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Response to Neoadjuvant Chemotherapy in Locally Advanced Cervical Cancer: The Role of Immune-related Factors

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Abstract. *Background/Aim: Treatment of locally advanced cervical cancer (LACC) consists of concomitant chemoradiation or neoadjuvant chemotherapy (NACT) plus radical surgery (RS). This study analyzed the prognostic role of neutrophil to lymphocyte ratio (NLR), platelet to lymphocyte ratio (PLR), tumor infiltrating lymphocytes (TILs), and PD-L1 expression in LACC patients, treated with NACT+RS. Patients and Methods: We prospectively analyzed 37 LACC patients treated from December 2016 to September 2019. Patients were submitted to pelvic examination, biopsy and imaging. Results: In 65% of cases, a nodal involvement was present at pre-treatment MRI. All cancers showed the presence of stromal TILs and PD-L1 staining of inflammatory cells. No significant correlations were found between clinicopathological parameters and the number of TILs and PDL-1 at baseline. After NACT, 29 patients (78%) were submitted to RS; 28% of patients showed pathological complete response, 62% partial response and 10% stable disease. Seven (24%) patients reported a positive node. Patients with high levels of stromal TILs and low NLR and PLR showed a significantly better response to NACT. No significant correlation was observed between PD-L1 expression and response to NACT. Conclusion: The number of TILs, the expression of PDL1, and NLR and PLR ratios correlate significantly with the response of LACC patients to NACT.*

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Locally advanced cervical cancer (LACC) patients who do not respond to primary treatment, either with concomitant chemoradiation (CTRT) or neoadjuvant chemotherapy (NACT) plus radical surgery (RS), represent a challenge for physicians. Alongside the known negative prognostic factors, such as FIGO stage, tumor volume, nodal metastasis, smoking, and anaemia, there is an urgent need to understand why, among patients affected by the same cervical cancer, some respond to treatment while others do not (1-5).

Thus, new therapeutic strategies are under evaluation for the treatment of this tumor, with particular attention to immunotherapy strategies that might represent a valid alternative, as they have been shown to significantly improve the management of many malignancies in the last decade.

In this scenario, some "immunologic markers" have been recently studied in some cancer patients. and correlated to the prognosis and to the efficacy of immunotherapy, particularly immune checkpoint inhibitors.

Several studies have shown that high neutrophil-to-lymphocyte ratio (NLR) and platelet-to-lymphocyte ratio (PLR) are markers of host inflammation and are associated with worse overall survival (OS) (6-8); high eosinophils both in tumor tissue as well as in peripheral blood, have been reported to be prognostic markers for a better outcome in some solid tumors (9-11); tumor infiltrating lymphocytes (TILs) and programmed death ligand 1 (PD-L1) can predict response to chemotherapy and immunotherapy, and reflect the immune response in the tumor microenvironment (12-16).

The aim of the present study was to analyze the prognostic effect, in terms of response to therapy, neutrophil to lymphocyte ratio, platelet to lymphocyte ratio, TILs and PD-L1 expression in cervical cancer patients at diagnosis, treated by NACT+RS.

Patients and Methods

The institutional review board (IRB) approved this study. Inclusion criteria were: age≥18 years, Eastern Cooperative Oncology Group Performance Status of 2 or less, histologically proven squamous cervical cancer, locally advanced disease (FIGO stage IB2-IVa), absence of renal (excluded postrenal failure), hepatic, respiratory, and cardiac failure, adequate bone marrow reserve, no concomitant or previous cancer. All patients provided written informed consent to treatment and to use clinical and pathological data for scientific purposes.

All patients, at admission, were submitted to clinical staging with gynecological pelvic examination and large cervical biopsy, abdomino-pelvic MRI, pelvic 3D ultrasound and chest CT-scan.

Cystoscopy or proctoscopy were performed if required. Clinical staging at diagnosis was performed according to the 2018 FIGO classification (17). Biopsy was sent to the pathologist to assess histotype, depth of invasion and grading together with TILs and PD-L1 expression. Methods for evaluation of stromal TILs and PD-L1 have been described elsewhere (18). Briefly, a minimum of 200 neoplastic cells should be present in each biopsy sample.

Hematoxylin-eosin stained slides were evaluated for the presence of stromal TILs according to Salgado *et al*. (19); for the statistical correlations we used the following definitions: <10% for low levels of stromal TILs; 10%-40% for moderate stromal TILs levels and >40% for high stromal TILs; TILs distribution in the stroma was also analyzed. Serial sections were obtained from each paraffin block for immunophenotyping of the inflammatory infiltrate.

The definitions used for the statistical analysis were: <1% for low PDL-1 expression on both inflammatory and cancer cells, 1%-49% for moderate PDL-1 expression and >49% in case of high PDL-1 expression on cells. Cell blood count was retrieved from each patient at diagnosis in order to assess neutrophil/lymphocyte ratio (NLR), platelet/lymphocyte ratio (PLR), eosinophil/lymphocyte ratio (ELR) and eosinophil neutrophil/lymphocyte ratio (ENLR).

Pre-treatment abdomino-pelvic MRI and 3-D pelvic ultrasound was performed in all patients. Before treatment, all the patients were submitted to MRI examination using 3T Magnet (GE Discovery MR 750, Rome, Italy) with phased-array coil 32 channels. Before the examination, the antiperistaltic agent Hyoscine N-butylbromide (Buscopan 20 mg/ml, Boehringer Ingelheim, Rome, Italy) was administered by intravenous injection to reduce normal bowel peristaltic artefacts. The protocol study included multiplanar T2 weighted High Resolution sequences in sagittal and orthogonal planes to the tumor, T1 turbo spin echo (TSE) and Diffusion Weighted Imaging (DWI) on axial plane using two b values (0, 1,000), apparent diffusion coefficient (ADC) map was calculated by drawing a circular region of interest (ROI) positioned inside the solid component of the tumor. The ROI size ranged from 50 to 75 mm^2. Three measurements were performed, and a median value was calculated.

Treatment consisted of 3 cycles of NACT with cisplatin and paclitaxel in a period of 9 weeks followed by RS in responders' patients. After chemotherapy, all patients were submitted again to gynecological examination, abdomino-pelvic MRI and pelvic 3D ultrasound to assess response and operability. Clinical objective tumor responses were determined according to the Response Evaluation Criteria in Solid Tumors criteria (Recist 1.1) (21). RS was performed as previously described (22). Pathological response (complete, pCR; partial, pPR; progression disease, PD) was obtained from the histological reports of the surgical specimens' post-therapy. A complete pathological response (pCR) was defined as the complete disappearance of invasive tumor cells from the cervix and regional lymph nodes, regardless of the presence of residual carcinoma *in situ* (ypT0/is, ypN0). pPR occurred when the residual tumor was larger than 1 millimeter. PD was defined as the increase in tumor size and/or the appearance of new nodal metastases.

Clinical, laboratory, surgical and pathological data were prospectively collected and then statistically analyzed for correlations.

In the descriptive analysis, quantitative variables were described as mean and range, while qualitative variables were reported as number and percentage. Univariate associations between clinicopathological features and pathological response (pCR, pPR, PD) were evaluated using the ANOVA one-way test, χ^2 test or Pearson correlation coefficient. Statistical significance was set at $p<0.05$. All analyses were performed using IBM SPSS Statistics 25 (IBM Corp. 2017, Rome, Italy). A non-parametric test was used for the evaluation of the ROI.

Results

From December 2016 to September 2019, 37 patients fulfilling the inclusion criteria referring to the Department of Maternal-Child and Urological Sciences of Sapienza University, Umberto I Hospital in Rome, were enrolled in the study.

Clinicopathological data (including NLR, PLR, ELR and ENLR calculated from cell blood count at diagnosis) are presented in Table I. The median age at diagnosis was 54 years (range=31-76 years), the majority of the tumors (23/38, 61%) were poorly differentiated and all the neoplasms were squamous histotype. In 24/37 patients (65%) a nodal involvement was documented at pre-treatment MRI.

Data concerning TILs numbers are shown in Table II. Briefly, all tumors showed stromal TILs, with a median percentage of 40% (range=2-80%) and with a more frequent multifocal distribution. Stromal TILs expression was low in 2/37 (6%) patients, moderate in 19/37 (51%) patients and high in 16/37 (43%) patients. ROI positively correlates with the number of TILS.

Data concerning PD-L1 expression are also shown in Table II. Briefly, membrane staining of neoplastic cells was present in 35/37 patients (95%), and in 10 patients (10/35, 27%) the expression was high; the membrane staining was scored 1+ in 10/35 (23%), 2+ in 7/35 (20%), and 3+ in 20/35 cases (57%).

PD-L1 staining of inflammatory cells was present in all the biopsies (100%). In 5 out of 37 patients (15%) the percentage of PD-L1 inflammatory cells was positive with high expression (>49%) with a prevalent (19/37, 51%) moderate staining. PDL-1 expression increased with the increasing TILs expression in tumor cells (Pearson correlation coefficient 0.38, $p=0.017$). No statistically significant correlations were found between clinicopathological parameters and the number of TILs and the expression of PDL-1 in the biopsies at baseline, as shown in Table III.

After NACT, 29/37 patients (78%) were submitted to radical surgery. Among the 8 patients not submitted to

Table I. *Patient characteristics*.

Number of patients	37
Patient characteristics	
Median age (range)	54 years (31-76)
Median BMI (range)	23 (17.3-37.6)
Cardiovascular diseases	9 (24%)
Metabolic diseases	5 (14%)
HIV infection	1 (3%)
HBV/HCV infection	2 (5%)
Median NLR (range)	3.05 (1.06-20.76)
Median PLR (range)	163,500 (59,000-632,000)
Median ELR (range)	0.1 (0.01-1.00)
Median ENLR (range)	0.44 (0.05-8.31)
Tumor characteristics	
FIGO stage	
Ib2-IIA	4 (11%)
IIB-IV	33 (89%)
Histotype	
Squamous	37 (100%)
Adenocarcinoma	0 (0%)
Others	0 (0%)
Grading	
G1-2	15 (39%)
G3	22 (61%)
Nodal status at RMI	
N–	13 (35%)
N+	24 (65%)

NLR: Neutrophil to lymphocyte ratio; PLR: platelet to lymphocyte ratio; ELR: eosinophil/lymphocyte ratio; ENLR: eosinophil neutrophil/lymphocyte ratio; RMI: resonant machine.

Table II. *TILs and PDL-1 expression characteristics*.

	Number of patients (%)
Stromal TIL levels	
<10%	2 (6)
10-40%	19 (51)
>40%	16 (43)
PD-L1 intensity staining on inflammatory cells	
1+	//
2+	19/37 (51)
3+	//
PDL-1 expression on inflammatory cells	
<1% negative	//
1-49% positive	32 (86)
>49% positive with high expression	5 (14)
PD-L1 intensity staining on neoplastic cells	
1+	10/35 (23)
2+	7/35 (20)
3+	20/35 (57)
PDL-1 expression	
<1% negative	2 (5)
1-49% positive	25 (68)
>49% positive with high expression	10 (27)

TILs: Tumor infiltrating lymphocytes.

surgery, 1 patient reported a myocardial infarction during treatment and was therefore sent to radiotherapy, 3 refused treatment, and 4 patients experienced PD.

Pathological response to NACT were as follows: pCR in 8 cases (28%), pPR in 18 (62%) cases and stable disease (SD) in 3 cases (10%). Seven (24%) patients showed positive node at pathological evaluation. Patients with high numbers of stromal TILs and low NLR and PLR showed a significantly better response to NACT (p=0.024, p=0.032, p=0.26). No significant correlation was observed for PD-L1 expression and response to NACT, Table IV.

The Kruskal-Wallis test also showed that there was at least one statistically significant difference in the percentage of TILS between the various pathological responses (p=0.049); in particular, no difference was observed between pCR and pPR (p=0.639) and between pCR and PD (p=0.163), but a statistically significant difference was observed between pPR and PD (p=0.019).

Discussion

LACC is generally treated by a multimodality approach including chemotherapy, radiotherapy and surgery. However, a consistent portion of patients with negative prognostic factors will recur or will not respond to primary treatment. Unfortunately, there is little possibility of salvage therapy for persistent/recurrent disease and, surely, a more precise knowledge of the prognostic factors could help the gynecologic oncologist in tailoring the treatment and predict the outcomes.

In recent years many studies in solid tumors (breast, lung, *etc*.) have shown that the presence of TILs is predictive of good response to therapy and overall survival (23-26). On the opposite few data are available for cervical cancer patients (27, 28).

Data presented in this study are consistent with the literature, showing that the presence of high levels of stromal TILs in cervical cancer biopsies before treatment significantly correlates with a better response to treatment. In addition, the number of TILs does not seem to be related to any clinical characteristic of the patients, and is therefore independent of age, BMI, *etc*. Instead, it seems that MRI can identify TILs.

There is still little information regarding the expression of PD-L1 and the number of TILs in cervical cancer. The expression levels of PD-1/PD-L1 and HPV status in cervical lesions has been analyzed. In 2010 Brismar *et al*. demonstrated that there was no difference in the mRNA expression of PDL1 between HPV DNA-positive and -negative women (29). However, PD-L1 expression was correlated with HPV-positivity and increased with CIN grade, and tumor metastasis in cervical cancer (30). The presence of PD-1/PD-L1 expression levels in TILs in patients with cervical cancer has also been investigated. For example, PD-L1 positivity in TILs

Table III. *Correlation between TILS levels and clinical pathological parameters.*

	TILS's levels		*p*-Value
	Low-Intermediate grade	High grade	
Age	56 (31-76)	53 (40-71)	0.53
BMI	23 (18-38)	22 (17-33)	0.44
PD-L1 expression on inflammatory cells	22 (2-60)	40 (10-60)	0.013
PD-L1 expression on neoplastic cells	20 (0-60)	30 (3-85)	0.095
ROI	0.84 (0.64-1.14)	1.11 (0.25-1.70)	**0.003**
NLR	3.32 (1.26-20.7)	2.67 81.06-5.72)	0.44
PLR	154,000 (59,000-632,000)	164,000 (89,000-538,000)	0.84
ELR	0.09 (0.02-0.57)	0.13 (0.01-1.00)	0.27
ENLR	0.44 (0.05-0.31)		0.64
Maximum diameter tumor	50 mm (20-90)	48 mm (35-80)	0.87
FIGO stage			
IB1-IIB	3 (8)	1 (3)	0.30
IIB-IV	18 (49)	15 (40)	
Grading			
G1-2	7 (19)	8 (21.5)	0.31
G3	14 (38)	8 (21.5)	
Nodal status at RMI			
N–	6 (16)	7 (19)	0.34
N+	15 (41)	9 (24)	

BMI: Body mass index; NLR: neutrophil to lymphocyte ratio; PLR: platelet to lymphocyte ratio; ELR: eosinophil/lymphocyte ratio; ENLR: eosinophil neutrophil/lymphocyte ratio; RMI: resonant machine; ROI: region of interest. Bold value is statistically significant.

Table IV. *Correlation between response to NACT and TILs, serum ratios, and PDL-1.*

	Complete/Partial response	Progression	*p*-Value
Tils levels (%)			
0-40%	15 (50%)	4 (100%)	**0.024**
>40%	15 (50%)	0 (0%)	
PDL1 expression on inflammatory cells			
0-49%	26 (86.7%)	4 (100%)	0.444
>49%	4 (13.3%)	0 (0%)	
PDL1 expression on cancer cells			
0-49%	22 (73.3%)	2 (50%)	0.343
>49%	8 (26.7%)	2 (50%)	
ROI	0.93 (0.25-1.70)	0.90 (0.72-1.02)	0.22
NLR	2.8 (1.06-20.76)	4.41 (2.14-9.34)	0.032
PLR	148,000 (59,000-632,000)	178,000 (164,000-286,000)	0.026
ELR	0.1 (0.01-1)	0.90 (0.04-0.57)	0.79
ENLR	0.36 (0.05-8.31)	0.57 (0.09-2.66)	0.68

BMI: Body mass index; NLR: neutrophil to lymphocyte ratio; PLR: platelet to lymphocyte ratio; ELR: eosinophil/lymphocyte ratio; ENLR: eosinophil neutrophil/lymphocyte ratio; ROI: region of interest. Bold value is statistically significant.

was found to be higher in cervical tumors than in ovarian and endometrial cancers (30), indicating a role in predicting response to anti-PD-L1 therapies (31, 32).

In 2009, Karim *et al*. showed that the expression of PD-L1 did not have a direct impact on patient survival, instead, patients with a relative excess of infiltrating regulatory T cells displayed a better survival when the tumor was PD-L1 positive (33). Moreover, disease-free rates were significantly poorer in patients with diffuse PD-L1 expression compared with patients with marginal PD-L1 expression on the interface between tumor and stroma. Disease-specific survival was worse in cervical adenocarcinoma patients with PD-L1-positive tumor-associated macrophages compared with adenocarcinoma patients without PD-L1-positive

macrophages. However, the expression of PD-L1or the density of CD8+ T cells before treatment was not associated with progression-free or overall survival in patients with advanced cervical cancer (34).

Similarly, other studies on breast and lung tumors have also shown that the ratio NLR and PLR also correlates with a better response to treatment, indicating that inflammation represents an important moment in neoplastic progression (35).

Another important assessment is the correlation between the increase in the percentage of TILs and the statistically significant increase (p=0.017) in the expression of PD-L1 on cancer cells. These data are very substantial in view of the potential application of immunotherapy also in cervical tumors (36).

Among the limitations of the present study, the small sample size and the failure to evaluate TILs, PDL1 and ratios at the end of the treatment must certainly be mentioned, and could be evaluated in a future study.

In addition, the role of PDL-1 in cervical cancer has been analyzed in studies carried out with pembrolizumab (37). Pembrolizumab is a selective, fully humanized monoclonal antibody that prevents the interaction between PD-1 and its ligands, both PD-L1 and PD-L2 (38). The KEYNOTE-028 trial showed that, in 24 cervical cancer patients treated with pembrolizumab 10 mg/kg every 2 weeks for up to 24 months, the overall response rate was 17% (95%CI=5-37%). In particular, four patients (17%) achieved partial response, and three patients (13%) had stable disease. The median duration of response in the cohort with partial response was 5.4 months (range=4.1-7.5 months) (39). Also, in KEYNOTE-158 study with 98 patients, a phase II basket study, patients received pembrolizumab 200 mg every 3 weeks for 2 years or until progression, intolerable toxicity, or physician or patient decision. Eighty-two patients (83.7%) had programmed death-ligand 1 (PD-L1)–positive tumors (combined positive score ≥1). The overall response rate was 12.2% (95%CI=6.5-20.4%), with three patients with complete response and nine with partial responses, respectively (37). Treatment-related adverse events occurred in 65.3% of patients, (hypothyroidism 10.2%, decreased appetite 9.2%, and fatigue 9.2% (37). Based on these studies h the US Food and Drug Administration granted accelerated approval of pembrolizumab for patients with advanced PD-L1–positive cervical cancer who experienced progression during or after chemotherapy. Therefore, pembrolizumab was approved by the FDA on 12 June 2018 for recurrent or metastatic cervical cancer progressing after chemotherapy and PDL-1 positive patients. In conclusion, the levels of TILs, the expression of PDL1, and the ratio of NLR and PLR correlate statistically significantly with the response to treatment in patients with LACC undergoing NACT plus RS. New therapeutic strategies will be sought in the field of LACC treatment to improve the outcome of these patients in terms of both survival and quality of life. Among the new perspectives, the advent of cancer immunotherapy has shown encouraging results in the management of many malignancies through the use of agents (often monoclonal antibodies) that inhibit proteins that suppress the adaptive immune system (23, 24).

As also reported in a recent review by Gadducci *et al*., cervical cancer therapy is still a challenge. The two main options are concomitant CT-RT and NACT followed by radical surgery. The chemotherapy options are also different. However, especially for the locally advanced stages, survival must be improved (40). Vast and well-designed studies will certainly be needed to establish which patients can benefit from these treatments, in an increasingly modern perspective of personalized medicine.

Conflicts of Interest

The Authors declare that there are no conflicts of interest that could be perceived as prejudicing the impartiality of this study.

Authors' Contributions

All the Authors made substantial contributions to the conception and design, and/or acquisition of data, and/or analysis and interpretation of data. Also, all the Authors participated in drafting the article or revising it critically for important intellectual content, and gave final approval of the version to be submitted and any revised version.

References

1 Cohen PA and Jhingran A, Oaknin A and Denny L: Cervical cancer. Lancet *393*: 169-182, 2019. PMID: 30638582. DOI: 10.1016/S0140-6736(18)32470-X

2 Marth C, Landoni F, Mahner S, McCormack M, Gonzalez-Martin A and Colombo N; ESMO Guidelines Committee: Cervical cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol *28*: iv72-iv83, 2017. PMID: 28881916. DOI: 10.1093/annonc/mdx220

3 Widschwendter P, Janni W, Scholz C, De Gregorio A, De Gregorio N and Friedl TWP: Prognostic factors for and pattern of lymph-node involvement in patients with operable cervical cancer. Arch Gynecol Obstet *300*: 1709-1718, 2019. PMID: 31696367. DOI: 10.1007/s00404-019-05341-3

4 Takekuma M, Kasamatsu Y, Kado N, Kuji S, Tanaka A, Takahashi N, Masakazu A and Hirashima Y: The issues regarding postoperative adjuvant therapy and prognostic risk factors for patients with stage I-II cervical cancer: a review. J Obstet Gynaecol Res *43*: 617-626, 2017. PMID: 29353973. DOI: 10.21147/j.issn.1000-9604.2017.06.05

5 Fang JH, Yu XM, Zhang SH and Yang Y: Effect of smoking on high-grade cervical cancer in women on the basis of human papillomavirus infection studies. J Cancer Res Ther *14*: S184-S189, 2018. PMID: 2578171. DOI: 10.4103/0973-1482.179190

6 Huang H, Liu Q, Zhu L, Zhang Y, Lu X, Wu Y and Liu L: Prognostic value of preoperative systemic immune-inflammation index in patients with cervical cancer. Sci Rep *9*: 3284, 2019. PMID: 30824727. DOI: 10.1038/s41598-019-39150-0

7 Ma JY, Ke LC and Liu Q: The pretreatment platelet-to-lymphocyte ratio predicts clinical outcomes in patients with cervical cancer: A meta-analysis. Medicine (Baltimore) *97*: e12897, 2018. PMID: 30412089. DOI: 10.1097/MD.0000000000012897

8 Ethier JL, Desautels DN, Templeton AJ, Oza A, Amir E and Lheureux S: Is the neutrophil-to-lymphocyte ratio prognostic of survival outcomes in gynaecologic cancers? A systematic review and meta-analysis. Gynecol Oncol *145*: 584-594, 2017. PMID: 28222899. DOI: 10.1016/j.ygyno.2017.02.026

9 Holub K and Biete A: New pre-treatment eosinophil-related ratios as prognostic biomarkers for survival outcomes in endometrial cancer. BMC Cancer *18*: 1280, 2018. PMID: 30577833. DOI: 10.1186/s12885-018-5131-x

10 Mingomataj MC: Eosinophil-induced prognosis improvement of solid tumors could be enabled by their vesicle-mediated barrier permeability induction. Med Hypotheses *70*: 582-584, 2008. PMID: 17689197. DOI: 10.1016/j.mehy.2007.06.017

11 Gatault S, Legrand F, Delbeke M, Loiseau S and Capron M: Involvement of eosinophils in the anti-tumor response. Cancer Immunol Immunother *61*: 1527-1534, 2012. PMID: 22706380. DOI: 10.1007/s00262-012-1288-3

12 Yu X, Zhang Z, Wang Z, Wu P, Qiu F and Huang J: Prognostic and predictive value of tumor-infiltrating lymphocytes in breast cancer: a systematic review and meta-analysis. Clin Transl Oncol *18*: 497-506, 2016. PMID: 26459255. DOI: 10.1007/s12094-015-1391-y

13 Meng Y, Liang H, Hu J, Liu S, Hao X, Sze Ka Wong M, Li X and Hu L: PD-L1 expression correlates with tumor infiltrating lymphocytes and response to neoadjuvant chemotherapy in cervical cancer. J Cancer *9*: 2938-2945, 2018. PMID: 30123362. DOI: 10.7150/jca.22532

14 Wang Q, Lou W, Di W and Wu X: Prognostic value of tumor PD-L1 expression combined with CD8+tumor infiltrating lymphocytes in high grade serous ovarian cancer. Int Immunopharmacol *52*: 7-14, 2017. PMID: 28846888. DOI: 10.1016/j.intimp.2017.08.017

15 Zhang Y, Zhu W, Zhang X, Qu Q and Zhang L: Expression and clinical significance of programmed death-1 on lymphocytes and programmed death ligand-1 on monocytes in the peripheral blood of patients with cervical cancer. Oncol Lett *14*: 7225-7231, 2017. PMID: 29344157. DOI: 10.3892/ol.2017.7105

16 Yang W, Song Y, Lu YL, Sun JZ and Wang He: Increased expression of programmed death (PD)-1 and its ligand PD-L1 correlates with impaired cell-mediated immunity in high-risk human papillomavirus-related cervical intraepithelial neoplasia. Immunology *139*: 512-522, 2013. PMID: 23521696. DOI: 10.1111/imm.12101

17 Bhatla N, Berek JS, Cuello Fredes M, Denny LA, Grenman S, Karunaratne K, Kehoen St,Konishi I, Olawaiye AB, Prat J, Sankaranarayann R, Brierley J, Mutch D, Queleu D, Cibula D, Quinn M, Botha H, Sigurd L, Rice L, Ryu HS, Ngan H, Maenpaa J, Andrijono A, Purwoto G, Maheshwari A, Bafna UD, Plante M and Natarajan J: Revised FIGO staging for carcinoma of the cervix uteri. Int J Gynecol Obstet *145*: 129-135, 2019. PMID: 30656645. DOI: 10.1002/ijgo.12749

18 D'Alessandris N, Palaia I, Pernazza A, Tomao F, Di Pinto F, Musacchio L, Leopizzi M, Di Maio V, Pecorella I, Benedetti Panici P and Della Rocca C: PD-L1 expression is associated with tumor infiltrating lymphocytes that predict response to NACT in squamous cell cervical cancer. Virchows Arch *145*: 129-135, 2020. PMID: 32915266. DOI: 10.1007/s00428-020-02922-5

19 Salgado R, Denkert C, Demaria S, Sirtaine N, Klauschen F, Pruneri G, Wienert S, Eynden Vd, Baehner FL, Penault- Llorca F, Perez EA, Thompson EA, Symmans WF, Richardson AL, Brock J, Criscitiello C, Bailey H, Ignatiadis M, Floris G, Sparano J, Kos Z, Nielsen T, Rimm DL, Allison KH, Loibl S; Sotirou C, Viale G, Badve S, Adams S, Willard-Gallo K and Loi S: The evaluation of tumor-infiltrating lymphocytes (TILs) in breast cancer: recommendations by an International TILs Working Group 2014. Ann Oncol *26*: 259-271, 2015. PMID: 25214542. DOI: 10.1093/annonc/mdu450

20 Pawelczyk K, Piotrowska A, Ciesielska U, Jablonska K, Plucinska NG, Grzegrzolka J, Podhorska Okolow M, Dziegel P and Nowinska K: Role of PD-L1 expression in non-small cell lung cancer and their prognostic significance according to clinicopathological factors and diagnostic markers. Int J Mol Sci *20*: 824, 2019. PMID: 30769852. DOI: 10.3390/ijms20040824

21 Eisenhauer EA, Therasseb P, Bogaertsc J, Schwartz LH, Sargent D, Ford R, Dancey J, Arbuck S, Gwyther S, Mooney M, Rubinstein L, Shankar L, Dodd L, Kaplan R, Lacombe D and Verweij J: New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). Eur J Cancer *45*: 228-247, 2009. PMID: 19097774. DOI: 10.1016/j.ejca.2008.10.026

22 Benedetti-Panici P, Maneschi F, Cutillo G, Greggi S, Salerno MG, Amoroso M, Scambia G and Mancuso S: Modified type IV-V radical hysterectomy with systematic pelvic and aortic lymphadenectomy in the treatment of patients with stage III cervical carcinoma. Feasibility, technique, and clinical results. Cancer *78(11)*: 2359-2365, 1996. PMID: 8941007. DOI: 10.1002/(sici)1097-0142(19961201)78:11<2359::aid-cncr14>3.0.co;2-#.

23 Zhao Y, Ge X, Cheng Y, Wang Z, Wang J and Sun L: The prognostic value of tumor-infiltrating lymphocytes in colorectal cancer differs by anatomical subsite: a systematic review and meta-analysis. World J Surg Oncol *17*: 85, 2019. PMID: 31118034. DOI: 10.1186/s12957-019-1621-9

24 Darb Esfahani S, Kunze CA, Kulbe H, Sehouli J, Wienert S, Lindner J, Budczies J, Bockmayr M, Dietel M, Denkert C, Braicu I and Johrens K: Prognostic impact of programmed cell death-1 (PD-1) and PD-ligand 1 (PD-L1) expression in cancer cells and tumor-infiltrating lymphocytes in ovarian high grade serous carcinoma. Oncotarget *7*: 1486-1499, 2016. PMID: 26625204. DOI: 10.18632/oncotarget.6429

25 Cimino-Mathews A, Thopson E, Taube JM, Ye X, Lu Y, Meeker A, Xu H, Sharma R, Lecksell K, Cornish TC, Cuka N, Argani P and Emens LA: PD-L1 (B7-H1) expression and the immune tumor microenvironment in primary and metastatic breast carcinomas. Hum Pathol *47*: 52-63, 2016. PMID: 26527522. DOI: 10.1016/j.humpath.2015.09.003

26 Blackley EF and Loi S: Targeting immune pathways in breast cancer: review of the prognostic utility of TILs in early stage triple negative breast cancer (TNBC). Breast *48*: S44-S48, 2019. PMID: 31839159. DOI: 10.1016/S0960-9776(19)31122-1

27 Liang Y, Lu W, Zhang X and Lu B: Tumor-infiltrating CD8+ and FOXP3+ lymphocytes before and after neoadjuvant chemotherapy in cervical cancer. Diagn Pathol *13*: 93, 2018. PMID: 30474571. DOI: 10.1186/s13000-018-0770-4

28 Chen R, Cong Y, Zou D, Wang L, Yuan L and Zhou Q: Correlation between subsets of tumor-infiltrating immune cells

and risk stratification in patients with cervical cancer. Peer J *7*: e7804, 2019. PMID: 31616592. DOI: 10.7717/peerj.7804

29 Brismar Wendel S, Kaldensjo T, Peterson P, Andersson S, Broliden K and Hirbod T: Slumbering mucosal immune response in the cervix of human papillomavirus DNA-positive and -negative women. Int J Oncol *37*: 1565-1573, 2010. PMID: 21042726. DOI: 10.3892/ijo_00000810

30 Yang W, Lu YP, Yang YZ, Kang JR, Jin YD and Wang HW: Expressions of programmed death (PD)-1 and PD-1 ligand (PD-L1) in cervical intraepithelial neoplasia and cervical squamous cell carcinomas are of prognostic value and associated with human papillomavirus status. J Obstet Gynaecol Res *43*: 1602-1612, 2017. PMID: 28833798. DOI: 10.1111/jog.13411

31 Mezache L, Paniccia B, Nyinawabera A and Nuovo GJ: Enhanced expression of PD L1 in cervical intraepithelial neoplasia and cervical cancers. Mod Pathol *28*: 1594-1602, 2015. PMID: 26403783. DOI: 10.1038/modpathol.2015.108

32 Festino L, Botti G, Lorigan P, Masucci GV, Hipp J, Horak CE, Melero I and Ascierto PA: Cancer treatment with anti-PD-1/PDL1 agents: Is PD-L1 expression a biomarker for patient selection? Drugs *76*: 925-945, 2016. PMID: 27229745. DOI: 10.1007/s40265-016-0588-x

33 Karim R, Jordanova ES, Piersma SJ, Kenter GG, Chen L, Boer JM, Melief CJM and van der Burg SH: Tumor-expressed B7-H1 and B7-DC in relation to PD-1+ T-cell infiltration and survival of patients with cervical carcinoma. Clin Cancer Res *15*: 6341-6347, 2009. PMID: 19825956. DOI: 10.1158/1078-0432.CCR-09-1652

34 Enwere EK, Kornaga EN, Dean M, Koulis TA, Phan T, Kalantarian M, Kobel M, Ghatage P, Magliocco AM, Lees-Miller SP and Doll CM: Expression of PD-L1 and presence of CD8-positive T cells in pre-treatment specimens of locally advanced cervical cancer. Mod Pathol *30*: 577-586, 2017. PMID: 28059093. DOI: 10.1038/modpathol.2016.221

35 Diem S, Schmid S, Krapf M, Flatz L, Born D, Jochum W, Templeton AJ and Fruj M: Neutrophil-to-Lymphocyte ratio (NLR) and Platelet-to-Lymphocyte ratio (PLR) as prognostic markers in patients with non-small cell lung cancer (NSCLC) treated with nivolumab. Lung Cancer *111*: 176-181, 2017. PMID: 28838390. DOI: 10.1016/j.lungcan.2017.07.024

36 Heeren AM, Punt S, Bleekrer MC, Gaarenstroom KN, van der Velden J, Kenter GG, de Gruijl TD and Jordanova ES: Prognostic effect of different PD-L1 expression patterns in squamous cell carcinoma and adenocarcinoma of the cervix. Mod Pathol *29*: 753-763, 2016. PMID: 27056074. DOI: 10.1038/modpathol.2016.64

37 Chung HC, Ros W, Delord JP, Perets R, Italiano A, Shapira-Frommer R, Manzuk L, Piha-Paul SA, Xu L, Zeigenfuss S, Pruitt SK and Leary A: Efficacy and safety of pembrolizumab in previously treated advanced cervical cancer: Results from the phase II KEYNOTE-158 study. J Clin Oncol *37(17)*: 1470-1478, 2019. PMID: 30943124. DOI: 10.1200/JCO.18.01265

38 Khoja L, Butler MO, Kang SP, Ebbinghaus S and Joshua AM: Pembrolizumab. J Immunother Cancer *3*: 36, 2015. PMID: 26288737. DOI: 10.1186/s40425-015-0078-9

39 Frenel JS, Le Tourneau C, O'Neil B, Ott PA, Piha-Paul SA, Gomez-Roca C, van Brummelen EMJ, Rugo HS, Thoms S, Saraf S, Rangwala R and Varga A: Safety and efficacy of pembrolizumab in advanced, programmed death ligand 1-positive cervical cancer: Results from the phase Ib KEYNOTE-028 trial. J Clin Oncol *35*: 4035-4041, 2017. PMID: 29095678. DOI: 10.1200/JCO.2017.74.5471

40 Gadducci A and Cosio S: Neoadjuvant chemotherapy in locally advanced cervical cancer: Review of the literature and perspectives of clinical research. Anticancer Res *40(9)*: 4819-4828, 2020. PMID: 32878770. DOI: 10.21873/anticanres.14485

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