ESTROGEN RECEPTOR α EXPRESSION IN SALIVARY GLAND TUMORS

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ABSTRACT

Immunohistochemical detection of Estrogen Receptors (ER) α has demonstrated similarity between salivary glands and breast tumors. ER status is being routinely used for breast cancers in predicting response to therapy, suggesting their possible role in tumor management of SGTs. The aim of this study was to determine expression of estrogen receptor α in salivary gland tumors in both genders and various age groups.

The data of 57 already diagnosed cases of salivary gland tumors both benign and malignant were taken. They were examined microscopically for selecting the sections with maximum epithelial content for Immunohistochemical staining for ER α. Allred scoring system was used for assessing ER positivity.

Among 57 cases of salivary gland tumors 34 (59.65%) were benign and 23 (40.35%) were malignant. The ER α status was found positive in 31/57 (54.4%) of cases. Receptors positivity was present in 22 (65%) of benign and 9/23 (39%) of malignant tumors. The age range was 13-90 years. Maximum positivity for receptors (85.7%) was shown in the youngest age group (21-40 years) as compared to the oldest (above 60) which was 50.0%. Fourteen (56%) males and 17 (53.1%) females showed positivity for the ER expression.

Present study concludes that salivary gland tumors, benign or malignant, may express estrogen receptor α in any gender and age group irrespective of their histological and biological type.

Key Words: *Salivary gland tumors, Estrogen receptors, Allred scoring*

INTRODUCTION

Salivary gland tumors (SGTs) constitute 1% of all the body tumors and 3-6% of all the head and neck tumors. Distribution of SGTs among age and sex varies with different geographic locations. Universally about 80% of SGTs are benign while the rest belong to malignant category.[1-3]

Overall parotid gland is the commonest site (70-80%) for the occurrence of SGTs. However higher percentage of minor SGTs have been observed in African as compared to western countries.[2,4]

Studies show that ER-β is expressed by normal salivary gland tissue and oral mucosal epithelium which suggests a role of ER in these non-targeted tissues for maintaining their integrity.[5] A similarity has been demonstrated between salivary glands and breast tumors by immunohistochemical (IHC) detection of ER-α.[6] Presence of ER-α and absence of ER-β is associated with malignant potential of breast and other body tumors which suggests their possible role in pathogenesis of SGTs.[7,8]

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The mitogenic effect of estrogen is mediated by ER-α. Breast cancer expressing ER-α has been effectively treated by ER-α antagonists.[9] ER status not only determines prognosis for breast cancers but is also useful to predict tumor response to hormonal therapy. It may indicate the importance of estrogen hormone status regarding anti-hormonal therapy in management of SGTs.[10]

Although ER expression has been studied in a wide variety of SGTs, they show variable expression of these receptors. The ER positivity in SGTs ranges from either a few cases or none in benign tumors as well as in various malignant tumors. The conflicting results about ER expression may be related to ER subtypes and different criteria used to assess the presence of ER.[5,11,12]

The non-availability of uniform criteria has made scoring and standardization of threshold for ER positivity difficult. We can overcome these problems if a single criteria as negative or positive is used.[12] Once the tumor is diagnosed as ER positive, antihormonal therapy may be recommended.[13]

In the light of above discussion, this study has been designed to investigate expression of ER α in benign and malignant tumors of the salivary glands.

METHODS

This comparative cross-sectional study consisted of 57 cases of salivary gland tumors (both benign and malignant) referred to pathology department of Peshawar Medical College (PMC), Ayub Medical College (AMC) and City Medical Laboratory (CML) Peshawar Pakistan from the year 2011 to 2015. Patients receiving chemotherapy, radiotherapy and very small Trucut biopsies were excluded.

Microscopic examination of already diagnosed cases of SGTs was assessed for various histological types for selecting the sections with maximum epithelial content for Immunohistochemical staining for ER-α. At least two slides each of 4-5 microns thin were prepared from each block of selected cases. One was stained with H&E for histological study and the other was kept for immunolocalization of ER proteins. Additional 6 slides for positive control with known ER positive carcinoma breast were mounted batch wise.

IHC of the prepared slides was performed for ER-α with Dako EnVision™ FLEX detection system (Monoclonal Rabbit Antihuman Clone EP1). Prepared slides were divided into 6 batches for the procedure. One slide for positive control with known ER positive carcinoma breast was added to each batch.

ER positivity was assessed by using Allred Scoring System. Scoring was based on examination of all tumor cells on the slide. A Proportion Score (PS) was assigned representing the proportion of tumor cells with positive nuclear staining. An Intensity Score (IS) was assigned representing the average staining intensity of all positive tumor cells. A Total Score (TS) was the sum of PS plus IS (ranging from 0, 2-8). A positive result for ER was defined as TS more than 3, as per guidelines for ER/PR positivity mentioned in pharmDx™ manual.[14]

Statistical analysis was performed using the Statistical Package for Social Sciences (SPSS) version 19. Chi square test was used to compare categorical variables (gender, behavior of tumor and immunoreactivity). Fisher's exact test was applied where values were less than 5. Probability values of less than or equal to 0.05 ($P \leq 0.05$) was considered statistically significant.

RESULTS

Out of 57 diagnosed cases of salivary gland tumors 34 were benign and 23 were malignant. Pleomorphic Adenoma was commonest benign tumor. Among the malignant lesions, adenoid cystic carcinoma (ACC) was the commonest type followed by mucoepidermoid carcinoma (MEC) (table-1).

Gender wise 25 (43.8%) were males and 32 (56%) females. The age ranged from 13-90 years. Maximum patients (both males and females) were in the age group of 21-40 followed by 41-60 years. All 57 cases included in the study were evaluated for ER-α with IHC. Thirty one out of 57 (54.4%) cases were positive for ER-α. Age range of 1-20 years showed maximum ER positivity, i.e., 85.7% (table-2).

According to this study 14 (56%) males and 17 (53.1%) females showed positivity for the receptor expression (table-3).

Among 34 benign tumors, positivity was present in 22 (65%) cases, while 9/23 (39%) of malignant cases showed positivity for the ER-α (table-4).

The p values showing relationship between ER-α expression with gender, age and behavior of tumor were not significant.

DISCUSSION

SGTs are uncommon accounting for 1% of all the body tumors. The biologic role of ER in oral mucosa, salivary glands and its neoplastic growth is well known.[5] The similarity observed between salivary glands and breast tumors by IHC detection of ER-α and successful use of anti-hormonal therapy in ER positive breast cancer patients has led the investigators to study expression of these receptors in SGTs. Because of the geographic variation in age and sex distribution of these tumors and because of the unavailability of any published data regarding expression of hormonal receptors in

TABLE 1: GENDER DISTRIBUTION OF HISTOLOGICAL TYPE AND ER STATUS OF SGTS

S. No	Histological Type	M	F	Total	ER Status	
					Pos (%)	Neg (%)
1	Pleomorphic Adenoma	14	17	31	20 (64.5)	11 (35.5)
2	Myoepithelioma	02	01	03	02 (66.6)	01 (33.3)
3	ACC	02	07	09	05 (55.5)	04 (44.5)
4	MEC	02	04	06	02 (33.3)	04 (66.6)
5	PLGA	03	02	05	01 (20)	04 (80)
6	Carcinoma ex-PA	01	01	02	0.0	02 (100)
7	Myoepithelial ca	01	00	01	01 (100)	00
	Total	25	32	57	31 (54.4)	26(45.6)

TABLE 2: EXPRESSION OF ER-α IN VARIOUS AGE GROUPS

Age groups (in years)	Estrogen Receptors expression		p value
	Positive (%)	Negative (%)	
1-20	06 (86)	01 (14)	
21-40	13 (52.0)	12 (48.0)	0.34
41-60	11 (47.8)	12 (52.2)	
>60	01 (50.0)	01 (50.0)	

TABLE 3: EXPRESSION OF ER-α IN RELATION TO GENDER

Gender	Estrogen Receptors expression		p value
	Positive (%)	Negative (%)	
Male	14(56)	11 (44)	
Female	17 (53.1)	15(46.9)	0.83
Total	31(54.4)	26 (45.6)	

TABLE 4: EXPRESSION OF ER-α ACCORDING TO BIOLOGICAL BEHAVIOR

Type of tumor	Estrogen Receptors expression		p value
	Positive (%)	Negative (%)	
Benign	22 (64.7)	12 (35.3)	
Malignant	9 (39)	14(61)	0.06
Total	31 (54.4)	26 (45.6)	

Pakistan, we aimed to find ER-α expression in SGTs in Khyber Pakhtunkhwa Province Peshawar Pakistan to make it a base line study for future researches in the management of such cases.

In this study, evaluation of ER-α was done for all the cases of SGTs and we found 31 cases (54.4%) to be positive which is significantly higher than other international studies showing variable results.[12] This disparity in the results could be due to differences in antibodies used, method of antigen retrieval and diverse criteria used to assess the positivity of ER-α.

In our study expression of receptors were found to be slightly more in males (56%) as compared to females (53.1%). This is in contrast to studies by Leu et al which showed female predominance in ER-α expression which was insignificant.[7]

Maximum positivity of receptors was seen in the age group 1-20 years (85.7%) while more than 60 years showed minimum positivity (50%). In literature, we could not find data showing analysis of relationship of ER expression in SGTs in various age groups to compare it with the present study.

This study showed a higher percentage of ER expression in benign tumors (64.7%) as compared to malignant (39%) which is in contrast to Kolude who in his study obtained a higher percentage of ER expression in high grade malignant tumors as compared to low grade malignant and benign tumors.[15] This variation in the results could be attributed to the difference in the scoring method and selection/availability of tumor cases.

Among malignant tumors, ACC was commonest and showed positivity in 55.6% of cases which is lower than studies of Leo et al showing positivity in 75%.[7] The difference in results may be because they utilized polyclonal antibodies which have higher sensitivity as compared to monoclonal antibodies used in our study.

The status of hormone receptors plays a significant role in decision making about adjuvant hormonal therapy for which the clinicians need a clear message about positivity or negativity of the receptors for recommendation of antihormonal therapy. In this study Allred scoring was used, as it has the ability to clearly define a case as either positive or negative eliminating weak positive cases.[16]

CONCLUSION

This study concludes that SGTs benign or malignant may express ER-α in any gender and age group irrespective of their histological and biologic type. Allred scoring can provide a clear message to clinicians for recommendation of antihormonal therapy.

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