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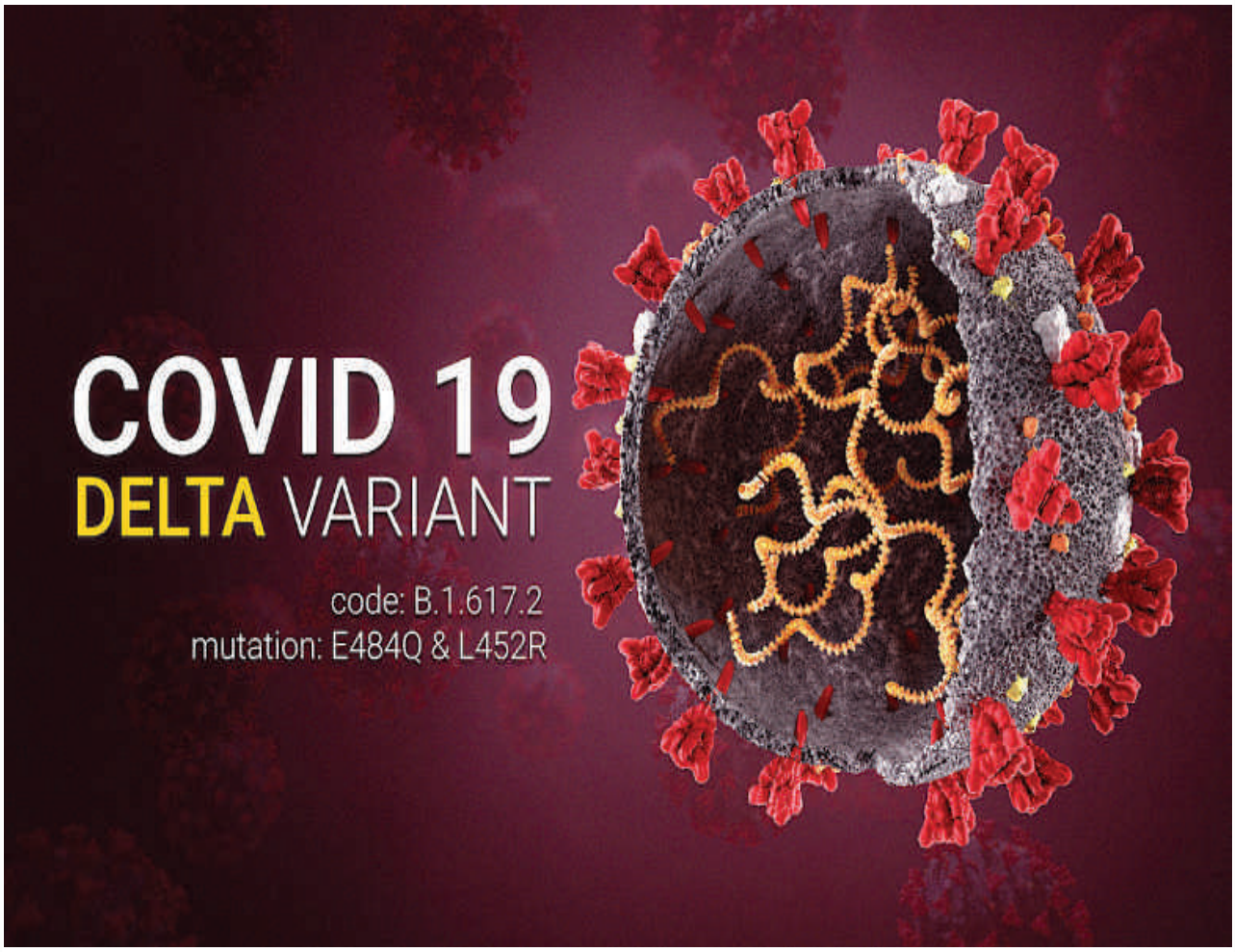

The Official Journal of the Gulf Federation For Cancer Control

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Original Article

Correlation Between Tumor Infiltration CD8+ T–cells And PD–L1 Expression In Laryngeal Cancer And Their Prognostic Significance: Prospective Non–interventional Trial

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Abstract

Introduction: Tumor microenvironment plays crucial role in cancer evolution. There is a dynamic and continuous relation between immune cells and cancer cells' resistance. Tumor infiltration CD8–lymphocytes and programmed death ligand–1 have proved important prognostic role in different malignancies. We aimed at evaluating this role in laryngeal cancer.

Patients and methods: We prospectively analyzed laryngeal cancer patients' specimens, to identify the CD8–lymphocytes and the PD–L1 expression. A total score formed of the sum of percentage and intensity of PD–L1. A final rate was considered as negative or low when combined percentage and intensity scores 0 to 4, and high when scores 5–7. CD8–lymphocyte infiltration was divided into strong (≥ 10/100 of epithelial cells or ≥20/100 stromal cell infiltration) or weak (< 10/100 epithelial cells or < 20/100 stromal cell infiltration).

Results: Forty patients were included; twelve had stage 1 or 2 and 28 with advanced stages. PD–L1 expression was positive in 92.5%. Neither the PD–L1 nor CD8–lymphocytes had overall survival impact, however high PD–L1 correlated with better survival in advanced stage subgroup (p = 0.036), high CD8–lymphocytes infiltration had better survival but did not reach significance. There was significant correlation between the CD8–lymphocyte infiltration; whether epithelial or stromal, and tumor PD–L1 expression; p–value of 0.001 and < 0.0001 respectively. Subgroup of patients with low CD8+ infiltration and low PD–L1 had the worst survival.

Conclusion: There is a correlation between CD8–lymphocytes infiltration and PD–L1 expression in laryngeal cancer and high PD–L1 expression is associated with better OS in advanced stages.

Key words: PD–L1, CD8, laryngeal cancer, tumor microenvironment

Introduction

Laryngeal cancer new cases estimated to be around 12.000 cases in the year 2019 with overall incidence of 0.6% of all cancer new cases. The 5–year survival rate is estimated to be around 60%; ranging between 33–77% based on the stage [1].

In comparison to normal cells, cancer cells are characterized by properties of autonomous hyper proliferative, invasive and limitless survival capacities. Tumor micro environment (TME) contains many distinct cell types, including fibroblasts, carcinoma–associated fibroblasts (CAFs), myofibroblasts, smooth muscle cells, endothelial cells and their precursors, pericytes, neutrophils, eosinophils, basophils, mast cells, T and B lymphocytes, natural killer cells (NKs) and antigen presenting cells (APC) such as macrophages and dendritic cells. Immune system plays a main role in protecting our bodies from cancer and rejecting it, however cancer cells affect the immune system and use its components to promote its growth and survival [2].

T– lymphocytes are considered the crucial component of antitumor immunity, with CD8+ T cells serving as cytotoxic effector cells and CD4+ Th1 cells serving to help and enhance the magnitude and duration of the antitumor responses. However, CD4+ Th2 cells and CD4+ T–regulatory cells can suppress effective CD8+ antitumor

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responses. Dysfunction of T–cell activation and effector function, leading to suppression of the immune system by the tumor [3].

Pretscher D et al concluded that suppression of local cellular immunity might be a mechanism by which tumor cells escape host immunity based upon tumor– leukocyte infiltration pattern varies between primary tumors and metastatic lymph nodes in HNSCC with a local decrease in the number of CD8+ T–cells and increase in CD20+ B–cells being the most relevant findings [4].

Patients with tumors expressing human papillomavirus–16 (HPV16) had a higher probability of infiltration by CD8+ T–cells compared with those patients with tumor negative for HPV or normal controls; and this may explain the better prognosis of HPV16 positive tumors [5].

The body response to foreign antigens including cancer cells is regulated through immune cells checkpoints. The most important defensive cells are the cytotoxic T–cells (CTLs). Activation of these cells occur through co–binding of T–cell receptors (TCR) to the foreign antigen presented to it through the antigen–presenting cells like dendritic cells and stimulation of one of the stimulatory receptors like CD28 by the B7 ligand on the dendritic cells [6].

CTLs has stimulatory ligands like CD28, CD27, CD137 and inhibitory or suppressor receptors like cytotoxic T–cell associated protein–4 (CTLA–4) and the programmed death receptor (PD–1). There is a continuous balance between the process of stimulation and suppression to prevent overstimulation of the immune system or occurrence of auto–immune diseases [6].

The CTLA–4 is formed early on the surface of T–cells in the process of immune response to infection (within 24–48 hours from CTLs activation) inside the lymph nodes before migrating to tissue [7].

CTLA–4 binds to the B7 strongly more than the CD28 leading to the displacement of CD28 and inactivation of the CTLs [8].

Like the CTLA–4 receptor, another receptor is found over the T–cells which is PD–1. It is found in the tissue and responsible for the long–term control of the immune response and inhibition of auto–immune disease. The PD–1 receptor is activated by 2 ligands PD–L1 or PD–L2 which is found on macrophages peripheral dendritic cells and many of cancer cells [8].

The activation of CTLA–4 through binding to the B7 leads to differential actions based on the type of stimulated cells; For example, inhibiting the CTLs and stimulating the T–suppressor cells (Treg) [9].

There is data suggesting that increasing the tumor microenvironment immune cells infiltration in head and neck squamous cell carcinoma is associated with improved survival and better prognosis [10].

Yang W et al [11] investigated the prognostic role of PD–L1 expression for survival in head and neck squamous cell carcinoma in a systematic review and meta–analysis included 23 studies which involved 3105 patients. There was no statistically significant difference in OS in patients with PD–L1 positive and negative HNSCC (HR: 0.98; 95% CI: 0.71–1.37; $p = 0.93$), whereas an improved PFS in advanced cases was observed in patients with positive PD–L1 expression (HR: 0.71; 95% CI: 0.55–0.93; $p = 0.01$). Also, they concluded that low CD8+ infiltration group of patients with high PD–L1 expression is associated with poor survival.

We aimed by this study to evaluate immune–histochemical expression of PD–L1 and CD8+ cells in laryngeal cancer and to determine the prognostic significance of these 2 markers in the tumor microenvironment.

Patient and methods

This study included 40 patients recently diagnosed with laryngeal squamous cell carcinoma irrespective of their stage. All patients should have had formalin–fixed and paraffin embedded tumor sections whether from laryngoscope or surgical specimen. Specimens were retrieved from the archives of pathology and clinical oncology department of Ain Shams University Hospitals, Cairo, Egypt.

Immuno–histochemical staining

Immunohistochemistry (IHC) staining was done on the paraffin embedded tissue sections with a labelled streptavidin– biotin–peroxidase complex technique using rabbit polyclonal PD–L1 antibody (Gene Tex: GTX104763) with concentrated dilution (25–100), and Mouse Monoclonal CD8 antibody (Thermo scientific: clone C8/144B) with dilution (1:25–50). Antigens were retrieved by microwaving in citrate buffer for 20 minutes for PD–L1. Counterstaining were performed using hematoxylin. After staining, the slides were washed, dehydrated in graded alcohol and xylene, mounted, and cover–slipped. To verify antibody specificity, placental tissue was used as a positive control for PD–L1 and tonsil for CD8.

Immuno–histochemical analysis

1. Immuno–histochemical scoring for PD–L1

- Cells with membranous expression of PD–L1 were considered positive and any cytoplasmic staining is disregarded.

- Scoring of PDL1 was done according to (Yu et al.,2019) as follows;
  - 0 point (positive rate <6%)
  - 1 point (positive rate 6%–25%)
  - 2 points (positive rate 26%–50%)
  - 3 points (positive rate 51–75%)
  - 4 points (positive rate >75%)
- Semi–quantitative staining intensity was divided into 4 grades:
  - 0 (negative), 1 (weak), 2 (moderate), 3 (strongly positive)
- A total score formed of the sum of percentage and intensity.
- A final comprehensive rate was considered as negative expression if score 0, low expression when combined percentage and intensity score range from 1 to 4, and high expression when combined percentage and intensity score range from 5–7 [12].

2. Immunohistochemical scoring for CD8:

CD8+T cell infiltration was evaluated. According to the distribution of tumor epithelial cells and peri–tumor stroma, the CD8+T cell infiltration was divided into strong infiltration (≥ 10/100 of epithelial cells; ≥20/100 stromal cell infiltration) and weak infiltration (< 10/100 epithelial cells; < 20/100 stromal cell infiltration).

Statistical methodology

A correlation between the PD–L1 expression with CD8+ T–cells in both the tumor epithelium and stroma was done and correlation of the PD–L1 expression and the different patient and tumor related prognostic factors was done.

Survival analysis of the population was done using Kaplan–Meir curve and statistical comparison to assess the prognostic impact of the PD–L1 and CD8+ expression on the event–free survival (EFS) and overall survival (OS) of the patients.

Results

Patients' characteristics

Forty patients were included in this study, their median age was 58 years (range 47–69). All of them were smokers or ex–smokers. The main presenting symptom of the disease was hoarseness of voice in 92% of patients. All of them were non–metastatic with good performance status ECOG 0–1 except one patient with ECOG 2. Two–thirds of patients were treated with laryngeal conservative management and one–third underwent total laryngectomy. Pathological and clinical staging are shown in table 1.

PD–L1 and CD8 staining results

Most of the patients (80%) showed strong membranous PD–L1 expression (high expression score 5–7) with 7.5% were negative (score 0) and 12.5% were weak (score 1–4).

Considering CD8 expression, the CD8+ infiltration in the tumor epithelium was seen in 22 patients (55%) and in the peri–tumor stroma in 24 patients (60%).

Correlation between PD–L1 and CD8+ expression

Using Fisher's exact test as show in table 2 and 3 showed a statistically significant correlation between the CD8+ T–cell infiltration whether in the tumor epithelium or stroma and the tumor cell PD–L1 expression with p values of 0.001 and < 0.0001, respectively. Most of the PD–L1 strong expression tumors have strong stromal and epithelial CD8+ infiltration (fig 1 and 2) (60% and 55%, respectively) while, All the negative or weak PD–L1 tumors have weak stromal and epithelial CD8+ cells expression (fig 3). The other group with strong tumor PD–L1 and weak epithelial and stromal CD8+ infiltration represented 25% and 20% respectively (fig 4).

Survival analysis

After a median follow up period of 28 months, the median progression free survival (PFS) and median overall survival (OS) were not reached. The 1–year and 2–year PFS rates were 84% and 67% respectively. The 1–year and 2–year OS rates were 89% and 77% respectively (fig 5 and 6).

Prognostic significance of PD–L1 and CD8+ tumor infiltration

There was no statistically significant correlation between the PD–L1 expression and survival in the whole population but there was significant correlation between PD–L1 expression and survival in advanced stage cases (stage III and IV) with p value of 0.191 and 0.036 respectively (fig 7). But the PFS was not significantly different between the different PD–L1 sub–groups (fig 8).

Considering CD8+ expression whether stromal or epithelial, there was no significant correlation with PFS or OS in the whole population or advanced cases (stage III or IV).

Age	Median age 58	Range (47–69)
Performance status (ECOG)	0–1	39 (97.5%)
	2	1 (2.5%)
Smoking history	Smokers or ex–smokers	40 (100%)
Presenting symptoms	Hoarseness of voice	37 (92.5%)
	Dysphagia	2 (5%)
	Difficulty in breathing	1 (2.5%)
Pathological type	Squamous cell carcinoma	40 (100%)
Grade	I	4 (10%)
	II	25 (62.5%)
	III	11 (27.5%)
Clinical tumor stage (cT)	1	1 (2.5%)
	2	13 (32.5%)
	3	16 (40%)
	4	10 (25%)
Clinical nodal stage (cN)	0	22 (55%)
	1	8 (20%)
	2	9 (22.5%)
	3	1 (2.5%)
Clinical stage	Stage I or II	12 (30%)
	Stage III or IV	28 (70%)
Laryngeal conservative management	Yes	27 (67.5%)
	Total laryngectomy	13 (32.5%)

Table 1: Patients' Characteristics
ECOG (Eastern cooperative oncology group)

		PD–L1 group		Total	Significance
		Negative or weak	Strongly positive		
CD8+ – epithelium	strong	0 (0%)	22 (55%)	22	Pearson Chi–square (Fisher's Exact test) 12.222 (0.001)
	weak	8 (20%)	10 (25%)	18	
Total		8	32	100%	

Table 2: Correlation between PD–L1 expression and CD8+ in the tumor epithelium
PD–L1 (Programmed death ligand–1)

		PD–L1 group		Total	Significance
		Negative or weak	Strongly positive		
CD8+ – stroma	strong	0 (0%)	24 (60%)	24	Pearson Chi–square (Fisher's Exact test) 15 (< 0.0001)
	weak	8 (20%)	8 (20%)	16	
Total		8	32	100%	

Table 3: Correlation between PD–L1 and CD8+ in tumor stroma
PD–L1 (Programmed death ligand–1)

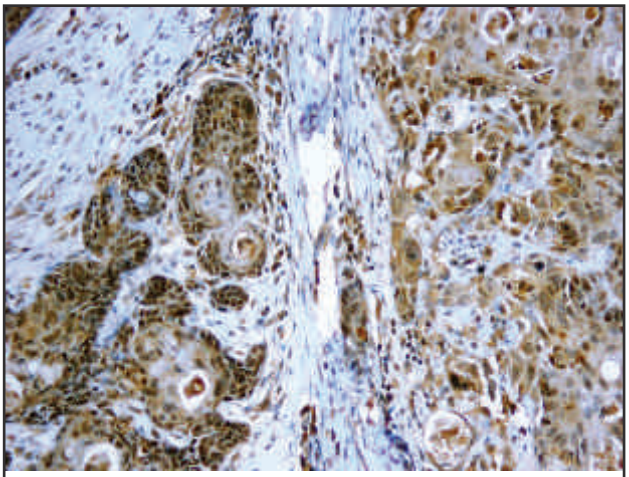

Figure 1 (a): PD–L1 expression in case of stage 3 moderately differentiated tumor showing moderate positive PD–L1 in 70% of malignant cells (high expression: score 5) (PDL1x200).

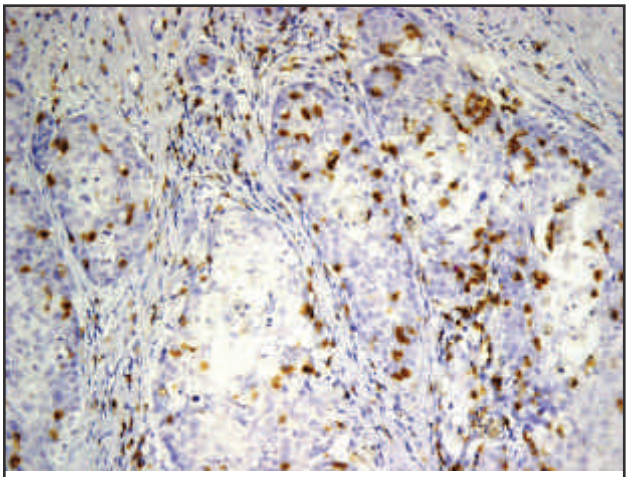

Figure 1 (b): same patient with strong CD8 epithelial (CD8>10/100 cells) (CD8x200)

Figure 1: a case of moderately differentiated laryngeal squamous cell carcinoma, stage III with positive high PD–L1 and strong epithelial CD8+ cells.

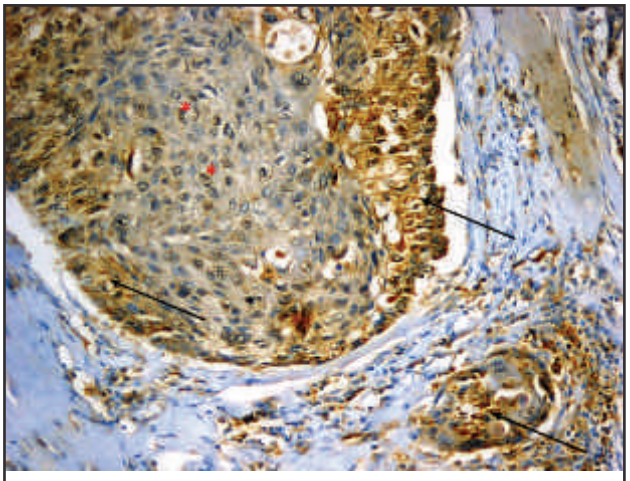

Figure 2 (a): A case of stage II moderately differentiated tumor with strong positive membranous PD–L1 expression in 20% of tumor cells (arrows). Other cell nests are negative for PD–L1 expression (*).(PD–L1x200)

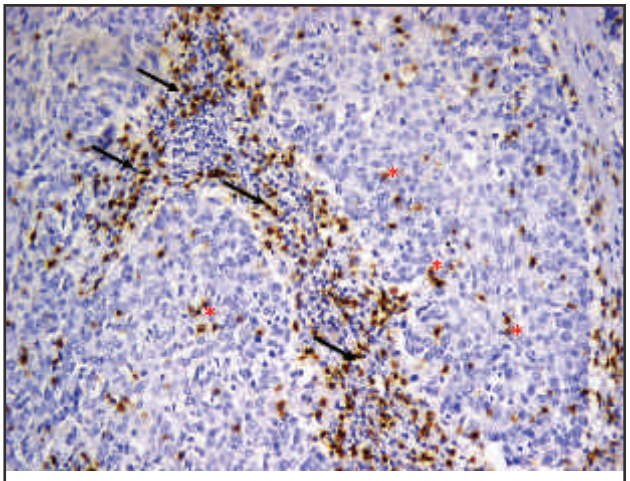

Figure 2 (b): CD8 in the same case showing high epithelial (CD8+ < 10/100 cells) (*) and high stromal expression (arrows) (CD8+ > 20/100cells) (CD8x200).

Figure 2: a case of stage II moderately differentiated tumor with strong PD–L1 expression and low epithelial and strong stromal CD8+ expression.

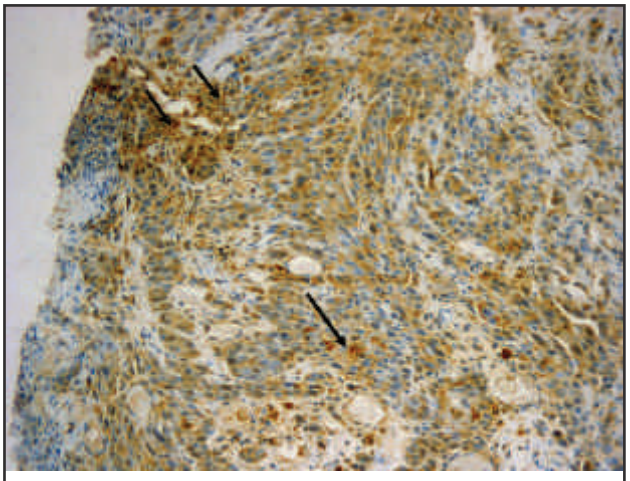

Figure 3 (a): a case of poorly differentiate stage 2 laryngeal cancer with low PD–L1 expression (score 3) with focal moderate membranous and non–specific cytoplasmic staining in 10 % of tumor cells "arrows". The remaining tumor cells show non–specific cytoplasmic staining without membranous staining (PDL1x200)

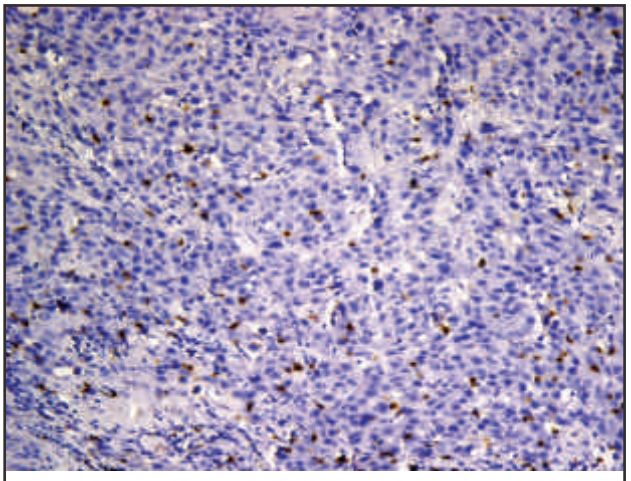

Figure 3 (b): same case showing high week epithelial (CD8<10/100 cells) and stromal expression (CD8<20/100cells) (CD8x200).

Figure 3: a case of stage II poorly differentiated laryngeal carcinoma with low PD–L1 expression (score 3), and weak epithelial and stromal CD8+ expression.

The patients in this study were categorized into 4 sub–groups according to the PD–L1 and CD8+ expression whether in the stroma or epithelium. Correlation for survival in the 3 other groups revealed better survival in patients with either positive PD–L1, CD8+ or both compared to patients with both negative PD–L1 and weak CD8 expression, but this correlation did not reach statistical significance (fig 9).

Discussion

Programmed death–1 (PD–1) is in dynamic status on the T–cell, it is expressed when the T–cells are active and not expressed when T–Cells are inactive [13] [14], also, activated T–cells express PD–L1 on its surface to escape from the deactivation by the tumor cells; immune cell escape [15].

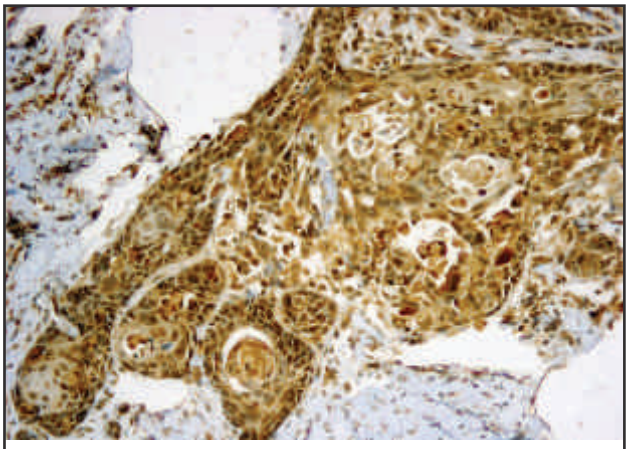

Figure 4 (a): a case of moderately differentiated stage 3 laryngeal cancer with high PD–L1 expression (score 7) with diffuse strong membranous and non–specific cytoplasmic staining in more than 75 % of tumor cells. (PD–L1x200)

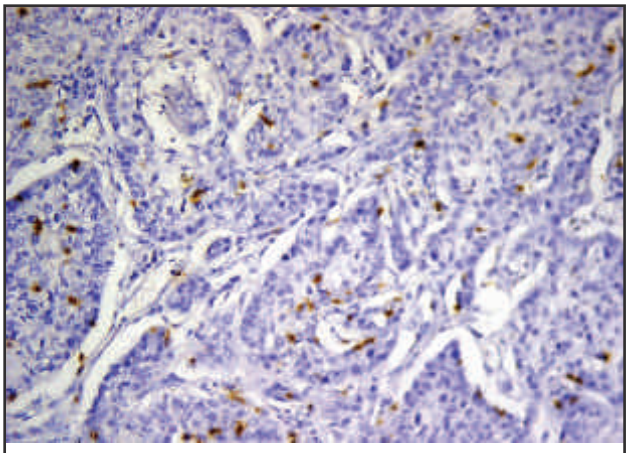

Figure 4 (b): same case showing weak CD8+ cells expression in the tumor epithelium <10/100 cells and weak CD8+ cells expression in the stroma < 20/100 (CD8x200)

Figure 4: a case of stage III moderately differentiated laryngeal carcinoma with strong PD–L1 expression in 75%, and weak epithelial and stromal CD8+ infiltration.

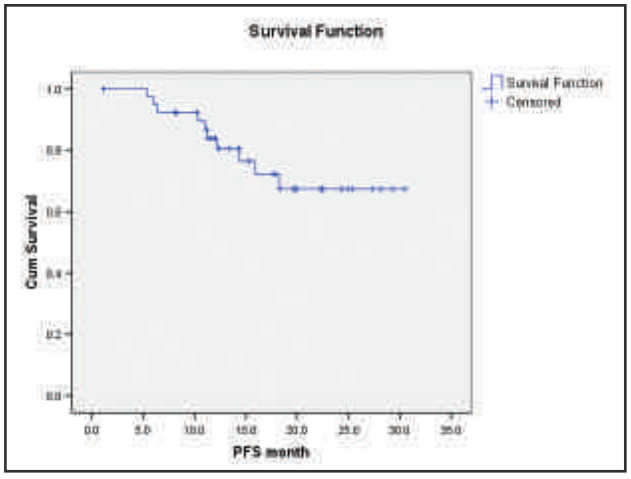

Figure 5: progression free survival (PFS) Kaplan–Meier curve of the whole population.

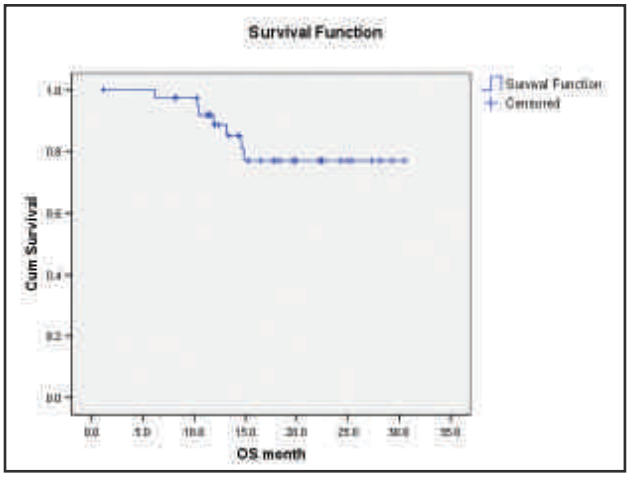

Figure 6: overall survival (OS) Kaplan–Meier curve of the whole population.

Also, several trials confirmed that PD–1 high expression on the CD8+ T–lymphocytes within the tumor microenvironment is associated with better overall survival (OS) [16] [17].

In case of an activated immune–system the tumor cells express more PD–L1 trying to escape the immune system (tumor escape immune system). In other words, the high expression of PD–L1 is reflecting that the immune–system is active in the tumor microenvironment and this may explain the better prognosis in case of high PD–L1 expression [18]. In our study, all the PD–L1 negative tumors were found weak for CD8+ infiltration and on the other hand, most of the PD–L1 positive tumors are associated with high CD8+ infiltration which goes with **Chen et al** [18] explanation.

In our study, the PD–L1 expression over the tumor cells was associated with a trend of better overall survival in the whole patients' population (P value, 0.191) and statistically significant better survival in advanced stage laryngeal cancer (stage III and IV) (p value, 0.036). This result goes with the results reported by **Chen et al** [18] and the meta–analysis which was done by Jia et al. [19]

Moreover, in this meta–analysis, the PD–L1 high expression was associated with better disease free survival (DFS) but, in our study the high expression of PD–L1 was associated with better progression free survival (PFS) but did not reach significance whether in the overall population or the advanced stage population (p values 0.521 and 0.237 respectively).

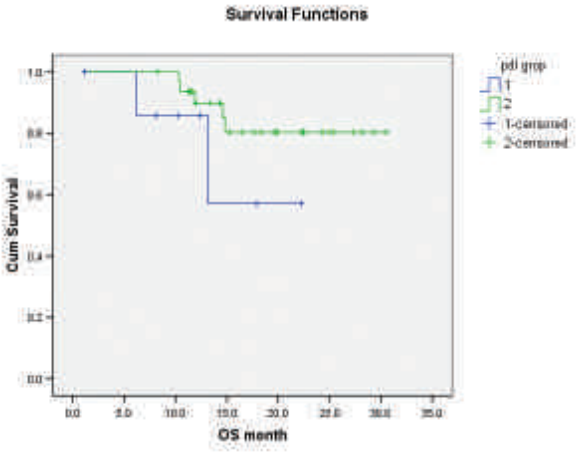

Figure 7 (a): OS survival correlation between weak or negative PD–L1 patients and strong PD–L1 patients showing non–significance (p value 0.191).

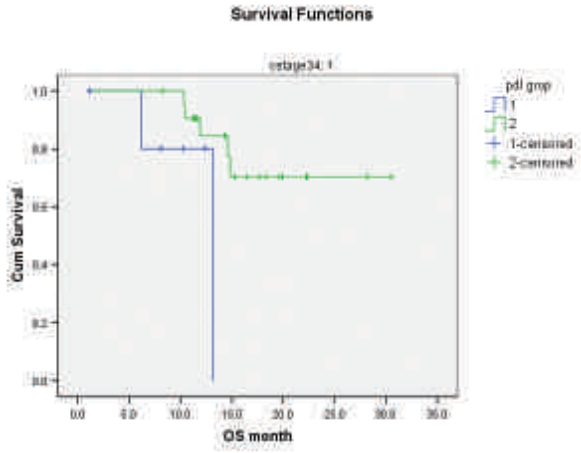

Figure 7 (b): OS survival correlation between weak or negative PD–L1 patients and strong PD–L1 patients in advanced stage disease (stage III and IV) showing significant better survival in strong PD–L1 patients (p value 0.036).

Figure 7: correlation between PD–L1 expression and overall survival (OS).

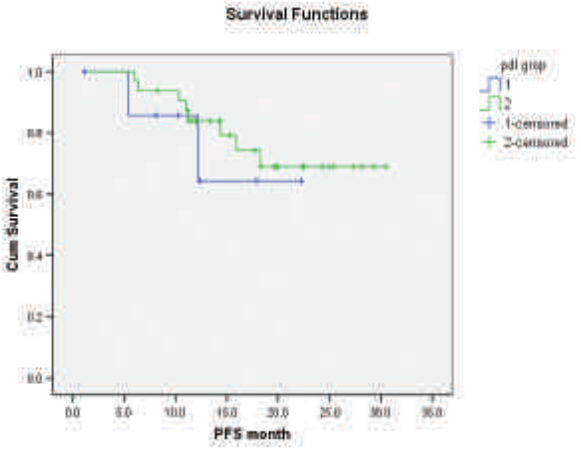

Figure 8 (a): PFS survival correlation between weak or negative PD–L1 patients and strong PD–L1 patients showing non–significance (p value 0.521).

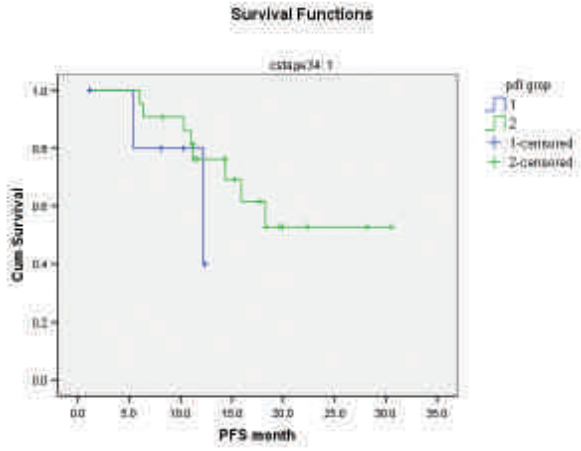

Figure 8 (b): PFS survival correlation between weak or negative PD–L1 patients and strong PD–L1 patients in advanced stage disease (stage III and IV) showing non–significance (p value 0.237).

Figure 8: correlation between PD–L1 expression and progression free survival (PFS).

Tumor microenvironment infiltration with CD8+ T–cells is associated with good prognosis and the PD–L1 expression on the peri–tumor macrophages induce cytolysis of the T–cells. Several trials reported that the high PD–L1 expression cells in the tumor stroma with low CD8+ infiltration is associated with the worst survival in hepatocellular cancer patients [20].

In our study, there was a strong correlation between tumor PD–L1 expression and CD8+ T–Cells infiltration whether in the tumor epithelium or peri–tumor stroma; this correlation is statistically significant (p value 0.001 and < 0.0001, respectively). Moreover, the patients with negative or low CD8+ and low or negative tumor PD–L1 expression had trend to be the worst survivors; indicating that active immune process in the tumor microenvironment providing better survival chances for laryngeal cancer patients.

Conclusion

Laryngeal cancer is a highly immunogenic tumor with most of the cases showing PD–L1 expression in the tumor cells and high CD8+ cells infiltration. There is a strong correlation between the CD8+ infiltration and the tumor PD–L1 expression in laryngeal cancer cases. Expression of PD–L1, CD8+ infiltration or both is associated with a trend of better overall survival (OS) compared to all

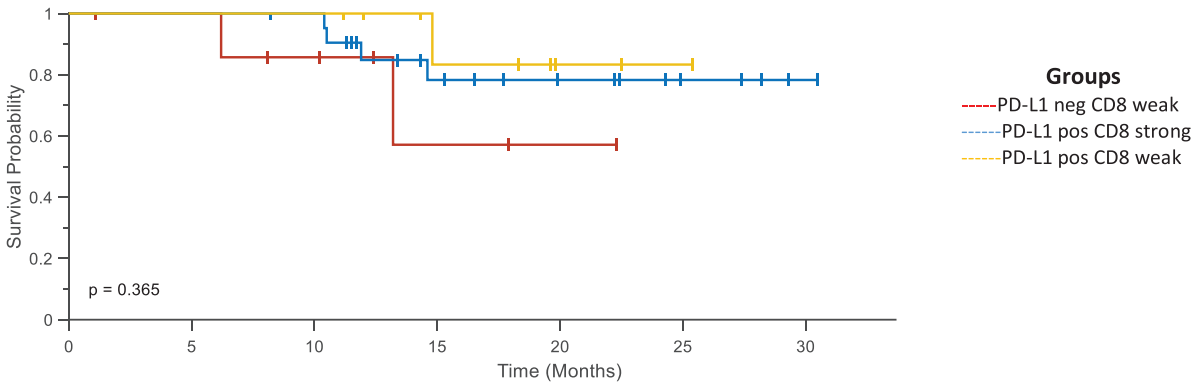

Figure 9 (a): OS of patient in the 3 subgroups with CD8+ expression in the tumor epithelium (p value 0.365).

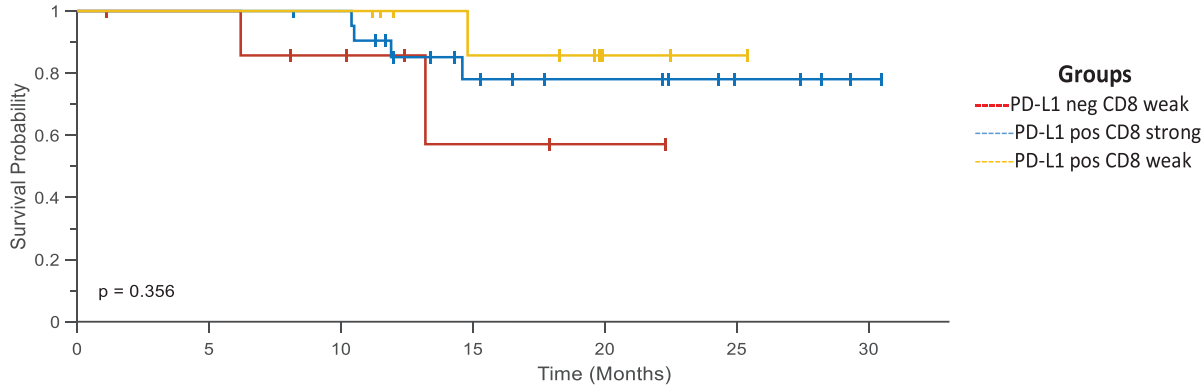

Figure 9 (b): OS of patient in the 3 subgroups with CD8+ expression in the tumor stroma (p value 0.356).

Figure 9: overall survival (OS) correlation between the different subgroups considering the CD8 and PD–L1 expression whether in the tumor epithelium or stroma.

negative tumors. Also in advanced laryngeal cancer, the expression of PD–L1 is associated with better OS.

Ethical approval and consent to participate

This study was presented to the research ethical committee of faculty of medicine, Ain Shams university, Cairo, Egypt and was approved on the 8th of June 2017 with reference number 115/2017. A verbal consent was taken from the patients to include their tumor specimen in this trial as this study was observational, non–interventional trial and work only on prognostic markers.

Consent for publication

All the authors agreed to publish this work in the Gulf Journal of oncology.

Availability of data and material

The patients' data and tumor specimens are available at Ain Shams faculty of medicine clinical oncology and pathology department archives.

Competing interest

All authors declared no conflict of interest.

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Authors' contribution

Research idea by MI and AG, data collection and treatment of patients by MI, HE and DA. Pathological analysis of specimens by MS. writing manuscript and responsibility carried out by AG. Revising, editing and data analysis by all the authors. All authors agreed on the final format of the manuscript and agreed for publication.

References

1. Siegel RL, Miller KD, Jemal A. Cancer statistics 2019. CA Cancer J Clin 2019; 69:7–34
2. Shaykhiev R and Bals R. Interactions between epithelial cells and leukocytes in immunity and tissue homeostasis. J Leukoc Biol 2007; 82(1):1–15.

3. Reichert TE, Strauss L, Wagner EM, et al. Signaling abnormalities, apoptosis, and reduced proliferation of circulating and tumor–infiltrating lymphocytes in patients with oral carcinoma. Clin Cancer Res 2002; 8 (10):3137–45

4. Pretscher D, Distel LV, Grabenbauer GG, et al. Distribution of immune cells in head and neck cancer: CD8+ T–cells and CD20+ B–cells in metastatic lymph nodes are associated with favourable outcome in patients with oro– and hypopharyngeal carcinoma. BMC Cancer 2009 ; 9: 292.

5. Hoffmann TK, Arsov C, Schirlau K, et al. T cells specific for HPV16 E7 epitopes in patients with squamous cell carcinoma of the oropharynx. Int J Cancer 2006; 118(8): 1984–91.

6. Mellman I, Coukos G, Dranoff G. Cancer immunotherapy comes of age. Nature 2011; 480 (7378): 480–9.

7. Jago CB, Yates J, Camara NO, et al. Differential expression of CTLA–4 among T cell subsets. Clin Exp Immunol 2004; 136(3): 463–71.

8. Pardoll DM. The blockade of immune checkpoints in cancer immunotherapy. Nat Rev Cancer 2012; 12(4): 252–64.

9. Elaine Vickers. A Beginner guide to targeted treatment. First edition, John Wiley & Sons Ltd 2018. Chapter 5, page162–169.

10. Spector ME, Bellile E, Amlani L, et al. Prognostic Value of Tumor–Infiltrating Lymphocytes in Head and Neck Squamous Cell Carcinoma. JAMA Otolaryngol Head Neck Surg 2019; https://doi.org/10.1001/jamaoto.2019.2427.

11. Yang WF, Wong MCM, Thomson PJ, et al. The prognostic role of PD–L1 expression for survival in head and neck squamous cell carcinoma: A systematic review and meta–analysis. Oral Oncol 2018 ; https://doi.org/10.1016/j.oraloncology.2018.09.016

12. Yu D, Cheng J, Xue K, et al. Expression of Programmed Death–Ligand 1 in Laryngeal Carcinoma and its Effects on Immune Cell Subgroup Infiltration. Pathol Oncol Res 2019; 25(4): 1437–1443.

13. Legat A, Speiser DE, Pircher H, et al. Inhibitory Receptor Expression Depends More Dominantly on Differentiation and Activation than "Exhaustion" of Human CD8T Cells. Front Immunol 2013; 4:455.

14. Saigusa S, Toiyama Y, Tanaka K, et al. Implication of programmed cell death ligand 1 expression in tumor recurrence and prognosis in rectal cancer with neoadjuvant chemoradiotherapy. Int J Clin Oncol 2016; 21(5): 946–952.

15. Keir ME, Butte MJ, Freeman GJ, Sharpe AH. PD–1 and its ligands in tolerance and immunity. Ann Rev Immunol 2008; 26: 677–704.

16. Birtalan E, Danos K, Gurbi B, et al. Expression of PD–L1 on Immune Cells Shows Better Prognosis in Laryngeal, Oropharygeal, and Hypopharyngeal Cancer. Appl Immunohistochem Mol Morphol 2018; 26(7): e79–e85.

17. Hsu MC, Hsiao JR, Chang KC, et al. Increase of programmed death–1 expressing intratumoral CD8 T cells predicts a poor prognosis for nasopharyngeal carcinoma. Mod Pathol 2010 ; 23(10):1393–403.

18. Chen SW, Li SH, Shi DB, et al. Expression of PD–1/PD–L1 in head and neck squamous cell carcinoma and its clinical significance. Int J Biol Markers 2019; 34(4): 398–405.

19. Jia YQ, Yang B, Wen LL, et al. Prognostic value of immune checkpoint molecules in head and neck cancer: a meta–analysis. Aging 2019; 11(2):501–522.

20. Liao H, Chen W, Dai Y, et al. Expression of Programmed Cell Death–Ligands in Hepatocellular Carcinoma: Correlation with Immune Microenvironment and Survival Outcomes. Front Oncol 2019; 9:883.