Original Article

Evaluation of serum sialic acid, fucose levels and their ratio in oral squamous cell carcinoma

Sangamesh Ningappa Chinnannavar, Lingappa Ashok[1], Kodige Chandrashekhar Vidya, Sunil Mysore Kantharaja Setty[2], Guru Eraiah Narasimha[3], Ranjana Garg[4]

Department of Oral Medicine and Radiology, Kalinga Institute of Dental Sciences, Bhubaneswar, Odisha, [1]Department of Oral Medicine and Radiology, Bapuji Dental College and Hospital, Davanagere, Karnataka, [2]Department of Oral Medicine and Radiology, Gurunanak Dev Dental College, Sunam, Punjab, [3]Department of Oral Medicine and Radiology, Vishnu Dental College, Bhimavaram, Andhra Pradesh, India, [4]Department of Oral Medicine and Radiology, SEGI University College, Petaling Jaya, Malaysia

Corresponding author (email: <csangs63@rediffmail.com>)
Dr. Sangamesh Ningappa Chinnannavar, Department of Oral Medicine and Radiology, Kalinga Institute of Dental Sciences, Bhubaneswar, Odisha, India.

Abstract

Background: Detection of cancer at the early stage is of utmost importance to decrease the morbidity and mortality of the disease. Apart from the conventional biopsy, minimally invasive methods like serum evaluation are used for screening large populations. Thus, this study aimed to estimate serum levels of sialic acid and fucose and their ratio in oral cancer patients and in healthy control group to evaluate their role in diagnosis. **Materials and Methods:** Serum samples were collected from 52 healthy controls (group I) and 52 squamous cell carcinoma patients (group II). Estimation of serum levels of sialic acid and fucose and their ratio was performed. This was correlated histopathologically with the grades of carcinoma. Statistical analysis was done by using analysis of variance (ANOVA) test and unpaired "*t*" test. **Results:** Results showed that serum levels of sialic acid and fucose were significantly higher in oral cancer patients compared to normal healthy controls ($P < 0.001$). The sialic acid to fucose ratio was significantly lower in cancer patients than in normal controls ($P < 0.01$). However, comparison with histological grading, habits, gender, and age group did not show any significant result. **Conclusion:** The mean serum sialic acid and fucose levels showed an increasing trend from controls to malignant group and their corresponding ratio showed decreasing trend from controls to malignant group. The ratio of sialic acid to fucose can be a useful diagnostic aid for oral cancer patients.

Key words: *Fucose, N-acetylneuraminic acid, oral cancer, oral squamous cell carcinoma, serum, sialic acid, sialic acid to fucose ratio*

INTRODUCTION

Cancer is one of the most formidable health problems facing mankind today.[1] Different cancers show different rates of incidence and distribution according to geographic parameters.[2] Cancers of the head and neck are heterogeneous group of neoplasms that display a wide range of biologic behaviors.[3] In the Indian subcontinent, it is observed that because of cultural, ethnic, and geographic factors and the popularity of the addictive habits, the frequency of oral cancer is one of the highest in the world.[1]

Substances changing quantitatively in the serum during cancer growth are known as tumor markers or

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biochemical serum markers. Classically, the marker is synthesized by the tumor and released into the circulation or expressed at the cell surface in large quantities by malignant cells as compared to normal cells.[4]

Tumor-specific substances are considered to be produced directly as a result of oncogenesis, while tumor-associated markers are various proteins and enzymes, hormones, and immunoglobulins which occur in the blood and may be mediated by the tumor itself or by the influence of the tumor on the involved tissue.[4-7]

The glycoproteins are usually defined as protein carbohydrate complexes in which oligosaccharides are joined by covalent linkage to specific amino acids of proteins. The carbohydrate portion contains amino sugars (sialic acid, etc.) and hexoses (galactose, etc.) or fucose. Increased quantities of glycoconjugates like sialic acid and fucose levels had been detected in the plasma or serum of the patients with different types of malignancies, indicating their usefulness in diagnosis and monitoring therapy.[7-10]

Serum sialic acid is the group name used for acetyl and glycolyl derivatives of neuraminic acid which are bound to alpha- and beta-globulins in the form of glycoproteins in large quantities and found free in the serum in small amounts.[11] Serum fucose is a monosaccharide, that is methylpentose, which is usually a terminal sugar in most plasma glycoproteins and is present in blood as well as in tissue glycoproteins.[12]

Hence, in the light of previous study, the present study was conducted to evaluate serum sialic acid and fucose levels and their ratio in oral cancer patients.

MATERIALS AND METHODS

The present study was conducted in the Department of Oral Medicine and Radiology, Bapuji Dental College and Hospital, Davanagere, Karnataka.

The study consisted of a total of 104 subjects. The study subjects were divided as group I and group II. Group I consisted of 52 age- and sex-matched, apparently healthy individuals and group II consisted of 52 oral cancer patients.

Inclusion criteria

All patients who were diagnosed clinically and histologically as oral squamous cell carcinoma were included.

Patients having tuberculosis, tubercular meningitis, chronic osteomyelitis, ulcerative colitis, cirrhosis of liver, or chronic uterine bleeding were excluded from the study.

Ethical clearance was obtained from the institutional ethical committee. An informed consent was also obtained from each patient for carrying out biopsy and serum evaluation.

Provisional diagnosis of oral malignancy was made on the basis of clinical examination and was confirmed histopathologically by performing biopsy.[9] Squamous cell carcinoma was clinically staged according to tumor nodal metastasis (TNM) status as per the international union against cancer [Union for International Cancer Control (UICC)].[10] Squamous cell carcinoma patients were graded histopathologically into well differentiated, moderately differentiated, and poorly differentiated.[11]

Method of collection of blood

In each case, 5 ml of venous blood was drawn from the medial cubital vein by using sterile syringe and standard tourniquet, which was then transferred to a test tube and allowed to clot. This was followed by centrifugation at 3000 rpm for 5 min. The extracted serum was subjected to biochemical estimation of sialic acid[12] by ninhydrin method, and fucose level was estimated in the serum by using orthocresol 20%.[13]

An incisional biopsy from the lesion was obtained following the methodology of Howe[9] for all the study subjects. Depending on the histopathological findings, squamous cell carcinoma was graded into grade I. II, and III.

Statistical analysis

One-way analysis of variance (ANOVA) was used for multiple group comparisons and unpaired "*t*" test for two-group comparison.

RESULTS

The mean serum sialic acid level in normal controls was 27.8 mg/100 ml, the mean fucose level was 7.3 mg/100 ml, and the mean serum sialic acid and fucose ratio was 3.9. The mean serum sialic acid level in the malignant group was 65.3 mg/100 ml, the mean fucose level was 21.2 mg/100 ml, and the ratio was 3.2 [Table 1].

In the malignant group, the mean serum sialic acid level in clinical stage III was 55.6 mg/100 ml, the mean fucose level was 21 mg/100 ml, and the ratio was 2.76.

The mean serum sialic acid level in clinical stage IV was 101.2 mg/100 ml, the mean fucose level was 21.8 mg/100 ml, and their ratio was 4.83 [Table 2].

The mean serum sialic acid level in histological grade I was 67.2 mg/100 ml, the mean fucose level was 21.07 mg/100 ml, and their ratio was 3.25. The mean serum sialic acid level in histological grade II was 63.5 mg/100 ml, the mean fucose level was 21.96 mg/100 ml, and their ratio was 3.14. The mean serum sialic acid level in histological grade III was 54.4 mg/100 ml, the mean fucose level was 20.41 mg/100 ml, and their ratio was 2.96.

There was statistically highly significant correlation in the mean serum sialic acid, fucose, and their ratio between controls and the malignant group.

There were statistically highly significant values of sialic acid and their ratio with clinical staging, whereas serum fucose was statistically not significant with clinical staging.

There were no statistically significant results found on comparison of the levels with age group, gender, habits, and histologic grading.

DISCUSSION

Although oral cavity is frequently examined by dentists and physicians, 60% of intraoral carcinomas are well advanced at the time of detection.[13,14]

Early detection of oral cancer offers the best prognosis with the least invasive treatment.[15] In the search for the possible causes of cancer, on one hand, and the need for a modality affording early diagnosis, on the other, attention was paid to the markers that have a diverse set of applications from detection of cancer to its therapy.[16]

Recently, tumor markers are receiving more attention in early detection as well as predicting the prognosis of the lesion. Considerable research has been done in the last few decades focused on defining the cell surface membrane molecule in neoplastic malignant transformation, particularly the cell surface glycoprotein which contributes to the malignant transformation of the cell.[17]

The reason for elevated serum glycoprotein levels in malignancies has not been clearly established, but various views have been put forward by several researchers, wherein they have reported that elevation above the normal level reflects the process of tissue destruction at the site and release of preformed glycoprotein from the tissue or it may be due to local synthesis and release of glycoprotein by the tumor cells or increased glycoprotein levels in diseases are, in whole or in part, associated with tissue proliferation rather than tissue destruction[16,18] or it may be due to overproduction of glycoprotein or due to polymerization of the ground substance of the connective tissue at the site of tumor invasion with release of solubilized component into the circulation.[17-19,24]

Increased levels of glycoproteins like serum sialic acid and serum fucose have been associated with different types of malignancies like lymphomas, malignant

Table 1: Correlation of sialic acid, fucose, and their ratio between controls and the experimental group

Group	Sialic acid		Fucose		Sialic acid/fucose ratio	
	Range	Mean±SD	Range	Mean±SD	Range	Mean±SD
I, controls (n=52)	15–38	27.8±6.5	5–9.3	7.3±1.2	2.1–7.2	3.9±1.1
II, patients (n=52)	35–182	65.3±17.9	12.8–33.3	21.2±4.4	1.3–8.2	3.2±1.5
t	9.43		22.14		2.74	
P	<0.001, HS		<0.001, HS		<0.001, S	

n=Number of cases, S=Significant, HS=Highly significant

Table 2: Correlation of sialic acid, fucose, and their ratio with clinical staging of oral cancer

Clinical staging	n	Sialic acid		Fucose		Sialic acid/fucose ratio	
		Range	Mean±SD	Range	Mean±SD	Range	Mean±SD
III	41	35–76	55.6±11.0	12.8–33.3	21.01±4.46	1.25–5.2	2.76±0.83
IV	11	66–182	101.2±41.2	12.8–29.0	21.80±4.07	2.90–8.2	4.83±2.11
t*		6.43		0.53		5.06	
P		<0.001, HS		0.60, NS		<0.001, HS	

*Unpaired t-test. n=Number of cases, S=Significant, HS=Highly significant

melanomas, cancer of prostate, breast cancer, bone and gastrointestinal tract (GIT) diseases including cancer of the oral cavity.[19,20]

In the present study, there was no significant correlation between age and serum sialic acid, fucose levels and their ratio.

The study done by Ghosh *et al*.[21] demonstrated significant difference between gender and serum sialic acid, fucose levels and their ratio. But in our study, there was no significant difference in the gender, which may be because of variation in sampling.

Shashikant and Rao[22] also showed no significant gender differentiation.

In the present study, we could not find significant results between habits and the levels of serum sialic acid and fucose and their ratio.

In our study, comparison of group I and group II showed highly significant results with a "*P*" value of <0.001.

These studies were consistent with the studies done by Macbeth and Bekesi,[23] Bradley *et al*.,[24] Ghosh and Nayak,[21] Shashikanth and Rao,[22] and Bose *et al*.[25]

On further comparison of serum levels of sialic acid, fucose, and their ratio with clinical stages of oral squamous cell carcinoma, we found highly significant results ($P < 0.001$).

Our findings with respect to serum fucose level were not similar to those of Dutta *et al*.,[26] Ghosh and Nayak,[21] Shashikanth and Rao,[22] and Bathi *et al*.[27] These differences in the markers could be due to the variations caused by the tumor or the tumor growth which results in synthesis of glycoprotein by the body as a response to local or systemic effect of tissue destruction, arising from the tumor itself and secretion into the blood stream.

Comparison of the histological grading of oral squamous cell carcinoma and serum sialic acid, fucose level, and their ratio showed no significant results. However, the study done by Rajpura *et al*.[28] showed statistically significant results on comparison with histological grades.

The variation in our study results may be due to the differences in the methodology and the small sample size.

CONCLUSION

The findings of this study revealed significant elevation of serum sialic acid and fucose levels with a corresponding decrease in their ratio in oral cancer patients and suggested potential utility of these parameters in diagnosis as well as determining the clinical stage of the malignant disease.

To the best of our knowledge, the reviewed literature showed only one study involving the estimation of sialic acid and fucose ratio. More studies needs to be carried out regarding estimation of the ratio of sialic acid to fucose to evaluate its use as marker for early detection of oral cancer.

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Conflicts of interest

There are no conflicts of interest.

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