

## RESEARCH ARTICLE

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# Galectin-3 Serum Levels Could Help Clinicians Screen for Salivary Gland Tumor Patients

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### Abstract

**Objective:** To identify serum levels of galectin-3 in salivary gland cancer and healthy populations; a prospective analysis was performed on serum specimens from 105 patients with salivary gland cancer and 56 healthy persons. **Methods:** Enzyme-linked immunosorbent assay (ELISA) was used to measure levels of galectin-3 (GAL-3). Serum levels were compared between patients with salivary gland tumors and healthy control. A total of 105 patients were enrolled in the study (55 men, 50 women). **Result:** Mean age was 45.5 years. Thirty-nine patients with malignant and 66 cases with benign tumors were compared with 56 healthy participants with a mean age of 51.7. No statistically significant differences were observed when comparing GAL-3 serum levels between malignant and benign salivary gland tumor patients, but a statistically significant difference was found between case and control patients with p-values of 0.02. Serum levels of galectin-3 protein were elevated in patients with salivary gland cancer compared with the healthy population. **Conclusion:** The difference between benign and malignant tumor patients was significant, but revealed no clinic pathological characteristics in malignant tumors. To the best of the authors' knowledge, this is the first time a study suggests that GAL-3 serum levels could help clinicians screen for salivary gland cancer.

**Keywords:** Salivary gland- cancer- serum- galectin-3- biomarker

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### Introduction

Galectins are multifunctional proteins that have an affinity for binding glycans consisting of b-galactoside. There are 15 types of galectins, and they are named by numbers based on the order of their discovery. Galectins play a principal role in cancer biology. Galectin-3 (Gal-3) is a 31kilodalton member of a family of non-integrin beta-galactosidase-binding lectins (Barondes et al., 1994).

Evidence has demonstrated that Gal-3 participates in several biological processes, such as cell growth and differentiation, angiogenesis, and apoptosis (Yang and Liu, 2003; D'Haene et al., 2013; Nangia-Makker et al., 2007). Altered expression of galectins has been associated with malignancy, progression, and invasion in several human tumors, including colon, prostate, thyroid, and breast cancer, and its altered expression correlates with the stage of tumor progression (Schoepfner et al., 1995; Berberat et al., 2001; Orlandi et al., 1998; Yamaki et al., 2013; Miyazaki et al., 2002). This multifunctional protein is expressed in normal and tumoral cells in a variety of tissue and cell types, mainly those found in the cytoplasm. Depending on cell type and proliferative state, a significant amount of this lectin can also be detected in the nucleus,

on the cell surface, or in the extracellular environment (Kasai and Hirabayashi, 1996; Hughes, 1999; Davidson et al., 2002).

Galectin has been implicated in a variety of biological events, including tumoral progression, participating in cellular transformation, and metastasis (Ellerhorst et al., 1999; Pacis et al., 2000; Nakamura et al., 1999; Takenaka et al., 2004). According to the evidence gathered by several studies, the serum level of Gal-3 may be an alarm biomarker for malignancy and metastasis. The significance of serum Gal-3 has been evaluated in many malignant tumors, and some investigators have examined the efficacy of Gal-3 as a diagnostic marker in such cancers as colorectal, gastric, lung, bladder, thyroid, and prostate, and in pancreatic carcinoma and head and neck squamous cell carcinoma (Iacovazzi et al., 2010; Iurisci et al., 2000; Xie et al., 2012; Išić et al., 2010; Balan et al., 2013; Saussez et al., 2008).

The current study was designed to identify and compare serum levels of galectin-3 in patients with salivary gland cancer (malignant and benign tumors) and healthy controls.

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## Materials and Methods

Prospective analysis was performed on serum specimens from 105 patients with salivary gland cancer confirmed by histopathology that referred to Khalili Hospital of Shiraz University of Medical Sciences, I.R. Iran for surgery and 56 healthy controls whose medical history including no cancer, thyroid or metabolic disease. Both groups were matched in age and gender. All patients with systemic disease, other kinds of cancer, or thyroid disease were excluded. Sampling was done after fully-informed consent was obtained from the patients or their guardians. This study conforms to the declaration of Helsinki and was approved by the ethics committee of Shiraz University of Medical Sciences. Blood samples were taken 2 days before surgery, and sera were separated, coded, and stored in aliquots at -20°C until further testing. Demographic and clinical information (such as sex, age, TNM stage, lymph node metastasis, invasion depth) was obtained from patients' medical records. Enzyme-linked immunosorbent assay (ELISA, DIAPRO, Italy) was used to measure levels of galectin-3 according to the manufacturer's instructions (anti-Human Gal-3 Ab, ELISA, Bender Med System, BMS279, Vienna, Austria). Serum levels were compared between patients with salivary gland tumors and healthy controls.

### Statistical analysis

Descriptive statistics were reported. Quantitative variables were expressed as mean  $\pm$  standard deviation (SD). The Student's t test, Kruskal-Wallis test, and Mann-Whitney U Test were used to compare findings between groups. A P-value  $<0.05$  was considered to be statistically significant. Statistical analysis was performed with SPSS software (version 18).

## Results

The case group comprised 39 patients with malignant tumors (24 males, 15 female) and 66 with benign tumors (31 males, 35 female) Mean age and standard deviation of this group was 45.5 $\pm$ 15.4 years. The control group comprised 56 (28 males, 28 female) healthy persons, and the mean age  $\pm$  SD was 51.7 $\pm$ 14.9 years. No statistically significant differences in Gal-3 serum levels between patients with malignant and benign salivary gland tumors were observed, but a statistically significant difference between case and control groups was found. The p-value was 0.02 for Gal-3 serum levels in case and control groups (Table 1).

Serum Gal-3 levels in the malignant salivary gland

Table 1. Mean  $\pm$  SD of Gal-3 Serum Levels between Salivary Gland Cancer Patients and Control Group

	N	Mean $\pm$ SD of Gal-3 serum levels	P-Value*
Benign Tumor	66	2.4 $\pm$ 2.1	$<0.02$
Malignant tumor	39	3.9 $\pm$ 1.4	
Control	56	0.9 $\pm$ 0.4	

\*Kruskal-Wallis Test, statistically significant if P-value  $<0.05$

Table 2. Serum Gal-3 Levels in Malignant Salivary Gland Cancer Patients Based on Clinic- Pathological Characteristics

	Number	Gal-3(ng/dl)	P-Value
TNM Stage			
1 + 2	15	3.2 $\pm$ 1.2	P=0.8
3 + 4	24	4.5 $\pm$ 2.4	
Tumor size			
T1+T2	29	3.8 $\pm$ 2.6	P=0.1
T3+T4	10	4.1 $\pm$ 1.4	
LN metastasis			
Negative	25	3.7 $\pm$ 2.2	P=0.1
Positive	14	4.4 $\pm$ 2.8	

cancer patients based on clinic-pathological characteristics are shown in Table 2.

## Discussion

Galectin-3 plays a role in cellular interactions and cell proliferation. It is also important in the control of cell growth. Accumulated evidence revealed that the overexpression of galectin-3 in some tissue is concomitant with neoplastic shift, progression, and invasion in tumors.

In previous studies, galectin-3 has been identified as a biomarker that can be used in the diagnosis of some cancers. It can also reveal the prognosis and stage of tumors. The essential role of the expression of Gal-3 in some cancers, including prostate and thyroid cancers, has been proven (Xu et al., 2000). Therefore, it may assist in decision-making regarding the management and appropriate treatment of cancer.

Iurisci et al., (2000) showed that serum levels of galectin-3 rise in patients with breast, ovary, lung, gastrointestinal, and melanoma cancers compared with healthy individuals. They also revealed that serum levels of galectin-3 were higher in patients with later stage tumors and distance metastasis than in those with localized tumors at earlier stages.

The present study found a significant difference in Gal-3 serum levels between malignant and benign salivary gland tumors (P $<0.02$ ).

Sakaki et al.,(2008) studied the expression and serum levels of galectin-3 in patients with bladder tumors. Their results demonstrated that both the expression and serum levels of galectin-3 were higher in patients with bladder tumors than in the control group, and these values were higher in patients in the metastatic stage.

Our study showed no significant difference in galectin-3 serum levels between later TNM (3+4) and earlier (1+2) stages in patients with malignant salivary gland tumors (P=0.8), although the serum level of Gal-3 was higher in the later TNM stage. In the present study, serum levels of Gal-3 in patients with large tumor size(T3+T4) and lymph node involved (positive) malignant cancers were higher than in those with small tumor size(T1+T2) and LN negative cancers.

Vereecken et al., (2006) showed that patients in the end stages of melanoma experienced a rise in galectin-3

serum levels.

Saussez et al., (2008) measured the levels of circulating galectin-1 and galectin-3 in HNSCC patients and claimed they could be used to monitor tumor progression and/or responses to therapy.

A study by Xu et al., (2008) demonstrated that the expression of galectin-3 in normal conditions is bound to salivary ductal cells; in benign and malignant tumors of the salivary glands, the researchers found changes in galectin-3 expression in ductal cells, which can help diagnose salivary gland masses.

Another study done by Teymoortash et al.,(2006) demonstrated that the expression of galectin-3 in adenoid cystic carcinoma in the head and neck had a correlation with the stage, prognosis, and survival rate of the tumor. They also mentioned that galectin-3 can be an alarm biomarker for metastasis.

In the recent article of present study authors, no clear-cut relationship was detected between MCP-1 levels and clinicopathologic factors, and MCP-1 was not a good marker for evaluating tumor dissemination. (Mardani et al., 2016).

In the present study, rises in serum levels of galectin-3 were seen in case patients compared with the control group, and the difference was statistically significant. Patients with malignant salivary gland tumors had higher galectin serum levels in later TNM stages, invasion depth, and LN metastasis characteristics, but the difference was not significant. The study sample size of malignant tumor is low, and this point may have caused the smaller difference. Serum levels of galectin-3 protein were elevated in patients with salivary gland cancer compared with the healthy population; however, the differences between the clinic-pathological characteristics of malignant tumors were not statistically significant.

To the best of the authors' knowledge, this study suggests for the first time that Gal-3 serum levels may help clinicians screen for salivary gland cancer. The authors suggest another study be performed with a larger sample size for better statistical evaluation.

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#### Statement conflict of Interest

The authors had no conflict of interest

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