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Relationship between ^{18}F-FDG uptake on positron emission tomography and molecular biology in malignant pleural mesothelioma

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Abstract *Background:* The usefulness of 2-[^{18}F]-fluoro-2-deoxy-D-glucose (^{18}F-FDG) positron emission tomography (PET) can help for predicting the therapeutic response and outcome in malignant pleural mesothelioma (MPM). However, no satisfactory biologic explanation exists for this phenomenon. The aim of this study is to investigate the underlying biologic mechanisms of ^{18}F-FDG uptake.

Methods: Twenty-one patients with MPM who underwent ^{18}F-FDG PET before treatment were included in this study. Tumour sections were stained by immunohistochemistry for glucose transporter 1 (Glut1); glucose transporter 3 (Glut3); hypoxia-inducible factor-1 alpha (HIF-1α); hexokinase I; vascular endothelial growth factor (VEGF); microvessels (CD34); epidermal growth factor receptor (EGFR); cell proliferation (Ki-67 labelling index); Akt/mTOR signalling pathway (PTEN, p-Akt, p-mTOR and p-S6K); cell cycle control (p53 and pRb); apoptosis marker (bcl-2). We also conducted an *in vitro* study of ^{18}F-FDG uptake in mesothelioma cell lines.

Results: ^{18}F-FDG uptake was significantly correlated with Glut1 ($p < 0.0001$), HIF-1α ($p = 0.006$), hexokinase I ($p = 0.0002$), VEGF ($p = 0.0013$), CD34 ($p = 0.0001$),

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Ki-67($p = 0.0047$), mTOR ($p = 0.00478$) and p53 ($p = 0.0004$). High uptake of ^{18}F-FDG was significantly associated with poor outcome in MPM. Our *in vitro* study showed that upregulation of Glut1 and HIF-1α was closely related with ^{18}F-FDG uptake into mesothelioma cell, and mTOR inhibitor induced a decrease in Glut1 expression and ^{18}F-FDG uptake.
Conclusion: The amount of ^{18}F-FDG uptake in MPM is determined by the presence of glucose metabolism, phosphorylation of glucose, hypoxia, angiogenesis, cell proliferation (Ki-67), cell cycle regulator, and mTOR signalling pathway.

1. Introduction

Malignant pleural mesothelioma (MPM) is an aggressive tumour with a poor prognosis and an increasing incidence in many countries. Recently, the usefulness of 2-[^{18}F]-fluoro-2-deoxy-D-glucose (^{18}F-FDG) positron emission tomography (PET) for the diagnosis of MPM has been investigated in some studies.[1–4] ^{18}F-FDG PET has proved useful in detecting malignant pleural lesions[5] and assessing treatment efficacy in MPM.[4] Moreover, ^{18}F-FDG PET is also described to help predicting the prognosis of MPM.[6] However, there is still no data about the possible mechanisms for ^{18}F-FDG uptake in MPM.

Determination of malignant lesions with ^{18}F-FDG PET is based on the glucose metabolism.[7,8] The overexpression of glucose transporter 1 (Glut1) has been shown to be closely related to ^{18}F-FDG uptake in human cancer.[7,8] Glut 1 is thought to be a possible intrinsic marker of hypoxia, and the expression of Glut 1 has been found to be regulated by hypoxia in a hypoxia inducible factor (HIF)-1-dependent way.[9,10] Previous studies suggest that hypoxic conditions correspond to a higher ^{18}F-FDG uptake.[11,12] In addition, several researchers described the relationship between ^{18}F-FDG uptake and the expression of vascular endothelial growth factor (VEGF) or micro-vessel density (MVD).[13,14] HIF-1α is considered to support tumour growth by the induction of angiogenesis via the expression of the VEGF and also by high and anaerobic metabolic mechanisms.[15] Recent preliminary report demonstrated that ^{18}F-FDG PET could be a valuable tool for assessing the effects of the mammalian target of rapamycin (mTOR) inhibition in lung cancer patients.[16] mTOR is a downstream component of the PI3K/AKT pathway involved in the regulation of cell proliferation, angiogenesis, and metabolism. However, there is no report about the relationship between ^{18}F-FDG uptake within tumour cells and PI3K/AKT/mTOR signalling pathway in human neoplasms. As many factors can influence the extent of ^{18}F-FDG uptake, the underlying mechanisms for ^{18}F-FDG accumulation are still a matter of debate in various human neoplasms. Defining a correlation between these biomarkers and ^{18}F-FDG uptake may lead to a better understanding and interpretation of ^{18}F-FDG PET scanning in MPM. Moreover, the molecular biology including epidermal growth factor receptor (EGFR), cell cycle control (p53 and Rb) and apoptosis (bcl-2), has been described to play an important role in the pathogenesis of MPM.[17] We conducted ^{18}F-FDG PET studies and immunohistochemical analyses in patients with MPM. *In vitro* studies were also performed to investigate the possible mechanisms of ^{18}F-FDG uptake.

2. Material and methods

2.1. Patients

Between August 2003 and May 2009, 25 consecutive patients with MPM who underwent ^{18}F-FDG PET were analysed in this study. Of these patients, four patients were excluded for further studies because the tissue specimen was not available. Thus, a total of 21 patients were analysed in the study. The study protocol was approved by the institutional review board.

The median age of the patients was 66 years (range, 19–79 years). Eighteen patients were men and three were women. Eleven of the 21 patients underwent surgical resection, six patients surgical biopsy and the remaining four patients only percutaneous needle-core biopsy. Disease stage was classified according to the TNM staging system proposed by the International Mesothelioma Interest Group (IMIG).[18] Sixteen patients had the histology of epithelial type, two of biphasic type, one of sarcomatous type, and two of desmoplastic type. Of the total patients, 8, 1, 5 and 7 had stage I, II, III and IV tumours, respectively. As the initial treatment, 11 patients underwent surgery, five patients underwent systemic chemotherapy, two underwent thoracic radiotherapy and three patients had best supportive care alone. If including neoadjuvant therapy or relapse after surgery, 17 of 21 patients had systemic chemotherapy.

2.2. ^{18}F-FDG PET imaging

Patients fasted for at least 4 h before ^{18}F-FDG PET examination. Patients received an intravenous injection of 200–250 MBq of fluoro-2-deoxy-D-glucose and then rested for approximately 1 h before undergoing imaging.[19] Image acquisition was performed using an Advance NXi PET scanner and Discovery PET-CT scanner (GE Medical Systems, Milwaukee, WI, United