Intratumoral Metabolic Heterogeneity for Prediction of Disease Progression After Concurrent Chemoradiotherapy in Patients with Inoperable Stage III Non-Small-Cell Lung Cancer

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Abstract

Purpose We evaluated the value of variable ^{18}F -FDG PET/CT parameters for the prediction of disease progression after concurrent chemoradiotherapy (CCRT) in patients with inoperable stage III non-small-cell lung cancer (NSCLC).

Methods One hundred sixteen pretreatment FDG PET/CT scans of inoperable stage III NSCLC were retrospectively reviewed (stage IIIA: 51; stage IIIB: 65). The volume of interest was automatically drawn for each primary lung tumor, and PET parameters were assessed as follows: maximum standardized uptake value (SUV_{max}), metabolic tumor volume (MTV) using the boundaries presenting SUV intensity exceeding 3.0, and the area under the curve of the cumulative

SUV-volume histograms (AUC-CSH), which is known to reflect the tumor heterogeneity. Progression-free survival (PFS), locoregional recurrence-free survival (LRFS), and distant metastasis-free survival (DMFS) were compared with each PET and clinical parameters by univariate and multivariate survival analysis.

Results In the ROC analysis, the optimal cutoff values of SUV_{max} , MTV (cm^3), and AUC-CSH for prediction of PFS were determined as 21.5, 27.7, and 4,800, respectively. In univariate analysis, PFS was statistically significantly reduced in those with $\text{AUC-CSH} < 4,800$ ($p = 0.004$). In multivariate analysis, AUC-CSH and SUV_{max} were statistically significant independent prognostic factors (HR 3.35, 95 % CI 1.79–6.28, $p < 0.001$; HR 0.25, 95 % CI 0.09–0.70, $p = 0.008$, respectively). Multivariate analysis showed that AUC-CSH was the most significant independent prognostic factor for LRFS and DMFS (HR 3.27, 95 % CI 1.54–6.94, $p = 0.002$; HR 2.79, 95 % CI 1.42–5.50, $p = 0.003$).

Conclusions Intratumoral metabolic heterogeneity of primary lung tumor in ^{18}F -FDG PET/CT can predict disease progression after CCRT in inoperable stage III NSCLC.

Keywords Non-small-cell lung cancer · Intratumoral heterogeneity · Prognosis · F-18 FDG PET

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Introduction

Lung cancer is a leading cause of cancer-related death worldwide [1], with non-small-cell lung cancer (NSCLC) accounting for 85 % of all lung cancer [2]. The majority of patients with locally advanced or advanced NSCLC are inoperable and incurable. Concurrent chemoradiotherapy (CCRT) is recommended

as the standard of care for patients with inoperable locally advanced NSCLC (stage III); however, its outcomes remain disappointing [3, 4]. Therefore, a more accurate prognostic factor is needed to select patients who are likely to benefit from treatment and to improve the outcome for NSCLC patients. Many prognostic factors, such as disease stage, performance status, and other clinical and therapeutic variables, have been suggested in stage III NSCLC [5]. Moreover, use of imaging parameters such as metabolic variables as predictors for stage III NSCLC patients have been suggested in pretreatment and posttreatment imaging scans. Although predicting response to therapy using the pretreatment imaging scan alone can allow the optimization of patient management, the amount of literature specifically dealing with pretreatment imaging parameters as prognostic factors in stage III NSCLC is limited.

^{18}F -fluorodeoxyglucose (FDG) positron emission tomography/computed tomography (PET/CT) has already been well established for the diagnosis and staging in variable cancers including NSCLC. Furthermore, FDG PET/CT has become an important tool used to predict prognosis. The intensity of the FDG uptake value such as the maximum standardized uptake value (SUV_{max}) and the metabolic tumor burden value such as the metabolic tumor volume (MTV) have demonstrated prognostic potential for the prediction of outcome [6–8]. However, according to a meta-analysis by Berghmans and colleagues, it remains unclear whether SUV is independent of other prognostic factors such as disease stage and performance status [6]. Moreover, recent studies have shown that the pretreatment FDG uptake value and metabolic tumor burden value of the primary tumor were not related to therapeutic response or outcome after radiotherapy (RT) or CCRT [9–11].

There is increasing interest in intratumoral metabolic heterogeneity on pretreatment FDG PET/CT to predict the therapeutic response in cancer [12]. However, the usefulness of intratumoral metabolic heterogeneity for prediction of prognosis in patients with NSCLC is unexplored. Thus, we evaluated the value of intratumoral metabolic heterogeneity analysis including other conventional PET parameters for the prediction of disease progression after CCRT in patients with inoperable stage III NSCLC.

Materials and Methods

Patients

We retrospectively reviewed primary tumors in pretreatment FDG PET/CT scans of pathologically confirmed inoperable stage III NSCLC between January 2005 and December 2010. The staging included pathologic examination from specimens obtained by transbronchial or transthoracic biopsies, torso FDG PET/CT, contrast-enhanced chest CT, and contrast-enhanced brain MRI. A disease stage was assigned according

to the American Joint Committee on Cancer (AJCC) staging system, 7th edition. The following are criteria for the inclusion of patients to this study: patients without a history of previous chemotherapy or thoracic radiotherapy due to another type of cancer; patients without a history of surgery for lung cancer treatment; patients with initial treatment with CCRT with or without consolidation chemotherapy. The exclusion criteria were as follows: primary tumors abutting FDG-avid lesions such as lymph nodes, inflammation, and atelectasis; primary tumors including empty lumens of the trachea or bronchus in the inner portion of the tumor; multifocal primary tumor lesions; a primary tumor less than 1.0 cm in diameter. In 49 patients, distinguishing the FDG accumulation of the primary tumor from that of lymph nodes, inflammatory lesions, or atelectasis was difficult because the primary tumor was abutting hypermetabolic lymph nodes or inflammatory lesions. In nine patients, distinguishing the MTV of the primary tumor was difficult because an empty lumen of the bronchus or trachea was located in the inner portion of the primary tumor. In six patients, choosing the primary tumor was difficult because there were multifocal primary tumors. In three patients, measurement of heterogeneity was inappropriate because of a partial volume effect in small-sized lesions. Finally, a total of 116 patients with inoperable locally advanced NSCLC were selected for this study.

Treatment

On day 1, thoracic radiotherapy was begun at a dose of 1.8–2.4 Gy/fraction, followed by daily administration of 25–36 fractions. The mean total dose of radiation administered was 68 Gy (dose range, 36–90 Gy). The chemotherapy consisted of cisplatin and paclitaxel concurrently administered on day 1, 8, 15, 22, 29, and 36. Some patients received consolidation chemotherapy after CCRT. After the treatment, a chest X-ray was performed monthly, a chest contrast-enhanced CT scan first at 1 month and after that every 3 months, and FDG PET/CT every 6 months.

Imaging Acquisitions

All patients underwent pretreatment FDG PET/CT scans. Scans were obtained using the Discovery ST PET/CT scanner (GE Medical Systems, Milwaukee, WI, USA). Patients were instructed to fast for at least 6 h before intravenous administration of ^{18}F -FDG. The serum glucose levels were found to be 8.3 mmol/l or less. The dose of injected ^{18}F -FDG was 7.4 MBq/kg body weight. Image acquisition for torso scanning was begun at about 1 h after the injection of ^{18}F -FDG. A low-dose CT scan was performed for attenuation correction from the skull base to the upper thigh. We used a standardized protocol for the CT using parameters with the following settings: 120 kV, from 10 to 130 mA, rotation time 0.7 s, field of view (FOV) 50 cm; scan length 40–50 s and slice thickness

Fig. 1 Measurement of intratumoral metabolic heterogeneity using VOI automatically produced by P-mode software (**a**) and CSH (**b**). Abbreviations: *VOI* volume of interest; *CSH* cumulative SUV-volume histogram; *SUV_{max}* maximum standardized uptake value

3.75 mm. Immediately after the CT, PET was performed with 15.7 cm axial FOV acquired in 2D mode with 150 s/bed position. The PET data sets were reconstructed iteratively with an ordered subset expectation maximization (OSEM) algorithm.

Image Analysis

Semiquantitative measurements of metabolic uptake in FDG-avid tumors in pretreatment scans were evaluated for their potential to predict the prognosis after CCRT. Image analysis

was performed using P-mode software (PMOD Technologies, Ltd., Zurich, Switzerland). The primary tumor lesion was used for evaluation of the *SUV_{max}*, MTV, and intratumoral metabolic heterogeneity.

SUV_{max} based on body weight, MTV, and intratumoral heterogeneity were measured with P-mode software. First, a volume of interest (VOI) encasing the primary tumor in the axial, coronal, and sagittal PET/CT images was drawn. The boundaries of voxels that have an SUV intensity greater than 3.0 were automatically produced (Fig. 1a). The *SUV_{max}* of the primary tumor was measured from the PET image. MTV,

Fig. 2 Tumor with a lower degree of heterogeneity (**a**) showed higher AUC-CSH (**b**) and tumor with a higher degree of heterogeneity (**c**) showed lower AUC-CSH (**d**). Abbreviations: *AUC-CSH* area under the curve of the cumulative SUV-volume histogram; *SUV_{max}* maximum standardized uptake value

Table 1 Patient characteristics of inoperable stage III non-small-cell lung cancer patients ($n=116$)

Characteristics	Number (%)
Gender	
Male	105 (91 %)
Female	11 (9 %)
Age (years)	
<65	37 (32 %)
≥ 65	79 (68 %)
Pathological type	
Squamous cell carcinoma	85 (73 %)
Adenocarcinoma	27 (23 %)
Large-cell carcinoma	1 (1 %)
Non-specified	3 (3 %)
ECOG	
0 or 1	105 (86 %)
2 or 3	11 (14 %)
CEA (ng/ml)	
≤ 4.7	65 (56 %)
> 4.7	33 (28 %)
Not checked	18 (16 %)
LDH (IU/l)	
≤ 472	98 (85 %)
> 472	16 (14 %)
Not checked	2 (%)
Smoking	
No	11 (10 %)
Yes	105 (91 %)
Radiation dose (Gy)	
$< 66^a$	34 (29 %)
$\geq 66^a$	82 (71 %)
Chemotherapy regimen	
Platinum-based	97 (84 %)
Non-platinum-based	19 (16 %)
Number of cycles of chemotherapy received	
3–5	29 (25 %)
6	87 (75 %)
Consolidation chemotherapy	
No	77 (66 %)
Yes	39 (34 %)
T stage ^b	
1 or 2	48 (41 %)
3 or 4	68 (59 %)
N stage ^b	
0, 1, or 2	74 (64 %)
3	42 (36 %)
TNM stage ^b	
IIIA	51 (44 %)
IIIB	65 (56 %)
Size (cm)	
$\leq 5.0^c$	104 (90 %)

Table 1 (continued)

Characteristics	Number (%)
$> 5.0^c$	12 (10 %)
SUV _{max}	
$\leq 21.5^c$	103 (89 %)
$> 21.5^c$	13 (11 %)
MTV	
$\leq 27.7^c$	38 (33 %)
$> 27.7^c$	78 (67 %)
AUC-CSH	
$< 4,800^c$	82 (71 %)
$\geq 4,800^c$	34 (29 %)

ECOG Eastern Cooperative Oncology Group, CEA carcinoembryonic antigen, LDH lactate dehydrogenase, TNM tumor-nodes-metastasis, SUV_{max} maximum standardized uptake value, MTV metabolic tumor volume, AUC-CSH area under the curve of cumulative SUV-volume histograms

^a The median value of the total dose of radiation

^b Staging by the American Joint Committee on Cancer staging system, 7th edition

^c The optimal cutoff values of size, SUV_{max}, MTV, and AUC-CSH in ROC analysis

defined as the total primary tumor volume, was automatically obtained using the boundaries of voxels presenting SUV intensity exceeding 3.0. In the case of presenting of any voxel showing lower SUV than 3.0 in the boundary of the primary tumor, MTV was obtained by the manual method based on the CT and PET images. Intratumoral metabolic heterogeneity was evaluated by the area under the curve of cumulative SUV-volume histograms (AUC-CSH), which was known to reflect the tumor heterogeneity [13]. A cumulative SUV-volume histogram (CSH) was obtained by plotting the percent volume of a tumor with an SUV above a certain threshold against that threshold, which was varied from 0 to 100 % of SUV_{max} (Fig. 1b). The area under the curve of these plots, AUC-CSH, is a quantitative index of heterogeneity in ¹⁸F-FDG uptake, with lower AUC corresponding to higher degrees of heterogeneity [13] (Fig. 2). For calculating the percent volume of the tumor, the threshold SUV intensity value exceeding 3.0 was used as a gross tumor volume.

Statistical Analysis

To evaluate the prognostic value of PET parameters in identifying the 2-year progression-free survival status, the optimal cutoff values of SUV_{max}, MTV, and AUC-CSH were determined in receiver-operating characteristic (ROC) curve analysis. Start dates for all survival estimates were the date of the pretreatment FDG PET/CT scan. Progression-free survival

(PFS) was defined as the interval from that date to the date of locoregional or systemic disease progression, death, or last follow-up; locoregional recurrence-free survival (LRFS) from that date to the date of locoregional recurrence (LR), death, or last follow-up; distant metastasis-free survival (DMFS) from that date to the date of distant metastasis (DM), death, or last follow-up. Disease progressions were defined as locoregional, with failure in or adjacent to the radiation planning target volume or in the ipsilateral hilus, mediastinum, or supraclavicular fossa; or distant, with failure at other sites. The disease progression was determined by using the Response Evaluation Criteria in Solid Tumors (RECIST). PFS, LRFS, and DMFS curves were produced using the Kaplan-Meier method. Log-rank and Cox regression analysis was used to develop the univariate and multivariate models describing the association of the independent variables with PFS, LRFS, and DMFS.

Results

Patient Characteristics

The patient characteristics in this study are listed in Table 1. The total of 116 patients (105 men and 11 women, mean age 66 years, range 44–83 years) consisted of 51 patients with stage IIIA and 65 patients with stage IIIB. On pathological examination, 85 were diagnosed with squamous cell carcinoma, 27 with adenocarcinoma, 1 with large-cell carcinoma, and 3 with non-specified NSCLC. The median total dose of radiation was 66 Gy (range, 36–90 Gy). In ROC analysis, the optimal cutoff values of size (cm), SUV_{max} , MTV (cm^3), and AUC-CSH were 5.0, 21.5, 27.7, and 4,800, respectively.

Survival Analysis

At the time of analysis, 76 patients had progressed or died, 34 had been in a progression-free state, and 6 were lost to follow-up. The median follow-up time for the surviving patients was 47.8 months, with a range of 16.0–87.4 months. The median PFS was 16.4 months, with a range of 2.7–75.4 months. The median PFS of patients with low AUC-CSH ($<4,800$) was significantly shorter than that of patients with high AUC-CSH ($\geq 4,800$) [11.8 vs. 43.7 months for progression ($p=0.004$)] (Fig. 3a). The median PFS of patients with low SUV_{max} (≤ 21.5) was also significantly shorter than that of patients with high SUV_{max} (>21.5) [15.6 vs. 60.0 months for progression ($p=0.026$)] (Fig. 3b). Median PFS between the patients with low MTV (≤ 27.7) and high MTV (>27.7) was not significantly different [28.1 vs. 12.2 months for progression ($p=0.154$)] (Fig. 3c).

Following univariate analysis, low AUC-CSH ($<4,800$), low SUV_{max} (≤ 21.5), and high LDH (>472) were significant predictors for poor PFS (Table 2). On multivariate analysis, low AUC-CSH ($<4,800$) [hazard ratio (HR) 3.35 for progression ($p<0.001$)] was the most statistically significant independent prognostic factor for poor PFS. High SUV_{max} (>21.5) [HR 0.25 for progression ($p=0.008$)] was associated with improved PFS. However, a high serum LDH level (>472) [HR 1.73 for progression ($p=0.097$)] and high MTV (>27.7) [HR 1.06 for progression ($p=0.879$)] were not independent prognostic factors for poor PFS (Table 2).

In LRFS analysis, univariate analysis showed that low AUC-CSH ($<4,800$) and high LDH (>472) were significant predictors for poor LRFS (Fig. 4, Table 2). On multivariate analysis, low AUC-CSH ($<4,800$) [hazard ratio (HR) 3.27 for locoregional progression ($p=0.002$)] was the most statistically significant independent prognostic factor for poor LRFS.

Fig. 3 Kaplan-Meier analyses of progression-free survival according to AUC-CSH (a), SUV_{max} (b), and MTV (c) for inoperable stage III non-small-cell lung cancer patients with concurrent chemoradiotherapy.

Abbreviations: AUC-CSH area under the curve of cumulative SUV-volume histograms; SUV_{max} maximum standardized uptake value; MTV metabolic tumor volume

Table 2 Univariate and multivariate survival analyses of factors associated with survival in a cohort of 116 inoperable stage III non-small-cell lung cancer patients

	Progression-free survival		Locoregional-recurrence-free survival		Distant metastasis-free survival	
	HR (95 % CI)	<i>P</i>	HR (95 % CI)	<i>P</i>	HR (95 % CI)	<i>P</i>
Univariate analysis						
Gender						
Male	0.93 (0.46–1.86)	0.832	1.91 (0.69–5.29)	0.214	0.96 (0.44–2.13)	0.924
Age (years)						
≥65	0.83 (0.51–1.34)	0.441	0.73 (0.43–1.25)	0.253	0.77 (0.46–1.30)	0.329
Diagnosis						
Adenocarcinoma	1.15 (0.69–1.92)	0.594	0.84 (0.45–1.56)	0.571	1.64 (0.94–2.85)	0.081
ECOG						
2 or 3	1.57 (0.78–3.17)	0.207	1.57 (0.71–3.48)	0.265	1.02 (0.44–2.39)	0.963
CEA (ng/ml)						
>4.7	1.42 (0.86–2.37)	0.175	1.16 (0.63–2.12)	0.634	1.39 (0.79–2.47)	0.255
LDH (IU/l)						
>472	1.95 (1.06–3.56)	0.031	2.09 (1.08–4.06)	0.029	1.46 (0.69–3.11)	0.322
Smoking						
Yes	0.99 (0.48–2.08)	0.998	1.01 (0.43–2.35)	0.983	1.11 (0.48–2.59)	0.812
Total radiation dose (Gy)						
<66 ^a	1.33 (0.82–2.17)	0.249	1.30 (0.74–2.28)	0.357	1.37 (0.80–2.35)	0.258
Chemotherapy regimen						
Non-Platinum-based	1.07 (0.56–2.02)	0.846	1.02 (0.48–2.16)	0.960	1.12 (0.55–2.28)	0.752
Number of cycles of chemotherapy received						
3–5	1.18 (0.71–1.95)	0.525	1.26 (0.70–2.22)	0.459	1.03 (0.58–1.83)	0.932
Consolidation chemotherapy						
No	1.33 (0.81–2.18)	0.255	1.52 (0.86–2.71)	0.153	1.06 (0.63–1.81)	0.820
T stage ^b						
3 or 4	0.96 (0.61–1.51)	0.845	0.95 (0.56–1.60)	0.837	0.79 (0.48–1.32)	0.371
N stage ^b						
3	0.87 (0.54–1.40)	0.577	0.60 (0.34–1.06)	0.077	1.07 (0.63–1.81)	0.802
TNM stage ^b						
IIIB	0.87 (0.55–1.38)	0.561	0.65 (0.38–1.10)	0.107	0.82 (0.49–1.40)	0.470
Size (cm)						
>5.0 ^c	1.45 (0.92–2.29)	0.108	1.54 (0.91–2.59)	0.107	1.05 (0.63–1.75)	0.858
SUV _{max}						
>21.5 ^c	0.32 (0.12–0.88)	0.026	0.50 (0.18–1.38)	0.179	0.20 (0.05–0.81)	0.024
MTV						
>27.7 ^c	1.43 (0.87–2.34)	0.154	1.37 (0.78–2.41)	0.269	1.14 (0.67–1.94)	0.635
AUC-CSH						
<4,800 ^c	2.27 (1.30–3.97)	0.004	2.84 (1.42–5.68)	0.003	1.66 (0.92–2.99)	0.094
Multivariate analysis						
Diagnosis						
Adenocarcinoma	1.47 (0.84–2.59)	0.182	0.99 (0.51–1.93)	0.985	1.92 (1.04–3.54)	0.036
LDH (IU/l)						
>472	1.73 (0.91–3.30)	0.097	2.07 (1.05–4.08)	0.035	1.13 (0.51–2.49)	0.760
Total radiation dose (Gy)						
<66 ^a	1.59 (0.93–2.70)	0.088	1.46 (0.82–2.61)	0.200	1.27 (0.71–2.26)	0.427

Table 2 (continued)

	Progression-free survival		Locoregional-recurrence-free survival		Distant metastasis-free survival	
	HR (95 % CI)	<i>P</i>	HR (95 % CI)	<i>P</i>	HR (95 % CI)	<i>P</i>
N stage ^b						
3	0.92 (0.56–1.52)	0.746	0.61 (0.34–1.10)	0.098	1.25 (0.71–2.18)	0.438
Size (cm)						
>5.0 ^c	0.96 (0.58–1.58)	0.876	2.06 (0.92–4.64)	0.080	0.73 (0.41–1.30)	0.284
SUV _{max}						
>21.5 ^c	0.25 (0.09–0.70)	0.008	0.38 (0.13–1.10)	0.074	0.15 (0.04–0.65)	0.011
MTV						
>27.7 ^c	1.06 (0.53–2.11)	0.879	0.77 (0.41–1.46)	0.420	1.24 (0.58–2.66)	0.582
AUC-CSH						
<4,800 ^c	3.35 (1.79–6.28)	<0.001	3.27 (1.54–6.94)	0.002	2.79 (1.42–5.50)	0.003

HR hazard ratio, CI confidence interval, ECOG Eastern Cooperative Oncology Group, CEA carcinoembryonic antigen, LDH lactate dehydrogenase, TNM tumor-nodes-metastasis, SUV_{max} maximum standardized uptake value, MTV metabolic tumor volume, AUC-CSH area under the curve of the cumulative SUV-volume histograms

^a The median value of the total radiation dose

^b Staging by the American Joint Committee on Cancer staging system, 7th edition

^c The optimal cutoff values of size, SUV_{max}, MTV, and AUC-CSH in ROC analysis

High serum LDH levels (>472) [HR 2.07 for locoregional progression ($p=0.035$)] was also an independent prognostic factor for poor LRFS. However, a high SUV_{max} (>21.5) was not a significant prognostic factor for locoregional prognosis [HR 0.38 for locoregional progression ($p=0.074$)] (Table 2).

In DMFS analysis, univariate analysis showed that the high SUV_{max} was a significant predictor of poor DMFS (Fig. 5, Table 2). On multivariate analysis, a low AUC-CSH (<4,800) [hazard ratio (HR) 2.79 for distant area progression ($p=0.003$)] was the most statistically significant independent prognostic factor for poor DMFS. High SUV_{max} (>21.5) [HR 0.15 for progression ($p=0.011$)] was associated with improved DMFS. Adenocarcinoma [hazard ratio (HR) 1.92 for distant area progression ($p=0.036$)] was also an independent prognostic factor for poor DMFS (Table 2).

Correlation of the variable PET parameters was calculated with the Pearson coefficient. SUV_{max} and AUC-CSH showed a moderate negative correlation (Pearson correlation $r=-0.66$). MTV and size were not significantly correlated with AUC-CSH ($r=-0.49$ and -0.43).

Discussion

This study showed that intratumoral metabolic heterogeneity of the primary tumor in pretreatment FDG PET/CT can predict disease progression after CCRT in inoperable locally advanced NSCLC patients. In our study, AUC-CSH, which is a quantitative index of intratumoral heterogeneity, is a strong independent prognostic factor for disease progression.

Moreover, AUC-CSH is better than SUV_{max}, MTV, or clinical factors predicting PFS, LRFS, and DMFS.

Several investigators have evaluated the usefulness of pretreatment FDG PET/CT for predicting outcomes for variable stage patients with NSCLC, and several cutoff SUV levels have been proposed [14, 15]. However, other recent studies [9–11] including variable stage patient groups have found that pretreatment FDG uptake of the primary tumor was not related to the therapeutic response or outcome after RT or CCRT. These discrepancies were also seen in a few studies including only locally advanced NSCLC patients. Ohno et al. demonstrated that the median PFS and OS period of patients with a pretreatment SUV_{max} of primary tumors <10.0 was significantly longer than that of patients with a SUV_{max} ≥ 10.0 [15]. However, Lopez Guerra et al. found that the pretreatment SUV_{max} of the primary tumor was not associated with any survival outcome after definitive RT [10]. Although we observed that the median PFS and DMFS period of patients with a high SUV_{max} (>21.5) was significantly longer than that of patients with a low SUV_{max} (≤ 21.5), the optimal cutoff value was extremely high so the number of high SUV_{max} patients was only 13.

Furthermore, the MTV of the primary tumor at the pretreatment FDG PET scan in advanced NSCLC patients as well as the SUV_{max} has not shown a significant difference between the responders and nonresponders in recent studies [9]. Consistent with these findings, we did not observe a significant relationship between the pretreatment MTV of the primary tumor and PFS in locally advanced NSCLC.

The intratumoral heterogeneity of malignancy can provide a novel, independent predictor of outcome. This supposition is

Fig. 4 Kaplan-Meier analyses of locoregional recurrence-free survival according to AUC-CSH (a), SUV_{max} (b), and MTV (c) for inoperable stage III non-small-cell lung cancer patients with concurrent chemoradio-

therapy. Abbreviations: AUC-CSH area under the curve of the cumulative SUV-volume histograms; SUV_{max} maximum standardized uptake value; MTV metabolic tumor volume

supported by several recent reports. Ganeshan et al. found that assessment of tumor heterogeneity by a CT texture analysis of non-contrast-enhanced images was a significant independent predictor of survival for patients with NSCLC [16]. Tixier et al. demonstrated that textural features of tumor metabolic distribution allow for the best stratification of esophageal carcinoma patients in the context of therapy-response prediction [12].

The underlying mechanisms, which might explain why heterogeneous tumors would have a worse initial prognosis in some kinds of malignancies, are not well established. Resistance to radiotherapy or chemotherapy is known to be associated with intracellular hypoxia and cancer stem cells [17, 18]. Intratumoral heterogeneity of FDG distribution, meanwhile, can be affected by many factors at the cellular level. Especially in an intracellular hypoxic condition, FDG preferentially accumulates in macrophages and granulation tissues surrounding necrotic foci [19]. However, FDG accumulates less in necrotic infiltration than in tumor cells [20]. Further-

more, cancer stem cells showed a significantly lower glucose uptake than tumor cells [21]. FDG uptake is also related to the expression of GLUT and hexokinase, vascularization, and tumor cell proliferation [22–24]. Therefore, intratumoral metabolic heterogeneity could reflect the physiologic processes related to response to CCRT.

In the present study, the cutoff value of SUV_{max} was relatively higher than in other studies. Lee et al. revealed that the initial SUV_{max} was greater in squamous cell carcinoma than in non-squamous cell carcinoma in stage IIIA non-small-cell lung cancer [25]. The relatively high proportion of squamous cell carcinoma patients compared to other histopathological subtypes might explain the reason for the high cutoff SUV_{max} value in the present study.

The prognostic value of LDH levels has been shown in several studies [25, 26]. Consistent with these studies, we observed that high serum LDH levels at diagnosis may predict a short LRFS time after CCRT. LDH was considered a marker of tumor hypoxia or increased anaerobic metabolism [27]. The

Fig. 5 Kaplan-Meier analyses of distant metastasis-free survival according to AUC-CSH (a), SUV_{max} (b), and MTV (c) for inoperable stage III non-small-cell lung cancer patients with concurrent chemoradio-

therapy. Abbreviations: AUC-CSH area under the curve of the cumulative SUV-volume histograms; SUV_{max} maximum standardized uptake value; MTV metabolic tumor volume

high LDH level is frequently associated with the aggressiveness of tumors and poor outcomes.

Our study has several limitations. First, we sometimes use partially manually drawn VOIs in some cases when using automated software for measuring MTV. With the primary tumor we encountered adjacent FDG-avid lesions (inflammation, atelectasis, and lymph nodes) or structures (myocardium, liver). If we could easily identify the adjacent FDG-avid lesion or structure using anatomic information obtained from PET/CT, we manually subtracted them and let the case be included in the inclusion criteria. However, if we could not easily identify the adjacent FDG-avid lesions or structures, we excluded the case. We also encountered the primary tumor presenting any voxel showing a lower SUV than 3.0 in the boundary of the primary tumor. In that portion, the MTV boundary was obtained by a partially manual method based on the enhanced CT and PET images.

Second, because of the retrospective study design, different detailed treatment regimens could affect the treatment outcome. However, the AUC-CSH was the most significant prognostic factor regardless of the regimen or the number of cycles of chemotherapy and radiation dose.

Conclusion

We demonstrated that intratumoral metabolic heterogeneity of the primary lung tumor in pretreatment FDG PET/CT might have better potential than FDG uptake intensity, metabolic tumor burden, or clinical factors for prediction of disease progression after CCRT in patients with inoperable stage III NSCLC. The AUC-CSH, which is a quantitative index of intratumoral heterogeneity, is a strong independent prognostic factor for disease progression after CCRT. Lower AUC-CSH can predict early disease progression in stage III NSCLC. These results suggest that intratumoral metabolic heterogeneity might reflect the physiological processes related to response to CCRT.

Conflict of Interest The authors declare no conflict of interest.

References

1. World Health Organization. The global burden of disease: 2004 update (2008); Available at http://www.who.int/healthinfo/global_burden_disease/GBD_report_2004update_part2.pdf. Accessed 7 Feb 2011.
2. American Cancer Society (2010) Cancer Facts and Figures 2010. American Cancer Society. Available at <http://www.cancer.org/acs/groups/content/@nho/documents/document/acspc-024113.pdf>. Accessed 7 Feb 2011.
3. Aupérin A, Le Péchoux C, Rolland E, Curran WJ, Furuse K, Fournel P, et al. Meta-analysis of concomitant versus sequential radiochemotherapy in locally advanced non-small-cell lung cancer. *J Clin Oncol*. 2010;28:2181–90.
4. O'Rourke N, Roqué I, Figuls M, Farré Bernadó N, Macbeth F. Concurrent chemoradiotherapy in non-small cell lung cancer. *Cochrane Database Syst Rev*. 2010;16:CD002140.
5. Berghmans T, Paesmans M, Sculier JP. Prognostic factors in stage III non-small cell lung cancer: a review of conventional, metabolic and new biological variables. *Ther Adv Med Oncol*. 2011;3:127–38.
6. Berghmans T, Dusart M, Paesmans M, Hossein-Foucher C, Buvat I, Castaigne C, et al. Primary tumor standardized uptake value (SUV_{max}) measured on fluorodeoxyglucose positron emission tomography (FDG-PET) is of prognostic value for survival in non-small cell lung cancer (NSCLC). A systematic review and meta-analysis (MA) by the European Lung Cancer Working Party for the IASLC lung cancer staging project. *J Thorac Oncol*. 2008;3:6–12.
7. Liao S, Penney BC, Wroblewski K, Zhang H, Simon CA, Kampalath R, et al. Prognostic value of metabolic tumor burden on ¹⁸F-FDG PET in nonsurgical patients with non-small cell lung cancer. *Eur J Nucl Med Mol Imaging*. 2012;39:27–38.
8. Yoo SW, Kim J, Chong A, Kwon SY, Min JJ, Song HC, et al. Metabolic tumor volume measured by F-18 FDG PET/CT can further stratify the prognosis of patients with stage IV non-small cell lung cancer. *Nucl Med Mol Imaging*. 2012;46:286–93.
9. Huang W, Zhou T, Ma L, Sun H, Gong H, Wang J, et al. Standard uptake value and metabolic tumor volume of ¹⁸F-FDG PET/CT predict short-term outcome early in the course of chemoradiotherapy in advanced non-small cell lung cancer. *Eur J Nucl Med Mol Imaging*. 2011;38:1628–35.
10. Lopez Guerra JL, Gladish G, Komaki R, Gomez D, Zhuang Y, Liao Z. Large decreases in standardized uptake values after definitive radiation are associated with better survival of patients with locally advanced non-small cell lung cancer. *J Nucl Med*. 2012;53:225–33.
11. Hoang JK, Hoagland LF, Coleman RE, Coan AD, Herndon 2nd JE, Patz Jr EF. Prognostic value of fluorine-18 fluorodeoxyglucose positron emission tomography imaging in patients with advanced-stage non-small-cell lung carcinoma. *J Clin Oncol*. 2008;26:1459–64.
12. Tixier F, Le Rest CC, Albarghach N, Pradier O, Metges JP, Corcos L, et al. Intratumor heterogeneity characterized by textural features on baseline ¹⁸F-FDG PET images predicts response to concomitant radiochemotherapy in esophageal cancer. *J Nucl Med*. 2011;52:369–78.
13. Velden FH, Cheebsumon P, Yaqub M, Smit EF, Hoekstra OS, Lammertsma AA, et al. Evaluation of a cumulative SUV-volume histogram method for parameterizing heterogeneous intratumoural FDG uptake in non-small cell lung cancer PET studies. *Eur J Nucl Med Mol Imaging*. 2011;38:1636–47.
14. Lee HY, Lee KS, Park J, Han J, Kim BT, Kwon OJ, et al. Baseline SUV_{max} at PET-CT in stage IIIA non-small-cell lung cancer patients undergoing surgery after neoadjuvant therapy: prognostic implication focused on histopathologic subtypes. *Acad Radiol*. 2012;19:440–5.
15. Ohno Y, Koyama H, Yoshikawa T, Matsumoto K, Aoyama N, Onishi Y, et al. Diffusion-weighted MRI versus ¹⁸F-FDG PET/CT: performance as predictors of tumor treatment response and patient survival in patients with non-small cell lung cancer receiving chemoradiotherapy. *Am J Roentgenol*. 2012;198:75–82.
16. Ganeshan B, Panayiotou E, Burnand K, Dizdarevic S, Miles K. Tumour heterogeneity in non-small cell lung carcinoma assessed by CT texture analysis: a potential marker of survival. *Eur Radiol*. 2012;22:796–802.
17. Wouters A, Pauwels B, Lardon F, Vermorken JB. Review: implications of in vitro research on the effect of radiotherapy and chemotherapy under hypoxic conditions. *Oncologist*. 2007;12:690–712.
18. O'Flaherty JD, Barr M, Fennell D, Richard D, Reynolds J, O'Leary J, et al. The cancer stem-cell hypothesis: its emerging role in lung cancer biology and its relevance for future therapy. *J Thorac Oncol*. 2012;7:1880–90.
19. Kubota R, Kubota K, Yamada S, Tada M, Ido T, Tamahashi N, et al. Microautoradiographic study for the differentiation of intratumoral

- macrophages, granulation tissues and cancer cells by the dynamics of fluorine-18-fluorodeoxyglucose uptake. *J Nucl Med.* 1994;35:104–12.
20. Brown RS, Leung JY, Fisher SJ, Frey KA, Ethier SP, Wahl RL, et al. Intratumoral distribution of tritiated fluorodeoxyglucose in breast carcinoma. I. Are inflammatory cells important? *J Nucl Med.* 1995;36:1854–61.
 21. Vlashi E, Lagadec C, Vergnes L, Matsutani T, Masui K, Poulou M, et al. Metabolic state of glioma stem cells and nontumorigenic cells. *Proc Natl Acad Sci USA.* 2011;108:3983–8.
 22. Rajendran JG, Schwartz DL, O'Sullivan J, Peterson LM, Ng P, Scharnhorst J, et al. Tumour hypoxia imaging with ^{18}F fluoromisonidazole positron emission tomography in head and neck cancer. *Clin Cancer Res.* 2006;12:5435–41.
 23. Kunkel M, Reichert TE, Benz P, Lehr HA, Jeong JH, Wieand S, et al. Over expression of Glut-1 and increased glucose metabolism in tumours are associated with a poor prognosis in patients with oral squamous cell carcinoma. *Cancer.* 2003;97:1015–24.
 24. Swinson DE, Jones JL, Richardson D, Cox G, Edwards JG, O'Byrne KJ. Tumour necrosis is an independent prognostic marker in non-small cell lung cancer: correlation with biological variables. *Lung Cancer.* 2002;37:235–40.
 25. Albain KS, Crowley JJ, LeBlanc M, Livingston RB. Survival determinants in extensive-stage non-small-cell lung cancer: the Southwest Oncology Group experience. *J Clin Oncol.* 1991;9:1618–26.
 26. O'Connell JP, Kris MG, Gralla RJ, Groshen S, Trust A, Fiore JJ, et al. Frequency and prognostic importance of pretreatment clinical characteristics in patients with advanced non-small-cell lung cancer treated with combination chemotherapy. *J Clin Oncol.* 1986;4:1604–14.
 27. Firth JD, Ebert BL, Ratcliffe PJ. Hypoxic regulation of lactate dehydrogenase A. Interaction between hypoxia-inducible factor 1 and cAMP response elements. *J Biol Chem.* 1995;270:21021–7.