divided into two groups: those with and those without oral cancer. The blood was drawn and examined for HSV-1 Immunoglobulin G levels using the Herpe Select-1 ELISA kit. A sandwich ELISA was also employed to study IL-6 levels.

Results: HSV-1 incidence in these oral squamous cell carcinoma grade groups increased significantly. This research found that the OSCC group had somewhat greater IL-6 levels than the control groups.

Conclusion: HSV-1 is not carcinogenic in its own right; however, it is linked to an increased risk of oral squamous cell carcinoma. In addition, the present study discovered that the pro-inflammatory cytokine IL-6 was present in greater quantities in patients' blood than in controls. Interleukin-6 was identified as a potentially hazardous factor in the development of oral cancer and has the potential to function as a valuable biomarker for the assessment of OSCC severity.

Introduction

The sixth most common cancer is oral cancer worldwide. Mouth cancer remains a deadly and disfiguring illness in Iraq despite regional differences. The progression of OSCC is modulated by viral infections, genetic variations, and environmental factors [1]. Based on the findings from the in vitro cell cultures and patient examinations it is evident that HSV-1 can contribute to cancer development by enhancing the tumorigenic potential of other risk factors and there is evidence that HSV-1 antibody levels are related to oral cancer [2]. It may increase the incidence of head and neck cancer by inducing chromosomal mutation, gene amplification and oncogene overexpression in neoplastic tissue [3, 4]. Although HSV-1 is not thought to be an oncogenic virus by itself, it may predispose to malignant development in the research by Brown *et al.*, and others [5-8]. This is because the virus remains in the oral mucosa and stimulates host cell DNA synthesis and repair.

The term "host cutoff" in the context of viral infections generally refers to the point at which a virus can no longer effectively infect a host or replicate within a host organism. This can occur due to (immune response, viral adaptation, host factors, and therapeutic interventions) [7].

Cytokines play a crucial role in managing the tumor microenvironment and preventing long-term pro-tumorigenic inflammation. Polypeptides, which are low molecular weight proteins produced by the immune system, tissue, and tumor cells, are involved in the regulation of cellular behavior, growth, differentiation, and function; however, their production during inflammation or carcinogenesis can be detrimental to physiological activities [9]. In lung cancer, hepatocellular carcinoma, colorectal cancer and oral squamous cell carcinoma (OSCC), the progression of these cancers has been associated with proinflammatory cytokines, as studied. IL-6 can interact with cell receptors via autocrine or paracrine pathways, facilitating the transmission and exchange of intercellular signals, potentially completing cell biological activities. IL-6 is linked to tumor formation, differentiation, apoptosis, immune response, and treatment resistance, according to a recent investigation [10].

One of the indications indicated as being useful for early diagnosis and predicting prognosis in OSCC patients is interleukin (IL-6) expression. Various investigations confirmed that those who suffer from OSCC have higher IL-6 levels than healthy controls [11, 12]. As with Harini *et al.*, it is revealed that the fundamental oncogene in the composition of malignancies such as OSCC is IL-6. Therefore, IL-6 can be considered beneficial for diagnosis or prognosis [13]. Moreover, Xiao *et al.*, confirmed that IL-6 is the main oncogene for the OSCC biological processes. Thus, it was suggested as a potential biomarker for identifying OSCC [14]. The investigation was an endeavor for assessing the blood IL-6 levels effects on the differential diagnosis of OSCC.

In this present study, the levels of HSV-1 IgG in the blood of thirty persons suffering from OSCC and thirty healthy controls are examined. Both groups were from the same age and lived in the same area. Then, Moreover, the identified concentrations of IL-6 in the blood of both groups were examined.

Methods

The study identified 60 individuals (30 with OSCC and 30 controls) using Human Anti-Herpes Simplex Virus Type 1 IgG ELISA Kits, Abcam, UK. IL-6 serum expression levels were evaluated employing a human Sandwich IL-6-ELISA Kit application (purchased from Abcam, UK) (Cat. No.: ab178013). The assessment was performed in compliance with the manufacturer's guidelines.

Statistical Analysis:

To perform statistical analysis on the data collected in this investigation, the program SPSS (version 24) was applied. The variables' significance was assessed using Chi-Square, and the mean was calculated. Statistical tests were conducted to confirm noteworthy alterations in categorical variables among groups and ROC analysis was used to establish the ideal cutoff for OSCC patients, which demonstrates excellent specificity and sensitivity.

Results

Assessment of Serum Interleukin-6 in study groups:

The study showed that oral squamous cell carcinomas have different levels of mean IL-6 in different stages of the disease; well, moderately, poorly and undifferentiated. Table 1 presents the mean serum IL-6 concentrations for OSCC and healthy controls, revealing a difference in serum interleukin-6 levels in OSCC tissues of all grades associated with the Healthy individual group. Diagnostic efficacy of the IL-6 (ROC) test is illustrated in Table 2 and Figure 1, with a positive predictive value (PPV) of 86.4%, a negative predictive value (NPV) of 100.0%, sensitivity of 100.0%, specificity of 83.0%, accuracy of 92.0%, and an area under the ROC curve (AUC) of 0.949. The genuine positive rate is signified by sensitivity, while the specificity means the true negative rate of IL-6. However, AUC > 0.8 to IL-6, indicates excellent model performance to use it as a good marker for diagnostic OSCC.

Cytokine	Study Subjects	OSCC Group
IL-6 pg/mL	Well-differentiated	20.9±2.85
	Moderately differentiated	33.2±5.32
	Poorly differentiated	40.7±9.74
	Undifferentiated	43.3±5.99
	Control	11.3±3.12
	LSD	8.37

Table 1: The mean serum IL-6 concentrations for OSCC and healthy controls.
(Phases of OSCC: well differentiated (low grade), moderately differentiated (intermediate grade), poorly differentiated (high grade), undifferentiated (high grade). Healthy person: Control group.

Diagnostic Values of IL-6	Percentage
Cutoff point	13.42
Area under ROC curve (AUC)	0.949
Sensitivity	100.0%
Specificity	83.0%
Accuracy	92.0%
Positive predictive value (PPV)	86.4%
Negative predictive value (NPV)	100.0%

Table 2: The Diagnostic Value of Serum Interleukin-6.
(AUC: Area under ROC (receiver operating characteristic) curve, PPV: Positive predictive value, NPV: Negative predictive value)

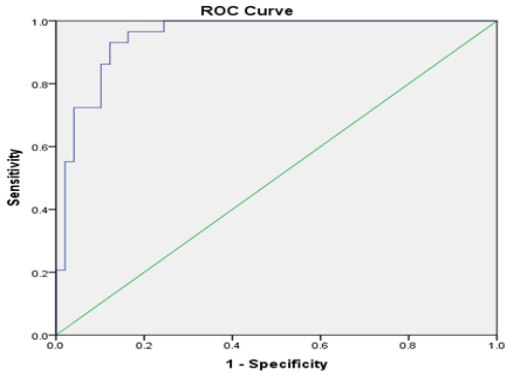

Figure 1: Evaluation of IL-6's Diagnostic Value Using ROC (receiver operating characteristic) Curve Analysis.
(The Area under ROC curve AUC = 0.949, sensitivity = 100.0%, specificity = 83.0%, accuracy = 92.0%, positive predictive value (PPV) = 86.4%, and negative predictive value NPV = 100.0%)

Detection of HSV-1 IgG in Studied groups:

In the current investigation, significant differences were observed between the OSCC groups and the healthy control group (P<0.05). Among the 30 OSCC patients studied 6.67%, 10.0%, 26.67%, and 20.0% tested positive for HSV1 antibodies in well, moderately, poorly, and undifferentiated grades of OSCC, respectively. At the same time, the seronegative percentages were (13.33%, 13.33%, 6.67%, and 3.33%) for well, moderately, poorly, and undifferentiated grade groups of OSCC. Besides that, among the 30 control participants, 7 (23.33%) were HSV1 antibody-positive, while 23 (76.67%) were seronegative for HSV1, as illustrated in Table 3.

Discussion

ROC curve analysis, specificity, and sensitivity calculations suggest that serum IL-6 is the most precise predictor of oral cancer. This and previous studies show that OSCC and other malignancies may be diagnosed using IL-6.

Study group	Grading of OSCC	HSV-1 seropositive		HSV-1 seronegative	
		N	%	N	%
OSCC	Well-differentiated	2	6.67	4	13.33
	Moderately differentiated	3	10.0	4	13.33
	Poorly differentiated	8	26.67	2	6.67
	Undifferentiated	6	20.0	1	3.33
	Total patients	30 (100 %)			
Control	Total patients	7	23.33	23	76.67
		30 (100 %)			

Table 3: The HSV-1 IgG levels based on grading of OSCC.
Phases of OSCC: well-differentiated (low grade), moderately differentiated (intermediate grade), poorly differentiated (high grade), undifferentiated (high grade). Control group: healthy person, HSV-1 Seropositive: Patients having HSV-1 in their blood samples, HSV-1 Seronegative: Patients not having HSV-1 in their blood samples

High levels of IL-6 have been associated with a poor prognosis in many types of cancer, including cancers of the stomach, kidney, colon, prostate, and non-small cell lung, as well as melanomas, skin, head, and neck cancers, and hematologic malignancies such as myeloma and non-Hodgkin's lymphoma. Researchers have found a link between elevated IL-6 levels and Hodgkin's lymphoma. Many studies have shown a connection between a person's blood IL-6 genotype and the risk of infected cancer [15-20].

Serum IL-6 can start a tumor by paracrine or autocrine mechanisms and impede the immune response against it. Therefore, it likely contributes to or indicates how cancer grows and operates [21]. IL-6 stops dendritic cells from growing, which makes the immune system less sensitive to cancer and speeds up the spread of metastatic disease [22]. Even though it has been shown that most serum comes from tumors, it has also been found that monocytes in people with head and neck SCC release more than in healthy people [23]. Reduced cellular immunity and abnormal monocyte function are well-known to be frequent and early characteristics of OSCC patients [24, 25].

This study, like others, demonstrates that IL-6 levels can be correlated with the aggressiveness and severity of a disease. The current investigation found that these pro-inflammatory cytokines are greater in OSCC patients' blood than in controls, suggesting they have diagnostic and/or prognostic value, which large multicenter studies must verify.

Recent research has looked at the effect of histopathological variables on OSCC patients' overall survival, with tumor differentiation impacting clinical behavior. Since the 1960s, HSV1 has been thought to contribute to the development of OSCC. Only some epidemiological studies have been done to test this idea. The Herpes simplex virus is a virus that replicates by copying its DNA in an envelope. When it invades a cell, it triggers a series of intricate reactions. HSV-1 usually causes infections in the mouth. It has been investigated as a potential method for HSV

transformation to induce cellular proteins (such as heat shock proteins) [26]. Another significant component brought on by HSV is known to be the host cell cutoff. The infected cell ceases protein synthesis, and cell RNA is rapidly destroyed. Another HSV activity that may be connected to cell transformation is viral replication stimulation. An enhancement in immune response can be attributed to the higher levels of IL-6 and HSV-1 infections in OSCC patients in the current investigation. Also, it has been thought that HSV1's ability to cause mutations could directly change cells into cancerous ones as in the results of this study possibly by causing chromosomal rearrangements or amplifying host genes that cause cancer [27, 28].

Jalouli *et al.*, used biopsy to find out how often HSV caused OSCC. These results demonstrate the prevalence of HSV infections, which may have consequences for oral health and the emergence of malignancy [29]. This study matches these findings. Eglin and coworkers examined OSCC biopsy samples for HSV1-complementary RNA using in situ hybridization. Approximately 50% of oral squamous cell carcinomas had HSV RNA [30]. For the first time, Delavarian *et al.*, investigated OSCC viruses in 21 formalin-fixed, paraffin-embedded sections of adolescent Iranian patients [31].

Finally, this study has also offered some data on HSV-1 infection in OSCC patients. This is because HSV-1 is involved in OSCC due to the significant variance in IL-6 levels between control and patient groups. More data is, however, desired to completely comprehend the HSV-1's role in OSCC.

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Competing Interest

The authors declare that there is no conflict of interest.

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