

学位論文

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PET volume parameters in p16-negative oropharyngeal,
hypopharyngeal, and supraglottic squamous cell carcinoma

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Predictive value of local control by 4'-[methyl-¹¹C]-thiotymidine PET volume parameters in p16-negative oropharyngeal, hypopharyngeal, and supraglottic squamous cell carcinoma

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Purpose We investigated the potential of baseline 4'-[methyl-¹¹C]-thiothymidine ([¹¹C]4DST) PET for predicting loco-regional control of head and neck squamous cell carcinoma (HNSCC).

Methods A retrospective analysis was performed using volumetric parameters, such as SUVmax, proliferative tumor volume (PTV), and total lesion proliferation (TLP), of pretreatment [¹¹C]4DST PET for 91 patients with HNSCC with primary lesions in the oral cavity, hypopharynx, supraglottis, and oropharynx, which included p16-negative patients. PTV and TLP were calculated for primary lesions and metastatic lymph nodes combined. We examined the association among the parameters and relapse-free survival and whether case selection focused on biological characteristics improved the accuracy of prognosis prediction.

Results The area under the curves (AUCs) using PTV and TLP were high for the oropharyngeal/hypopharyngeal/supraglottis groups (0.91 and 0.87, respectively), whereas that of SUVmax was 0.66 ($P < 0.01$).

On the other hand, the oral group had lower AUCs for PTV and TLP (0.72 and 0.77, respectively). When all cases were examined, the AUCs using PTV and TLP were 0.84 and 0.83, respectively.

Conclusion Baseline [¹¹C]4DST PET/CT volume-based parameters can provide important prognostic information with p16-negative oropharyngeal, hypopharyngeal, and supraglottic cancer patients. *Nucl Med Commun XXX: XXXX–XXXX* Copyright © 2024 Wolters Kluwer Health, Inc. All rights reserved.

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Keywords: [¹¹C]4DST PET, head and neck carcinoma, proliferative tumor volume, relapse-free survival, total lesion proliferation

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Introduction

Head and neck cancers are responsible for more than 880 000 cases and approximately 450 000 deaths annually worldwide [1]. Estimates of the male-to-female ratio range from 2:1 to 4:1. Approximately 90% of all head and neck cancers are head and neck squamous cell carcinomas (HNSCCs). By incidence worldwide, HNSCC is the sixth leading cancer. Most HNSCCs are formed in the epithelial lining of the oral cavity, oropharynx, larynx, and hypopharynx [2].

Although they are the same squamous cell carcinoma, treatment strategies and prognosis vary depending on the location of the tumor. However, because lymphatic flow and anatomical function (swallowing, breathing, and effect on quality of life) of the oropharynx, hypopharynx, and supraglottic sites are identical, their treatment strategies are similar [3]. The prognosis of oropharyngeal carcinoma differs depending on whether it is human papillomavirus (HPV)-associated or not, and it is classified

according to p16 status, a surrogate marker for HPV. p16-positive oropharyngeal carcinoma is considered to have a good prognosis. Surgery is recommended for oral cancer, and radiotherapy (RT) and surgery are often the treatment of choice for pharyngeal and laryngeal cancer. In addition, the recommended extent of neck dissection for the treatment of cervical lymph node metastasis, which is associated with prognosis, is different for oral cavity cancer and pharyngeal/laryngeal cancer due to differences in lymphatic flow. RT is often selected when both preservation of organ function and radical cure are expected, whereas surgery is often chosen when the lesion is small and functional impairment is minimal [4] or radical cure with RT is difficult.

4'-[methyl-¹¹C]-thiothymidine ([¹¹C]4DST) is a tracer for PET developed by Toyohara *et al.* [5] that is resistant to thymidine phosphatase and is incorporated into DNA. Because of its characteristics, [¹¹C]4DST PET has been suggested to be superior to

2-deoxy-2-[¹⁸F]-fluorodeoxyglucose (¹⁸F]FDG) PET, which determines sites of cellular glucose metabolism, in determining cell proliferation and as an indicator for predicting therapeutic effect [6].

A histopathological study reported that overexpression of the nuclear proliferation marker Ki-67 was strongly associated with recurrence and independently predicted poor relapse-free survival (RFS, defined as the interval between the date of the initial medical examination and the date of detection of recurrent/metastatic disease clinically or by follow-up CT or other imaging modalities) [7]. However, the histopathology of cancer is heterogeneous; thus, results may depend on the location that is sampled. A PET study reported that [¹¹C]4DST PET showed a higher correlation with cell proliferation, as evaluated by Ki-67 index, than [¹⁸F]FDG PET in non-small cell lung cancer (NSCLC); thus, this finding suggests that [¹¹C]4DST PET/CT is a useful noninvasive modality for imaging DNA synthesis by NSCLC [8]. The present study aimed to evaluate the overall malignancy of cancerous tissue, including metastatic lesions, using pre-treatment [¹¹C]4DST PET/CT and investigate RFS prediction accuracy of [¹¹C]4DST PET/CT.

Our previous study reported that [¹¹C]4DST PET may be more useful than [¹⁸F]FDG PET for predicting clinical outcome of head and neck cancer, using volumetric parameters such as metabolic/proliferative tumor volume (PTV) and total lesion glycolysis/proliferation [9]. It included all HNSCC cases including oropharyngeal, hypopharyngeal, laryngeal, oral, and maxillary carcinomas. We hypothesized that focusing on hypopharyngeal, supraglottic, and p16-negative oropharyngeal cancers, which are assumed to have similar biological characteristics, would be more useful for improving the accuracy of prognosis prediction than analyzing all head and neck cancers together, as few reports have examined these cancers together by primary site.

The aim of the present study was to investigate the prediction accuracy of RFS in each primary site (especially in oropharynx/hypopharynx/supraglottis) for volumetric parameters of pre-treatment [¹¹C]4DST PET.

Material and methods

Patients

We conducted a retrospective analysis of prospectively collected data. This retrospective cohort study evaluated the prediction accuracy of RFS using maximum standardized uptake value (SUV_{max}), PTV, and total lesion proliferation (TLP) of [¹¹C]4DST PET in patients with newly diagnosed HNSCC. We included patients who received initial treatment at Kagawa University Hospital between May 2011 and March 2020. Other inclusion criteria were: (1) histologically proven squamous cell carcinoma without distant metastasis; (2) head and neck cancer with primary sites in the oral cavity, oropharynx,

hypopharynx, or supraglottis; (3) no prior treatment for head and neck cancer; (4) stage III/IV disease (classified by UICC 7th edition); and (5) [¹¹C]4DST PET performed prior to treatment. For cancers of the oropharynx, the exclusion criterion was patients confirmed to be p16-positive at the time of diagnosis.

Image analysis

The SUV was calculated using the following formula: $SUV = c_{dc}/(d_i/w)$, where c_{dc} is the decay-corrected tracer tissue concentration (Bq/g); d_i is the injected dose (Bq); and w is the patient's body weight (g). PTV values were computed from attenuation-corrected PET data using a commercial software package (Synapse Vincent, Fujifilm Medical, Tokyo, Japan) for each of primary tumor and metastatic lymph nodes. The software automatically measured the PTV of the tumor and metastatic lymph nodes using the selected SUV threshold within the region of interest contoured by a board-certified nuclear medicine physician. The contouring margins around the tumor and lymph nodes were defined using SUVs of 2.5. TLP was calculated for [¹¹C]4DST PET using the following formula: (PTV) × (SUV_{mean}).

Regional lymph node metastases not confirmed pathologically were diagnosed by enhanced CT or [¹¹C]4DST PET/CT. Criteria for detecting regional lymph node metastases by CT were as follows: (1) lymphadenopathy with a minimum diameter of 10 mm or more, (2) lymph nodes with necrosis, and (3) the appearance of crushed lymph nodes. The criterion for detection of lymph node metastasis by [¹¹C]4DST PET was visual confirmation. Higher [¹¹C]4DST PET uptake in ipsilateral lymph nodes than in surrounding tissue was considered positive. High [¹¹C]4DST PET uptake without positive findings on enhanced CT compared to the contralateral neck was considered negative.

Relapse was defined as exhibiting recurrence or disease progression histologically or by follow-up CT or other imaging modalities. Relapse-free was defined as having no local recurrence or distant metastasis.

Treatment and follow-up

RT using 4 MV photons was administered at the primary tumor site and the neck once daily, at paired bilaterally opposed fields in the upper neck, and an anterior port at the lower neck. As a radical treatment, patients were irradiated with a total dose of 66–70 Gy in 2 Gy fractions once daily. After 40 Gy had been administered, the clinical target volume was reduced to encompass only the primary tumor and the involved cervical lymph nodes.

In surgical treatment (ST) cases, the primary lesion was resected with the goal of preserving function after resection as much as possible, while maintaining a radical cure. The necessity of neck dissection and the extent of dissection were determined according to the presence or

absence of cervical lymph node metastasis and the extension of the primary lesion. When reconstructive surgery was required, we collaborated with plastic and gastrointestinal surgeons as necessary.

After completion of treatment, patients were followed up every 1–2 months for the first 2 years, and then every 3 months thereafter. In cases of cancer of the pharynx and larynx, pharyngolaryngoscopy was performed at each visit. In addition, CT or MRI imaging studies were performed every 3 months in the first year, and then every 4–6 months after the second year.

Radiotracer synthesis and PET/CT imaging

All imaging was performed using a Biograph mCT 64-slice PET/CT scanner (Siemens Medical Solutions USA Inc., Knoxville, TN, USA) with an axial field of view of 21.6 cm.

[¹¹C]4DST was produced using an automated synthesizer equipped with an HM-18 cyclotron (QUPID; Sumitomo Heavy Industries Ltd., Tokyo, Japan) as synthesized using the method described in Toyohara *et al.* [5]. [¹¹C]4DST emission data were acquired from mid-cranial to proximal thigh (2 min per bed position) at 15 min after intravenous injection of 4DST (7.4 MBq/kg). Unenhanced low-dose CT of the same region was performed for attenuation correction and image fusion; PET data were reconstructed with an ordered subset expectation maximization algorithm incorporating a Gaussian filtered point spread function (PSF) and time-of-flight model correction (two iterations, 21 subsets). Quantification was based on PSF-reconstructed data. [¹¹C]4DST PET/CT was taken within 3 weeks prior to treatment initiation.

Statistical analysis

The area under the curve (AUC) for predicting RFS for each parameter (SUVmax, PTV, and TLP) were calculated for the oral cavity, oropharynx, hypopharynx, oropharynx/hypopharynx/supraglottis, and all cases. Cutoff values were also calculated. Statistical differences between relapse cases and relapse-free cases were calculated using Mann–Whitney *U* test. Subsequently, univariate analyses in predicting RFS were performed in the lesion of oropharynx/hypopharynx/supraglottis cases. Statistical differences between relapse and relapse-free cases were calculated using Fisher's exact test.

Survival was estimated by the Kaplan–Meier method and analyzed using log-rank test for SUVmax, PTV, and TLP for oropharynx/hypopharynx/supraglottis cases. To evaluate the survival impact of these parameters, we estimated the hazard ratio (HR) and 95% confidence interval using multivariate Cox proportional hazards models. Confounding variables considered in the multivariate analyses were age (<65 vs. ≥65 years), treatment modality (RT vs. ST), and clinical T stage (T2/3 vs. T4).

Stratification was performed by dichotomized confounding variables. All tests were 2-sided, and *P*-values < 0.05 were considered significant for Mann–Whitney *U* test, univariate analyses using Fisher's exact test, and log-rank test. In the Cox proportional hazards models, *P*-values < 0.0167 were considered significant according to the Bonferroni method, as the three variables (SUVmax, PTV, and TLP) were evaluated equally. All statistical analyses were performed using EZR (<https://www.jichi.ac.jp/saitama-sct/SaitamaHP.files/statmed.html>) [10].

Results

The present study included 259 consecutive untreated patients with primary head and neck carcinoma who underwent [¹¹C]4DST PET examination at our institution, of whom, 91 patients met the eligibility criteria and were enrolled in this study. Age, sex, primary tumor site, TNM stage, and treatment are summarized in Table 1. Representative [¹¹C]4DST PET images of recurrent and non-recurrent cases are shown in Fig. 1.

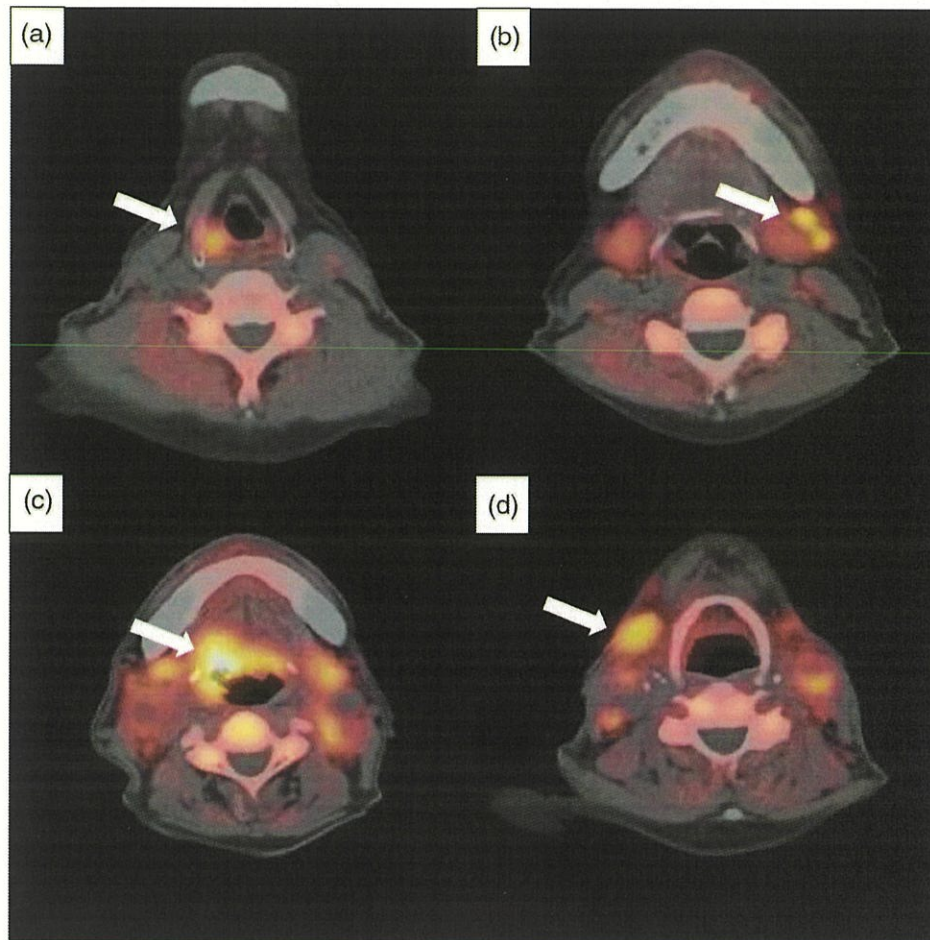
Of these, 64 patients had no recurrence and 27 had recurrence. The median observation period of relapse-free cases was 60.5 months (range 22–104 months), and that for relapse cases was 7.0 months (range 3–25 months). Focally increased [¹¹C]4DST uptake was observed in all primary lesions.

In the RT cases, four patients received RT alone, and 39 patients received concurrent chemoradiotherapy. Chemotherapy agents and administration routes consisted

Table 1 Patient characteristics

Characteristics	Value
Age (years)	
Median	68
Range	46–85
Sex (n)	
Male	80
Female	11
Tumor site (n)	
Oral cavity	26
Oropharynx	24
Hypopharynx	25
Supraglottic	16
T category (n)	
T2	29
T3	32
T4a	30
N category (n)	
N0	22
N1	11
N2 (a:b:c)	57 (2:36:19)
N3	1
TNM stage (n)	
III	22
IV	69
Primary treatment (n)	
Surgical therapy	
Surgery alone	37
Post-CRT	11
Radiotherapy	
Chemoradiotherapy	39
Radiotherapy	4

Fig. 1



$[^{11}\text{C}]4\text{DST-PET}$ images of a patient with cT4aN2cM0 hypopharyngeal cancer. Pre-treatment $[^{11}\text{C}]4\text{DST-PET}$ images show low-level increased metabolism in the primary site (a) and metastatic lymph nodes (b). $[^{11}\text{C}]4\text{DST-PET}$ parameters of all lesions are as follows: SUV max 9.45, PTV 4.86, and TLP 21.26. The patient is alive 52 months after receiving concomitant chemoradiotherapy (70 Gy/35 Fr irradiation and cisplatin 75 mg/m², 5-FU 1000 mg/m², two cycles). (c) and (d) are $[^{11}\text{C}]4\text{DST-PET}$ images of a patient with cT4aN2cM0 oropharyngeal (right side of frontal wall) cancer. Pre-treatment $[^{11}\text{C}]4\text{DST-PET}$ images show relatively high metabolism in the primary site (c). Metastatic lymph nodes (d) show mixed accumulation with high metabolism or no accumulation (suspected necrotic tissue). $[^{11}\text{C}]4\text{DST-PET}$ parameters of all lesions are as follows: SUV max 8.36, PTV 54.42, and TLP 242.18. Three months after receiving concomitant chemoradiotherapy (70 Gy/35 Fr irradiation and 200 mg/m² total dose of cisplatin), pleural metastasis and massive pleural effusion appear. The patient died 7 months after definitive therapy.

of intravenous cisplatin (CDDP; 100 mg/m²) (N = 21), nedaplatin (80 mg/m²) and S-1 (100 mg/day for 14 days) (N = 13), CDDP (70 mg/m²) and 5-fluorouracil (1000 mg/m² continuous infusion for 5 days) (N = 3), intraarterial cisplatin (100 mg/m²) twice during RT (N = 1), six courses of cetuximab (initial doses were 400 mg/m² and subsequent doses were 250 mg/m²) (N = 1).

In ST cases, the primary lesions were resected as follows: total laryngopharyngoesophagectomy (N = 4), partial hypopharyngectomy (N = 1), total laryngectomy and partial hypopharyngectomy (N = 1), total laryngectomy (N = 8), radical tonsillectomy (N = 7), partial oropharyngectomy (N = 3), partial glossectomy (N = 2), hemiglossectomy (N = 7), subtotal glossectomy (N = 5), segmental mandibulotomy (N = 6), marginal

mandibulotomy (N = 1), partial maxillectomy (N = 2), and endoscopic laryngo-pharyngeal surgery (N = 1). In some cases, resection methods were duplicated. Of these, bilateral neck dissection was performed in 33 cases and unilateral dissection in 11 cases. In addition, 34 patients required reconstructive surgery during primary lesion resection. The following types of flaps were used for reconstruction: anterolateral femoral flap (N = 19), rectus abdominis musculocutaneous flap (N = 5), fibula flap (N = 4), free jejunum autograft (N = 4), forearm flap (N = 1), and pectoralis major musculocutaneous flap (N = 1). Eleven cases received post-operative RT because of close surgical margins for the primary site or extra-nodal extension of cervical lymph node metastasis. Post-operative treatment consisted of concurrent

chemoradiotherapy (N = 9) (66–70 Gy irradiation in all cases; cisplatin = 8 cases, nedaplatin and S-1 = 1 case), and RT alone (N = 2).

The AUCs for prediction of relapse using SUVmax, PTV, and TLP for the oral cavity, oropharynx, hypopharynx, oropharynx/hypopharynx/supraglottis, and all cases are shown in Table 2. The AUCs of PTV and TLP were higher than those of SUVmax except for the primary site of the oral cavity. In the oral cavity, the AUCs of SUVmax, PTV, and TLP were relatively low (0.76, 0.72, and 0.77, respectively). The AUCs of PTV and TLP in the oropharynx cases were high, indicating high accuracy. In the oropharynx/hypopharynx/supraglottis cases, the AUCs calculated using PTV and TLP were 0.91 and 0.87, respectively, whereas that using SUVmax was 0.66. There was a significant difference between PTV and TLP for predicting relapse ($P < 0.01$). Comparing the oropharynx/hypopharynx/supraglottis cases with all cases, the AUCs calculated using PTV and TLP for oropharynx/hypopharynx/supraglottis cases had better results than those for all patients (0.84 and 0.83, respectively).

Univariate analysis for predicting relapse was performed for the oropharynx/hypopharynx/supraglottis cases. The

cutoff values of SUVmax, PTV, and TLP were applied and calculated from ROC curves in Table 2. There were 13 cases of recurrence, suggesting that T stage, SUVmax, PTV, and TLP may be significant prognostic factors (Table 3).

Figure 2 shows the RFS curves of SUVmax, PTV, and TLP for oropharynx/hypopharynx/supraglottis cases. There were significant differences in RFS for all three parameters. After adjustment for age, T stage, and treatment modality, only PTV and TLP were significant prognostic factors (Table 4).

Discussion

In this study, we evaluated correlations among [11C]4DST PET parameters and RFS for each primary site in HNSCC. As we hypothesized, the AUC of PTV was extremely high even when the three sites (oropharynx/hypopharynx/supraglottis) were evaluated together. Cox regression hazard analysis showed PTV and TLP were independent predictive markers compared to other clinical factors.

Hanamoto *et al.* [11] reported a correlation between [18F] FDG PET parameters and treatment response for each

Table 2 [11C]4DST PET/CT parameters of relapse and relapse-free patients

Site	Parameter	Relapse-free median (range)	Relapse median (range)	AUC	95% CI	P-value	Cutoff value
Oropharynx + supraglottis + hypopharynx	SUVmax	7.41 (2.67–15.38)	8.27 (5.63–13.49)	0.66	0.51–0.81	0.085	7.15
	PTV	11.44 (1.32–37.11)	39.54 (11.14–96.08)	0.91	0.82–1	<0.001	21.11
	TLP	44.23 (4.19–192.24)	149.22 (46.81–468.66)	0.87	0.78–0.97	<0.001	71.40
Oropharynx	SUVmax	7.58 (4.53–12.57)	8.79 (7.15–10.10)	0.66	0.42–0.90	0.198	8.36
	PTV	17.04 (3.97–37.11)	55.20 (22.63–96.08)	0.92	0.82–1	0.001	22.60
	TLP	71.16 (20.82–192.24)	173.88 (92.28–468.66)	0.87	0.74–1	0.003	92.28
Hypopharynx	SUVmax	6.79 (4.37–13.20)	7.72 (5.63–13.49)	0.64	0.35–0.93	0.50	7.29
	PTV	6.76 (2.68–25.11)	35.84 (11.14–41.71)	0.91	0.76–1	0.005	21.11
	TLP	28.72 (10.02–114.48)	149.22 (46.81–192.05)	0.85	0.76–1	0.007	46.81
Oral cavity	SUVmax	6.84 (4.0–8.76)	8.02 (6.62–14.36)	0.76	0.56–0.95	0.027	6.76
	PTV	16.12 (1.57–50.17)	30.98 (7.74–107.28)	0.72	0.52–0.92	0.057	13.67
	TLP	64.06 (4.46–178.8)	135.62 (29.4–429.74)	0.77	0.58–0.95	0.021	62.37
All	SUVmax	7.35 (2.67–15.38)	8.15 (5.63–13.36)	0.68	0.57–0.79	0.007	6.62
	PTV	11.44 (1.32–50.17)	38.37 (7.74–107.28)	0.84	0.76–0.93	<0.001	21.00
	TLP	44.23 (4.19–192.24)	149.22 (29.40–468.66)	0.83	0.75–0.92	<0.001	62.37

SUVmax, maximum standardized uptake value; PTV, proliferative tumor volume; TLP, total lesion proliferation; AUC, area under the curve; CI, confidence interval.

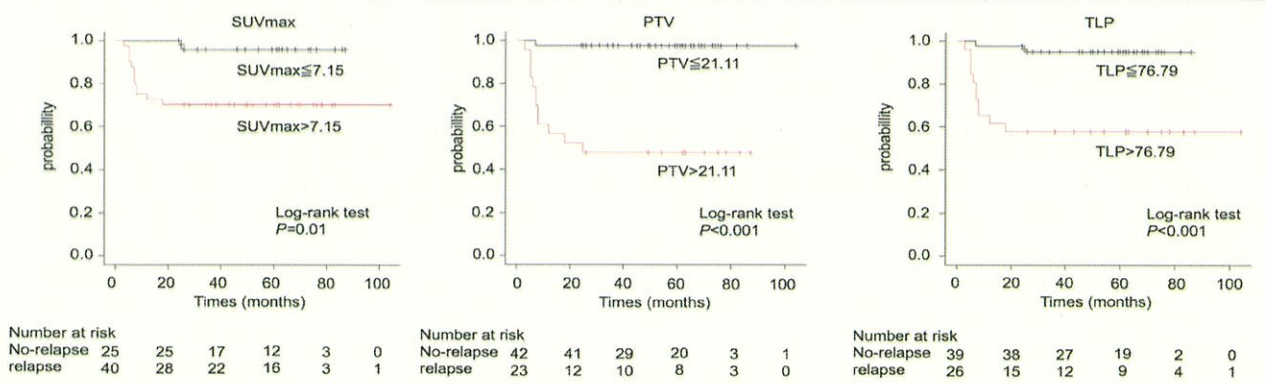
Table 3 Univariate analysis for relapse in oropharyngeal, supraglottic, and hypopharyngeal cancer

Factor	Level	Relapse rate		Odds ratio	95% CI	P-value
		N	%			
Age	<65 (N = 30)	5	17	1.47	0.37–6.53	0.76
	≥65 (N = 35)	8	23			
T stage	T2,3 (N = 48)	6	13	4.75	1.10–21.59	0.03
	T4 (N = 17)	7	41			
	Treatment	Surgery (N = 25)	7			
SUVmax	CRT (N = 40)	6	15	0.46	0.11–1.87	0.22
	<7.15 (N = 25)	1	4			
PTV	≥7.15 (N = 40)	12	30	10.01	1.30–45.66	0.01
	<21.11 (N = 42)	1	2			
TLP	≥21.11 (N = 23)	12	52	41.62	5.2–19.33	<0.01
	<76.79 (N = 39)	2	5			
	≥76.79 (N = 26)	11	42	12.97	2.42–134.4	<0.01

CRT, chemoradiotherapy.

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Fig. 2



Relapse-free survival curves of SUVmax, PTV, and TLP for oropharynx/hypopharynx/supraglottis cases. There was a difference between the two groups when divided by the cutoff value.

Table 4 Multivariate Cox regression analysis of RFS

Variables	SUV model		PTV model		TLP model	
	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value	Odds ratio (95% CI)	P-value
Age >65 years	1.45 (0.46–4.57)	0.53	3.57 (1.07–11.90)	0.38	1.92 (0.61–6.07)	0.29
RT or ST	0.85 (0.27–2.68)	0.77	1.21 (0.35–4.22)	0.76	1.01 (0.33–3.11)	0.99
T4	4.03 (1.27–12.79)	0.02	3.64 (0.99–13.32)	0.05	3.06 (0.97–9.62)	0.06
SUV 7.15	7.90 (1.02–61.31)	0.05				
PTV21.11			34.69(4.31–279.30)	<0.001		
TLP76.79					8.61 (1.85–40.08)	0.006

CI, confidence interval; RT, radiotherapy; ST, surgical treatment.

primary site in HNSCC. In laryngo-hypopharyngeal cancer (LHC), AUC ranged from 0.71 to 0.90, indicating high accuracy of PET/CT parameters to discriminate between complete and partial responders. Moreover, AUCs of metabolic tumor volume (MTV) and total lesion glycosys (TLG) were significantly higher than those of SUVs. Multivariate analysis of MTV and TLG in LHC revealed that they were independent, significant predictors. Based on these results, we hypothesized that more accurate prognosis could be predicted in [¹¹C]4DST PET by focusing on sites with similar biological characteristics, such as oropharyngeal, hypopharyngeal and supraglottic cancers, and examining them together to predict prognosis with a high degree of accuracy. The present findings support this hypothesis.

In the present study, the AUCs of PTV and TLP in supraglottic/hypopharyngeal cancer were 0.85 and 0.89, respectively. HPV-negative oropharyngeal AUCs of PTV and TLP were also high, indicating high accuracy. On the other hand, AUCs of PTV and TLP were higher than those of SUVmax except for the primary site of the oral cavity. Future large-scale prospective studies are needed to evaluate each cancer and confirm whether accuracy could be maintained by further narrowing down the sites.

In this study, the accumulation of primary and metastatic lymph nodes were combined to calculate each PET parameter using the methodology of Ito *et al.* [12] Many previous reports using [¹¹C]4DST/[¹⁸F]FDG and [¹⁸F]FDG have calculated PET parameters based on the primary tumor alone [6,11,13–15]. However, as the presence or absence of regional lymph node metastasis is considered to have an impact on RFS, the findings of the present study, in which the aggregation of all lesions with similar biological characteristics was analyzed, are useful.

PET tracers may be useful as cell proliferation imaging agent based on their ability to be incorporated into DNA. [¹⁸F]FLT TLP was an independent factor for loco-regional control, as were [¹⁸F]FLT SUVmax and PTV for overall survival (OS) [16]. However, [¹⁸F]FLT is not incorporated into DNA [17], because it lacks a 30-hydroxyl, unlike thymidine. [¹¹C]4DST was developed as a cell proliferation imaging agent based on its ability to be incorporated into DNA [18,19]. Therefore, in the present study we used [¹¹C]4DST as a tracer of cell proliferation instead of FLT.

Minamimoto *et al.* [8] reported that [¹¹C]4DST PET showed a higher correlation with cell proliferation as

evaluated by Ki-67 index in NSCLC than [¹⁸F]FDG PET, confirming [¹¹C]4DST PET/CT as a valuable noninvasive modality for imaging DNA synthesis by NSCLC. They also examined [¹⁸F]FDG and [¹¹C]4DST PET/CT for evaluating response to platinum-based doublet chemotherapy in advanced NSCLC. Consequently, PTV of baseline [¹¹C]4DST PET/CT along with MTV of baseline [¹⁸F]FDG PET/CT represent promising predictors of progression-free survival (PFS), and PTV of baseline [¹¹C]4DST PET/CT along with MTV and TLG of baseline [¹⁸F]FDG PET/CT are possible predictors of OS in patients with advanced NSCLC [20, 21]. Hotta *et al.* [6] reported the utility of [¹¹C]4DST and [¹⁸F]FDG PET parameters in patients with esophageal cancer. They reported that higher [¹⁸F]FDG TLG, [¹¹C]4DST SUVmax and TLP were associated with poor PFS in Kaplan–Meier analyses. Moreover, only the [¹¹C]4DST PET parameters showed significant HR in Cox regression hazard model. In head and neck cancer, Ito *et al.* [12] reported that [¹¹C]4DST TLP strongly correlated with [¹⁸F]FDG TLG. ROC analysis showed that TLG3.0 and TLP2.5 had the highest prognostic ability for local recurrence and metastasis. They concluded that [¹⁸F]FDG TLG3.0 had a slightly better prognostic capacity than [¹¹C]4DST TLP2.5. Our previous study showed that [¹⁸F]FDG MTV, [¹¹C]4DST PTV, and TLP were significant independent factors for RFS. Therefore, our findings suggest that [¹¹C]4DST volumetric parameters are superior to those of [¹⁸F]FDG for providing important prognostic information [9]. However, these studies have several limitations. Notably, the study population had heterogeneous tumor etiologies and was too small to enable data analysis according to primary tumor site.

Ihara-Nishishita *et al.* [14] reported the relationship between texture indices of [¹¹C]4DST PET and [¹⁸F]FDG PET in oropharyngeal squamous cell carcinoma. They concluded that texture indices of the primary lesion on [¹¹C]4DST PET may have predictive value for p16 status for targeted patients. This analysis method can measure the uniformity of intra-tumor metabolism and it can be combined with proliferative parameters to improve prognostic accuracy further.

Generally, the oropharynx is a hypermetabolic region where FDG accumulates physiologically, creating a high background that may artificially elevate SUV. Lim *et al.* [22] examined 176 patients with oropharyngeal cancer and reported that elevated MTV and TLG were independent predictors of death after adjustment for tumor stage. However, adjustments were not made for HPV status and treatment modality, and the patients were treated with a diversity of regimens of chemo- and/or bioradiotherapy using an unreported range of intensities.

Regarding HPV status, a previous study of 201 oropharyngeal cancer patients, including 109 HPV-positive

patients, reported that HPV-negative tumors had a significantly higher SUVmax, SUVpeak, MTV, and TLG. In univariable analysis, all PET variables were significantly associated with local control, OS, and disease-free survival (DFS). In multivariable analysis, TLG was significantly associated with DFS in patients with HPV-positive oropharyngeal cancer [23]. Another study also reported that TLG was significantly associated with loco-regional RFS, whereas stage was the most significant prognostic factor for distant metastasis-free survival and OS [24].

Although there are scattered reports of PET parameters for HPV-positive oropharyngeal cancer, there are few reports on treatment outcomes and PET parameters for HPV-negative oropharyngeal cancer. Moan *et al.* [25] reported that the prognostic role of [¹⁸F]FDG PET in head and neck cancer depends on HPV status. Of the 166 patients included, 48 had loco-regional and 23 had a metastatic recurrence. Only in the subgroup of HPV-unrelated head and neck carcinoma (HPV-negative oropharyngeal cancer and non-oropharyngeal cancer; n = 73) could the multivariate model be improved by including MTV. However, the correlation between prognosis and PET parameters in HPV-negative oropharyngeal cancer is unclear because only 22% of oropharyngeal cancer cases were included in the HPV-negative head and neck carcinoma group.

Pathogenesis factors, lymph node metastasis rates, and prognosis of HPV-negative oropharyngeal cancer are thought to be similar to those of laryngeal (especially supraglottic) and hypopharyngeal carcinoma. Our results support this hypothesis, and the information obtained from the volumetric parameters of [¹¹C]4DST PET suggest that at least these three heterogeneous populations could be analyzed as a homogeneous population.

This study had several limitations. This study had a small number of cases (65 cases of three primary sites), which made data analysis difficult. Especially, recurrence occurred in only 13 cases in oropharynx/hypopharynx/supraglottis; thus, the number of events was insufficient to accurately perform Cox proportional hazards regression. Further case accumulation and extension of the observation period are needed to strengthen the findings of this study.

Conclusion

Baseline [¹¹C]4DST PET volumetric parameters can provide important information on local control in p16-negative oropharyngeal/hypopharyngeal/supraglottic cancer patients.

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Availability of data and materials: The datasets generated and/or analyzed during the study are available from the corresponding author on reasonable request after approval by the Ethics Committee of our institution.

Ethics approval and consent to participate: The study protocol was approved by the Kagawa University Ethical Committee (approval number: 2021-056). This study was conducted in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki and its later amendments. Written informed consent was obtained from all individual participants included in the study. Data were entered into a password-protected database and analyzed by descriptive statistics. Data anonymity was guaranteed by the use of fully de-identified database records. Informed consent was waived due to its retrospective nature.

Conflicts of interest

There are no conflicts of interest.

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