Differentiation between malignant transformation and tumour recurrence by ^{68}Ga-bombesin and ^{18}F-FDG-PET, in patients with low grade gliomas

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Abstract

Treatment of gliomas is multimodal. Magnetic resonance imaging (MRI) in the posttreatment course is of limited value due to therapy-induced changes. In low-grade gliomas (LGG) malignant transformation is of special interest. Our *patients and methods* were as follows: In nine consecutive patients with LGG we examined the role of bombesin labelled with gallium-68 (^{68}Ga-bombesin) studied with positron emission tomography (PET), in addition to fluoro-18-fluorodeoxyglucose (^{18}F-FDG) in the differential diagnosis of tumour recurrence versus malignant transformation. We used ^{68}Ga-bombesin combined with ^{18}F-FDG-PET in these patients with suspicious new contrast enhancement at the original tumour site or resection cavity in MRI. Eight patients were operated. In one patient, tumour recurrence was most likely as shown by the PET findings and chemotherapy was administered. *Our results* have shown that in this last mentioned patient after the follow-up period, MRI contrast enhancement was definitively regressive. In the operated patients the tumour was graded as glioblastoma multiforme, gliosarcoma and WHO grade III tumour. In two patients histological grading confirmed the PET findings without malignant transformation. In all of the 9 patients the combination of ^{68}Ga-bombesin and ^{18}F-FDG-PET predicted correctly malignant transformation or recurrence of the initial tumour grade which shows that ^{68}Ga-bombesin-PET can provide additional important information to detect a malignant transformation. *In conclusion* it is crucial for the patient to differentiate the nature of the new lesion in order to endorse an aggressive or non-aggressive treatment.

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Introduction

Treatment of gliomas is multimodal and consists of surgical resection, external or local radiation, local or systemic chemotherapy. Regular follow-up examination with magnetic resonance imaging (MRI) is essential in the post treatment course. However, in some cases due to therapy-induced changes it is difficult to differentiate by MRI between radiation induced changes and tumour recurrence or progression [1,2]. There is particular interest to discern malignant transformation in initial low-grade gliomas (LGG), (WHO grade II) in the post treatment phase, in order to tailor the appropriate adjuvant treatment since LGG have a high probability to transform into high-grade gliomas within a variable and unpredictable period of time, within a median range of 5–7 years [3].

The initial treatment of LLG is surgical resection whenever feasible or stereotactic biopsy to confirm the diagnosis, followed by external radiation, chemotherapy or in some cases watchful waiting [4,5]. Upon follow-up examination, lesions with edema and/or contrast enhancement in MRI may occur, both due to radiation induced changes as well as to recurrent tumour mass [6]. Therefore, re-treatment may not always be indicated as it carries a risk which can outweigh the potential therapeutic benefit. Furthermore, already limited therapeutic options in glioma treatment are additionally shortened if treatment is unnecessary.

Hence we wanted to determine in this study of nine consecutive patients the role of gallium-68-bombesin, positron emission tomography (^{68}Ga-bombesin PET) in addition to fluoro-18 fluorodeoxyglucose (^{18}F-FDG-PET) in the differential diagnosis of tumour recurrence versus malignant transformation, in patients with initially diagnosed LGG. This differentiation is crucial for the patient as it determines further treatment and prognosis.

Patients and methods

Patients

Nine consecutive patients were included in this study. All patients had initially a LGG grade II according to WHO classification [7]. Three patients had an oligodendroglioma, one patient had an oligoastrocytoma, and six patients had harboured pure astrocytic tumours. Initial treatment was applied after stereotactic biopsy in two cases. In one of these two cases, biopsy was followed by stereotactic seed implantation and in the other by external radiation treatment. Out of the remaining seven patients the tumour was removed surgically "en gross" in five and subtotally removed in two patients. One patient received external radiation treatment after tumour recurrence and also a second tumour resection. All patients underwent MRI scans every three to six months. In three patients clinical symptoms with focal seizures led to an earlier MRI scan, while the other six patients were in an asymptomatic course. All nine patients showed new contrast enhancing lesions in their follow-up MRI scans. Median time to the diagnosis of progression or transformation by the new contrast enhancement scan was two years (range 1-18 years). PET was performed in all patients for better determination of the new contrast enhancing lesions (Fig. 1).

Written informed consent was obtained from each patient and also permission to release their medical records. The study was performed in accordance with the institutional review board (2008-208S-MA).

^{18}F-FDG and ^{68}Ga-bombesin PET studies

We used for the first time ^{68}Ga-bombesin, in terms of a pan-bombesin analog, the peptide BZH_3. The pan-bombesin analog BZH_3 was labeled with the positron emitter ^{68}Ga with a half life of 68.3 min suitable for PET studies [8]. BZH_3 binds to at least three receptor subtypes: the BB_1 (also known as neuromedin B), the BB_2 (also known as gastrin releasing peptide or GRP), and the BB_3 (bombesin receptor subtype 3).

For ^{18}F-FDG studies, all patients were nil per os (NPO) for at least 4 hours before PET, blood glucose levels were measured immediately prior to PET and were within normal level (<130 mg/dl). ^{18}F-FDG was prepared according to the method described by Toorongian et al (1990) [9].

PET studies were performed for 60 min following the intravenous application of 300-370 MBq ^{18}F-FDG and 150-210 MBq ^{68}Ga-BZH_3 (3 nmol) on two consecutive days. A dedicated PET system (ECAT EXACT HR+, Siemens Co, Erlangen, Germany) with an axial field of view of 15.3 cm, operated in septa extended two-dimensional mode, was used for patient studies. We used simultaneous acquisition of 63 transaxial slices and theoretical slice thickness of 2.4 mm. Transmission scans for a total of 10 min were obtained prior to radionuclide application, for attenuation correction of the acquired emission tomographic images. An image matrix of 256 x 256 pixels was used for iterative image reconstruction. Reconstructed images were converted to standardized uptake value (SUV) images based on the formula: SUV = tissue concentration (Bq/g) / (injected dose (Bq) / body weight (g)) [10].

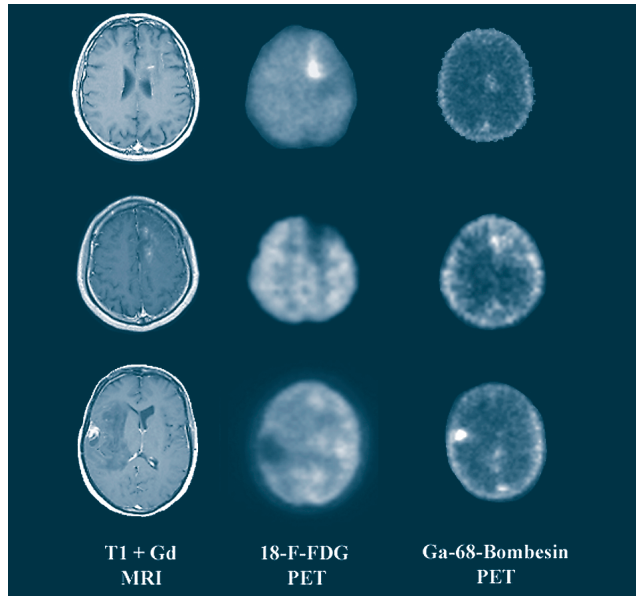

Figure 1. Examples of patients with suspicious lesions, in MRI follow up and the corresponding PET-findings: *Patient 1 (upper row)*:Transversal T1 MR with gadolinium (left) shows new contrast enhancement in a patient with an oligodendroglioma WHO grade II. Transversal PET image (middle) 55-60 min after the i.v. injection of ^{18}F-FDG, enhanced ^{18}F-FDG-uptake in a lesion located in the left frontal lobe with an average SUV of 19.1 and a decreased ^{68}Ga-Bombesin uptake (right). *Patient 2 (middle row)*:T1 MR showed new contrast enhancement in a patient with a suspicious recurrent astrocytoma. ^{18}F-FDG showed low ^{18}F-FDG-uptake in the suspicious lesion located left frontal with an average SUV of 4.6. Bombesin PET in the same patient. Enhanced uptake of ^{68}Ga-BZH3 with an average SUV of 1.12. Again, the lesion can be delineated due to the high contrast. *Patient 3 (bottom row)*: T1 MR shows new contrast enhancement in a patient with initial astrocytoma WHO grade II. ^{18}F-FDG showed low ^{18}F-FDG-uptake in the suspicious lesion located in the right temporo-parietal region with an average SUV of 2.2 and increased bombesin uptake with an average SUV of 0.7.

The SUV 55-60 min post injection was used for the analysis of both tracers.

For evaluation of the dynamic PET data the software package PMod (PMOD, Technologies Ltd., Adliswil, Switzerland) was used. Quantitative evaluation of enhanced tracer uptake on transaxial, coronal and sagittal images was performed using volume of interest (VOIs) consisting of several regions of interest (ROI) over the target area. Irregular ROI were drawn manually.

Results

Examination with PET

SUV of the tracers in suspect areas were compared with mean uptake values in an arterial vessel.

Six patients showed a decrease or a slight increase in ^{18}F-FDG metabolism and an overall increase in bombesin uptake. In one patient there was a decrease in bombesin and ^{18}F-FDG uptake, in another patient there was no bombesin or ^{18}F-FDG uptake at all (Patient 6, Fig. 2). A patient with an initial oligodendroglioma WHO grade II, had a slight decrease in bombesin uptake and a massively increased ^{18}F-FDG metabolism (Fig. 2).

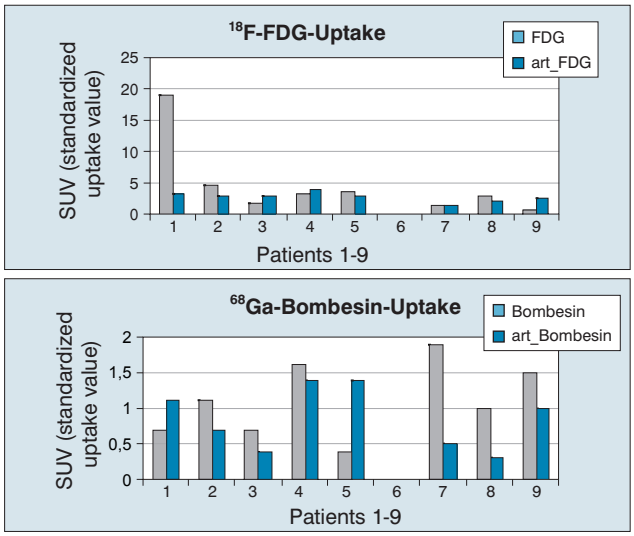

Figure 2. ^{68}Ga-bombesin and ^{18}F-FDG-uptake in the PET-examination.

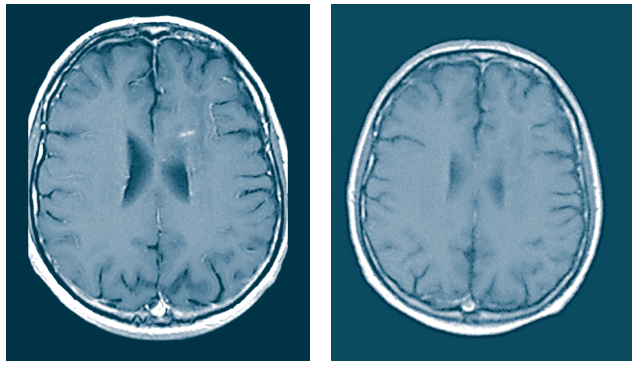

Figure 3. *Patient 1* with an initial oligodendroglioma WHO grade II and a new suspicious contrast enhancement in T1-weighted image after Gd-application (left) and complete regression of contrast enhancement, following chemotherapy with temozolomide (right).

Table 1. *Initial histology and final grading after re-operation.*

Patients	Initial histology	Final grading
1	Oligodendroglioma WHO grade II	(*)
2	Astrocytoma WHO grade II	Glioblastoma multiforme WHO grade IV
3	Astrocytoma WHO grade II	Glioblastoma multiforme WHO grade IV
4	Oligoastrocytoma WHO grade II	Anaplastic oligodendroglioma WHO grade III
5	Astrocytoma WHO grade II	Astrocytoma WHO grade II
6	Astrocytoma WHO grade II	Astrocytoma WHO grade II
7	Astrocytoma WHO grade II	Gliosarcoma WHO grade IV
8	Oligoastrocytoma WHO grade II	Anaplastic oligodendroglioma WHO grade III
9	Oligodendroglioma WHO grade II	Anaplastic oligodendroglioma WHO grade III

(*) No histological confirmation, but complete regression of contrast enhancement after chemotherapy.

Treatment:

Seven patients were operated and the suspected area was surgically removed. In one asymptomatic patient stereotactic biopsy from the suspected area around the right insular region, was performed. The asymptomatic patient with the initial oligodendroglioma received chemotherapy with temozolomide (200mg/m^2 for 5 days in a 28 days cycle), according to the PET findings suggesting tumour recurrence without malignant transformation.

Histological examination and follow-up:

All six patients with an increase in bombesin uptake showed malignant transformation histologically. Two patients had a glioblastoma multiforme, one patient had a gliosarcoma and the three other patients had anaplastic oligodendrogliomas. The two patients with decreased bombesin and ^{18}F-FDG uptake and no bombesin or ^{18}F-FDG uptake at all, showed no evidence for malignant transformation and the diagnosis of WHO grade II astrocytoma was confirmed (Table 1).

The patient with the initial oligodendroglioma and decreased bombesin uptake received four cycles of chemotherapy with temozolomide. Follow-up MRI scanning showed completely regressive contrast enhancement (Fig. 3). The patients with malignant transformation received a concomitant radiochemotherapy scheme according to Stupp et al (2005) [11]. The other two patients with recurrent low grade tumours received external radiation treatment.

Discussion

This study demonstrates the value of PET examination in the follow-up of patients with LGG where equivocal MRI scans show new gadolinium contrast enhancement. In this particular scenario, it is crucial to differentiate between treatment induced changes, tumour recurrence or malignant transformation for further management strategies.

Since MRI in already treated patients is particularly difficult to interpret [12], further diagnostic tools are required to differentiate between transformers and non-transformers. MR-spectroscopy has shown promising results in detecting malignant progression in gliomas [13], validation of this method in clinical routine, however, is still in process. PET more than single-photon emission tomography (SPET) is used instead to fill this diagnostic gap. Limitations for PET diagnostics are the dependence on cyclotron produced tracers, limited availability and high costs. While PET has a moderate spatial resolution up to 5mm, it is even more limited in SPET (8-10mm). However, the latter is available in almost every nuclear medicine department and the investigation is considerably cheaper than PET examinations [14]. Using PET for further diagnostics significant correlation has been reported for ^{18}F-FDG uptake and histological grading of gliomas [15]. ^{18}F-FDG has a sensitivity of 75% and a specificity of 81% to distinguish recurrent tumour from radiation necrosis [16] and it can detect recurrent tumour or transformation by the newly appearing hypermetabolism [17,18]. On the contrary, in our series we found a decrease or only a slight increase ^{18}F-FDG metabolism in all pa-

tients with malignant transformation. Uptake of ^{68}Ga-bombesin was instead increased in all of these patients, which was in excellent accordance with the histological evidence of malignant transformation in each case. Furthermore in cases with decreased bombesin or no bombesin uptake there was no histological evidence for malignant transformation. Therefore bombesin might be a promising tracer for malignant brain tumour cells which are not yet picked up by ^{18}F-FDG-PET.

Contrarily to the aforementioned patients ^{18}F-FDG metabolism was massively increased while bombesin uptake was slightly decreased in one patient with an initially diagnosed oligodendroglioma WHO grade II.

In oligodendrogliomas WHO grade II variability in ^{18}F-FDG metabolism (hypo- and hypermetabolism) is reported [18]. Ginsberg et al (1998) describe that a lack of contrast enhancement on initial MRI scanning is not a reliable criterion for differentiation between high-grade and low-grade tumours; up to 40% of high-grade gliomas showed their presence without enhancement [19]. In a larger series of 314 patients malignancy in non-enhancing supratentorial gliomas was shown in approximately 30% [20]. Especially in oligodendrogliomas the presence of contrast enhancement in MRI scans is not statistically significant in differentiation between high-grade and low-grade tumours [21]. In our patient with new contrast enhancement and an underlying oligodendroglioma we opted for chemotherapy instead of a second surgical intervention, on the basis of an increased ^{18}F-FDG metabolism and decreased ^{68}Ga-bombesin uptake. Although no definite histological proof exists, MRI scanning showed regression of contrast enhancement after chemotherapy, implying appropriate adjuvant treatment. In low-grade astrocytomas, decreased ^{18}F-FDG metabolism is common [18]. Therefore we think that in early stages of transformation ^{18}F-FDG metabolism might still be reduced while ^{68}Ga-bombesin already binds to the above mentioned receptors of malignant glioma cells and might therefore be a more sensitive parameter in interpreting newly detected suspected lesions. This was our rationale to treat those patients with an increased ^{68}Ga-bombesin uptake more aggressively. This treatment strategy was confirmed by the lack of malignant transformation in the two patients with decreased bombesin-uptake.

However, this study is limited by the small number of patients prohibiting to draw a definite conclusion from our findings. Larger patient series are required to better characterize the role of ^{68}Ga-bombesin in PET diagnostics of suspicious lesions in MRI in patients with LGG. Such a study is already on the way.

In conclusion a multimodal diagnostic workup is important in patients with suspicious MRI findings on follow-up examination, after treatment of LGG. It is crucial to differentiate the dignity of a new lesion as it should determine the decision for aggressive or non-aggressive management of these patients. Therefore with regard to the results of this series ^{68}Ga-bombesin-PET seems to provide additional important information in this process and may represent a suitable tool to detect those patients with early malignant transformation that will be missed by ^{18}F-FDG-PET.

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