Two or four hour [18F]FMISO-PET in HNSCC

When is the contrast best?

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Keywords

FMISO PET, head and neck cancer, tumour-to-muscle-ratio, contrast-to-noise-ratio, bio-imaging, radiotherapy treatment planning

Summary

[18F]Fluoromisonidazole positron emission tomography (FMISO-PET) is a non invasive imaging technique that can assist detecting intra tumour regions of hypoxia. FMISO-PET evinces comparatively low signal-to-noise-ratio (SNR) and may be acquired dynamically or after different uptake times post injection (p. i.). The aim of this study was to identify, if static images acquired two hours (MISO2) or four hours (MISO4) p. i. reveal higher contrast. Patients, methods: As part of a prospective trial, 23 patients with cancers of the head and neck underwent [18F]fluorodeoxyglucose (FDG) PET before and during curative radiochemotherapy. Additionally, FMISO-PET studies 2 h and 4 h p. i. were done before treatment and after a mean dose of 11 Gy, 23 Gy and 57 Gy during RCT. After coregistration, a dedicated software was used to define the gross tumour volume (GTV) by FDG PET for the primary tumour. This volume was overlaid to the FMISO images and hypoxia within the GTV was determined. The contrast between hypoxia determined by MISO2 and by MISO4 was investigated and analysed with the Wilcoxon-matched-pairs test. Results: Mean SUV_{max} in tumours of all examinations was 2.2 (stdev: 0.4, min: 1.3, max: 3.4) after 2 h and 2.4 (stdev: 0.7, min: 1.1, max: 4.4) after 4 h. In the neck musculature the mean SUV_{max} was 1.5 at both time points and the mean SUV_{mean} decreased from 1.2 after 2 h to 1.1 after 4 h, respectively. These effects resulted in significantly rising contrast ratios from MISO2 to MISO4. The differently defined contrasts revealed significantly higher values for examinations 4 h p. i. ($p < 0.002$). Conclusion: Data acquisition of [18F]FMISO should be done 4 h p. i. to gather the optimal contrast, preferably allowing further analysis, e. g. hypoxic sub volume definition for therapy planning.

Schlüsselwörter

FMISO PET, Kopf/Hals-Tumoren, Tumor-zu-Muskel-Signalverhältnis, Kontrast-zu-Rausch-Verhältnis, Bioimaging, Strahlentherapieplanung

Zusammenfassung

Die [18F]Fluormisonidazol-Positronenemissionstomographie (FMISO-PET) ist ein nicht invasives Bildgebungsverfahren, das hypoxische Subvolumina in Tumoren detektieren kann. Die FMISO-PET kann dynamisch oder statisch nach unterschiedlichen Uptakezeiten post injectionem (p. i.) akquiriert werden, hat aber ein vergleichsweise niedriges Signal zu Rausch Verhältnis (SNR). Ziel dieser Studie war es für spätere Analysen zu klären, ob der Bildkontrast in statisch aufgenommenen Untersuchungen nach einer Uptakezeit von zwei Stunden (MISO2) oder vier Stunden (MISO4) p. i. höher ist. Patienten, Methoden: Bei einer Subgruppe von 23 Patienten einer prospektiven Studie zur kurativen Radiochemotherapie (RCT) von Plattenepithelkarzinomen des Hals-Nasen-Rachen-Raumes (HNSCC) wurden vor und während der Therapie [18F]Fluordeoxyglukose (FDG-)PET-Untersuchungen durchgeführt. Zusätzlich wurden bei diesen Patienten FMISO-PET-Aufnahmen zwei und vier Stunden p. i. nach Strahlentherapiedosen von im Mittel 11 Gy, 23 Gy und 57 Gy während der RCT akquiriert. Nach Koregistrierung aller PET- und CT-Datensätze wurde die Rover-Software (ABX, Radeberg) verwendet, um das aus der FDG-PET abgeleitete „gross tumour volume" der Primärtumoren festzulegen. Diese Volumina wurden in die FMISO-Datensätze kopiert um Hypoxie innerhalb des Primärtumors zu definieren. Der Kontrast zwischen hypoxischen Regionen in den Aufnahmen MISO2 und MISO4 wurde untersucht und mit dem Wilcoxon-Rangsummen-Test auf signifikante Unterschiede geprüft. Ergebnisse: Der mittlere SUV_{max} der Primärtumoren aller Untersuchungen war 2.2 (stdev: 0.4, min: 1.3, max: 3.4) nach 2 h p. i. und 2.4 (stdev: 0.7, min: 1.1, max: 4.4) nach 4 h p. i.. Der mittlere SUV_{max} in der Nackenmuskulatur war zwei und vier Stunden p. i. 1.5 und der mittlere SUV_{mean} fiel von 1.2 nach 2 h auf 1.1 nach 4 h ab. Diese geringen Veränderungen bedingten aber einen steigenden Kontrast von MISO2 nach MISO4. Für die unterschiedlich definierten Kontraste ergab der Wilcoxon-Rangsummen-Test signifikant höhere Werte in den Untersuchungen vier Stunden p. i. ($p < 0.002$). Schlussfolgerung: Die Datenakquisition für die [18F]FMISO-PET sollte vorzugsweise vier Stunden p. i. erfolgen, da der Kontrast zwei Stunden p. i. schlechter ist. Diese Datensätze eignen sich deshalb besser für weitere Analysen, z. B. für die verbesserte Definition hypoxischer Tumorsubvolumina zur Strahlentherapieplanung.

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Zwei oder vier Stunden [18F]FMISO-PET in Kopf-Hals-Karzinomen: Wann ist der Bildkontrast am höchsten?
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Independent of treatment, tumour hypoxia is an important prognostic factor for various cancers (1, 11). It was shown that tumour hypoxia expressed by an oxygen partial pressure (pO_2) < 10 mmHg can be noninvasively visualized by positron emission tomography (PET) (23) using the hypoxia tracer [18F]fluoromisonidazole (FMISO) (28). FMISO is thought to visualize preferably chronic hypoxia, which is assumed to be of particular importance for outcome of radiotherapy. Tackling hypoxia related poor prognosis (25) by boosting radiotherapy during standard radiochemotherapy (RCT) is subject of current research (4, 15). However, several limitations of FMISO-PET related to limited reproducibility are currently discussed (18). Ongoing research utilizes imaging flow dynamics (29) using 4D-PET during the first minutes post injection (p. i.) and static PET images commonly acquired between two and four hours p. i. (6). FMISO data might be segmented to provide information on hypoxic sub volumes in tumours and may be integrated into radiotherapy planning since hypoxia has been shown to be a significant determinant of local control after fractionated irradiation (31–32). Clinical investigations utilizing FMISO were initially performed in patients with cancers of the head and neck region (10, 24) and only few centres promote research on FMISO-PET (14). Unfortunately, FMISO-PET does not provide high signal-to-noise-ratio (SNR) and by far not as high contrast-to-noise-ratio (CNR) as supplied by FDG-PET. Given the discussed restrictions, to optimize hypoxic sub volume delineation, imaging should evince high CNR and even small improvements in CNR are valuable.

The aim of this study was to decide, whether FMISO data sets acquired two or four hours p. i. show higher CNR and are therefore more suitable for further analysis and integration into therapeutic decisions, e. g. radiotherapy treatment planning.

Patients, material, methods

Patients

The imaging data included into this analysis has been acquired from a subset of patients accrued for an ongoing prospective study on head and neck squamous cell carcinomas (HNSCC) imaged between July 2006 and August 2009. Major inclusion criteria:

1. non operable HNSCC,
2. curative therapeutic intent,
3. standard RCT and
4. WHO health status 0–2.

We included and analysed imaging data of 23 patients (3 women, median age 55 years) in clinical stages III (n = 9) and IV (n = 14). The prospective trial was approved by the institutional ethics committee and the Federal Office for Radiation Protection of Germany. All patients were informed about the aims of the study and gave their written informed consent.

Data acquisition

PET data acquisition was done using a Biograph16 PET/CT (Siemens Medical Solutions Inc., Knoxville, TN, USA). Before RCT an initial diagnostic whole body FDG-PET/CT 76±19 min p. i. of 335±25 MBq [18F]FDG (GlucoRos®, Research-Centre Dresden-Rossendorf, Germany) and 120 ml contrast material (Ultravist370®, Bayer, Germany) was acquired for staging (▶Fig. 1a). The pretherapeutic FMISO-PET imaging was acquired two to four days after the initial FDG-PET. Static FMISO images were acquired 126±11 min (MISO2) and 241±16 min (MISO4) p. i. of 256±37 MBq (▶Fig. 1b). For each data set one field of view (FOV; 16 cm in z-axis) was scanned for 15 min as three-dimensional emission scan. The patients were imaged before RCT, after a mean dose of 11±4 Gy, 23±9 Gy and 57±5 Gy during RCT. Due to clinical issues, imaging was not possible at the same day during the course of RCT in every patient. During repetitive imaging, patients were immobilized with an individually fitted thermoplastic face mask (additec GmbH, Markt Indersdorf, Germany) in a supine position on a radiotherapy board. This ensured reproducible positioning not only during radiation but also during consecutive FMISO- and FDG-PET/CT. FMISO-PET tracer was delivered from Research Centre Dresden-Rossendorf, Germany and was synthesized as published before (26). For different clinical reasons we were unable to acquire all MISO2 and MISO4 PETs in all of the patients. A total of 61 out of 92 pairs of MISO2 + MISO4 data sets, exemplarily given in ▶Figure 2, were available for the present study.

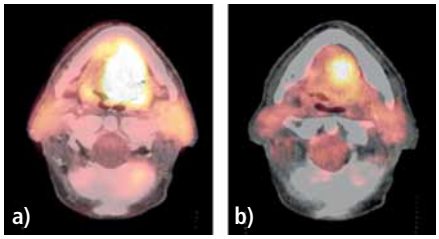

Fig. 1 A patient with an oropharyngeal HNSCC before treatment, investigated with two days interval: **a)** fused FDG-PET/CT; **b)** FMISO-PET/CT

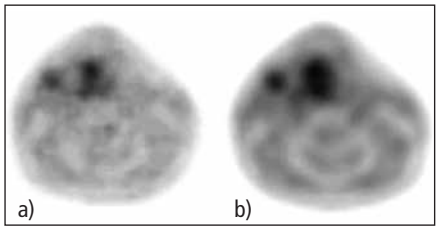

Fig. 2 A patient with an hypopharyngeal HNSCC and lymph node metastasis before treatment (representative axial slices)
a) MISO2; **b)** MISO4

Tab. 1 Treatment and diagnostic schedule of the prospective study with curative therapeutic intent using radiochemotherapy (pre-RCT: before radiochemotherapy; ● FMISO-PET/CT; ❒ FDG-PET/CT)

	pre-RCT	week					
		1	2	3	4	5	6
chemotherapy		×	×	×	×	×	×
radiotherapy		|||||	|||||	|||||	||||| |||||	||||| |||||	||||| ||||| boost
PET-CT	●❒	●	●				●❒

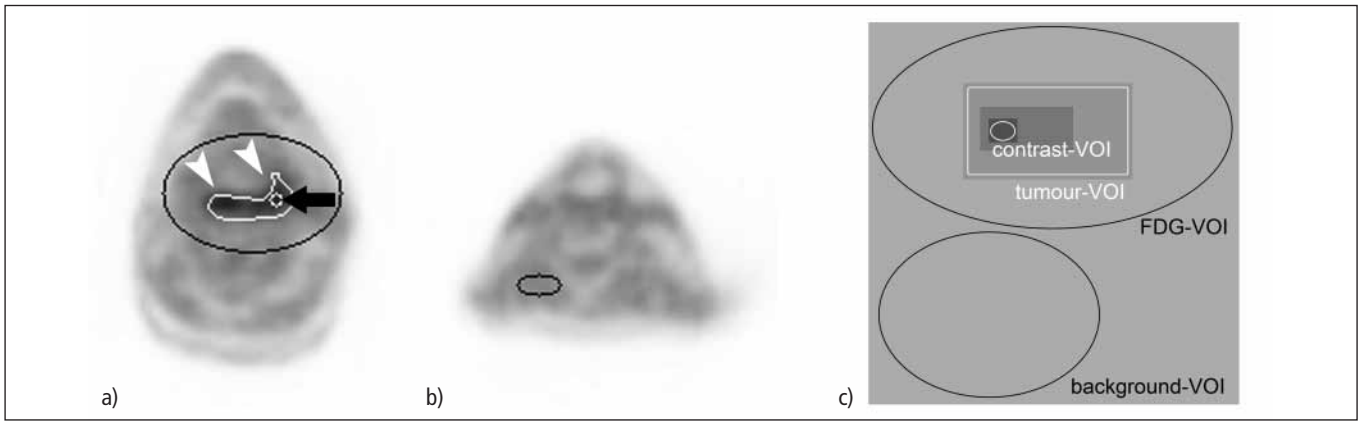

Fig. 3 Axial slices of an FMISO-PET data set showing VOI definitions for volume analysis
a) hypopharyngeal region (→ contrast-VOI, white triangles: tumour-VOI, black ellipse: FDG-VOI)
b) upper cervical region (black ellipse: background-VOI as defined by CT and copied into the FMISO-PET)
c) schematic illustration of VOIs clarifying nomenclature

Data analysis

FMISO data sets were reconstructed using the attenuation maps provided by the low dose CT-data set measured immediately before FMISO image acquisition at the same scanner. The images were also corrected for scatter, randoms, dead time and frame duration by the reconstruction workstation of the scanner. All FMISO and FDG images were registered to the CT data set gathered in tandem with the initial FDG-PET using Rover-Software (ABX, Radeberg, Germany) using rigid registration. Registration errors between all PET images were in the range of the voxel size, i. e. 5 mm³. Tumour volume was defined on the FDG-PET data set by applying a semi-automatic source-to-background-algorithm within a manually defined volume of interest (VOI) that was called FDG-VOI on the FDG-PET provided by Rover-Software previously described in (13) and applied in (9). The Rover-Software is capable to handle all VOIs in every registered data set. Lymph node metastases, if present, were excluded from analysis. The resulting FDG-based segmented volume served as VOI (tumour-VOI) in the registered FMISO-data sets and was not modified during therapy. Additionally, a 3.5 ml small ellipsoid (contrast-VOI) was located at the FMISO activity hot spot position inside the tumour volume. Another 5 to 10 ml large ellipsoid (background-VOI) delineated using the low dose CT in neck muscle (erector spine muscle) was defined as reference volume for background activity analysis of FMISO-PET. All manually defined VOIs were determined by a physician with more than three years of experience analysing FMISO-PET/CT data (NA). VOIs applied to an exemplar patient data set and a schematic overview is illustrated in ▶Figure 3. The resulting activity parameters of all VOIs were assessed using Rover-Software and mean values, standard deviation and maxima are given. The standard uptake values (SUV) of these parameters where calculated including decay correction using activity at injection and time interval between injection and acquisition start time as well as the body weight.

Analysis of hypoxic subvolumes and their modification during therapy will be analysed in further investigations.

Statistical analysis

Based on the performed measurements, at least two contrast definitions are possible:

- the tumour-to-muscle-ratio (TMR),
- the contrast-to-noise-ratio (CNR).

TMR is defined as SUV of the tumour divided by the SUV_{mean} in the background-VOI:

$$TMR = \frac{SUV_{tumour}}{SUV_{mean\ background}}$$

CNR is defined as SUV of the tumour minus SUV_{mean} in the background-VOI and the result being divided by the standard deviation (SD) inside the background-VOI:

$$CNR = \frac{(SUV_{tumour} - SUV_{mean\ background})}{SUV_{SD\ background}}$$

These two equations combined with the two ways of identifying uptake (mean and maximum) lead to four contrast definitions. The resulting values are named TMR_{mean}, TMR_{max}, CNR_{mean} and CNR_{max} and were calculated for contrast-VOI and tumour-VOI separately. Statistical analysis for comparison of the contrasts of MISO2 and MISO4 data sets was done using the Wilcoxon-matched-pairs test.

Results

Mean SUV_{max} in tumours of all examinations was 2.2 after 2 h and 2.4 after 4 h. In the neck musculature the mean SUVmax was 1.5 at both time points and the mean SUV_{mean} decreased from 1.2 after 2 h to 1.1 after 4 h, respectively. These effects resulted in significantly rising contrast ratios from MISO2 to MISO4. The differently defined contrasts revealed significantly higher values for examinations four hours p. i. by applying the Wilcoxon-matched-pairs test ($p < 0.002$).

Count rate statistics of the investigated VOIs are given in ▶Table 2. Count rates are reduced by a factor of two due to the decay during waiting time between MISO2 and MISO4, which is in the range of [18F] half life. Exemplarily, the mean activity in the

Tab. 2 VOIs from FMISO-PET data sets 2 h and 4 h after tracer injection; stdev: standard deviation of count rate or SUV averaged over the respective data sets; min/max: minimum/maximum values

FMISO-PET		MISO2 (stdev, min, max)			MISO4 (stdev, min, max)		
		mean	max	stdev	mean	max	stdev
count rate (Bq/ml)	contrast-VOI	3606.7 (1038.3, 1688.9, 6338.5)	4217.7 (1216.2, 1891.5, 7141.4)	95.8 (148.6, 64, 802.5)	1760.2 (600.2, 790.9, 3510.3)	2266.3 (805.5, 944.2, 4391)	221.7 (112.5, 57, 608.4)
	tumour-VOI	3071.7 (803.2, 1390.1, 5133.6)	4256.7 (1193.8, 1975.8, 7141.4)	459.4 (207.5, 142.9, 1029.2)	1518.6 (481.6, 660.9, 3171.2)	2274.3 (774.3, 950.3, 4491.9)	251 (122.2, 89.7, 775.4)
	background-VOI	2374.2 (681.3, 1191.4, 4485.4)	3030.2 (737.5, 1546.6, 4878.8)	286.6 (107.2, 122, 545)	1080.6 (341, 499, 2625.8)	1456.4 (411.1, 718, 3209.8)	123.1 (31.9, 58.4, 200.6)
SUV	contrast-VOI	1.8 (0.4, 1.1, 2.7)	2.2 (0.4, 1.3, 3.4)	0.2 (0.1, 0.1, 0.4)	1.8 (0.5, 0.9, 3.2)	2.4 (0.7, 1.1, 4.4)	0.2 (0.1, 0.1, 0.6)
	tumour-VOI	1.6 (0.3, 0.9, 2.2)	2.2 (0.4, 1.2, 3.4)	0.2 (0.1, 0.1, 0.5)	1.6 (0.3, 0.8, 2.5)	2.4 (0.7, 1.3, 4.3)	0.3 (0.1, 0.1, 0.7)
	background-VOI	1.2 (0.2, 0.7, 1.7)	1.5 (0.2, 1.1, 2.3)	0.1 (0.1, 0.1, 0.3)	1.1 (0.2, 0.8, 1.5)	1.5 (0.2, 1.1, 2)	0.1 (0, 0.1, 0.2)

contrast VOI decreases from 3607 Bq/ml in MISO2 to 1760 Bq/ml in MISO4. These are still enough events for further analysis. The resulting SUV of the parameters averaged over all patients are shown in ▶Table 2.

The rising contrast from MISO2 to MISO4 is independent of the choice of the presented definitions of contrast. This is expressed by rising TMR and CNR between MISO2 and MISO4 indicating a more convincing analysis on MISO4 images. Wilcoxon-matched-pairs test results shown in ▶Table 3 suggest a highly significant difference of contrasts between these two data sets.

TMR and CNR values for contrast-VOI and tumour-VOI as well as the respective p-values are shown in ▶Table 3. TMR_{max} and CNR_{max} as measured in the contrast-VOI and tumour-VOI revealed the highest differences between MISO2 and MISO4 indicating that further usage of MISO4 for clinical application is favourable.

By visual analysis of the 23 data pairs, the spatial distribution of the intratumoural FMISO uptake remains constant between the investigated time points MISO2 and MISO 4 (▶Fig. 2). To provide an overview, ▶Figure 4 shows the comparison of selected analysed parameters after two and four hours p. i.

Tab. 3 Calculated tumour-to-muscle- (TMR) and contrast-to-noise-ratios (CNR) from FMISO-PET data sets 2 h and 4 h after tracer injection and p-values of Wilcoxon-matched-pairs test for MISO2 and MISO4 data sets: The values in CNR_{max} differ minimally since few hot spots were not in the main uptake region of FMISO, which was thought to be clinically more relevant.

		TMR		CNR	
		mean	max	mean	max
MISO2	contrast-VOI	1.6	1.8	4.7	7.0
	tumour-VOI	1.3	1.8	2.6	7.2
MISO4	contrast-VOI	1.7	2.2	5.7	9.9
	tumour-VOI	1.4	2.2	3.6	10.0
p values	contrast-VOI	1.6e-03	7.3e-08	1.9e-03	4.3e-08
	tumour-VOI	4.1e-05	3.1e-09	5.7e-06	1.8e-08

Discussion

The comparison of 61 pairs of two and four hour FMISO-PETs revealed that images acquired after four hours of uptake and wash out provided higher contrast between hypoxia within the HNSCC and the background in the neck muscles. Decay time of [18F] is approximately the time between the two FMISO measurements and TMR and CNR are increasing during this period. The four hour images are preferably used for further analysis, especially if spatial information is to be derived, which may be relevant if hypoxia information is used for radiation therapy planning. The pathophysiological background is that major amounts of the injected FMISO are bound intracellular in hypoxic tissues whereas FMISO will be washed out from normoxic tissues via blood pool exchange, hence increasing contrast over time (19–20). Because of the lipophilicity and the comparatively slow adduct-formation and clearance of unbound tracer, imaging with FMISO remains challenging (2, 21).

Based on the imaging performed, we decided to use the FDG data to define the tumour volume, as it has been done before (3). The semi-automated source-to-background algorithm applied by the Rover software excluded interobserver variation during visual segmentation and a similar strategy was applied for FMISO-PET analysis previously (27). The reasons for this volume definition were to exclude specific epithelial cellular FMISO uptake not related to hypoxia and to exclude evaluation of hypoxia outside of the tumour related to changes of surrounding normal tissues during therapy. Studies designed to evaluate spatial information of FMISO data sets may preferably apply a similar definition. We used the initial FDG tumour volume for all subsequent images in one patient during therapy. Since tumour shrinkage during therapy on one side and increasing mucosal uptake on the other side will unpredictably modify FDG uptake, this seemed to be the most appropriate approach.

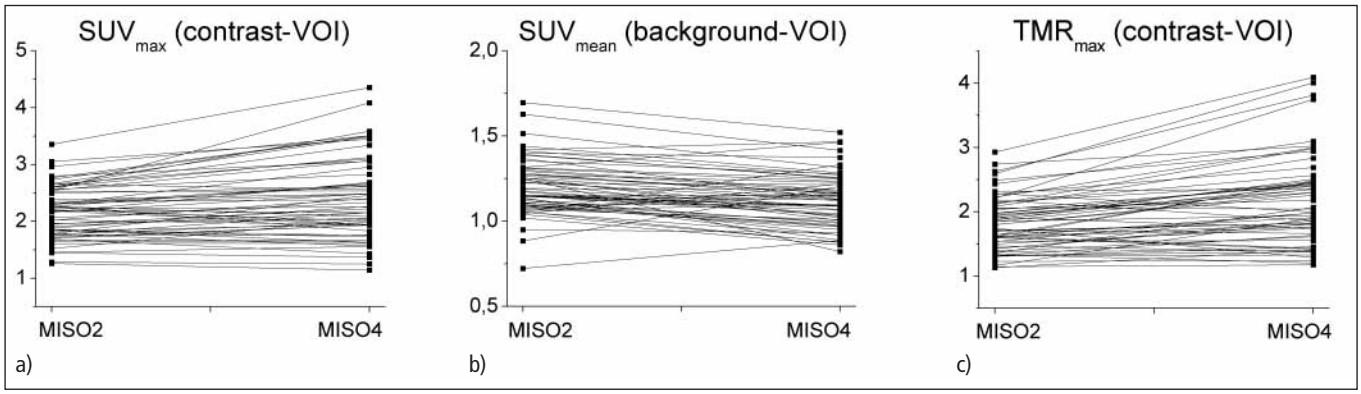

Fig. 4 Comparison of SUV_{max} in the contrast-VOI, SUV_{mean} in the background-VOI and TMR_{max} in the contrast-VOI between MISO2 and MISO4: Despite decay during a complete half life between MISO2 and MISO4, SUV_{max} in the contrast VOI shows an upward trend, while SUV_{mean} in the background-VOI shows a downward trend. Taken together, these two trends lead to a significant increase of the TMR_{max} in the contrast-VOI.

The evaluation of contrast in our study included several general definitions that are provided herein to ensure wide comparability. To select a threshold to define hypoxia authors have applied measurements of blood activity and have chosen thresholds of tissue-to-blood activity ratios between 1.2 and ≥ 1.4 at 120–160 min p. i. (15, 22–23). In contrast to these studies, we and others did not include blood sampling and therefore utilized background measurements in muscle to define a threshold that would define hypoxia. This background signal, measured in muscle and segmented on the plain CT to exclude fat and bone, served as reference in this contrast analysis study. To use the SUV_{mean} of the background VOI for further analysis is straightforward. In comparison, quantification of the tumour hypoxia is more complex even though, only the determination of contrast was aimed for. Inserting small ellipsoid VOIs on hot spots for maximum activity measurement in low contrast phantom data was applied before (12). Alternatively it is possible to identify the SUV_{max} of the tumour as done in clinical routine. Furthermore, the analysis of the SUV_{mean} in such a volume will reduce the influence of noise in the image, which is comparatively high in FMISO PET. Both approaches are common and differences and consequences are discussed between various user groups. Therefore, in our work we studied combinations of these parameters observing a reproducible result: independent from the chosen equation, contrast increased from MISO2 to MISO4 with high but varying significance. The variation underlines the importance of the choice of contrast definition for research and clinical routine. TMR_{max} and CNR_{mean} showed smaller changes from MISO2 to MISO4 than CNR_{max}. On the one hand this is due to the definition of the ratio. TMR is a relative value depending directly on the mean background activity, which is falling due to decay and wash out. CNR depends directly on the noise inside the background. For threshold based segmentation algorithms noise is more important than the ratio between tumour and background activity. Two data sets with identical TMR might differ in CNR. Most automatic threshold segmentation algorithms will fail, if a low CNR indicates that the tumour hides in the noise of the background. But TMR is not able to numerate this objective. CNR should therefore always be regarded when analysing low contrast PET images. On the other hand mean and maximum values differ between the VOIs. It seems not meaningful to analyse mean values of the tumour-VOI because of the reproducibility issues, e. g. visual segmentation of VOI and biological variability (18). The increased contrast of four hour FMISO-PET images is mainly influenced by processes related to the faster wash-out of tracer out of the normal tissues as compared to hypoxic tumours, a mechanism that is well known from other methods used in nuclear medicine. Dubois et al. (5) suggested the same presumption analysing measurements of ^{18}F-(EF3), another suggested PET tracer for tumour hypoxia. Despite decay during a complete half life between MISO2 and MISO4, SUV_{max} in the contrast VOI shows an upward trend, while SUV_{mean} in the background-VOI shows a downward trend. Taken together, these two trends lead to a highly significant increase of the TMR_{max} and the CMR_{max} in the contrast-VOI and the tumour-VOI in MISO4. Already in 1987 Grunbaum and colleagues suggested from preclinical investigations that imaging with FMISO would be most favourable at 4 h p. i. (8). As with FDG-PET in clinical application, we are aware of summing up several effects by investigating patients with FMISO. It was not intended in this study to investigate further influences of biological and technical variability, e.g. correlation of hypoxic voxels in repetitive FMISO-PETs or relevance of FMISO-PET for planning a radiotherapy boost volume as others have done (16, 18). Further correlations with additional tracers not related to hypoxia might be useful (17).

Several imaging analysis tools exist that are applicable for PET imaging, but most of them are not optimized for this special kind of application. Only few software packages are available that are focussed on quantifying PET examinations beyond clinical routines and most researchers have designed custom made programs, e. g. for neurological or cardiac applications. These and oncologic analysis tools rely on measured activities or SUVs and apply contouring algorithms utilizing thresholds, source-to-background or gradient based information. Available software packages are very well suited to analyse FDG-PET data but will typically fail in low contrast data sets as seen in FMISO.

The increased overall quality of the images derived with FMISO delivering a lower signal-to-noise and contrast-to-noise-ratio as compared to FDG-PET was our aim. Our eventual goal is to include FMISO data sets into radiation treatment planning. For this, a threshold

separating the background signal from hypoxic tumour sub volumes is needed. Values for such a fixed threshold had been published with magnitudes varying between 1.2 and 1.5 (7, 30, 33). As shown in our results, SUV_{mean} values in the background-VOI have not changed significantly between two and four hour examination, while contrast and noise are changing continuously. Therefore, the definition of a threshold to allow for hypoxic tumour sub volume segmentation will have to be adapted to the individual imaging protocol. In our analysis, this threshold might not have been constant between both examinations.

Limitations

Even though only few centres are investigating FMISO the amount of available literature steadily increases. FMISO is not available everywhere and imaging tumour patients during therapy is as challenging as analysing low contrast data sets. The authors are aware of the organisational challenges appearing in clinical routine imaging related to increasing time slots between injection and data acquisition.

Conclusion

The authors recommend investing the additional two hours to increase image quality and it is assumed that clinical impact will rise accordingly.

> Our data suggest, that for volume analyses of FMISO data sets, for example for radiotherapy planning, 4 h data sets are more suitable than 2 h data sets.

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Conflict of interest

The authors declare, that there is no conflict of interest.

References

1. Brizel DM, Sibley GS, Prosnitz LR et al. Tumor hypoxia adversely affects the prognosis of carcinoma of the head and neck. Int J Radiat Oncol Biol Phys 1997; 38: 285–289.
2. Busk M, Horsman MR, Jakobsen S et al. Can hypoxia-PET map hypoxic cell density heterogeneity accurately in an animal tumor model at a clinically obtainable image contrast? Radiother Oncol 2009; 92: 429–436.
3. Daisne JF, Duprez T, Weynand B et al. Tumor volume in pharyngolaryngeal squamous cell carcinoma: comparison at CT, MR imaging, and FDG PET and validation with surgical specimen. Radiology 2004; 233: 93–100.
4. Dirix P, Vandecaveye V, De Keyzer F et al. Dose painting in radiotherapy for head and neck squamous cell carcinoma: value of repeated functional imaging with ^{18}F-FDG PET, ^{18}F-fluoromisonidazole PET, diffusion-weighted MRI, and dynamic contrast-enhanced MRI. J Nucl Med 2009; 50: 1020–1027.
5. Dubois L, Landuyt W, Cloetens L et al. [^{18}F]EF3 is not superior to [^{18}F]FMISO for PET-based hypoxia evaluation as measured in a rat rhabdomyosarcoma tumour model. Eur J Nucl Med Mol Imaging 2009; 36: 209–218.
6. Eschmann SM, Paulsen F, Bedeshem C et al. Hypoxia-imaging with ^{18}F-Misonidazole and PET: changes of kinetics during radiotherapy of head-and-neck cancer. Radiother Oncol 2007; 83: 406–410.
7. Eschmann SM, Paulsen F, Reimold M et al. Prognostic impact of hypoxia imaging with ^{18}F-misonidazole PET in non-small cell lung cancer and head and neck cancer before radiotherapy. J Nucl Med 2005; 46: 253–260.
8. Grunbaum Z, Freauff SJ, Krohn KA et al. Synthesis and characterization of congeners of misonidazole for imaging hypoxia. J Nucl Med 1987; 28: 68–75.
9. Hentschel M, Appold S, Schreiber A et al. Serial FDG-PET on patients with head and neck cancer: implications for radiation therapy. Int J Radiat Biol 2009; 85: 796–804.
10. Hicks RJ, Rischin D, Fisher R et al. Utility of FMISO PET in advanced head and neck cancer treated with chemoradiation incorporating a hypoxia-targeting chemotherapy agent. Eur J Nucl Med Mol Imaging 2005; 32: 1384–1391.
11. Hockel M, Knoop C, Schlenger K et al. Intratumoral pO_2 predicts survival in advanced cancer of the uterine cervix. Radiother Oncol 1993; 26: 45–50.
12. Hofheinz F , Dittrich S, Pötzsch C et al. Effects of cold sphere walls in PET phantom measurements on the volume reproducing threshold. Phys Med Biol 2010; 55: 1099–1113.
13. Hofheinz F, Poetzsch C, van den Hoff J. Quantitative 3D ROI volume delineation in PET: algorithm and validation. J Nucl Med 2007; 48: 1709.
14. Kotzerke J, Oehme L, Lindner O, Hellwig D. Positron emission tomography 2008 in Germany. Nuklearmedizin 2010; 49: 58–64.
15. Lee NY, Mechalakos JG, Nehmeh S et al. Fluorine-18-labeled fluoromisonidazole positron emission and computed tomography-guided intensity-modulated radiotherapy for head and neck cancer: a feasibility study. Int J Radiat Oncol Biol Phys 2008; 70: 2–13.
16. Lin Z, Mechalakos J, Nehmeh S et al. The influence of changes in tumor hypoxia on dose-painting treatment plans based on ^{18}F-FMISO positron emission tomography. Int J Radiat Oncol Biol Phys 2008; 70: 1219–1228.
17. Linecker A, Kermer C, Sulzbacher I et al. Uptake of ^{18}F-FLT and ^{18}F-FDG in primary head and neck cancer correlates with survival. Nuklearmedizin 2008; 47: 80–85 .
18. Nehmeh SA, Lee NY, Schroder H et al. Reproducibility of intratumor distribution of ^{18}F-fluoromisonidazole in head and neck cancer. Int J Radiat Oncol Biol Phys 2008; 70: 235–242.
19. Nunn A, Linder K, Strauss HW. Nitroimidazoles and imaging hypoxia. Eur J Nucl Med 1995; 22: 265–280.
20. Padhani AR, Krohn KA, Lewis JS, Alber M. Imaging oxygenation of human tumours. Eur Radiol 2007; 17: 861–872.
21. Rajendran JG, Krohn KA. Imaging hypoxia and angiogenesis in tumors. Radiol Clin North Am 2005; 43: 169–187.
22. Rajendran JG, Wilson DC, Conrad EU et al. [^{18}F]FMISO and [^{18}F]FDG PET imaging in soft tissue sarcomas. Eur J Nucl Med Mol Imaging 2003; 30: 695–704.
23. Rasey JS, Koh WJ, Evans ML et al. Quantifying regional hypoxia in human tumors with positron emission tomography of [^{18}F]fluoromisonidazole. Int J Radiat Oncol Biol Phys 1996; 36: 417–428.
24. Rischin D, Hicks RJ, Fisher R et al. Prognostic significance of [^{18}F]-misonidazole positron emission tomography-detected tumor hypoxia in patients with advanced head and neck cancer randomly assigned to chemoradiation with or without tirapazamine. J Clin Oncol 2006; 24: 2098–2104.
25. Schuetze C, Bergmann R, Mosch B et al. The impact of pretreatment [^{18}F]FMISO hypoxic volume on local control after single dose irradiation in FaDu HNSCC in nude mice. Radiother Oncol 2009; 90: 55.
26. Tang G, Wang M, Tang X et al. Fully automated one-pot synthesis of [^{18}F]fluoromisonidazole. Nucl Med Biol 2005; 32: 553–558.
27. Thorwarth D, Eschmann S-M, Scheiderbauer J et al. Kinetic analysis of dynamic ^{18}F-fluoromisonidazole PET correlates with radiation treatment outcome in head-and-neck cancer. BMC Cancer 2005; 5: 152.
28. Troost EG, Laverman P, Philippens ME et al. Correlation of [^{18}F]FMISO autoradiography and pimonidazole immunohistochemistry in human head and neck carcinoma xenografts. Eur J Nucl Med Mol Imaging 2008; 35: 1803–1811.
29. Wang W, Lee NY, Georgi JC et al. Pharmacokinetic analysis of hypoxia ^{18}F-fluoromisonidazole dynamic PET in head and neck cancer. J Nucl Med 2010; 51: 37–45.
30. Wyss MT, Honer M, Schubiger PA, Ametamey SM. NanoPET imaging of [^{18}F]fluoromisonidazole uptake in experimental mouse tumours. Eur J Nucl Med Mol Imaging 2006; 33: 311–318.
31. Yaromina A, Thames HD, Zhou X et al. Radiobiological hypoxia, histological parameters of tumour microenvironment and local tumour control after fractionated irradiation. Radiother Oncol 2010; 96: 116–122.
32. Yaromina A, Zips D, Thames HD et al. Pimonidazole labelling and response to fractionated irradiation of five human squamous cell carcinoma (hSCC) lines in nude mice. Radiother Oncol 2006; 81: 122–129.
33. Zimny M, Gagel B, DiMartino E et al. FDG – a marker of tumour hypoxia? A comparison with [^{18}F]fluoromisonidazole and pO_2-polarography in metastatic head and neck cancer. Eur J Nucl Med Mol Imaging 2006; 33: 1426–1431.