Comparison of ^{18}F-FLT PET and ^{18}F-FDG PET in Esophageal Cancer

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^{18}F-FDG PET has gained acceptance for staging of esophageal cancer. However, FDG is not tumor specific and false-positive results may occur by accumulation of FDG in benign tissue. The tracer ^{18}F-fluoro-3′-deoxy-3′-L-fluorothymidine (^{18}F-FLT) might not have these drawbacks. The aim of this study was to investigate the feasibility of ^{18}F-FLT PET for the detection and staging of esophageal cancer and to compare ^{18}F-FLT PET with ^{18}F-FDG PET. Furthermore, the correlation between ^{18}F-FLT and ^{18}F-FDG uptake and proliferation of the tumor was investigated. **Methods:** Ten patients with biopsy-proven cancer of the esophagus or gastroesophageal junction were staged with CT, endoscopic ultrasonography, and ultrasound of the neck. In addition, all patients underwent a whole-body ^{18}F-FLT PET and ^{18}F-FDG PET. Standardized uptake values were compared with proliferation expressed by Ki-67 positivity. **Results:** ^{18}F-FDG PET was able to detect all esophageal cancers, whereas ^{18}F-FLT PET visualized the tumor in 8 of 10 patients. Both ^{18}F-FDG PET and ^{18}F-FLT PET detected lymph node metastases in 2 of 8 patients. ^{18}F-FDG PET detected 1 cervical lymph node that was missed on ^{18}F-FLT PET, whereas ^{18}F-FDG PET showed uptake in benign lesions in 2 patients. The uptake of ^{18}F-FDG (median standardized uptake value [SUV_{mean}], 6.0) was significantly higher than ^{18}F-FLT (median SUV_{mean}, 3.4). Neither ^{18}F-FDG maximum SUV (SUV_{max}) nor ^{18}F-FLT SUV_{max} correlated with Ki-67 expression in the linear regression analysis. **Conclusion:** In this study, uptake of ^{18}F-FDG in esophageal cancer is significantly higher compared with ^{18}F-FLT uptake. ^{18}F-FLT scans show more false-negative findings and fewer false-positive findings than do ^{18}F-FDG scans. Uptake of ^{18}F-FDG or ^{18}F-FLT did not correlate with proliferation.

Key Words: esophageal cancer; ^{18}F-fluoro-3′-deoxy-3′-L-fluorothymidine; ^{18}F-FDG; staging

J Nucl Med 2005; 46:400–404

Most patients with esophageal cancer are treated in specialized institutes and staged by endoscopic ultrasonography (EUS), CT of the chest and abdomen, and ultrasound examination (US) of the cervical region (*1*). However, these traditional methods for staging esophageal cancer have limited sensitivity and specificity. The presence of distant metastases before surgery, which is not detected by conventional imaging techniques, is relatively high, as indicated by detection of metastases during surgery in approximately 25% of the patients (*2*).

PET using ^{18}F-FDG is a noninvasive metabolic imaging technique and its usefulness has been established for several malignancies (*3*). ^{18}F-FDG is the most widely used tracer for staging tumors with PET (*3*). FDG is a glucose analog that enters the cells via the same membrane transporters as glucose. Glucose as well as ^{18}F-FDG are phosphorylated by the enzyme hexokinase. In contrast to glucose-6-phosphate, ^{18}F-FDG-6-phosphate is not a substrate for further metabolism in the glycolytic pathway. Therefore, ^{18}F-FDG-6-phosphate is trapped in the cells in proportion to their glycolytic activity (*3,4*).

There is evidence for improved preoperative staging of esophageal cancer with ^{18}F-FDG PET. Sensitivities of 67%–74% have been reported, especially with regard to the detection of nonregional lymphatic or hematogenic disease (*5,6*). Although these results may indicate an important role for ^{18}F-FDG PET, FDG is not a tumor-specific tracer and false-positive results may occur (*7,8*). For example, macrophages and neutrophils can demonstrate increased ^{18}F-FDG uptake, which can lead to false-positive results (*9,10*).

^{18}F-Fluoro-3′-deoxy-3′-L-fluorothymidine (^{18}F-FLT) was introduced as a PET proliferation tracer by Shields et al., which might not have these drawbacks (*11,12*). ^{18}F-FLT is monophosphorylated by thymidine kinase 1 (TK1), which leads to intracellular trapping. Since the TK1 concentration is especially increased during the S phase of the cell cycle, the uptake of ^{18}F-FLT is believed to depend on proliferation (*12*).

Received Aug. 13, 2004; revision accepted Sep. 30, 2004.

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The aim of this study was to investigate the feasibility of ^{18}F-FLT PET for the detection and staging of esophageal cancer compared with ^{18}F-FDG PET. Furthermore, the correlation between uptake of ^{18}F-FLT or ^{18}F-FDG and proliferation of the tumor was investigated.

MATERIALS AND METHODS

Patients

This prospective study consisted of 10 patients with biopsy-proven malignancy of the esophagus or gastroesophageal junction. All patients were staged with multidetector CT (Somatom Sensation; Siemens Medical Systems) of the chest and abdomen, EUS (GF-UM20, 7.5–12 MHz; Olympus), and US of the cervical region. Patients were included from November 2003 until February 2004.

All patients gave written informed consent. Only patients with liver and kidney functions and hematologic parameters (hemoglobin, hematocrit, erythrocytes, thrombocytes, leukocytes, and white cell count) within normal limits were included because of the toxicity of FLT in high concentrations. The Medical Ethics Committee of Groningen University Hospital approved the study protocol.

FDG and FLT Synthesis

FDG was produced according to the method described by Hamacher et al. using the coincidence ^{18}F-FDG synthesis module (*13*). Synthesis of ^{18}F-FLT was performed according to the method of Machulla et al. (*14*). ^{18}F-FLT was produced by ^{18}F-fluorination of the 4,4′-dimethoxytrityl–protected anhydrothymidine, followed by a deprotection step. After purification by reversed-phase high-performance liquid chromatography, the product was made isotonic and passed through a 0.22-μm filter. ^{18}F-FLT was produced with a radiochemical purity of >95% and a specific activity of >10 TBq/mmol. The radiochemical yield was 6.7% ± 3.7% (decay corrected).

PET

PET studies were performed using an ECAT EXACT HR+ scanner (Siemens/CTI, Inc.). Before PET, patients were instructed to fast for at least 6 h to keep both study protocols comparable. Patients were also instructed to drink 500 mL of water before imaging to stimulate ^{18}F-FDG and ^{18}F-FLT excretion from the renal calyces and to stimulate subsequent voiding.

Data acquisition started 90 and 60 min after injection of ^{18}F-FDG and ^{18}F-FLT, respectively. Scans were performed in whole-body mode, for 5 min per bed position from femur to the crown. Transmission imaging was obtained for 3 min per bed position for attenuation correction. Images were reconstructed using an iterative reconstruction technique and were read from computer monitors (*15*).

Pathologic Evaluation

Tissue was fixed in 4% buffered formalin, routinely processed, and embedded in paraffin. Subsequently, 4-μm sections were cut. For morphology, slides were routinely stained with hematoxylin and eosin. Proliferating cells were detected using the monoclonal antibody MIB-1, which recognizes an epitope of the Ki-67 nuclear antigen that is present during DNA synthesis (*16*). For this immunohistochemistry, slides were pretreated for 30 min in Tris buffer (pH 9.5) at 98°C. Staining was performed using the automated immunohistochemistry slide-staining system NexES (Ventana Medical Systems Inc.). As the first step, monoclonal antibody MIB-1 (DakoCytomation BV) detection of the cell proliferation marker Ki-67 was applied. As the second step, a basic 3,3′-diaminobenzidine detection system was used (Ventana Medical Systems Inc.). All reagents and equipment were used according to the instructions of the suppliers.

The MIB-1 score was estimated by counting the percentage of MIB-1–positive cell nuclei per 1,000 tumor cells in the region of the tumor with the greatest density of staining, which, in most instances, corresponds to areas with the highest mitotic activity. The pathologist was unaware of the results of the PET images.

Data Analysis

Patients were staged according to the tumor, node, metastasis (TNM) staging system of the International Union Against Cancer on the basis of CT, EUS, and US (*17*). The gold standard for the presence or absence of metastases was either histopathologic examination or follow-up. If this information was not available, other staging modalities were used as a reference. Both ^{18}F-FDG PET and ^{18}F-FLT PET scans were interpreted independently by experienced nuclear physicians who were unaware of clinical data and information from the other PET scan.

Three-dimensional regions of interest (ROIs) were placed semiautomatically using a dedicated software program over the primary tumor on multiple slices, with a threshold of 70% of the maximum pixel value within the tumor. The maximum standardized uptake value (SUV_{max}) and the mean SUV (SUV_{mean}) were calculated according to the equation:

$$SUV = \frac{C_i}{A/M},$$

where C_i is the activity concentration, A is the injected radioactivity, and M is the body mass. SUV_{max} denotes the maximum SUV value within the tumor ROI, and SUV_{mean} denotes the mean value averaged over all voxels.

Statistical Analysis

The results of the visually interpreted PET images were compared with the histologic data or dedicated radiographic imaging, which were used as the standard. ^{18}F-FDG and ^{18}F-FLT uptake was compared using the Wilcoxon signed rank test. The amount of Ki-67–positive cells and SUVs for ^{18}F-FDG and ^{18}F-FLT were compared using linear regression analysis. Two-tailed P values < 0.05 were considered significant.

RESULTS

Patients

Ten patients were included (median age, 61 y; range, 48–75 y). Patient characteristics are summarized in Table 1. Patients received ^{18}F-FDG with a median dose of 368 MBq (250–750 MBq) and received ^{18}F-FLT with a median dose of 410 MBq (340–450 MBq). Eight patients underwent esophagectomy and 2 patients received an expendable metal stent because of an irresectable T4 tumor on preoperative staging in patient 5 and an irresectable tumor encountered during surgical exploration in patient 7.

Detection of Esophageal Cancer

^{18}F-FDG PET visualized all primary tumors, whereas ^{18}F-FLT visualized 8 of 10 esophageal cancers (Table 1). In

TABLE 1
Patient Characteristics and Staging Results

Patient no.	Sex	Age (y)	Localization	Histology	Treatment	Staging				^{18}F-FDG PET		^{18}F-FLT PET		Proliferation: Ki-67 (% ± SE)
						EUS	CT	^{18}F-FDG PET	^{18}F-FLT PET	Surgery or PA	SUV_{max}	SUV_{mean}	SUV_{max}	SUV_{mean}	
1	M	73	Distal	AC	Esophagectomy	T3 N1 M0	T3 N1 M0	T+ N1 M0	T+ N1 M0	T3 N1 M0	7.82	6.22	3.85	3.15	78 ± 1.97
2	M	75	Mid	AC	Esophagectomy	T3 N1 M0	T3 N1 M0	T+ N0 M0	T+ N0 M0	T3 N1 M0	8.90	7.24	4.93	3.85	65 ± 2.19
3	M	68	GEJ	AC	Esophagectomy	T3 N1 M0	T3 N1 M0	T+ N0 M0	T+ N0 M0	T3 N1 M0	12.16	9.79	2.85	2.32	85 ± 3.55
4	M	70	Distal	AC	Esophagectomy	T1 N1 M0	Tx N1 M0	T+ N0 M1a	T+ N0 M0	T2 N1 M0	6.99	5.71	NA	NA	76 ± 2.56
5	F	57	Mid	AC	Stent	T3 N1 M0	T4 N1 M1b	T+ N1 M1b	T+ N0 M0	NA	5.92	4.74	5.11	4.09	68 ± 2.89
6	M	48	GEJ	AC	Esophagectomy	T3 N0 M0	T3 N0 M0	T+ N0 M0	T+ N0 M0	T3 N0 M0	5.67	4.59	4.41	3.58	77 ± 3.35
7	M	56	Mid	SCC	Stent	T3 N1 M0	T3 N0 M0	T+ N0 M0	T+ N0 M0	T4 N1 Mx	14.04	11.50	5.25	4.27	62 ± 5.15
8	F	50	Distal	AC	Esophagectomy	T3 N1 M0	T3 N0 M1	T+ N0 M0	T+ N0 M0	T3 N1 M0	5.50	4.62	2.89	2.30	72 ± 2.49
9	F	57	Mid	SCC	Esophagectomy	T3 N0 M0	T3 N1 M0	T+ N0 M0	T+ N1 M0	T3 N1 M0	8.12	6.76	3.63	2.99	74 ± 5.85
10	M	65	Distal	AC	Esophagectomy	T3 N1 M0	T4 N0 M0	T+ N1 M0	T− N0 M0	T4 N1 M0	4.40	3.58	NA	NA	57 ± 6.61

PA = pathology; AC = adenocarcinoma; mid = mid-esophagus; GEJ = gastroesophageal junction; NA = not applicable; SCC = squamous cell carcinoma.

FIGURE 1. ^{18}F-FDG PET (A) and ^{18}F-FLT PET (B) of patient 2 with long esophageal tumor.

patients 4 and 10, no uptake of ^{18}F-FLT was observed. Therefore, the SUV could not be calculated for ^{18}F-FLT in these 2 patients.

Staging of Esophageal Cancer with ^{18}F-FDG PET and ^{18}F-FLT PET

Pathology for assessment of lymph nodes was available in 9 patients. ^{18}F-FDG PET and ^{18}F-FLT PET were comparable with regard to the detection of regional lymph nodes. Both ^{18}F-FDG PET and ^{18}F-FLT PET correctly detected regional lymph node metastases in only 2 of 8 patients.

^{18}F-FDG PET showed false-positive uptake in the celiac trunk region in patient 4, whereas all other staging modalities, including ^{18}F-FLT PET, did not show any abnormality. Pathologic examination revealed cellular reactivity in the celiac trunk lymph nodes in this patient, and the uptake on ^{18}F-FDG PET was scored as a false-positive result. In patient 5, ^{18}F-FDG PET and CT showed a cervical lymph node metastasis. ^{18}F-FLT PET did not detect this metastasis and this was scored as a false-negative result.

Comparison Between ^{18}F-FDG and ^{18}F-FLT Uptake

The median SUV_{max} and median SUV_{mean} for ^{18}F-FDG were 7.4 and 6.0 and for ^{18}F-FLT were 4.1 and 3.4. Uptake of ^{18}F-FDG was significantly higher than ^{18}F-FLT, whether expressed in SUV_{max} ($P = 0.012$) or SUV_{mean} ($P = 0.012$). Figure 1 shows ^{18}F-FDG PET and ^{18}F-FLT PET of patient 2.

Correlation of ^{18}F-FDG and ^{18}F-FLT Uptake with MIB-1 Score

All tissue specimens contained immunoreactivity to Ki-67 antigen. Ki-67 positivity ranged from 57% to 85%, with a median of 73% (Table 1). Linear regression analysis indicated no correlation between ^{18}F-FDG SUV and Ki-67 or between ^{18}F-FLT SUV and Ki-76 (^{18}F-FDG SUV_{max} vs.

Ki-67, $r = 0.14$; ^{18}F-FLT SUV_{max} vs. Ki-76, $r = -0.76$; ^{18}F-FDG SUV_{mean} vs. Ki-67, $r = 0.13$; ^{18}F-FLT SUV_{mean} vs. Ki-76, $r = -0.74$).

Additional Findings

In patient 6, ^{18}F-FDG PET showed uptake in the rectosigmoid. However, ^{18}F-FLT PET did not show any abnormality in this region. Additional sigmoidoscopy revealed diverticulitis. In patient 10, a hypermetabolic lesion in the ascending colon was found on ^{18}F-FDG PET and proven to be a carcinoma by colonoscopy. However, ^{18}F-FLT PET did not detect this synchronous neoplasia.

DISCUSSION

This pilot study was conducted on 10 patients and showed that ^{18}F-FDG PET could detect all esophageal cancers, whereas ^{18}F-FLT PET visualized the tumor in 8 patients. Both ^{18}F-FDG PET and ^{18}F-FLT PET detected lymph node metastases in 2 of 8 patients. The uptake of ^{18}F-FDG (median SUV_{mean}, 6.0; range, 3.6–11.5) in esophageal cancer was significantly higher than that of ^{18}F-FLT (median SUV_{mean}, 3.4; range, 2.3–4.3). Furthermore, neither ^{18}F-FDG uptake nor ^{18}F-FLT uptake reflects proliferation as determined by Ki-67 immunostaining.

^{18}F-FDG PET was able to detect all primary esophageal cancers, whereas ^{18}F-FLT PET missed 2 of them. This fact may be related to the lower uptake of ^{18}F-FLT compared with ^{18}F-FDG, which has been reported earlier for several other tumors (*18–21*). The ^{18}F-FLT phosphorylation rate in vitro is known to be about 30% of the phosphorylation rate of serum thymidine by TK1, which could explain the low ^{18}F-FLT uptake in the tumor (*22,23*). Although plasma levels are low, thymidine may compete with ^{18}F-FLT for the active site of nucleoside carriers in cell membranes and also for the active site of the trapping enzyme TK1. Moreover, the affinity of human TK1 for thymidine has been reported to be 4-fold higher than the affinity for ^{18}F-FLT (*22,24*).

Both ^{18}F-FDG PET and ^{18}F-FLT PET had low sensitivity for the detection of regional lymph node metastases (2 of 8 patients). Several studies have reported the moderate sensitivity of ^{18}F-FDG PET for detection of regional lymph node metastases, which ranges from 8% to 67% (*25–27*). ^{18}F-FLT PET did not improve the regional staging of esophageal cancer. This can be explained by low tissue uptake of ^{18}F-FLT (as described) or by the detection limit of PET for small tumor deposits (*28*).

A strong correlation between ^{18}F-FLT uptake and proliferation expressed as Ki-67–positive cells was found for lung cancer and sarcoma (*18,29*). However, we did not find a correlation between ^{18}F-FLT uptake and Ki-67 or between ^{18}F-FDG uptake and Ki-67. A correlation between ^{18}F-FLT uptake and proliferation was not reported for breast cancer or thoracic tumors (*18,21,30*). The rationale of ^{18}F-FLT uptake in malignant tissue is based on TK1 dependence of proliferation (*12,18*). However, tumors vary in the relative contribution of de novo and salvage nucleotide biosynthesis. Dominance of de novo pathways, although uncommon, would mask proliferation-dependent increases in TK1 activity (*31*). Furthermore, in cells for which proliferation is less dependent on TK1, the correlation between tracer uptake and TK1 activity was poor (*18,31*). We did not obtain full kinetic parameters of ^{18}F-FLT, which might be explain why a correlation between ^{18}F-FLT and proliferation was not found. For example, the correlation between the rate of phosphorylation of ^{18}F-FLT and SUV should be investigated to assess proliferation (*32*). In addition, Ki-67 is not a perfect measure of DNA synthesis, since it just measures the number of cells in a proliferating state (*16*). Moreover, Ki-67 was assessed in a proliferating part of the tumor and was compared with the SUV value of a tumor volume. This comparison might be flawed.

Its small sample size and the absence of evaluation after therapy limit drawing solid conclusions from this study. ^{18}F-FDG PET is able to identify nonresponders early during neoadjuvant chemoradiotherapy for esophageal cancer (*33*). Therefore, it will be worthwhile to investigate the ability of ^{18}F-FLT PET in identifying nonresponders to neoadjuvant treatment regimens.

At present, ^{18}F-FDG is the tracer of choice for the staging of esophageal cancer. Despite the lower incidence of false-positive results with ^{18}F-FLT, false-negative results will increase by using ^{18}F-FLT, which is a major disadvantage for the staging of esophageal cancer.

CONCLUSION

In this feasibility study, ^{18}F-FLT uptake in esophageal cancer is significantly lower compared with ^{18}F-FDG uptake. ^{18}F-FLT PET has more false-negative findings and fewer false-positive findings compared with ^{18}F-FDG PET. In addition, ^{18}F-FLT uptake and ^{18}F-FDG uptake in esophageal cancer do not reflect proliferation in this population.

ACKNOWLEDGMENT

This study was supported by a ZonMw program for Health Care Efficiency Research and the Dutch Cancer Foundation.

REFERENCES

1. Rice TW. Clinical staging of esophageal carcinoma. CT, EUS, and PET. *Chest Surg Clin N Am.* 2000;10:471–485.
2. Clements DM, Bowrey DJ, Havard TJ. The role of staging investigations for oesophago-gastric carcinoma. *Eur J Surg Oncol.* 2004;30:309–312.
3. Czernin J, Phelps ME. Positron emission tomography scanning: current and future applications. *Annu Rev Med.* 2002;53:89–112.
4. Phay JE, Hussain HB, Moley JF. Strategy for identification of novel glucose transporter family members by using internet-based genomic databases. *Surgery.* 2000;128:946–951.
5. Flamen P, Lerut A, Van Cutsem E, et al. Utility of positron emission tomography for the staging of patients with potentially operable esophageal carcinoma. *J Clin Oncol.* 2000;18:3202–3210.
6. Heeren PA, Jager PL, Bongaerts F, Van Dullemen H, Sluiter W, Plukker JT. Detection of distant metastases in esophageal cancer with ^{18}F-FDG PET. *J Nucl Med.* 2004;45:980–987.
7. Strauss LG. Fluorine-18 deoxyglucose and false-positive results: a major problem in the diagnostics of oncological patients. *Eur J Nucl Med.* 1996;23:1409–1415.

8. van Westreenen HL, Heeren PA, Jager PL, van Dullemen HM, Groen H, Plukker JT. Pitfalls of positive findings in staging esophageal cancer with F-18-fluorodeoxyglucose positron emission tomography. *Ann Surg Oncol.* 2003;10:1100–1105.
9. Ishimori T, Saga T, Mamede M, et al. Increased ^{18}F-FDG uptake in a model of inflammation: concanavalin A-mediated lymphocyte activation. *J Nucl Med.* 2002;43:658–663.
10. Kubota R, Yamada S, Kubota K, Ishiwata K, Tamahashi N, Ido T. Intratumoral distribution of fluorine-18-fluorodeoxyglucose in vivo: high accumulation in macrophages and granulation tissues studied by microautoradiography. *J Nucl Med.* 1992;33:1972–1980.
11. Mier W, Haberkorn U, Eisenhut M. [^{18}F]FLT: portrait of a proliferation marker. *Eur J Nucl Med Mol Imaging.* 2002;29:165–169.
12. Shields AF, Grierson JR, Dohmen BM, et al. Imaging proliferation in vivo with [F-18]FLT and positron emission tomography. *Nat Med.* 1998;4:1334–1336.
13. Hamacher K, Coenen HH, Stocklin G. Efficient stereospecific synthesis of no-carrier-added 2-[^{18}F]-fluoro-2-deoxy-D-glucose using aminopolyether supported nucleophilic substitution. *J Nucl Med.* 1986;27:235–238.
14. Machulla HJ, Blocther A, Kuntzsch M, Piert M, Wei R, Grierson JR. Simplified labeling approach for synthesizing 3′-deoxy-3′-[F-18]fluorothymidine ([F-18]FLT). *J Radiochem Nucl Chem.* 2000;243:843–846.
15. Lonneux M, Borbath I, Bol A, et al. Attenuation correction in whole-body FDG oncological studies: the role of statistical reconstruction. *Eur J Nucl Med.* 1999;26:591–598.
16. Scholzen T, Gerdes J. The Ki-67 protein: from the known and the unknown. *J Cell Physiol.* 2000;182:311–322.
17. Sobin LH, Wittekind C. *TNM Classification of Malignant Tumours.* 6th ed. New York, NY: John Wiley & Sons; 2003:60–64.
18. Buck AK, Halter G, Schirrmeister H, et al. Imaging proliferation in lung tumors with PET: ^{18}F-FLT versus ^{18}F-FDG. *J Nucl Med.* 2003;44:1426–1431.
19. 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;45:226–231.
20. Francis DL, Visvikis D, Costa DC, et al. Potential impact of [^{18}F]3′-deoxy-3′-fluorothymidine versus [^{18}F]fluoro-2-deoxy-D-glucose in positron emission tomography for colorectal cancer. *Eur J Nucl Med Mol Imaging.* 2003;30:988–994.
21. Smyczek-Gargya B, Fersis N, Dittmann H, et al. PET with [^{18}F]fluorothymidine for imaging of primary breast cancer: a pilot study. *Eur J Nucl Med Mol Imaging.* 2004;31:720–724.
22. Munch-Petersen B, Cloos L, Tyrsted G, Eriksson S. Diverging substrate specificity of pure human thymidine kinases 1 and 2 against antiviral dideoxynucleosides. *J Biol Chem.* 1991;266:9032–9038.
23. Toyohara J, Waki A, Takamatsu S, Yonekura Y, Magata Y, Fujibayashi Y. Basis of FLT as a cell proliferation marker: comparative uptake studies with [^{3}H]thymidine and [^{3}H]arabinothymidine, and cell-analysis in 22 asynchronously growing tumor cell lines. *Nucl Med Biol.* 2002;29:281–287.
24. Kong XB, Zhu QY, Vidal PM, et al. Comparisons of anti-human immunodeficiency virus activities, cellular transport, and plasma and intracellular pharmacokinetics of 3′-fluoro-3′-deoxythymidine and 3′-azido-3′-deoxythymidine. *Antimicrob Agents Chemother.* 1992;36:808–818.
25. Jager PL, Que TH, Vaalburg W, Pruim J, Elsinga P, Plukker JT. Carbon-11 choline or FDG-PET for staging of oesophageal cancer? *Eur J Nucl Med.* 2001;28:1845–1849.
26. Kato H, Kuwano H, Nakajima M, et al. Comparison between positron emission tomography and computed tomography in the use of the assessment of esophageal carcinoma. *Cancer.* 2002;94:921–928.
27. Yoon YC, Lee KS, Shim YM, Kim BT, Kim K, Kim TS. Metastasis to regional lymph nodes in patients with esophageal squamous cell carcinoma: CT versus FDG PET for presurgical detection—prospective study. *Radiology.* 2003;227:764–770.
28. Cobben DC, Jager PL, Elsinga PH, Maas B, Suurmeijer AJ, Hoekstra HJ. 3′-^{18}F-Fluoro-3′-deoxy-L-thymidine: a new tracer for staging metastatic melanoma? *J Nucl Med.* 2003;44:1927–1932.
29. Cobben DC, Elsinga PH, Suurmeijer AJ, et al. Detection and grading of soft tissue sarcomas of the extremities with ^{18}F-3′-fluoro-3′-deoxy-L-thymidine. *Clin Cancer Res.* 2004;10:1685–1690.
30. Dittmann H, Dohmen BM, Paulsen F, et al. [^{18}F]FLT PET for diagnosis and staging of thoracic tumours. *Eur J Nucl Med Mol Imaging.* 2003;30:1407–1412.
31. Schwartz JL, Tamura Y, Jordan R, Grierson JR, Krohn KA. Monitoring tumor cell proliferation by targeting DNA synthetic processes with thymidine and thymidine analogs. *J Nucl Med.* 2003;44:2027–2032.
32. Visvikis D, Francis D, Mulligan R, et al. Comparison of methodologies for the in vivo assessment of ^{18}FLT utilisation in colorectal cancer. *Eur J Nucl Med Mol Imaging.* 2004;31:169–178.
33. Wieder HA, Brucher BL, Zimmermann F, et al. Time course of tumor metabolic activity during chemoradiotherapy of esophageal squamous cell carcinoma and response to treatment. *J Clin Oncol.* 2004;22:900–908.