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Clinical Practice

Increased mu-opioid receptor expression is associated with reduced disease-free and overall survival in laryngeal squamous cell carcinoma

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Abstract

Background: Expression of the mu-opioid receptor (MOR) is associated with poor long-term outcomes in various types of cancer. The association between MOR expression and clinical outcomes in laryngeal squamous cell carcinoma (LSCC) is not clear.

Methods: This retrospective study included patients who underwent laryngectomy for LSCC. The expression pattern of the MOR protein and *OPRM1* gene in tumours and corresponding adjacent non-carcinoma specimens was measured. Propensity score matching was used to minimise bias. The primary endpoints were overall survival (OS) and disease-free survival (DFS). The secondary endpoints were intraoperative sufentanil consumption, grade of surgical complications according to the Clavien-Dindo classification, and hospital length of stay.

Results: A total of 207 LSCC patients were enrolled. After propensity score matching, there was a significant difference in DFS between groups at 1, 3, and 5 yr (60.2% *vs* 81.2%, $P=0.019$; 39.4% *vs* 50.2%, $P=0.026$; 37.5% *vs* 42.5%, $P=0.023$, respectively) in patients with high MOR expression. The OS rates at 1, 3, and 5 yr were significantly lower in the high MOR expression group (81.2% *vs* 93.2%, $P=0.027$; 57.7% *vs* 78.3%, $P<0.001$; 42.5% *vs* 60.3%, $P<0.001$, respectively). The multivariate analysis indicated that high MOR expression was associated with worse DFS and OS (hazard ratio: 1.52, 95% confidence interval: 1.07, 2.25, $P=0.034$; hazard ratio: 1.42, 95% confidence interval: 1.17, 2.34, $P=0.032$).

Conclusion: High MOR expression may be associated with poor prognosis in patients with LSCC, suggesting that MOR could be used as a valuable molecular biomarker to predict prognosis of LSCC patients.

Keywords: disease-free survival; laryngeal squamous cell carcinoma; mu-opioid receptor; OPRM1; overall survival

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Editor's key points

- Accumulating evidence suggests that expression of mu-opioid receptors (MORs) is associated with poor long-term outcomes in cancer.
- The authors retrospectively examined the association between MOR expression and clinical outcomes in laryngeal squamous cell carcinoma.
- Increased MOR expression in tumour tissue was associated with reduced disease-free survival and overall survival.
- MOR expression might provide a valuable molecular biomarker or actionable target in the prognosis and treatment of laryngeal squamous cell carcinoma.

Laryngeal squamous cell carcinoma (LSCC) is one of the most prevalent types of head and neck malignancy worldwide.[1] The main options for initial treatment for LSCC include surgery, radiotherapy, and chemotherapy, of which surgery is the most commonly used therapeutic approach.[2] Opioids such as fentanyl, sufentanil, and morphine are the mainstay analgesics during and after surgery, with the mu-opioid receptor (MOR) being the main target for opioids.[3]

Evidence suggests that increased expression of MOR in cancer cells is associated with poor prognosis in various types of cancer, such as prostate cancer, lung cancer, and hepatocellular carcinoma.[4–6] Potential mechanisms to explain the negative impact of MOR in the survival of patients with those cancers include facilitation of tumour growth by increased angiogenesis and cell cycle progression, promotion of metastasis by stimulating cell migration and invasion, and modulation of the tumour microenvironment.[7] However, expression of MOR was not associated with negative prognosis in oesophageal squamous cell carcinoma or colorectal cancers.[8,9]

Two retrospective studies have indicated that opioids are associated with poor prognosis in patients with LSCC.[10,11] However, it is unclear whether the level of expression of MOR is associated with long-term oncologic outcomes in LSCC. Therefore, we conducted a retrospective study to investigate the association between MOR expression and disease-free (DFS) and overall survival (OS) in patients with LSCC. Furthermore, the differences in expression of MOR between tumour and adjacent tissue, and the relationship between tumour MOR levels and intraoperative opioid consumption, postoperative complications, and hospital length of stay (LOS) were evaluated.

Methods

Study population

This study was approved by the Ethics Committee of Fudan University Shanghai Cancer Centre (FUSCC), China (Protocol #1901208-3). From January 2014 to December 2017, patients undergoing laryngectomy for LSCC with complete clinical characteristic data, OS, and DFS records were enrolled in this retrospective cohort. All patients enrolled signed a consent form for data use for research before receiving treatment. The exclusion criteria included previous or other concomitant cancers, distant cancer metastasis, preoperative radiotherapy, chronic inflammatory diseases, and infections including autoimmune diseases, and a loss of contact during follow-up. Data were collected from the database of the FUSCC clinical information system. The medical information of each patient was reviewed and recorded, which included demographic information, medical history, primary diagnosis, operative details, tumour differentiation and pathological staging, and DFS and OS time (Fig. 1a).

Primary outcome

The primary outcomes of this study were DFS and OS. DFS was defined as the interval between the date of surgery and the date of tumour recurrence or December 31, 2018. OS was defined as the length of time from the date of surgery to the date of death or the last follow-up date.

Secondary outcomes

Secondary endpoints included intraoperative sufentanil consumption, grade of surgical complications according to the Clavien-Dindo classification, and hospital LOS.

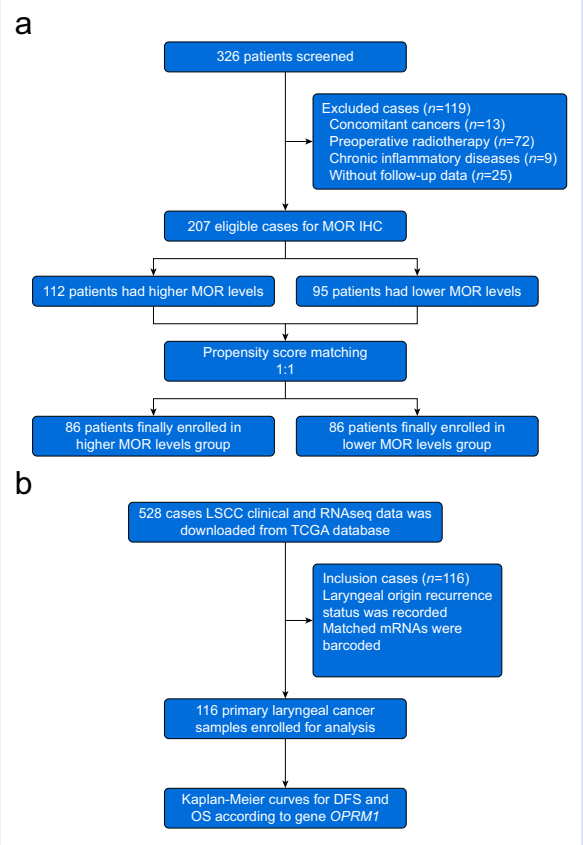

Fig 1. Flow diagram detailing the selection process for patients included in this retrospective analysis. (a) Flow diagram of LSCC patients enrolled in this study; (b) flow diagram of 528 cases LSCC clinical and RNAseq data from the TCGA database. DFS, disease-free survival; IHC, immunohistochemistry; LSCC, laryngeal squamous cell carcinoma; MOR, mu-opioid receptor; OS, overall survival; TCGA, The Cancer Genome Atlas.

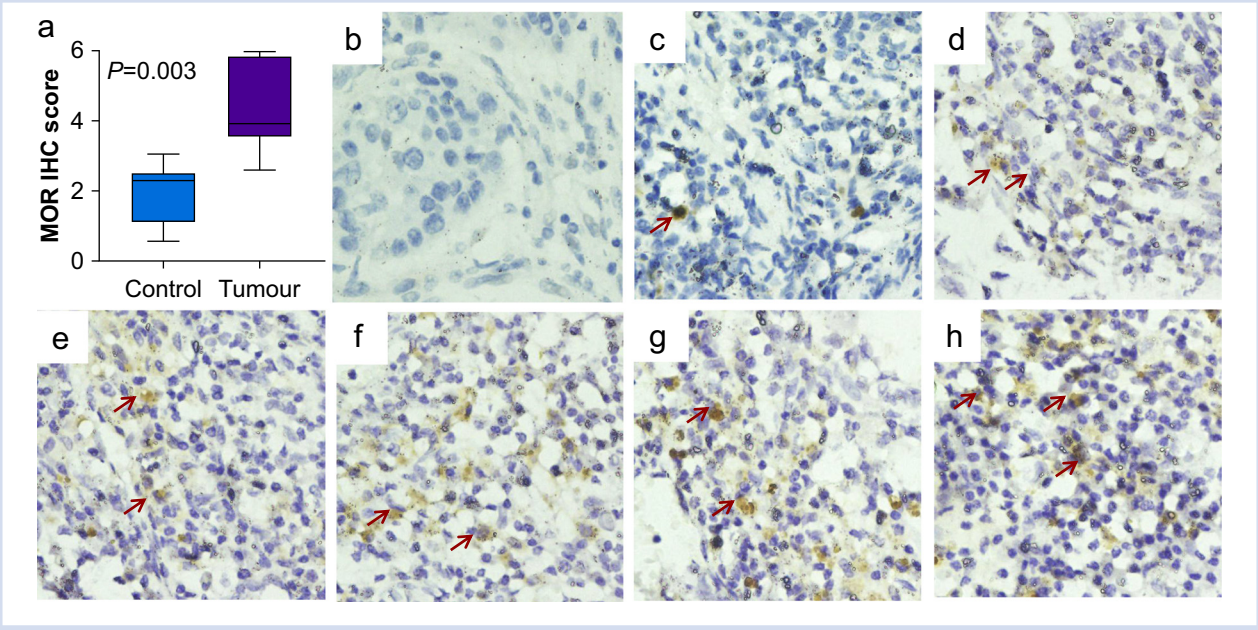

Fig 2. Representative image of immunohistochemistry sample to describe scoring and MOR labelling. All images are at 400×. (a) MOR IHC score in laryngeal squamous cell carcinoma tumour tissue and adjacent non-tumour tissue; (b) score 0; (c) score 1; (d) score 2; (e) score 3; (f) score 4; (g) score 5; (h) score 6. ICH, immunohistochemistry; MOR, mu-opioid receptor.

Survival data analysis from The Cancer Genome Atlas database

LSCC clinical and level 3 RSEM normalised RNAseq data were downloaded from The Cancer Genome Atlas (TCGA) database (http://firebrowse.org/). Samples of laryngeal cancer were selected according to the following criteria: (1) laryngeal origin; (2) recurrence status was recorded; and (3) matched mRNAs were barcoded. In total, 116 primary laryngeal cancer samples were selected from 528 LSCC cases. The optimal cut-off was defined as the point with the most significant (log-rank test) split for DFS and OS (Fig. 1b).

Anaesthesia care

All patients were monitored according to ASA monitoring standards. In all patients, general anaesthesia was induced with propofol (target-controlled infusion, effect-site concentration: 3.0–4.0 μg ml^{-1}), sufentanil (0.3–0.5 μg kg^{-1}), and rocuronium (0.6 mg kg^{-1}). After tracheal intubation, general anaesthesia was maintained with sevoflurane 2.0–3.0 vol% in an oxygen/air mixture. Repeated bolus injections of sufentanil and rocuronium were given as necessary throughout the operation. Flurbiprofen 50 mg i.v. was used for postoperative rescue analgesia.

Immunohistochemistry

Immunohistochemical staining was performed using a horseradish peroxidase method. Paraffin sections from 207 patients with LSCC were selected to detect expression of MOR in tumour tissue and adjacent non-tumour tissue. Anti-MOR antibody (UMB3)-C-terminal (ab134054) was used overnight at 4°C at a working concentration of 1:200.[12–14] The secondary antibody was goat anti-rabbit IgG H&L (HRP) (ab205718). To minimise sampling bias, five regions of each sample were selected and the number of positive cells per region was estimated using Image Pro plus 6.0 (Media Cybernetics Inc., Bethesda, MD, USA).

Evaluation of staining of MOR

All slides for immunohistochemistry were independently and blindly assessed and scored by two investigators. An immunohistochemistry score for MOR ranging from 0 to 6 was determined as the sum of staining intensity and proportion of immunopositive cancer cells according to previously published criteria.[9] Staining intensity was scored as 0 (no staining), 1 (weakly stained), 2 (moderately stained), or 3 (strongly stained). The percent positivity was scored as 0 (<5%, negative), 1 (5–25%, sporadic), 2 (25–50%, focal), or 3 (>50%, diffuse). If disagreement between the two pathology investigators occurred, the slides were re-examined to obtain a final consensus. The expression of MOR was scored by adding the intensity staining scores and the proportion of positive cancer cells stained. The total ranged from 0 to 6 (Fig. 2 b–h).

Statistical analysis

We compared different clinical and pathologic factors between high and low MOR expression groups. We also compared intraoperative sufentanil consumption and LOS between both groups. Descriptive statistics including mean and standard deviation were used for continuous variables. Frequency counts and percentages were calculated for categorical variables such as sex, ASA physical status, surgical procedure, histological grade, primary tumour site, T classification, clinical stage, tumour size, lymph node status, postoperative radiotherapy, and grade of surgical complications.

Table 1 Correlation of mu-opioid receptor expression with clinical pathological features of patients before and after propensity score matching.

Variable	MOR expression(before matching)		P	MOR expression (after matching)		Standard difference (%)
	Low (*n*=95)	High (*n*=112)		Low (*n*=86)	High (*n*=86)	
Age (*n*, %)			0.061			2.89
<60 yr	59 (62.1)	55 (49.1)		55 (63.9)	54 (62.8)	
≥60 yr	36 (37.9)	57 (50.9)		31 (36.1)	32 (37.2)	
Sex (*n*, %)			0.554			–
Male	89 (93.7)	107 (95.5)		82 (95.3)	83 (96.5)	
Female	6 (6.3)	5 (4.5)		4 (4.7)	3 (3.5)	
Alcohol consumption			0.416			–
Yes	42 (45.2)	54 (48.2)		39 (45.3)	40 (46.5)	
No	40 (42.1)	49 (43.6)		37 (43.0)	39 (45.3)	
Unknown	13 (13.7)	9 (8.2)		10 (11.7)	7 (8.2)	
Smoking history			0.748			–
Yes	43 (45.3)	56 (50.0)		41 (47.7)	41 (47.7)	
No	44 (46.3)	46 (41.1)		39 (45.3)	38 (44.2)	
Unknown	8 (8.4)	10 (8.9)		6 (7)	7 (8.1)	
ASA physical status (*n*, %)			0.367			–
1–2	77 (81.0)	96 (85.7)		71 (82.6)	72 (83.7)	
3	18 (19.0)	16 (14.3)		15 (17.4)	14 (16.3)	
Surgical procedure (*n*, %)			0.298			4.62
Partial laryngectomy	50 (52.6)	67 (59.8)		46 (53.5)	45 (52.3)	
Total laryngectomy	45 (47.4)	45 (40.2)		40 (46.5)	41 (47.7)	
Histological grade (*n*, %)			0.013			3.65
G1	48 (50.5)	31 (27.7)		44 (51.2)	30 (34.9)	
G2+G3	47 (49.5)	81 (72.3)		42 (48.8)	56 (65.1)	
Primary tumour site (*n*, %)			0.200			2.98
Glottic	59 (62.1)	79 (70.5)		56 (65.1)	57 (66.3)	
Others	36 (37.9)	33 (29.5)		30 (34.9)	29 (33.7)	
T classification			0.043			5.15
T1+T2	55 (57.9)	49 (43.6)		49 (57.0)	47 (54.7)	
T3+T4	40 (42.1)	63 (56.4)		37 (43.0)	39 (45.3)	
Clinical stage (*n*, %)			0.009			4.89
I–II	52 (54.7)	41 (36.6)		48 (55.8)	40 (46.5)	
III–IV	43 (45.3)	71 (63.4)		38 (44.2)	46 (53.5)	
Tumour size (*n*, %)			<0.001			3.75
≤2 cm	72 (75.8)	67 (59.8)		66 (76.7)	63 (73.3)	
>2 cm	23 (24.2)	45 (40.2)		20 (23.3)	23 (26.3)	
Lymph node status			<0.001			5.68
N0	57 (60.0)	49 (43.6)		50 (58.1)	47 (54.7)	
N+	38 (40.0)	63 (56.4)		36 (41.9)	39 (45.3)	
Postoperative radiotherapy			0.694			6.25
Yes	76 (80.0)	92 (82.1)		68 (79.0)	67 (77.9)	
No	19 (20.0)	20 (17.9)		18 (21.0)	19 (22.1)	

Data shown as mean (standard deviation) or *n* (%).
MOR, mu-opioid receptor.
$P \leq 0.05$ was considered statistically significant.

The χ^2 test was used to evaluate the association between two categorical variables. The Kaplan–Meier method was used for time-to-event analysis, including DFS and OS for MOR and *ORPM1* analysis. The median time to the event in months with a 95% confidence interval (CI) was calculated. The log-rank test was used to evaluate the difference in time-to-event endpoints between groups. Univariate Cox proportional hazards models were fitted to evaluate the effects of continuous variables on time-to-event outcomes. Multivariable Cox proportional hazard models were used for multivariate analysis to include important and significant covariates. We performed propensity score matching analysis to reduce selection bias by building a matched group of patients to compare OS and DFS between patients with high MOR expression and those with low MOR expression. Nine variables were entered in our propensity model: age, surgical procedure, histological grade, primary tumour site, T classification, clinical stage, tumour size, lymph node status, and administration of postoperative radiotherapy. Patients were matched using a 5-to-1 digit greedy match algorithm. Cut-off values of MOR expression used for survival analysis were determined by X-tile 3.6.1 software[15] (Yale University, New Haven, CT, USA). Statistical analyses were performed with SPSS version 17.0 (SPSS Inc., Chicago, IL, USA). $P<0.05$ was considered statistically significant.

Results

A total of 207 patients who underwent laryngectomy for LSCC were enrolled in this study. MOR levels in tumour tissues were

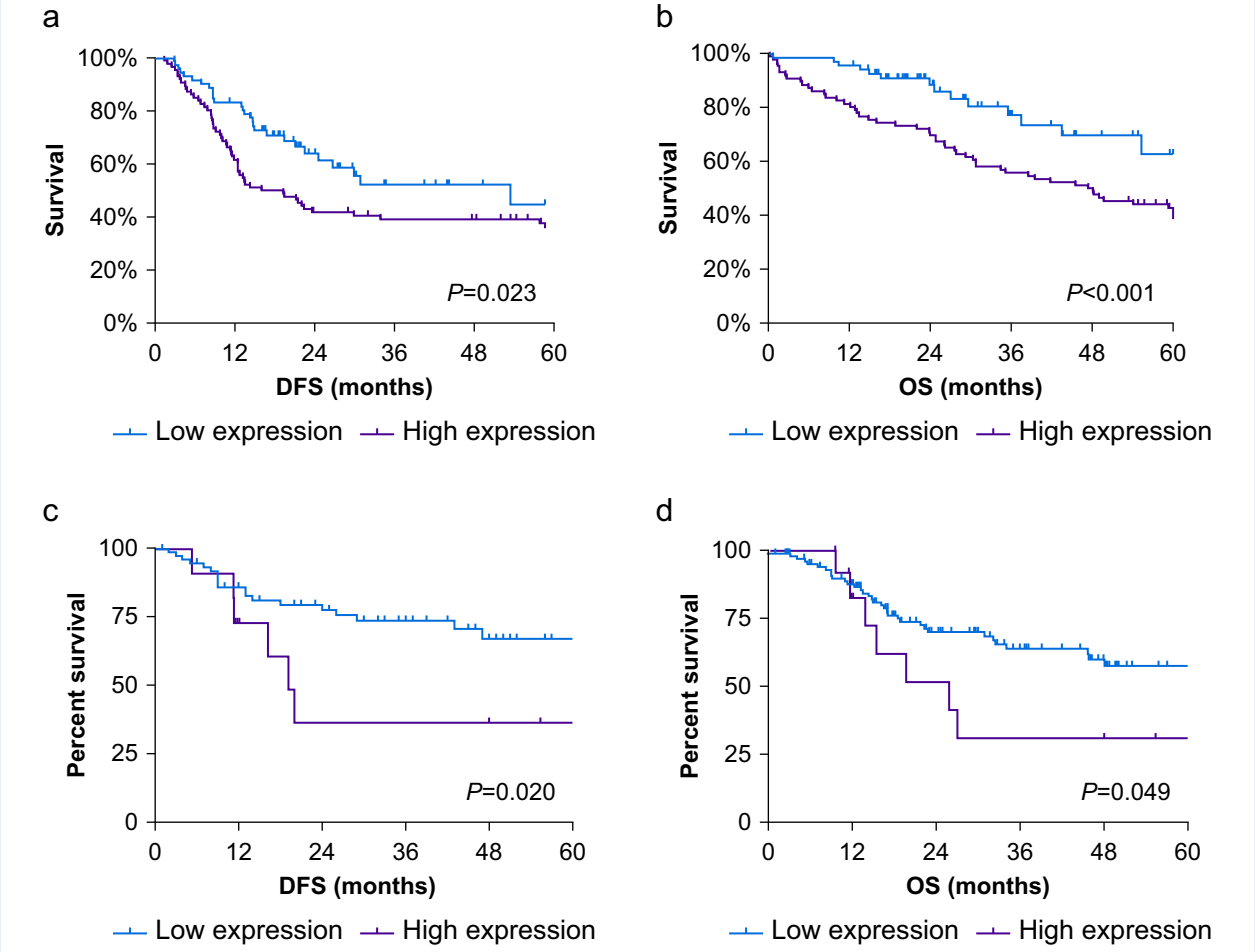

Fig 3. (a) DFS curves from the date of surgery according to expression of MOR; (b) OS curves from the date of surgery according to expression of MOR; (c) DFS curves from the date of surgery according to expression of *OPRM1* from the TCGA database; (d) OS curves from the date of surgery according to expression of *OPRM1* from the TCGA database. DFS, disease-free survival; MOR, mu-opioid receptor; OS, overall survival; TCGA, The Cancer Genome Atlas.

significantly higher than those in adjacent non-tumour tissue (P=0.003, Fig. 1a). The level of MOR was positively correlated with histological grade (G1 *vs* G2+G3, P=0.013), T classification (T1+T2 *vs* T3+T4, P=0.043), clinical stage (I–II *vs* III–IV, P=0.009), lymph node status (P<0.001), and tumour size (P<0.001). However, MOR level was not significantly associated with age (P=0.061), sex (P=0.554), alcohol consumption (P=0.416), smoking history (P=0.748), ASA physical status (P=0.367), surgical procedure (P=0.298), and primary tumour site (P=0.200) (Table 1).

Primary endpoint

The median follow-up time for all patients was 46.6 months (95% CI: 43.7–60.0). After propensity score matching, Kaplan–Meier survival analyses are shown in Figure 3. Compared with the low MOR group, DFS in the high MOR group at 1, 3, and 5 yr after surgery was significantly lower (1-yr DFS: 81.2% *vs*. 60.0%, P=0.019; 3-yr DFS: 50.2% *vs* 39.2%, P=0.026; 5-yr DFS: 42.5% *vs* 37.2%, P=0.023, respectively, Fig. 3a). Univariate Cox regression analysis showed that histological grade (P=0.010), T classification (P=0.003), clinical stage (P=0.021), lymph node status (P=0.023), no postoperative radiotherapy (P<0.001), and high MOR expression (P<0.001) were associated with worse DFS (Table 2). In the multivariate Cox proportional hazards model, the following covariates were significantly associated with shorter DFS: higher histological grade (HR: 2.20, 95% CI: 2.13–2.71, P<0.001), T classification (HR: 1.27, 95% CI: 1.14–1.85, P=0.035), clinical stage (HR: 2.32, 95% CI: 1.76–2.66, P=0.026), positive lymph node status (HR: 1.32, 95% CI: 1.16–1.86, P=0.015), no postoperative radiotherapy treatment (HR: 1.89, 95% CI: 1.15–2.84, P=0.002). The association between high MOR expression and low DFS was statistically significant in the model (HR: 1.64, 95% CI: 1.04–2.39, P=0.025, Table 3). After propensity score matching, the association between high MOR expression and low DFS remained statistically significant (HR: 1.52, 95% CI: 1.07–2.25, P=0.034, Table 3).

Kaplan–Meier curves for OS indicated that lower MOR expression was associated with improved OS. After matching, OS at 1, 3, and 5 yr after surgery was significantly higher in patients with lower MOR expression than with higher MOR expression (1-yr OS: 93.2% *vs* 81.2%, P=0.027, 3-yr OS: 78.3% *vs*

Table 2 Cox univariate analysis of contributory factors to DFS and OS among patients.

Variables	OS		DFS	
	HR (95% CI)	P-value	HR (95% CI)	P-value
Age (yr)	1.12 (0.93–2.73)	0.523	1.34 (0.97–3.60)	0.325
Sex (male)	0.86 (0.71–1.14)	0.640	0.90 (0.83–1.18)	0.462
Alcohol consumption (yes)	1.32 (0.68–2.15)	0.665	1.46 (0.73–1.59)	0.764
Smoking history (yes)	1.07 (0.49–1.27)	0.237	1.17 (0.53–1.39)	0.072
ASA (3)	1.22 (0.73–1.53)	0.171	1.44 (0.84–1.98)	0.426
Surgical procedure (total laryngectomy)	1.16 (0.92–1.42)	0.975	1.55 (0.90–2.67)	0.115
Histological grade (G2+G3)	2.36 (1.72–2.70)	<0.001	2.70 (1.78–3.83)	0.010
T classification (T3+T4)	1.45 (1.83–2.53)	<0.001	2.58 (1.34–4.21)	0.003
Clinical stage (III–IV)	2.89 (1.14–3.14)	0.013	2.97 (1.17–4.54)	0.021
Tumour size (>2 cm)	1.10 (0.41–1.97)	0.845	1.34 (0.52–1.61)	0.545
Lymph node status (N+)	1.45 (1.22–1.92)	0.011	1.69 (1.63–2.90)	0.023
Postoperative radiotherapy (no)	2.15 (1.52–3.52)	<0.001	2.85 (2.26–3.52)	<0.001
MOR expression (high)	1.85 (1.38–3.63)	<0.001	2.05 (1.92–3.63)	<0.001

CI, confidence interval; HR, hazard ratio; DFS, disease-free survival; MOR=mu-opioid receptor; OS, overall survival.
P≤0.05 was considered statistically significant.

57.6%, P<0.001, 5-yr OS: 60.3% *vs* 42.5%, P<0.001, respectively, Fig. 3b). The univariate Cox regression analysis showed that histological grade (P<0.001), T classification (P<0.001), clinical stage (P=0.013), lymph node status (P=0.011), no postoperative radiotherapy (P<0.001), and higher MOR expression (P<0.001) were associated with shorter OS (Table 2). Multivariate analysis showed that the following covariates were significantly associated with reduced OS: high histological grade (HR: 2.12, 95% CI: 2.07–2.62, P<0.001), T classification (HR: 1.22, 95% CI: 1.08–2.11, P=0.022), clinical stage (HR: 2.16, 95% CI: 1.76–2.32, P=0.026), positive lymph node status (HR: 1.26, 95% CI: 1.16–1.44, P=0.019), no postoperative radiotherapy treatment (HR: 1.85, 95% CI: 1.34–2.86, P=0.004). The association between high MOR expression and reduced OS was statistically significant in the model (HR: 1.55, 95% CI: 1.04–2.56, P=0.039, Table 3). After matching, the association between high MOR expression and reduced OS was still statistically significant (HR: 1.42, 95% CI: 1.17–2.34, P=0.032, Table 3).

Using the TCGA database, we investigated the impact of expression of the *OPRM1* gene on our primary endpoints. Kaplan–Meier curves for DFS and OS indicated that high *OPRM1* expression in laryngeal cancer is an indicator of poor prognosis (DFS: $OPRM1^{high}$ *vs* $OPRM1^{low}$, P=0.020, OS: P=0.049, respectively, Fig. 3c and d).

Secondary outcomes

The mean intraoperative sufentanil consumption was significantly lower in the low MOR expression group (40.2 [5.6] μg) than in the high MOR expression group (56.2 [5.8] μg) (P<0.001, Fig. 4a). Postoperative complications after surgery according to the Clavien-Dindo classification were no different between high MOR expression and low MOR expression groups (Grade I, 22.3% *vs* 25.4%, Grade II 12.6% *vs* 12.3%, Grade III 5.3% *vs* 5.2%, P=0.492, Fig. 4b). In terms of hospital LOS, the median duration (inter-quartile range) in the high MOR expression group was 8.4 (7.5–8.9) days, whereas in the low MOR expression group, mean LOS was 8.1 days (6.5–8.3) (P=0.120, Fig. 4c).

Discussion

We aimed to determine the association between MOR and *OPRM1* expression and DFS and OS in LSCC patients. The results showed that expression of MOR was higher in LSCC tumour tissue than in adjacent non-tumour tissue. The analysis also indicated that higher expression of MOR or *OPRM1* in tumour tissue was independently associated with shorter DFS and OS. Our findings are in line with other studies. Zylla and colleagues[4] found that for every 1% increase in MOR

Table 3 Cox multivariable analysis of contributory factors to DFS and OS among patients.

Variables	OS (before match)		OS (after match)		DFS (before match)		DFS (after match)	
	HR (95% CI)	P-value	HR (95% CI)	P-value	HR (95% CI)	P-value	HR (95% CI)	P-value
Histological grade (G2+G3)	2.12 (2.07–2.62)	<0.001	2.02 (1.95–2.43)	<0.001	2.20 (2.13–2.71)	<0.001	2.10 (2.07–2.32)	<0.001
T classification (T3+T4)	1.22 (1.08–2.11)	0.022	1.14 (1.07–1.62)	0.027	1.27 (1.14–1.85)	0.035	1.18 (1.10–1.65)	0.038
Clinical stage (III–IV)	2.16 (1.76–2.32)	0.026	1.72 (1.67–2.12)	0.032	2.32 (1.76–2.66)	0.026	2.24 (1.87–2.54)	0.035
Lymph node status (N+)	1.26 (1.16–1.44)	0.019	1.13 (1.06–1.54)	0.024	1.32 (1.16–1.86)	0.015	1.22 (1.17–1.61)	0.026
Postoperative radiotherapy (no)	1.85 (1.34–2.86)	0.004	1.68 (1.47–2.30)	0.015	1.89 (1.15–2.84)	0.002	1.62 (1.17–2.60)	0.016
MOR expression (high)	1.55 (1.12–2.56)	0.039	1.42 (1.17–2.34)	0.032	1.64 (1.04–2.39)	0.025	1.52 (1.07–2.25)	0.034

CI, confidence interval; DFS, disease-free survival; HR, hazard ratio; MOR, mu-opioid receptor; OS, overall survival.
P≤0.05 was considered statistically significant.

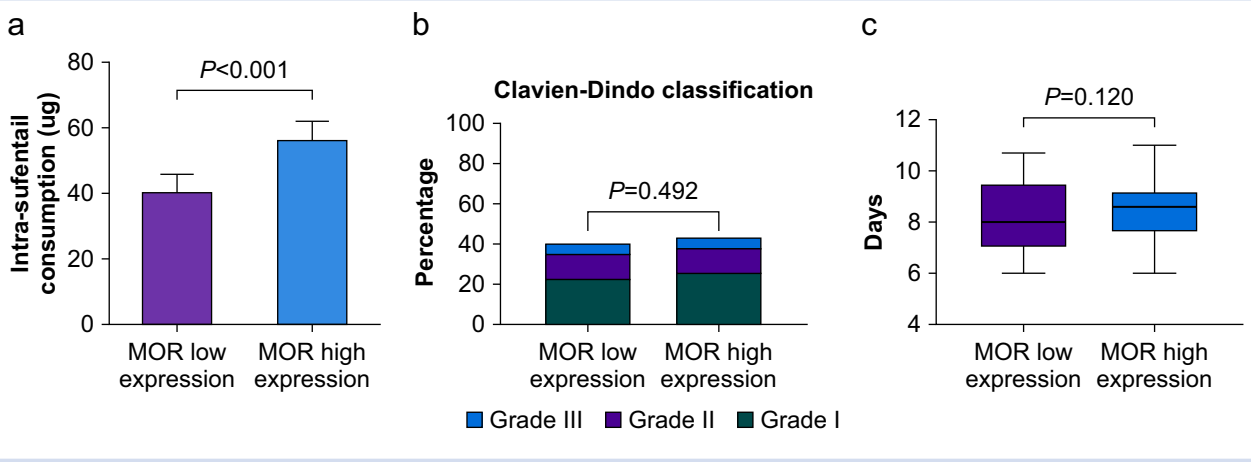

Fig 4. (a) Intraoperative sufentanil consumption between groups; (b) percentage of postoperative complications between groups according to the Clavien-Dindo classification; (c) hospital length of stay between groups. MOR, mu-opioid receptor. *P<0.001.

expression there was a 65% and a 55% higher risk of progression/death or death, respectively, in patients with prostate cancer. In patients with non-small cell lung cancer, higher expression of MOR was found in patients with metastasis to lymph nodes than those without metastatic disease, also suggesting a role of the receptor in cancer aggressiveness.[5]

Opioids are commonly used to provide analgesia during general anaesthesia, and are the widely used agents for treatment of acute pain in the immediate postoperative period. Laryngectomy is a lengthy procedure with great surgical trauma that might require administration of a large amount of opioid analgesics.[16] Cata and colleagues[10] investigated the impact of opioid administration during laryngectomy for LSCC. They reported that patients requiring high dosages of opioids had worse survival after adjusting for relevant clinical and histological variables associated with cancer progression.[10,11]

We also observed that patients with increased opioid requirements showed higher levels of MOR. In fact, tumours with high MOR expression were larger, had a higher T score, and had positive metastatic lymph nodes. We speculate that patients with extensive surgical resections require larger doses of opioids as a result of more aggressive tumours. However, the association between MOR expression and survival remained statistically significant after matching for those important prognostic factors. Alternatively, it can be hypothesised that high concentrations of opioids can trigger overexpression of MOR in cancer cells. Opioid receptors have been implicated in hepatocarcinoma progression, both directly and indirectly. Activation of opioid receptors can directly regulate the function of hepatic stellate cells, leading to liver cirrhosis, and play an important role in cholestasis, which indirectly promotes hepatocarcinoma formation.[17,18] Levins and colleagues[19] showed that opioid-sparing analgesia reduced MOR expression in breast cancer. In laryngeal cancers, Shoffel-Havakuk and colleagues[11] showed increased expression of MOR in the larynx of heroin drug users. In comparison to control subjects, heroin users showed dysregulated expression of *OPRM1* splice variant mRNAs in the medial prefrontal cortex, again suggesting that opioids could induce overexpression of MOR.[20]

The impact of anaesthesia on the long-term prognosis of cancer patients who undergo surgery is increasingly recognised as important. However, these results are conflicting. Current evidence suggests that anaesthetics can modulate inflammatory endpoints, the immune response, mechanisms linked with cell proliferation, migration and invasion, stemness, and angiogenesis.[21] However, they cannot fully explain cancer recurrence months to years after surgery. In fact, a recent RCT showed that regional anaesthesia did not impact survival or recurrence after breast cancer surgery.[22]

A delay in postoperative recovery and return to intended oncological therapies (RIOT) as a result of postoperative complications can have a negative impact on cancer progression.[23,24] Our results indicate that complications and recovery were nearly identical in patients with high and low MOR expression. Therefore, we conclude that neither a delay in recovery nor complications impacted the different oncological outcomes between groups.

Some limitations must be acknowledged: (1) the retrospective design of the study is associated with biases that may have influenced our analysis and findings; (2) although this is the largest study of the association between MOR expression and laryngeal cancer recurrence, the low rate of events limits the statistical power of any association; (3) lack of evaluation of the *OPRM1* gene variant polymorphisms, which have been shown to affect survival in other cancers; and (4) the fact that only perioperative opioid use was recorded.

In conclusion, MOR expression was increased in LSCC tissue and was associated with reduced DFS or OS. Our results suggest that MOR is a valuable molecular biomarker or an actionable target in the prognosis and treatment, respectively, of LSCC. Further investigations are warranted to evaluate whether blockade of the receptor during the perioperative period might confer survival benefits in patients with LSCC.

Authors' contributions

Study design: HZ, JPC, WKC, CHM
Coordination: DZ, ZRS, AG
Data acquisition: HZ, WKC, AG, MLS
Data interpretation: DZ, ZRS, AG, WAZ

Drafting: HZ, WKC, JPC
Final approval of manuscript: all authors

Declarations of interest

The authors declare that they have no conflicts of interest.

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