

**Table S1. Search strategies.**

Database	
Pubmed	<p>#1 "head"[MeSH Terms] OR "head"[All Fields] OR ("hnsccs"[All Fields] OR "squamous cell carcinoma of head and neck"[MeSH Terms] OR ("squamous"[All Fields] AND "cell"[All Fields] AND "carcinoma"[All Fields] AND "head"[All Fields] AND "neck"[All Fields]) OR "squamous cell carcinoma of head and neck"[All Fields] OR "hnscc"[All Fields]) OR ("mouth"[MeSH Terms] OR "mouth"[All Fields] OR "oral"[All Fields]) OR ("tongue"[MeSH Terms] OR "tongue"[All Fields] OR "tongues"[All Fields] OR "tongue s"[All Fields]) OR ("mouth"[MeSH Terms] OR "mouth"[All Fields] OR "mouths"[All Fields] OR "mouth s"[All Fields] OR "mouthed"[All Fields] OR "mouthful"[All Fields] OR "mouthfuls"[All Fields] OR "mouthing"[All Fields]) OR ("nasalance"[All Fields] OR "nasality"[All Fields] OR "nasalization"[All Fields] OR "nasalized"[All Fields] OR "nasally"[All Fields] OR "nose"[MeSH Terms] OR "nose"[All Fields] OR "nasal"[All Fields] OR "nasals"[All Fields]) OR "sinonasal"[All Fields] OR "sino-nasal"[All Fields] OR "paranasal"[All Fields] OR ("pharynx"[MeSH Terms] OR "pharynx"[All Fields] OR "pharynxes"[All Fields] OR "pharynxes"[All Fields]) OR ("pharyngeals"[All Fields] OR "pharynges"[All Fields] OR "pharynx"[MeSH Terms] OR "pharynx"[All Fields] OR "pharyngeal"[All Fields]) OR ("oropharynx"[MeSH Terms] OR "oropharynx"[All Fields] OR "oropharynxes"[All Fields]) OR ("larynx"[MeSH Terms] OR "larynx"[All Fields] OR "larynxes"[All Fields]) OR ("larynges"[All Fields] OR "laryngitis"[MeSH Terms] OR "laryngitis"[All Fields] OR "laryngitides"[All Fields] OR "larynx"[MeSH Terms] OR "larynx"[All Fields] OR "laryngeal"[All Fields]) OR ("hypopharynx"[MeSH Terms] OR "hypopharynx"[All Fields] OR "hypopharynges"[All Fields] OR "hypopharynxes"[All Fields]) OR ("hypopharynx"[MeSH Terms] OR "hypopharynx"[All Fields] OR "hypopharyngeal"[All Fields]) OR ("vocal"[All Fields] OR "vocales"[All Fields] OR "vocalic"[All Fields] OR "vocalisation"[All Fields] OR "vocalisations"[All Fields] OR "vocalised"[All Fields] OR "vocalising"[All Fields] OR "vocalization"[All Fields] OR "vocalizations"[All Fields] OR "vocalize"[All Fields] OR "vocalized"[All Fields] OR "vocalizer"[All Fields] OR "vocalizers"[All Fields] OR "vocalizes"[All Fields] OR "vocalizing"[All Fields] OR "vocally"[All Fields] OR "vocals"[All Fields]) OR ("cone rod dystrophies"[MeSH Terms] OR ("cone rod"[All Fields] AND "dystrophies"[All Fields]) OR "cone rod dystrophies"[All Fields] OR "cord"[All Fields] OR ("cord s"[All Fields] OR "cords"[All Fields])</p> <p>#2 "CD274"[All Fields] OR "B7-H"[All Fields] OR "B7H1"[All Fields] OR "PD-L1"[All Fields] OR "PDCD1LG1"[All Fields] OR "PDL1"[All Fields] OR "hPD-L1"[All Fields] OR "programmed cell death-ligand 1"[All Fields] OR "programmed cell death-ligand 1"[All Fields]</p> <p>#3 #1 AND #2</p> <p>#4 "B7-H1 Antigen"[MeSH Terms] OR "cd274 protein human"[Supplementary Concept]</p> <p>#5 "Squamous Cell Carcinoma of Head and Neck"[MeSH Terms]</p> <p>#6 #4 AND #5</p> <p>#3 OR #6</p>
Embase	<p>#1 'head'/exp OR head OR 'hnscc'/exp OR hnscc OR oral OR 'tongue'/exp OR tongue OR 'mouth'/exp OR mouth OR nasal OR sinonasal OR 'sino nasal' OR paranasal OR 'pharynx'/exp OR pharynx OR pharyngeal OR 'oropharynx'/exp OR oropharynx OR oropharyngeal OR 'larynx'/exp OR larynx OR laryngeal OR 'hypopharynx'/exp OR hypopharynx OR hypopharyngeal OR vocal OR cord OR cords</p> <p>#2 cd274 OR 'b7 h' OR b7h1 OR 'pd l1' OR pdcd1l1 OR pdcd1lg1 OR pdl1 OR 'hpd l1' OR 'programmed cell death-ligand 1' OR 'programmed cell death ligand 1'</p> <p>#3 #1 AND #2 AND [embase]/lim NOT [medline]/lim</p>

Table S2. Summary of included studies.

Author, (Country)	year	N cases	Sites	Therapy	Scoring system	Clone and platform	Main results	Other results and limitations	
Clone E1L3N (6 studies)									
Delafoy, (France) [32]	2021	44 (91% M, median age 65 y)	HNSCC NOS (Bx and Rx) (FU 24 months)	Concurrent cisplatin and RT	TC and IC scored separately, positive if >1%	E1L3N on Ventana Benchmark XT	<ul style="list-style-type: none"><li>In 36 paired specimens before and after RT-CHT</li><li>Statistically significant correlation between the PD-L1 on the initial and relapsing tumor, both on TC or IC (p = 0.001)</li><li>Change of PD-L1 expression in 36.1%, among whom 7 (19.4%) had positivization on TC and negativization in 6 (16.7%) at recurrence</li><li>Decrease in PD-L1 expression after radiation of 36.1% on TC and 19.4% on IC</li></ul>	<ul style="list-style-type: none"><li>No prognostic value of PD-L1 expression (P&gt;0.7) for OS and PFS</li><li>No correspondence between Bx and Rx nor separation of data</li></ul>	
Doescher, (Germany) [33]	2020	67 (mean age 62 y)	HNSCC excluded sinonasal and rhinopharyngeal (Bx) (median FU 26 months)	Concurrent platinum-based CHT and RT	TC and IC scored separately with H-score	E1L3N on Dako Autostainer Universal Staining System	<ul style="list-style-type: none"><li>No significant changes in the expression of PD-1 on neither tumor nor TIL during therapy</li><li>No significant changes by therapy and no significant differences for PD-L1 expression on TC overall (p=0.28)</li><li>PD-L1 expression on IC of non-responders was significantly lower before RT-CHT (p=0.008)</li></ul>	<ul style="list-style-type: none"><li>Adverse prognostic value of PD-L1, PD-1 and CD27</li><li>No use of established CPS cutoffs as in trials</li><li>Small sample size</li><li>Heterogeneity in tumor locations, fixation and storage of tissue blocks and treatments with no stratification</li></ul>	
Gomez-Roca, (France) [34]	2020	26 (81% M, median age 59.5 y)	Oral, OP, L, HP (Bx and Rx)	Inhibitor of apoptosis protein Debio 1143 with and without cisplatin	TC and IC as % positive cells	E1L3N on Dako Autostainer EnVision FLEX System	<ul style="list-style-type: none"><li>Increased PD-L1 levels in immune cells with neoadjuvant target therapy with inhibitor of apoptose protein and in combination with cisplatin, but not with cisplatin alone</li><li>PD-L1 levels remained at baseline levels of increased not significantly in TC</li></ul>	<ul style="list-style-type: none"><li>Pharmacodynamic and pharmacogenomic study</li><li>No use of established CPS cutoffs as in trials</li></ul>	
Leduc, (France) [36]	2018	21 (67% M, mediana age 54 y)	Oral and L (Bx and Rx)	Docetaxel, platinum and fluorouracil (TPF)	TC and IC scored separately with >5% as positive	E1L3N on Ventana Discovery Ultra autostainer and HQ-amplification kit	<ul style="list-style-type: none"><li>Significant increase of PD-L1 expression in IC after therapy (71%, p=0.003)</li><li>Significant increase in PD-L1 expression also in TC after therapy (38%, p=0.005)</li></ul>	<ul style="list-style-type: none"><li>Therapy also induce increase of CD8 TILs (p=0.01)</li><li>Small sample size</li></ul>	
Long, [39]	2021 (China)	24 (75% M, median age 47 y)	NP	Concurrent and cisplatin	IMRT	TPS, positive if >1%, CPS	E1L3N, platform not stated, and 22C3 for imagery	<ul style="list-style-type: none"><li>PD-L1 expression unchanged in 7 (29%), increased in 10 (42%), and decreased in 7 patients (29%)</li><li>PD-L1 expression with CPS unchanged in 4 (17%), increased in 8</li></ul>	<ul style="list-style-type: none"><li>PD-L1 expression correlated with better clinical response</li><li>Not full correspondence of data in tables and text of the article</li></ul>

									(33%), and decreased in 12 patients (50%)	
Ock, 2017 [44]	(Korea)	35 M, mean age 63 y)	(89%)	OP mainly	Docetaxel, platinum and fluorouracil (TPF) regimens or cisplatin-based regimens and concurrent RT	TPS>5% positive	as	E1L3N on Ventana Benchmark XT	<ul style="list-style-type: none"><li>Statistically significant up-regulation of PD-L1 in 69% of originally negative cases (p=0.003)</li><li>18% of positive cases were negative after therapy, with no statistical significance (p=0.72)</li><li>Radiotherapy significantly induces up-regulation of PD-L1 (p&lt;0.001)</li></ul>	<ul style="list-style-type: none"><li>Small sample size</li><li>Down-regulation of EMT markers after therapy</li><li>Unclear detailing of IHC protocol</li></ul>
Clone SP142 (2 studies)										
Chan, 2017 [36]	(China)	161 M, median age 53 y)	(73%)	NP	IMRT and CHT with cisplatin	TC and IC with cutoff 1%		SP142 on Ventana Benchmark	<ul style="list-style-type: none"><li>Reduction of PD-L1 expression in 56% cases in IC and in 33% in TC after treatment</li><li>Median decrease lower than 3%</li><li>47% positive cases changed to negative, 12% became positive at cutoff 1%</li><li>No influence of type of therapy</li></ul>	<ul style="list-style-type: none"><li>No separated data for patients receiveinfg radiotherapy alone</li><li>Similar changes for CD8</li><li>No correlation of PD-L1 with OS or PFS</li></ul>
So, 2020 [45]	(Korea)	42 M, mean age 59.5 y)	(74%)	Oral cavity, L, OP, HP, sinuses (Bx) (FU 61 months)	RT only	TC and IC positive if >10%, automated scoring on WSI digitized slides		SP142 on Ventana Benchmark XT with Optiview DAB detection kit and Optiview Amplification kit	<ul style="list-style-type: none"><li>Ratio of recurrent (R) vs initial (I) PD-L1 expression was &lt;1 in 69% cases with TC turning negative in 23% and positive in 9%</li><li>Similar results for ratio of CD8 TILs</li></ul>	<ul style="list-style-type: none"><li>No prognostic role of PD-L1 decrease, while patients with decrease of CD8 TILs showed worse prognosis (p=0.003)</li><li>Reported limitations: small sample size, retrospective design, and different treatment characteristics among patients</li></ul>
Other clones (7 studies, a clone each)										
Karabajakian, 2021 (France) [35]		35 M, median age 64 y)	(71%)	Oral, HP, L (Bx and Rx)	OP, RT-CHT NOS	CPS		QR1 on Ventana Ultra View	<ul style="list-style-type: none"><li>At CPS1 and CPS20 64% and 67% concordance between initial and recurrent tumor</li><li>Majority of samples positive at CPS1 remained positive (76%), while majority of negative CPS samples at diagnosis became positive at relapse (75%)</li><li>At CPS20 majority of negative CPS samples at diagnosis remained negative at relapse (71%)</li></ul>	<ul style="list-style-type: none"><li>No special limitations; additional strong correlation between gene expression profile of PD-L1 with mRNA and CPS scoring</li></ul>
Naruse, (Japan) [41]	2020	121 M)	(76%)	Tongue	Platinum-based CHT	TC only, positive if >5%		Abcam ab156361	<ul style="list-style-type: none"><li>No marked difference in expression of PD-L1 in recurrent patients who had surgery alone or CHT plus surgery</li><li>Higher expression in recurrent patients treated with CHT alone</li><li>Lower expression in CHT treated patients without local recurrence</li></ul>	<ul style="list-style-type: none"><li>No use of CPS or approved assay</li><li>No reporting of CI or p values for comparisons; only reporting for correlation with clinicopathological variables and prognosis</li><li>No adjustments reported nor FU time</li></ul>

Ono, 2020 (Japan) [42]	30 M, mean age 66 y)	(90%	L, OP, HP	Cisplatin-based RT- CHT	TPS and IC density score	clone D3 on Ventana BenchMark Ultra with Ultraview detection kit	<ul style="list-style-type: none"> <li>No significant changes in PD-L1 expression after RT-CHT (p&gt;0.41)</li> <li>Same results for PD-L2 and other immune markers</li> <li>Positive correlation between increased CD8 TILs and PD-L1 up-regulation after therapy (p=0.007)</li> </ul>	<ul style="list-style-type: none"> <li>Small sample size</li> </ul>
Pflumio, 2021 (France) [37]	100 M, mean age 62.4 y)	(79%	Oral, OP, L, HP (Rx) (FU median 18 months)	RT-CHT NOS	TPS>1%	SP263	<ul style="list-style-type: none"> <li>CD3+ TIL significantly lower in intratumoral and stromal components in the irradiated area cohort (p=0.001)</li> <li>No significant difference between CD8+ TIL in the two cohorts (p=0.273)</li> <li>Significantly lower percentage of PD-L1+ TC within the irradiated area cohort than the de novo cohort (p=0.001)</li> <li>Significantly fewer tumors with PD-L1+ IC in the irradiated area cohort (p=0.01)</li> </ul>	<ul style="list-style-type: none"> <li>Indirect comparison, as 50 irradiated cancers are compared with 50 de novo cancers</li> <li>No CPS used</li> </ul>
Seki-Soda, 2021 (Japan) [43]	71 M)	(68%	Oral cavity (Bx and Rx)	Preoperative fluorouracil-based CHT	TC	28-8, platform not stated	<ul style="list-style-type: none"> <li>PD-L1 positivity in TC significantly decreased by CHT irrespective of clinical response</li> </ul>	<ul style="list-style-type: none"> <li>No use of established CPS cutoffs as in trials</li> </ul>
Shen, 2020 (China) [40]	47 M, median age 64 y)	(100%	HP (Bx and Rx)	Platinum-based CHT and RT	TC	66248-1-Ig, Proteintech, platform not stated	<ul style="list-style-type: none"> <li>PD-L1 expression increased after RT</li> </ul>	<ul style="list-style-type: none"> <li>Not stated a clear cutoff for positivity, only 10% for low vs high</li> <li>Only 25 cases with available pre and post RT samples</li> </ul>
Wolf, 2020 (USA) [46]	96 M, mean age 60.2 y)	(72%	Oral cavity (bx and Rx)	Multi-cytokine preparation with cyclophosphamide	IC and H-score, CPS	Multiplex fluorescence immunohistochemistry	<ul style="list-style-type: none"> <li>No significant changes in PD-L1 expression after CHT</li> <li>Trend to positization in 38% of negative cases before CHT with add of multicytokine preparation</li> </ul>	<ul style="list-style-type: none"> <li>Small sample size (only 28 paired specimens for complete analysis)</li> </ul>

Bx, biopsy specimen; CHT, chemotherapy; CI, confidence interval; CPS, combined positive score; EMT, epithelia-mesenchymal transition; FU, follow-up; HNSCC, head and neck squamous cell carcinoma; HP, hypopharynx; IC, immune cells; IHC, immunohistochemistry; IMRT, intensity modulated radiation therapy; L, laryngeal; M, male; NOS, not otherwise specified; NP, nasopharyngeal; OP, oropharyngeal; OS, overall survival; PFS, progression-free survival; RP, rhinopharynx; RT-CHT, radio-chemotherapy; Rx, resection specimen; TC, tumor cells; TILs, tumor-infiltrating lymphocytes; TPS, tumor proportion score; WSI, whole-slide imaging; y, years.