Available online at www.sciencedirect.com

ScienceDirect

journal homepage: http://www.elsevier.com/locate/rpor

Original research article

Effectiveness of PET/CT with ^{18}F-fluorothymidine in the staging of patients with squamous cell head and neck carcinomas before radiotherapy

Radovan Vojtíšek[a,*], *Jiří Ferda*[b], *Jindřich Fínek*[a]

[a] *Department of Oncology and Radiotherapy, University Hospital in Pilsen, alej Svobody 80, 304 60 Pilsen, Czech Republic*

[b] *Department of Imaging Methods, University Hospital in Pilsen, alej Svobody 80, 304 60 Pilsen, Czech Republic*

ARTICLE INFO

Article history:
Received 12 August 2014
Received in revised form
25 October 2014
Accepted 28 January 2015

Keywords:
^{18}F-fluorothymidine
PET/CT
Radiotherapy planning
Head and neck carcinomas

ABSTRACT

Aim: The aim of our study was to compare the staging of the disease declared before anticancer treatment was begun with the staging that was found after the planning PET/CT scanning with ^{18}F-FLT was performed.

Background: PET/CT in radiotherapy planning of head and neck cancers can facilitate the contouring of the primary tumour and the definition of metastatic lymph nodes.

Materials and methods: Between November 2010 and November 2013, 26 patients suffering from head and neck carcinomas underwent planning PET/CT examination with ^{18}F-FLT. We compared the staging of the disease and the treatment strategy declared before and after ^{18}F-FLT-PET/CT was performed.

Results: The findings from ^{18}FLT-PET/CT led in 22 patients to a change of staging: in 19 patients it led to upstaging of the disease and in 3 patients it led to downstaging of the disease. In one patient, a secondary malignancy was found.

Conclusions: We have confirmed in this study that the use of ^{18}F-FLT-PET/CT scanning in radiotherapy planning of squamous cell head and neck carcinomas has a great potential in the precise evaluation of disease staging and consequently in the precise determination of target volumes.

© 2015 Published by Elsevier Sp. z o.o. on behalf of Greater Poland Cancer Centre.

1. Background

Radiotherapy has an important role to play in the treatment of locally and locoregionally advanced head and neck carcinomas, with or without concomitant chemotherapy. Using intensity modulated radiotherapy (IMRT), which is used more and more in routine practice, it is possible to achieve much better conformity in dose distribution than using standard 3D conformal radiotherapy (3D CRT). In the treatment of head and neck carcinomas, this technique allows the delivery of high doses to target volumes with better sparing of

* *Corresponding author*. Tel.: +420 377105605.
E-mail address: vojtisekr@fnplzen.cz (R. Vojtíšek).

http://dx.doi.org/10.1016/j.rpor.2015.01.005
1507-1367/© 2015 Published by Elsevier Sp. z o.o. on behalf of Greater Poland Cancer Centre.

proximal organs at risk (spinal cord, parotid glands and swallowing structures).[1,2] Considering the high conformity of dose distribution and a steep dose gradient, both resulting from the use of the IMRT technique, it is very important to know the precise location and the exact boundaries of the primary tumour and metastatic lymph nodes. Positron emission tomography (PET) provides the biological information about the tumour that is complementary to the anatomical information obtained through computed tomography (CT) scanning. Thus, the combination of PET and CT scanning can significantly facilitate the contouring of the primary tumour as well as the definition of metastatic lymph nodes during radiotherapy planning of head and neck carcinomas.[3] Initial studies with the most commonly used radiopharmaceutical, ^{18}F-flurodeoxyglucose (^{18}F-FDG), showed that volumes irradiated at high doses can be reduced with consequent sparing of neighbouring organs at risk and that higher doses may thus be delivered to the target volume.[4–6]

The major limitations of PET/CT examination with ^{18}F-FDG are false-positive findings due to the accumulation of this radiopharmaceutical in inflammatory changed tissues or in reactively changed lymph nodes.[3] Unlike F-FDG, 3′-deoxy-3′-^{18}F-fluorothymidine (^{18}F-FLT), is a radiopharmaceutical that reflects DNA synthesis and is not as influenced by peritumourous inflammatory changes.[7] Inasmuch as squamous cell carcinomas of the head and neck show relatively intense accumulation of ^{18}F-FLT,[8] we decided to exploit PET/CT scanning with ^{18}F-FLT for radiotherapy planning of these tumours.

2. Aim

The aim of our representative study was to compare the staging of the disease declared before anticancer treatment was begun with the staging that was found after the planning PET/CT scanning with ^{18}F-FLT was performed. Furthermore, we investigated the impact of possible changes in the staging on the treatment strategy. To the best of our knowledge, no study has been published investigating radiotherapy planning of head and neck carcinomas using ^{18}F-FLT-PET/CT scanning.

3. Materials and methods

3.1. *Patients*

Between November 2010 and November 2013, 26 patients (24 men and 2 women, with a median age of 61, see details in Table 1) suffering from histologically proven squamous cell carcinoma of the head and neck referred for radical or adjuvant radiotherapy – either as a single method or in combination with concomitant chemotherapy – took part in our study. At the time of planning investigation, all patients were without proven distant metastatic spread (TNM category – M0). They were conventionally radiologically staged with the use of neck ultrasound and chest X-ray examination. All patients signed an informed consent.

Table 1 – Patients and disease characteristics.

All patients ($n = 26$)	
Sex	
Male	24 (92.3%)
Female	2 (7.7%)
Age at the time of diagnosis (median, range)	61 (46–82)
Histology	
Squamous cell carcinoma	26 (100%)
Grading	
G1	6 (23.1%)
G2	18 (69.3%)
G3	1 (3.8%)
G4	1 (3.8%)
Locality	
Oropharynx	11 (42.4%)
Hypopharynx	2 (7.7%)
Larynx	5 (19.2%)
Base of mouth	5 (19.2%)
Tongue	2 (7.7%)
Paranasal sinuses	1 (3.8%)
Radiotherapy purpose without the informations from ^{18}FLT-PET/CT	
Radical	20 (76.9%)
Adjuvant	6 (23.1%)
Radiotherapy purpose with the informations from ^{18}FLT-PET/CT	
Radical	18 (69.3%)
Adjuvant	1 (3.8%)
Palliative	6 (23.1%)
No radiotherapy indicated	1 (3.8%)

3.2. *Radiotherapy simulation and image acquisition*

All patients underwent initial preparation at the Department of Oncology and Radiotherapy, University Hospital in Pilsen according to our institutional standards, i.e. the choice of proper positioning (supine position with arms along the body) and immobilization using a customized thermoplastic mask by Efficast® (Orfit, Wijnegem, Belgium) for the head, neck and shoulders, determination of the reference plane on an X-ray simulator and drawing the projection points of the simulated isocentre on the mask. Planning PET/CT examination with ^{18}F-FLT was carried out at the Department of Imaging Methods at the same hospital and in the same manner as routine diagnostic examinations, the only difference being that patients were positioned in radiotherapy positions with dedicated immobilization devices ensuring the same position as during their initial examination on an X-ray simulator. The concavity of the examination table was compensated for with a radiolucent hand-made board. Patients were placed into the required position with the help of a laser positioning system, the projection points on the skin were marked with radiopaque marks.

PET scans were performed 70–80 min after intravenous administration of ^{18}F-FLT with a radioactivity level of about 80 MBq. CT scans were performed focusing on the head, neck and lungs on a Biograph HiRez/16 slice (Siemens, Forcheim, Germany) after intravenous administration of 80 ml of a non-ionic iodine contrast agent. CT scans were performed during inhalation; during PET acquisition the patient was instructed to breathe slowly and shallowly. CT and PET scans were then exported to our PlanW treatment planning system.

3.3. *Radiotherapy planning and treatment delivery*

Contouring of target volumes and organs at risk was carried out according to the recommendations of ICRU Reports 50[9] and 62[10] and based on the fusion of ^{18}F-FLT-PET and CT scans. No mathematical algorithm for automatic contouring of GTV was used. We only used detailed descriptions of pathological findings, consulted a radiologist in case of doubts and used all accessible clinical informations. Patients were treated either with a shrinking-field technique using 3D CRT (3D conformal radiotherapy) or with simultaneous integrated boost using IMRT (intensity modulated radiotherapy). Radiotherapy itself was carried out on an Elekta Synergy® (Elekta, Stockholm, Sweden) linear accelerator. Most patients underwent preventive introduction of PEG (percutaneous endoscopic gastrostomy) before the beginning of treatment.

3.4. *Compared parameters*

We compared the following parameters: the staging of the disease and the treatment strategy declared before and after ^{18}F-FLT-PET/CT.

4. Results

The findings obtained from planning ^{18}FLT-PET/CT examination led in 22 (84.6%) patients to a change of staging: in 19 (73.1%) patients it led to upstaging of the disease and in 3 (11.5%) patients it led to downstaging of the disease. In one (3.8%) patient a secondary malignancy was found in a different subregion of the head and neck area than the primary tumour. In 7 (26.9%) patients distant metastatic spread was found. When upstaging of the disease was determined, it was mainly due to category "N" (nodi), namely in 11 patients (42.3%), less due to categories "M" (metastases) and "R" (residual disease) – 7 (26.9%) and 1 (3.8%) patient, respectively. See details in Table 2.

The changes in staging of the disease led to a change in treatment strategy in 11 (42.3%) patients. In 6 (23.1%) patients with proven metastatic spread, radiotherapy was conducted for palliative purposes and radiotherapy was abandoned in 1 (3.8%) patient. In 4 (15.4%) patients, the original adjuvant treatment strategy was changed to a radical one. See details in Table 2.

5. Discussion

Currently, the most widely used radiopharmaceutical for radiotherapy planning PET/CT scanning, not only in head and neck tumours, is ^{18}F-FDG. The main problem connected with its use is possible false-positive findings due to the accumulation of this radiopharmaceutical either in inflammatory changed tissues or in reactively changed lymph nodes.[3] This is also one of the reasons why another radiopharmaceutical, ^{18}F-FLT, is increasingly being used both in diagnosis and radiotherapy planning. Squamous cell carcinomas of the head and neck show relatively intensive accumulation of the ^{18}F-FLT[8] used to image DNA synthesis and cell proliferation. After intravenous injection, the intracellular trapping of ^{18}F-FLT in the cytosole occurs after phosphorylation by thymidine kinase 1, which is a key enzyme in DNA synthesis with high enzymatic activity observed during the S phase of the cell cycle and low activity in the G0/G1 phase. During DNA synthesis, the activity of thymidine kinase 1 is increased almost tenfold.[11] ^{18}F-FLT uptake inside the cell has thus been found to be a useful marker for assessment of the proliferation of several types of cancer, including squamous cell carcinomas of the head and neck.[12,13]

To the best of our knowledge, no study has been published investigating radiotherapy planning of head and neck carcinomas using ^{18}F-FLT-PET/CT scanning. A modelling study comparing contouring of target volume GTV (gross tumour volume) in squamous cell carcinomas of the thoracic oesophagus according to PET/CT examination with ^{18}F-FDG and ^{18}F-FLT with consequent pathological postoperative findings was published in 2010 by Han et al.[14] In 22 patients referred to radical surgical resection with lymphadenectomy, the GTV was delineated using seven different methods in ^{18}F-FLT-PET/CT (visual interpretation, SUV 1.3, 1.4, 1.5, SUV 20%, 25% and 30%) and three different methods in ^{18}F-FDG-PET/CT (visual interpretation, SUV 2.5 and SUV 40%). The lengths of the GTVs were compared with the lengths found through histopathological examination after surgery. An SUV cut-off of 1.4 on ^{18}F-FLT-PET/CT and one of 2.5 on ^{18}F-FDG-PET/CT provided the closest estimation of pathologic GTV length. The difference between the lengths of FLT 1.4 and FDG 2.5 was significantly different ($p = 0.442$). A total of 14 nodes were falsely positive using ^{18}F-FDG but only 3 using ^{18}F-FLT, thus, 11 falsely positive nodes using ^{18}F-FDG-PET/CT were corrected by ^{18}F-FLT-PET/CT. A total of 8 falsely negative nodes were found using ^{18}F-FDG-PET/CT, 12 using ^{18}F-FLT-PET/CT and 19 using CT alone. The authors also compared two radiotherapy plans determined using the optimal threshold during the use of ^{18}F-FDG and ^{18}F-FLT. The differences in dosimetric values for the lungs V20 (bilateral lung volume receiving $\geq$20 Gy), for the heart V40 and Vmax (maximal dose) for the spinal cord were not statistically significant. But values of MLD (mean lung dose), V5, V10, V30, V40 and V50 for the lungs and MHD (mean heart dose) and V30 for the heart were significantly lower using ^{18}F-FLT than ^{18}F-FDG.

Some authors[15,16] have described the high percentage of falsely positive findings in lymph nodes in head and neck carcinomas as being due to increased ^{18}F-FLT uptake in the germinative centres of reactively changed lymph nodes. However, the above-mentioned study found markedly lower false positivity using ^{18}F-FLT as opposed to ^{18}F-FDG. Even though those tumours were oesophageal, they were likewise squamous cell carcinomas, whose biological behaviour in terms of radiopharmaceutical uptake should be similar. We were unable to evaluate false positivity/negativity in our group of patients because planning examinations were not followed up by surgical procedures with histopathological examination. In situations where increased uptake of the radiopharmaceutical occurred predominantly at the periphery along the circumference of the nodes and not in the centre, we considered it to be the activation of germinative zones. By contrast, in situations where increased uptake occurred at the core of lymph nodes, we considered it to be metastatically involved.

Table 2 – Detailed characteristics related to the changes of the staging and the treatment strategy of each patient.

Patient (no.)	Sex (M/F)	Age	Tumour site	Grade	Stage before ^{18}FLT-PET/CT	Stage after ^{18}FLT-PET/CT	Original purpose	Purpose after ^{18}FLT-PET/CT	Notes
1	M	57	Larynx	4	pT4apN2bM0	pT4aN2cM0	Adjuvant	Radical	
2	F	61	Base of mouth	2	pT3pN2bM0,R1	pT3N2bM0,R2	Adjuvant	Radical	
3	M	64	Oropharynx	2	T3N1M0	T3N2cM0	Radical	Radical	
4	M	60	Larynx	2	pT4apN2cM0,R2	pT4apN2cM1,R2	Radical	Palliative	Rib metastases
5	M	66	Base of mouth	1	pT4apN2bM0	pT4aN2cM0	Adjuvant	Radical	
6	F	48	Paranasal sinuses	3	T3N0M0	T3N2bM0	Radical	Radical	
7	M	62	Tongue	2	T3N2bM0	T3N2bM0, second tumour	Radical	Radical	Left paralaryngeal carcinoma in situ
8	M	65	Oropharynx	2	T4aN2bM0	T4aN2bM1	Radical	Palliative	
9	M	63	Oropharynx	2	T4bN2cM0	T4N2bM0	Radical	Radical	
10	M	52	Larynx	2	pT1pN2bM0	pT1N2cM1	Adjuvant	Palliative	Mediastinal metastases
11	M	82	Oropharynx	2	T2N0M0	T2N2bM1	Radical	Palliative	Lung hilar metastases
12	M	60	Larynx	1	pT2pN0M0	pT2N2cM0	Adjuvant	Adjuvant	
13	M	46	Oropharynx	1	rT3N0M0	rT3N0M0	Radical	Radical	Reirradiation
14	M	60	Hypopharynx	1	T3N2bM0	T3N0M0	Radical	Radical	
15	M	62	Hypopharynx	2	pT2pN0M0	pT2N1M0	Adjuvant	Radical	
16	M	78	Oropharynx	2	T3N1M0	T3N2cM0	Radical	Radical	
17	M	65	Base of mouth	1	T4aN2cM0	T4aN2bM1	Radical	Palliative	Mediastinal metastases
18	M	67	Oropharynx	2	T3N2cM0	T3N2cM0	Radical	Radical	
19	M	47	Oropharynx	2	T4aN2cM0	T4aN2cM1	Radical	Palliative	Lung metastases
20	M	61	Base of mouth	2	T3N2bM0	T3N2cM0	Radical	Radical	
21	M	52	Oropharynx	2	pT2pN2apM1 (soft neck tissue), R2	pT2pN2aM1	Radical	No RT	Lung and mediastinal metastases
22	M	73	Tongue	2	T3N2cM0	T3N2bM0	Radical	Radical	
23	M	49	Base of mouth	2	T4aN2bM0	T4aN2cM0	Radical	Radical	
24	M	79	Oropharynx	2	T3N2bM0	T3N2cM0	Radical	Radical	
25	M	53	Larynx	2	T2N3M0	T2N3M0	Radical	Radical	
26	M	59	Oropharynx	1	T3N0M0	T3N2cM0	Radical	Radical	

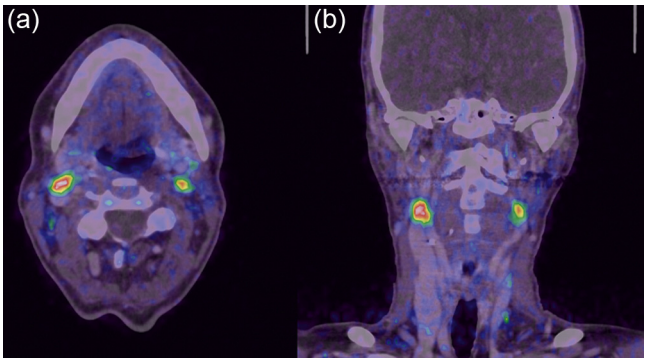

Fig. 1 – Patient (no. 6) with paranasal sinus carcinoma – axial (a) and coronal (b) view. According to the staging ultrasound investigation only benign cervical lymph nodes bilaterally. In the right upper jugular region increased FLT accumulation due to tumour infiltration, on the left side reactively changed node.

In our group of patients using planning ^{18}F-FLT-PET/CT, we saw upstaging of the disease in 19 (73.1%) patients, with 11 (42.3%) patients upstaged due to more advanced nodal involvement than originally appeared in CT and ultrasound examinations (Fig. 1). However, these findings did not lead to a change of treatment strategy, which remained radical. In seven (26.9%) patients, distant metastatic spread was found in the planning examination (one case of skeletal metastases, one of pulmonary hilar metastases, three of mediastinal metastases, one of lung metastases and one of mediastinal and lung metastases), but only in one case (mediastinal and lung metastases) was radiotherapy abandoned; in all other cases radiotherapy was carried out for palliative purposes. This was due to inaccurate staging of the disease consisting of neck ultrasound and chest X-ray examination, which may miss mediastinal, pulmonary hilar and small lung metastases. One (3.8%) patient (carcinoma of the edge of the tongue; Fig. 2) was suspected in the planning examination of having a secondary malignancy in the left paralaryngeal space. It was histologically proven by biopsy to be a squamous cell carcinoma in situ. However, this focus was then put into the GTV. Only three (11.5%) patients saw downstaging of the disease due to ^{18}F-FLT-PET/CT examination, in all cases in terms of "N" category, but even this change did not lead to a change of treatment strategy. It was of interest that in four (15.4%) patients our original treatment strategy was changed to a radical one because of findings of metastatic nodal involvement (two contralaterally, one ipsilaterally and one bilaterally).

Inasmuch as the activity of thymidine kinase 1 is extremely sensitive to ionizing radiation, the changes in ^{18}F-FLT uptake are considered to be the marker of a direct biological effect of radiation.[17] These changes also precede the changes in ^{18}F-FDG uptake; in other words, a decrease of cell proliferation precedes a change in cell metabolism.[18] This assumption has led to several experiments on the use of ^{18}F-FLT-PET/CT performed during radiotherapy in an effort to predict treatment outcomes, to identify subvolumes with higher proliferative activity and to the eventual use of adaptive radiotherapy during radiotherapy treatment with a boosted dose applied to the high-risk subvolume, as the surviving cell populations are much more capable of repopulation in the period between two radiation doses and this process of repopulation is often the cause of treatment failure.[19,20] Kishino et al.[21] compared the utility of ^{18}F-FLT-PET/CT with that of ^{18}F-FDG-PET/CT to assess the early locoregional clinical outcomes of chemoradiotherapy for head and neck squamous cell carcinomas in 28 patients. During mid-treatment imaging, ^{18}F-FDG-PET/CT had high sensitivity (100%), but low specificity (19%), low positive predictive value (17%) and low overall accuracy (30%). Mid-treatment ^{18}F-FLT-PET/CT showed significantly higher specificity (72%) and overall accuracy (74%) than ^{18}F-FDG-PET/CT ($p < 0.0001$ and 0.0001, respectively). According to the authors of the study, ^{18}F-FLT-PET/CT examination performed during radiotherapy and soon after has the potential to predict clinical outcomes and to identify patients who would require thorough follow up to detect persistent or recurrent disease. Another study, Hoeben et al.[22] monitored early clinical outcomes and also evaluated the association between PET parameters and clinical outcomes. 48 patients with squamous cell carcinomas of the head and neck underwent ^{18}F-FLT-PET/CT examination before and during radio(chemo)therapy (in the second and forth weeks) to assess whether early changes in ^{18}F-FLT uptake can predict long-term outcomes. A decrease of SUV max $\geq$45% during the first two weeks was associated with significantly better disease-free survival (DFS) (88% [95% CI, 75–100] vs. 63% [95% CI, 41–85], $p = 0.035$). In the fourth week of radiotherapy, SUV max in all patients decreased to such an extent

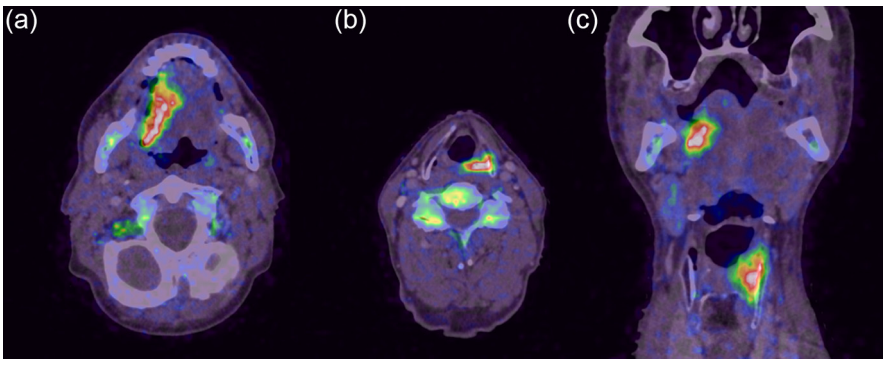

Fig. 2 – Patient (no. 7) with carcinoma of the right edge of the tongue (a, c) with secondary malignancy in the left paralaryngeal space (b, c). Axial (a, b) and coronal view (c).

that it was no longer useful for assessing outcome. Thus, a greater decrease in accumulation was a predictor of better clinical outcome (DFS), with a particularly strong early association in patients treated with radiochemotherapy. According to the results of this study, the evaluation is best done in the second week of treatment because later measurements did not add much information and the ^{18}F-FLT-PET signal became more difficult to quantify. Troost et al.[23] monitored early tumour response based on repetitive ^{18}F-FLT-PET/CT examination using volume and signal changes. They also assessed the heterogeneity of intratumourous distribution of the ^{18}F-FLT and identified subvolumes with high proliferative activity suitable for dose escalation and, thus, to adaptive radiotherapy. Ten patients with oropharyngeal tumours underwent three consecutive ^{18}F-FLT-PET/CT examinations (1 before and 2 during radiotherapy). In the primary tumours, the SUV max of the second ^{18}F-FLT-PET scan was significantly decreased relative to the first scan, and the SUV max decreased even further in the third (7.6 ± 2.6, 3.1 ± 1.7 and 1.7 ± 0.4). On average, the relative decrease in SUV max was 55% between the first and second scans and 34% between the second and third scans. In lymph node metastases, the relative decrease in SUV max was 44% between the first and second scans and 47% between the second and third scans. In all primary tumours a $GTV_{80\%}$ (subvolume with activity based on 80% isocontour) within GTV_{CT} (GTV delineated on CT) could be identified in the first and second ^{18}F-FLT-PET/CT scans. In the third scan, this identification was hampered by the relatively low ^{18}F-FLT uptake in the tumour relative to the background. In contrast to the early decrease of accumulation associated with treatment, significant changes in the size of GTV_{CT} were detected only in the fourth week (after 15–18 fractions). In several patients, the size and location of the subvolumes changed during the initial phase of treatment. Image acquisition late during treatment (e.g. in the fourth week) did not lead to useful results because signal intensities were low. Thus ^{18}F-FLT-PET/CT examination was useful neither in terms of response monitoring based on volumetric changes nor adaptation of the GTV delineation during treatment.

6. Conclusions

In conclusion, we have confirmed in this study that the use of ^{18}F-FLT-PET/CT scanning in radiotherapy planning of squamous cell head and neck carcinomas has a great potential in the precise evaluation of disease staging and, consequently, in the precise determination of target volumes. Furthermore, it has a considerable impact on possible changes in the therapeutic approach taken and, consequently, on the choice of an optimal treatment strategy.

Conflict of interest

None declared.

Financial disclosure

None declared.

REFERENCES

1. Feng FY, Kim HM, Lyden TH, et al. Intensity-modulated chemoradiotherapy aiming to reduce dysphagia in patients with oropharyngeal cancer: clinical and functional results. *J Clin Oncol* 2010;**28**(June (16)):2732–8.
2. Kam MK, Leung SF, Zee B, et al. Prospective randomized study of intensity-modulated radiotherapy on salivary gland function in early-stage nasopharyngeal carcinoma patients. *J Clin Oncol* 2007;**25**(November (31)):4873–9.
3. Troost EG, Schinagl DA, Bussink J, Oyen WJ, Kaanders JH. Clinical evidence on PET–CT for radiation therapy planning in head and neck tumours. *Radiother Oncol* 2010;**96**(September (3)):328–34.
4. Madani I, Duthoy W, Derie C, et al. Positron emission tomography-guided, focal-dose escalation using intensity-modulated radiotherapy for head and neck cancer. *Int J Radiat Oncol Biol Phys* 2007;**68**(May (1)):126–35.
5. Schwartz DL, Ford EC, Rajendran J, et al. FDG-PET/CT-guided intensity modulated head and neck radiotherapy: a pilot investigation. *Head Neck* 2005;**27**(June (6)):478–87.
6. Vanderstraeten B, Duthoy W, De Gersem W, De Neve W, Thierens H. [^{18}F]fluoro-deoxy-glucose positron emission tomography ([^{18}F]FDG-PET) voxel intensity-based intensity-modulated radiation therapy (IMRT) for head and neck cancer. *Radiother Oncol* 2006;**79**(June (3)):249–58.
7. Shields AF, Grierson JR, Dohmen BM, et al. Imaging proliferation in vivo with [F-18]FLT and positron emission tomography. *Nat Med* 1998;**4**(November (11)):1334–6.
8. Menda Y, Boles Ponto LL, Dornfeld KJ, et al. Kinetic analysis of 3′-deoxy-3′-(18)F-fluorothymidine ((18)F-FLT) in head and neck cancer patients before and early after initiation of chemoradiation therapy. *J Nucl Med* 2009;**50**(July (7)):1028–35.
9. ICRU Report 50. *Prescribing, recording and reporting photon beam therapy*. Bethesda: International Commission for Radiation Units and Measurements; 1993, 71 p.
10. ICRU Report 62. *Prescribing, recording and reporting photon beam therapy (Suppl. to ICRU Report 50)*. Bethesda: International Commission for Radiation Units and Measurements; 1999, 52 p.
11. Rasey JS, Grierson JR, Wiens LW, Kolb PD, Schwartz JL. Validation of FLT uptake as a measure of thymidine kinase-1 activity in A549 carcinoma cells. *J Nucl Med* 2002;**43**(September (9)):1210–7.
12. Yamamoto Y, Nishiyama Y, Ishikawa S, et al. Correlation of ^{18}F-FLT and ^{18}F-FDG uptake on PET with Ki-67 immunohistochemistry in non-small cell lung cancer. *Eur J Nucl Med Mol Imaging* 2007;**34**(October (10)):1610–6.
13. van Westreenen HL, Cobben DC, Jager PL, et al. Comparison of ^{18}F-FLT PET and ^{18}F-FDG PET in esophageal cancer. *J Nucl Med* 2005;**46**(March (3)):400–4.
14. Han D, Yu J, Yu Y, et al. Comparison of (18)F-fluorothymidine and (18)F-fluorodeoxyglucose PET/CT in delineating gross tumour volume by optimal threshold in patients with squamous cell carcinoma of thoracic esophagus. *Int J Radiat Oncol Biol Phys* 2010;**76**(March (4)):1235–41.
15. Troost EG, Vogel WV, Merkx MA, et al. ^{18}F-FLT PET does not discriminate between reactive and metastatic lymph nodes in primary head and neck cancer patients. *J Nucl Med* 2007;**48**(May (5)):726–35.
16. Cobben DC, van der Laan BF, Maas B, et al. ^{18}F-FLT PET for visualization of laryngeal cancer: comparison with ^{18}F-FDG PET. *J Nucl Med* 2004;**45**(February (2)):226–31.
17. Mankoff DA, Shields AF, Krohn KA. PET imaging of cellular proliferation. *Radiol Clin North Am* 2005;**43**(January (1)):153–67.
18. Yang YJ, Ryu JS, Kim SY, et al. Use of 3′-deoxy-3′-[^{18}F]fluorothymidine PET to monitor early

responses to radiation therapy in murine SCCVII tumours. *Eur J Nucl Med Mol Imaging* 2006;**33**(April (4)):412–9.
19. Maciejewski B, Withers HR, Taylor JM, Hliniak A. Dose fractionation and regeneration in radiotherapy for cancer of the oral cavity and oropharynx: tumour dose–response and repopulation. *Int J Radiat Oncol Biol Phys* 1989;**16**(March (3)):831–43.
20. Withers HR, Taylor JM, Maciejewski B. The hazard of accelerated tumour clonogen repopulation during radiotherapy. *Acta Oncol* 1988;**27**(2):131–46.
21. Kishino T, Hoshikawa H, Nishiyama Y, Yamamoto Y, Mori N. Usefulness of 3′-deoxy-3′-^{18}F-fluorothymidine PET for predicting early response to chemoradiotherapy in head and neck cancer. *J Nucl Med* 2012;**53**(October (10)):1521–7.
22. Hoeben BA, Troost EG, Span PN, et al. ^{18}F-FLT PET during radiotherapy or chemoradiotherapy in head and neck squamous cell carcinoma is an early predictor of outcome. *J Nucl Med* 2013;**54**(April (4)):532–40.
23. Troost EG, Bussink J, Hoffmann AL, Boerman OC, Oyen WJ, Kaanders JH. ^{18}F-FLT PET/CT for early response monitoring and dose escalation in oropharyngeal tumours. *J Nucl Med* 2010;**51**(June (6)):866–74.