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Original Research Article

Immunohistochemical expression of HER1, HER2, HER3 and HER4 in head and neck squamous cell carcinoma

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Abstract

Background: Lip, oral cavity cancers are ranked as the sixteenth most common cancer worldwide according to the latest GLOBOCAN estimates (2022). The determination of expression of ERBB receptors in head and neck squamous cell carcinoma (HNSCC) can help profile the patients, potentially devise newer treatment strategies and improve outcome for patients.
Aim and Objective: The aim was to study immunohistochemical expression of EGFR, HER2, HER3 and HER4 in HNSCC and association with clinical factors, prognostic factors and their co expression.
Materials and Methods: 100 cases of head and neck squamous cell carcinoma were included in the study. Tumour representative blocks were selected for immunohistochemistry for EGFR, HER2, HER3 and HER4. IHC scoring was done using H-score. Results: In our study, 49% patients showed EGFR expression, 22% showed HER2 expression. HER2 expression showed association with lymph node involvement and extra nodal extension. 94% patients showed HER3 expression and 58% showed HER4 over expression. HER4 over expression showed correlation with sex of patient.
Conclusion: There is a need to conduct more such studies and clinical trials about expression of ERBB receptors in HNSCC as there are targeted drugs available against these receptors. Also, a reporting protocol needs to be established for interpreting immunohistochemical expression of these markers.

Keywords: Head and neck squamous cell carcinoma (HNSCC), Head and neck cancers (HNC), EGFR, HER2, HER3, HER4.

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1. Introduction

Head and neck squamous cell carcinoma (HNSCC) constitutes neoplasms arising from squamous cell lining the oral cavity, hypopharynx, nasopharynx, oropharynx, lip, nasal cavity, paranasal sinuses, and salivary glands. Head and neck cancer (HNC) causes considerable morbidity and mortality worldwide.[1]

Lip, oral cavity cancers are the sixteenth most common cancers worldwide according to the latest GLOBOCAN estimates (2022). Globally this category accounts for 2% of all cancers and causes 1.9% of cancer deaths.[2]

India tops in the incidence rates where tobacco accounts for 80% of HNSCC cases.[3]

In central India, the incidence of head and neck cancers (HNC) in males and females is 28.3 per 100,000 and 9.4 per 100,000 respectively. The proportion of HNC to all site cancer in central India is highest in males (34.1%) and in fémales (10.6%).[4]

Tobacco, alcohol, areca nut, HPV infection, and Epstein–Barr virus (EBV) infection are the main risk factors associated with HNSCC.

HNC arise as a result of multistep process which leads to disruption of normal regulatory pathways causing unregulated cellular proliferation and growth.[5] A number of oncogenes and tumour suppressor genes such as p53, p16, cyclin D1, EGFR, HER2, HER3, HER4 have been implicated in carcinogenesis of HNSCC.[6,7]

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EGFR plays a major role in development and differentiation of mammalian cells. EGFR over expression is seen in over 90% of squamous cell carcinomas in head and neck and is associated with poor prognosis. This can be utilised as a target for anti EGFR therapy.[8-10]

HER2 (ERBB2) is also a member of EGFR family which on activation, is involved in normal tissue growth and differentiation.[11] It is most commonly overexpressed in a number of tumours especially breast carcinoma. Overexpression of its receptor ligands, cross talk with other ERBB family members can affect action of EGFR inhibitors.[12]

HER3 (ERBB3) is another member of ERBB family. Unlike EGFR, HER2 and HER4, it lacks intrinsic tyrosinase activity. It activates PI3K (Phosphoinositide 3- kinase) signaling pathway. HER3 is also seen to be activated in HNC and its over expression is associated with poor prognosis.[13-15]

HER4 (ERBB4) also activates PI3K pathway directly. There are very few studies which discuss role of HER4 in HNC. It is observed that its expression has a good prognosis in laryngeal squamous cell carcinoma (LSCC) and has a protective effect. But some studies have associated it with poor clinical result especially when co expressed with EGFR.[16,17]

The ERBB family receptors are involved in pathogenesis of number of solid malignancies. Interdependence and interaction amongst ERBB family members plays a role in prognosis of HNC. This can help in tailoring treatment strategies for patients requiring targeted immunotherapy.

Molecular subtyping is the gold standard for determining expression of EGFR, HER2, HER3 and HER4 in HNSCC. But molecular tests are expensive and not easily available. In a country like India this is not feasible.

Immunohistochemistry (IHC) expression cannot map out the exact molecular defect. But it offers a cheaper and surrogate alternative to molecular tests for ERBB receptors.

A combined study of ERBB receptors together has been studied in very few centres across India.

There is a dearth of published data of such studies in Central Indian population.

The determination of expression of ERBB receptors in HNSCC can help profile the patients, potentially devise newer treatment strategies and improve outcome for patients.

2. Aims and Objectives

The aim was to study immunohistochemical expression of ERBB receptors (HER1, HER2, HER3 AND HER4) in HNSCC and their association with clinical factors- age, sex, tobacco consumption, alcohol intake; association with prognostic factors such as tumour grade, tumour size, lymph node status and TNM stage; also to study expression of HER (ERBB) family receptors in various subsites of HNSCC and to study co- expression of HER (ERBB) family receptors.

3. Materials and Methods

This study was conducted in department of pathology, National Cancer Institute, Nagpur. It was retrospective and prospective type of study.

A total of 100 samples were studied. Samples received between January 2023 to May 2023 were included in study.

3.1. Inclusion criteria

Biopsy proven cases of primary squamous cell carcinoma of head and neck sites were taken for the study where surgical resection was done. Head and neck sites included were oral cavity, pharynx, larynx, tongue, lip, salivary gland and neck nodes.

3.2. Exclusion criteria

Small biopsy cases and resection specimen with inadequate tumour tissue were excluded from the study.

Relevant clinical details of patients were collected.

The resected specimen were preserved in 10% neutral buffered formalin. H & E stained slides of tumour were assessed for TNM staging. Blocks with adequate tumour tissue was selected for IHC.

Formalin fixed paraffin embedded (FFPE) blocks were stained immunohistochemically for EGFR, HER2, HER3 and HER4.

The antibodies used were-

1. EGFR- CONFIRM anti-Epidermal Growth Factor Receptor (3C6) Primary Antibody Mouse monoclonal (Roche),
2. HER2 - anti-HER2/ neu (4B5) Rabbit Monoclonal Primary antibody (Roche),
3. ErbB3/Her3 [p Tyr1328] antibody- polyclonal, host: rabbit.
4. ErbB4/Her4 antibody - polyclonal, host: rabbit.

FFPE blocks were cut at thickness of 3-4 micron. Dewaxing and hydration were done. Sections were taken on adhesive /charged slides. Slides were baked at 65 ℃ for one hour. Labels were generated for the slides. These slides were then loaded in trays on the Automated Roche Ventana Benchmark XT platform. Ultraview DAB detection kit was used.

The control taken for EGFR was normal skin tissue. Staining was seen in the basal layer of epithelium which was cytoplasmic. The control for HER2 was taken as HER2 equivocal breast tumour showing membranous staining. The control taken for HER3, HER4 was normal prostate tissue. In

HER3 IHC, the basal cells showed nuclear staining and weak nuclear stain was seen in luminal cells in control. In HER4 control, the basal cells showed nuclear staining and luminal cells showed cytoplasmic and/or membranous staining. The negative control for all markers were also done where the primary antibody was omitted.

IHC scoring
IHC scoring for all the four antibodies was done using H score.[18]
It was based on staining intensity and percentage positivity (0-100%).
Staining intensity was graded as
Negative staining=0, weak staining= 1+, moderate staining= 2+, strong staining=3+.
H score ranged from 0 to 300.
H score= (1x%1+)+(2x%2+)+(3x%3+).

A score of 0 was taken as negative. Score 1 and above was taken as positive for all markers. The slides were screened independently by three pathologists to eliminate bias.

Findings were recorded separately. An average of the three readings was taken and a consensus was made of final score. Data was transferred onto an excel sheet. Chi square test with and without Yate's correction was employed to compare various parameters. The various parameters with which immunohistochemical expression of ERBB receptors were compared were - age, sex, site of tumour, tobacco chewing, alcohol consumption, chemotherapy, immunotherapy, tumour grade, lymphovascular invasion (LVI), perineural invasion (PNI), tumour category, lymph node category, extranodal extension (ENE), worst pattern of invasion (WPOI), pattern of invasion(POI). P value <0.05 was taken as critical level of significance. SPSS version 29.0 was used for statistics.

4. Results

The age range in our study was 24-75 years (average: 48.5 years). Maximum cases of HNSCC belonged to 40-60 years of age group - 54%.(**Table 1**) The male to female ratio was 6:1. 86 patients (86%) gave history of tobacco consumption. Only 16 cases had received neoadjuvant chemotherapy and 3 cases had received immunotherapy. The most common site of tumour was oral cavity (64%), followed by tongue (33%), lip (2%) and larynx (1%). (**Table 2**)

59% of cases were moderately differentiated squamous cell carcinoma (MDSCC), 38% were well differentiated squamous cell carcinoma (WDSCC) and only 3% were poorly differentiated squamous cell carcinoma (PDSCC).

Maximum cases belonged to T3-T4 category (54%). 45 cases showed lymph node metastasis. Of these 14 cases showed extra nodal extension.

For interpretation of EGFR and HER2 on immunohistochemistry, cytoplasmic and membranous staining in tumour cells was seen and considered as positive. HER3 showed predominantly nuclear staining. Cytoplasmic and membranous staining was also seen. Keratin pearls did not take up HER3 staining.

HER4 also showed predominantly cytoplasmic and membranous staining. In the normal epithelium, the basal layers showed nuclear and cytoplasmic and membranous staining. Normal tissue such as muscle, salivary gland and inflammatory cells also took up staining. Few cases showed nuclear staining which was also considered as positive.

A score of 0 was taken as negative and score above 0 was positive. Only in case of HER4, negative score was not obtained. Hence, a mean of HER4 scores was taken, which was 230. A score above 230 was taken as over expression.

4.1. Correlation of EGFR with clinicopathological factors

49 cases (49%) were positive for EGFR while 51 cases (51%) did not show staining for EGFR. (**Figure 1** A,B) EGFR expression did not correlate with age, sex, tobacco chewing, alcohol consumption, site of tumour, neoadjuvant chemotherapy, immunotherapy, tumour grade, lymphovascular invasion (LVI), perineurial invasion (PNI), tumour category, lymph node status, extra nodal extension (ENE), worst pattern of invasion (WPOI) and depth of invasion (DOI).

4.2. Correlation of HER2 with clinicopathological factors

Only 22 cases (22%) were immunopositive for HER2. (**Figure 2** A,B) HER2 expression showed correlation with lymph node status and p value was statistically significant (p=0.02). It was observed that majority of cases with lymph node deposits were immunonegative for HER2. 40 cases (88.9%) out of 45 cases of N1-N3 category were negative for HER2.

HER2 also showed association with ENE. P value was statistically significant (p=0.035). Cases which did not show expression of HER2 did not have ENE.

HER 2 expression did not show correlation with rest of the clinicopathological factors.

4.3. Correlation of HER3 with clinicopathological factors

94 cases (94%) showed HER3 expression and 6 cases (6%) did not show HER3 staining. (**Figure 3** A,B) HER3 expression again did not show correlation with any of the clinicopathological factors.

4.4. Correlation of HER4 with clinicopathological factors

42 cases (42%) showed expression of HER4 and 58 cases (58%) showed overexpression of HER4.(**Figure 4** A, B, **Figure 5**)

HER4 expression showed association with sex. 12 out of 14 females (85.7%) showed HER4 over expression. 46 out of 86 males (53.5%) showed HER4 over expression. A higher percentage of female patients showed HER4 over expression in the tumour where p value was statistically significant (p=0.023). HER4 expression did not show association with other clinicopathological factors.

4.5. ERBB receptors expression across tumour grade

In WDSCC, 17 cases (17%) were positive for EGFR, 6 cases (06%) were positive for HER2, 36 cases (36%) were positive for HER3 and 23 cases (23%) were positive for HER4.

In MDSCC, 32%, 15%, 55% showed expression of EGFR, HER2, HER3 respectively. 34% showed HER4 over expression.

In PDSCC, none of the cases showed expression for EGFR. HER2 expression seen in only 1% of cases. HER3 expression was seen in 3% of cases. HER4 expression was seen in only 1% of cases.

Tumour grade and EGFR, HER2, HER3, HER4 immunohistochemical expression did not show any correlation individually and p value was not statistically significant. (**Table 3**)

4.6. Correlation between ERBB receptors

A correlation between immunohistochemical expression of EGFR, HER2, HER3 and HER4 was analysed. Overall, weak correlations between the studied variables (EGFR, HER2, HER3, and HER4) were seen. None of these values were statistically significant at the conventional level of significance ($p < 0.05$).

This suggested that there was no strong linear relationship between these variables in the given dataset.

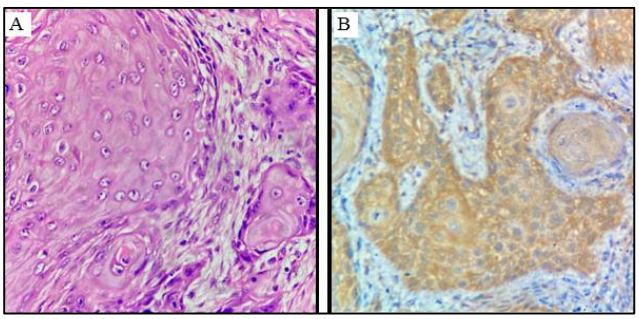

Figure 1: EGFR: (A): WDSCC, H & E stain (40x); (B): EGFR expression on immunohistochemistry in a case of WDSCC, showing cytoplasmic staining. (40x)

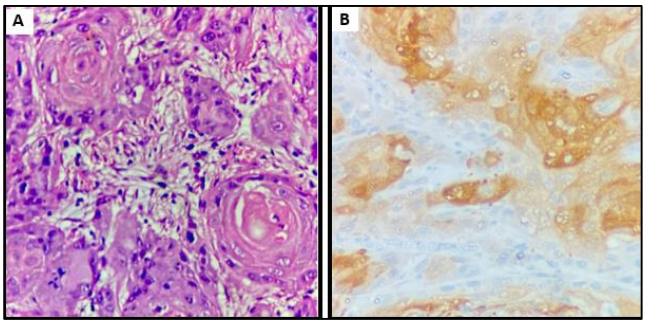

Figure 2: HER2: **(A):** WDSCC, H&E stain. (40X); **(B):** HER2 staining in WDSCC showing membranous and cytoplasmic staining. (40X)

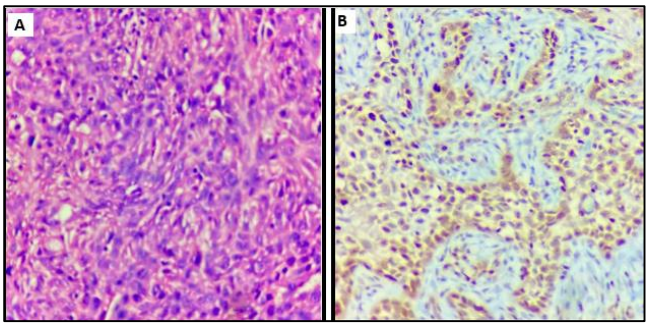

Figure 3: HER3: **(A):** PDSCC, H&E stain. (40x); **(B):** HER3 immunostain in PDSCC showing nuclear staining. (40x)

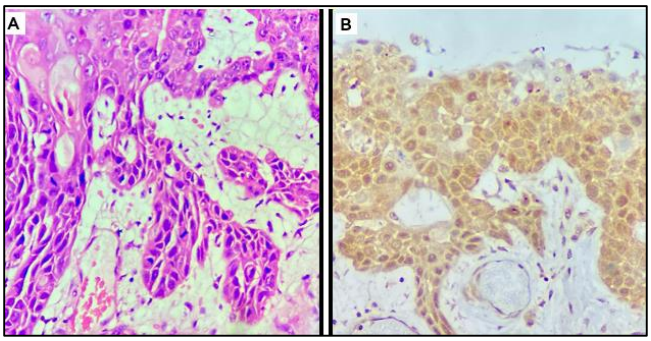

Figure 4: HER4: (A): MDSCC, H & E stain. (40x); **(B):** MDSCC: HER4 overexpression showing nuclear staining

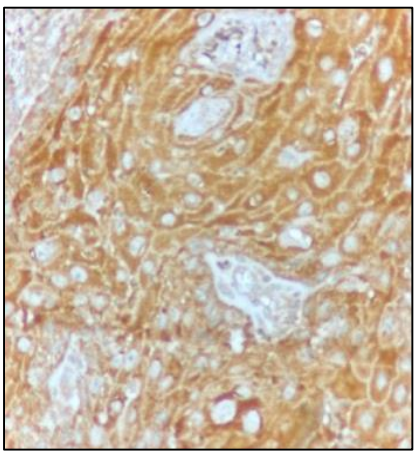

Figure 5: HER4 overexpression in a case of MDSCC showing cytoplasmic and membranous staining

Table 1: Age wise distribution of patients

	Age group (years)	Number	%
1	20-40	26	26.0
2	40-60	54	54.0
3	60-80	20	20.0

Table 2: Patient characteristics

		Frequency	Percentage (%)
Age groups (years)	20-40	26	26
	40-60	54	54
	60-80	20	20
Gender	F	14	14
	M	86	86
Site	Larynx	1	1
	Lip	2	2
	Oral cavity	64	64
	Tongue	33	33
Tobacco	No	14	14
	Yes	86	86
Alcohol	No	81	81
	Yes	19	19
NACT	No	84	84
	Yes	16	16
Immunotherapy	No	97	97
	Yes	3	3
Tumour grade	WDSCC	38	38
	MDSCC	59	59
	PDSCC	3	3
LVI	Absent	93	93
	Present	7	7
PNI	Absent	65	65
	Present	35	35
Tumour category	T1-T2	46	46
	T3-T4	54	54
Nodes	N0	55	55
	N1-N3	45	45
ENE	Absent	86	86
	Present	14	14
WPOI	WPOI 1-4	81	84.4
	WPOI 5	15	15.6
DOI	< 5 mm	16	17
	5-10 mm	42	44
	> 10 mm	38	39

Table 3: Expression of ERBB receptors across tumour grades

	EGFR			P value	HER2			P value
	Negative	Positive	Total	0.145	Negative	Positive	Total	0.463
WDSCC	21	17	38		32	6	38	
MDSCC	27	32	59		44	15	59	
PDSCC	3	0	3		2	1	3	
	HER3			P value	HER4			P value
	Negative	Positive	Total	0.420	Express-ion	Over Expression	Total	0.694
WDSCC	2	36	38		15	23	38	
MDSCC	4	55	59		25	34	59	
PDSCC	0	3	3		2	1	3	

Table 4: Comparison of EGFR expression in present study with other studies

Author	Year	EGFR expression on IHC-positive (%)
Laimer et al[19]	2006	73.2
Hiraishi et al[20]	2007	98
Sarkis et al[21]	2010	87.5
Hashmi et al[22]	2018	53.9
Verma et al[23]	2018	98
Present study	2024	49

Table 5: HER2 expression comparison of present study with previous studies

Author	Year	HER2 expression on IHC-positive (%)
Xia et al.[7]	1997	51
AJ Khan et al.[35]	2002	17
Cavalot et al.[34]	2007	39
Seifi et al.[11]	2009	17
Bernades et al.[31]	2013	2.2
Vats et al.[32]	2018	20
Present study	2024	22

5. Discussion

HNC pose a major problem worldwide. Resection of these tumours is challenge as the anatomy of head and neck is complex and involves many vital structures. Also these complicated surgeries lead to disfiguration and difficulty carrying out basic activities like eating, drinking, talking, etc.

TNM reporting is still important in staging, indicating prognosis and deciding patient's line of treatment.

5.1. EGFR

In our study, 49 cases (49%) showed immunohistochemical expression for EGFR while 51 cases (51%) did not show expression for EGFR. Laimer et al, Hiraishi et al, Sarkis et al, Hashmi et al and Verma et al reported 73.2%, 98%, 87.5%, 53.9% and 98% of cases positive for EGFR respectively.[19-23] (**Table 4**)

It has been observed that 40-80% of EGFR overexpression is seen in head and neck cancers.[24] Various studies showed differences in reporting EGFR expression. These variation in reporting percentage of EGFR expression could be due to different ways of interpreting immunohistochemical expression. There is no established protocol of EGFR IHC reporting in HNSCC.

In our study, EGFR did not show association with any of the clinicopathological factors. Hiraishi et al reported EGFR high expression association with tumour invasion but no correlation with other clinicopathologic factors.[20] Sarkis et al too did not find any significant association between EGFR expression with other clinicopathologic factors.[21] Zafar et al did not find any correlation of EGFR with tumour grade.[25] Verma et al reported EGFR expression association with tumour grade. But this study also did not find any association with tumour stage, lymph node metastasis.[23] Reimers et al and Glazers et al too did not reported any association with lymph node metastasis.[26,27]

EGFR is a glycoprotein which causes cellular proliferation and differentiation in epithelial, mesenchymal tissue. It plays an important role in carcinogenesis where it's over expression or mutations cause unregulated cellular proliferation. EGFR over expression is associated with more aggressive disease. EGFR overexpression, in majority of studies, does not show correlation with clinicopathologic factors. These clinicopathologic factors play a key role in designing treatment regimes. Anti EGFR therapeutic agents have been shown to improve patient outcome and survival. Cetuximab is one such anti EGFR agent, a monoclonal antibody, has shown therapeutic benefit and improved survival in HNSCC when used along with other chemotherapeutic agents and radiotherapy. Other anti EGFR agents have also shown similar good results in trials.[28-30]

5.2. HER2

In our study, only 22 cases (22%) were immunopositive for HER2 and 78 cases (78%) did not show expression for HER2

in present study. In various literature, HER2 positivity percentage in HNC shows great variation.

This could be due to differences in IHC methods, clone of antibody used, antigen retrieval method. Also there is no established method of reporting in HNC. Some studies used ASCO/CAP guidelines for HER2 reporting in breast cancer while some used expression in cytoplasm and/or membrane of tumour cells as positive. Bernardes et al reported 97.8% tumours negative for HER2.[31] In a study by Vats et al, 20% cases were HER2 positive.[32] Mirza et al reported only 0.72% of cases positive for HER2 expression.[33] These studies used ASCO/CAP guidelines for reporting. (**Table 5**)

In a study by Xia et al, tumours showing membranous and cytoplasmic staining were considered HER2 positive. Scoring was based on percentage and intensity of HER2 expression. 51% of tumours expressed HER2- 2+ and 3+ in this study.[7] Seifi et al reported 17% showing HER2 expression.[11] Cavalot et al reported, 39% of tumours showing over expression of HER2.[34] Khan et al also reported 17% of cases as HER2 positive on IHC. In this study, 1+ and 2+ staining was taken as positive for HER2 and only membranous staining was taken as positive.[35] Sardari et al in their study reported all cases showing membranous and/ or cytoplasmic staining except one case which showed only cytoplasmic staining.[36]

In our study, HER2 expression showed association with lymph node deposits and presence of ENE.

Xia et al too reported association of HER2 expression with nodal stage.[7] In a study by Cavalot et al, their multivariate analysis showed that HER2 expression and lymph nodal status were independent prognostic factors for disease free survival.[34]

However study by Khan et al did not show any correlation of HER2 with margin status, sex, age, T stage, N stage and tumour grade.[35] Study by Sardari et al also did not show any correlation of HER2 expression with age, gender, tumour size, lymph node and distant metastasis, tumour stage and histologic differentiation.[36] Mirza et al too reported no significant association of HER2 expression with clinicopathological parameters.[33]

There are phase II clinical trials and preclinical studies being conducted, where effect of anti HER2 antibodies against HNSCC is being evaluated. The results have been variable.[37] A phase II trial where trastuzumab was give in combination with anti EGFR showed no improvement in response to chemotherapy.[38] Another study showed addition of pertuzumab (humanised monoclonal antibody to HER2 receptor) to gefitnib in HNSCC cell lines overcame resistance against geftinib.[39]

5.3. HER3

In present study, 94 cases (94%) showed HER3 expression and 6 cases (6%) did not show HER3 staining. Nuclear staining was seen predominantly. This could be attributed to type of antibody used.

A study by Chang et al of expression of HER3 in cervical cancer also showed nuclear staining in tumour.[40]

Also in present study, HER3 overexpression did not show significant association with age, sex, site of tumour, tobacco intake, alcohol consumption, NACT, immunotherapy, LVI, PNI, tumour grade, tumour category, lymph node status, WPOI, ENE and DOI.

In a study by Takikita et al, HER3 membranous and cytoplasmic expression were reported separately. Takikita reported 8.8% of tumours showed membranous staining and 77.5% of tumours showed cytoplasmic staining. This study also assessed the prognostic relevance of HER3 expression and age, gender, tumour grade, site, lymph node status by using multivariate proportional hazard model, adjusting these factors. HER3 expression, age and site of tumour were independent prognostic factors.[41]

Wei et al did a study of HER3 expression in laryngeal carcinoma and reported 26.7% of cases showing strong staining.[42]

In a study by Deuss et al, HER3 immunohistochemistry showed membranous and cytoplasmic staining in 86% of cases and over expression in 17%.[43]

Studies have been done of correlation HER3 overexpression with survival in breast cancer, gastric cancer, colorectal cancer, pancreatic cancer, cervical cancer, melanoma and head and neck cancer. HER3 expression in gastric cancers have shown poor outcome whereas for other tumours the results have been variable.[44] Some studies have reported cytoplasmic staining in ovarian tumours, oesophageal tumours whereas few other studies observed cytoplasmic as well as membranous staining in gastric, colorectal and breast cancer.[45-49]

These various patterns of staining could be attributed to different clones of HER3 antibody used and different methods of IHC. Also there is no established reporting protocol of HER3 immunohistochemistry in HNSCC.

HER3 over expression in HNC has been associated with poor prognosis.[50] A number of anti HER3 drugs are being applied in preclinical, phase II and clinical trials. One approach is using monoclonal antibodies, another approach is using TKI (tyrosine kinase inhibitors).[51] One such is Duglitozumab, antibody against EGFR and HER3, in a phase Ib trial showed promising effect in HNSCC but another trial showed no significant response.[52,53]

5.4. HER4

In present study, for HER4 IHC, it was observed that the tumour showed strong cytoplasmic and membranous staining. 42 cases (42%) showed expression of HER4 and 58 cases (58%) showed over expression of HER4.

Membranous and cytoplasmic staining of HER4 in oropharyngeal squamous cell carcinoma (OPSCC) was seen ubiquitously in 97% and 27% of over expression in a study by Deuss et al. This study reported nuclear expression of HER4 separately which was infrequent in 45% of tumours with 10% showing over expression.[43]

In a study by Bussu et al, HER4 expression was seen in 43.3% of laryngeal SCC cases.[16]

In present study, HER4 expression showed significant correlation with sex. P value was statistically significant, (p=0.023). HER4 expression did not show correlation with age, site of tumour, tobacco intake, alcohol intake, NACT, immunotherapy, tumour grade, LVI, PNI, tumour category, lymph node status, ENE, DOI and WPOI.

Deuss et al stated in their study that membranous/ cytoplasmic expression of HER4 showed correlation with advanced tumour stage and lymph node deposits.[43]

There are very limited studies done on expression of HER4 and its role in head and neck cancer.

5.5. Correlation between ERBB receptors

In present study, correlation between immunohistochemical expression of EGFR, HER2, HER3 and HER4 showed weak correlations. None of these values were statistically significant at the conventional level of significance ($p < 0.05$).

This suggested that there was no strong linear relationship between these variables in the given dataset. There are very few studies discussing coexpression of ERBB receptors. Deuss et al reported high correlation between coexpression of EGFR with HER3, nuclear HER4. Also this study did not find any correlation between HER2 and EGFR expression. HER3 showed correlation with EGFR, nuclear HER4. Nuclear HER4 showed correlation with EGFR.[43] Bernardes et al too reported no significant association between EGFR and HER2.[31] In a study by Takikita et al, cytoplasmic as well as membranous HER3 did not show any association with membranous HER2 expression.[41]

6. Conclusion

In this study we evaluated the immunohistochemical expression of EGFR, HER2, HER3 and HER4 in HNSCC, their association with clinicopathological factors and also expression of ERBB receptors amongst various subsets of HNSCC.

There is a dearth of such studies being done in this region.

Staging and grading of surgically resected tumours of head and neck are still one of the most important factors in guiding further treatment and prognosis of patients.

We did not find EGFR expression associated with clinicopathological factors. There is increased interest in role of EGFR for immunotherapy. Patients with over expression of EGFR can be given targeted therapy against it.

HER2 expression in HNC has not shown promising results. Most studies found no association with clinicopathological factors. HER2 is seen to heterodimerize with EGFR. Hence, therapy can be designed to target HER2 in cases not responding to anti-EGFR therapy.

Studies of HER3 expression in HNC has shown poor prognosis. Again HER2 is seen to heterodimerize with HER3. Its role as an additional agent in immunotherapy can prove to be useful for patients. There are very limited studies of HER4 expression in HNC. Some studies state that HER4 expression is associated with poor prognosis whereas few others have observed better prognosis in HNSCC.

Immunohistochemistry is cheaper and more easily available than molecular tests. Molecular tests are can help design specific targeted therapy. We did not do molecular tests of ERBB receptors. This was the limitation of our study.

Thus, there is a need for more such studies and clinical trials to be done with greater sample size. Also, a reporting protocol needs to be established for uniform results as all studies evaluated IHC using various methods.

7. Source of Funding

None.

8. Conflict of Interest

None.

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