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# Molecular Imaging-Guided Radiotherapy for the Treatment of Head-and-Neck Squamous Cell Carcinoma: Does it Fulfill the Promises?

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**With the routine use of intensity modulated radiation therapy for the treatment of head-and-neck squamous cell carcinoma allowing highly conformed dose distribution, there is an increasing need for refining both the selection and the delineation of gross tumor volumes (GTV). In this framework, molecular imaging with positron emission tomography and magnetic resonance imaging offers the opportunity to improve diagnostic accuracy and to integrate tumor biology mainly related to the assessment of tumor cell density, tumor hypoxia, and tumor proliferation into the treatment planning equation. Such integration, however, requires a deep comprehension of the technical and methodological issues related to image acquisition, reconstruction, and segmentation. Until now, molecular imaging has had a limited value for the selection of nodal GTV, but there are increasing evidences that both FDG positron emission tomography and diffusion-weighted magnetic resonance imaging has a potential value for the delineation of the primary tumor GTV, effecting on dose distribution. With the apprehension of the heterogeneity in tumor biology through molecular imaging, growing evidences have been collected over the years to support the concept of dose escalation/dose redistribution using a planned heterogeneous dose prescription, the so-called "dose painting" approach. Validation trials are ongoing, and in the coming years, one may expect to position the dose painting approach in the armamentarium for the treatment of patients with head-and-neck squamous cell carcinoma.**

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## Introduction

Molecular imaging, also known as biological imaging or functional imaging, is the use of noninvasive imaging techniques that enable the visualization of various biological pathways and physiologic characteristics of tumors or normal tissues. In short, it mainly refers (but not only) to positron emission tomography (PET) and magnetic resonance imaging (MRI). Molecular imaging offers the unique opportunity to allow for earlier diagnosis and staging of the disease, to contribute to the selection and delineation of the optimal target volumes before and during (ie, adaptive treatment) radiotherapy and to a lesser extent before surgery, to monitor the response early on during the treatment or after its completion, and to help in the early detection of recurrence. From the viewpoint of experimental radiation oncology, molecular imaging may bridge radiobiological concepts such as tumor hypoxia, tumor proliferation, tumor stem cell density, and tumor radiosensitivity by integrating tumor biological heterogeneity into the treatment planning equation.

Typically, anatomical imaging modalities such as computed tomography (CT) and MRI, have always been the most widely used modalities for radiotherapy planning of head and neck (H&N) tumors. Over the last few years, however, molecular imaging and in particular PET and multiparametric MRI have become increasingly used. Providing appropriate tracers are

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selected, molecular imaging with PET enables the visualization of various molecular pathways including metabolism, proliferation, oxygen delivery and consumption, and receptor or gene expression, all of which may be important in the response to ionizing radiation.[1] On the other hand, diffusion-weighted MRI (DW-MRI) characterizes tissues by probing differences in the random mobility of water molecules related to tissue cellularity and cellular membrane integrity. Dynamic contrast-enhanced MRI provides biological information linked to tumor vasculature (permeability and flow). Proton MR spectroscopy provides information on the relative concentration of chemical substances, which has been shown to represent the chemical signature of tumor, such as an elevated choline-to-citrate ratio.[2]

In this context, this review will focus on the role of molecular imaging with PET and DW-MRI for planning radiotherapy treatment in H&N squamous cell carcinoma (HNSCC). It will successively review the technical and methodological issues related to image acquisition, reconstruction, and segmentation, the benefit of molecular imaging for target volume selection and delineation with FDG-PET and DW-MRI, and the "dose painting" and dose escalation concept. Although in principle covering the complete H&N area, this review will mainly focus on pharyngolaryngeal SCC for which primary radiotherapy is one of the main treatment modalities.

## Image Acquisition, Reconstruction and Segmentation With PET and MRI

PET and PET/CT have been routinely used as a diagnostic tool to detect and stage lesions for quite some time in oncology.[3] In radiotherapy, there is a growing trend to use PET in treatment planning, either to delineate the target volumes or to further investigate their heterogeneity.[4] These new usages have, however, much stronger requirements for image quality to reach acceptable quality. PET comes indeed with a couple of appealing characteristics, related to its functional nature, as well as intrinsic limitations, such as a rather low spatial resolution and a high level of noise.[5] For several physical and technical reasons, spatial resolution of PET is typically around half a centimetre, whereas (anatomical) CT and MRI do not exceed 1 mm.[3] This explains the blurry aspect of PET images. As PET is an emission modality, the activity of the injected tracer must be limited for obvious radioprotective reasons; this restricts the number of emitted and detected photons and thus leads to rather noisy images.[5]

When target delineation or heterogeneity assessment are aimed at resolution and noise should be carefully optimized when selecting or designing acquisition protocols and reconstruction procedures.[4] For instance, it is recommended to acquire images in 3-dimensional (3D) mode (not in 2D) and to correct for scatter, attenuation, random events and dead time.[3] If available, the use of time-of-flight measurements also increase quality.[5,6] New crystal scintillators and silicon photomultipliers improve both time and space resolution in recent PET systems.[6] Regarding reconstruction, iterative (statistical) algorithms are now the standard option.[3] Depending on hardware specificities, most vendors develop their own adapted reconstruction software. In some PET systems, resolution recovery is a feature that can compensate (partly) for blur.[7] Reconstruction speed is no longer an issue thanks to modern central and graphical processors.[6] In summary, there is a trade-off to attain between the injected tracer dose, the acquisition duration, patient comfort, and image quality.[3]

Depending on reconstruction options, PET images can be further processed afterwards, with denoising or deblurring filters.[8,9] Denoising reduces spurious random oscillations in the image, which can affect contrast. When image quality is essential as it is for automatic segmentation, edge-preserving filters are preferred to usual Gaussian smoothing, which degrades spatial resolution.[4] Image deblurring aims at resolution recovery and partial volume effect correction.[10] It restores sharp edges between regions of low- and high-tracer uptake. To some (limited) extent, deblurring methods can also recover some of the uptake heterogeneities occurring within the tumor. Such methods, however, require an accurate knowledge of the resolution characteristics of the PET camera.

Accurate delineation of the tumor volume and shape from PET images remains an open challenge.[4,11,12] Manual delineation by experienced physicians in nuclear medicine or radiation oncology remains widespread, although such methodology appears highly debatable. Variability in image display and interobserver or intraobserver variability are the most prominent caveats.[4,11,12] On the other hand, highly trained physicians can develop an expertise that can be difficult to translate into an automatic method. Multimodal delineation (PET fused with CT or MR) makes the process even more complicated to describe and formalize.

Variability in manual delineation has motivated the development of automatic delineation methods that are supposedly more objective and reproducible.[4,11,12] The simplest method relies on a fixed uptake threshold to separate tumor and surrounding healthy tissues. It can be expressed as an absolute value, in standardized uptake values (SUVs) for instance, or in a relative way, like the maximal uptake within the tumor ($SUV_{max}$) or some more elaborate statistic ($SUV_{peak}$). Common values are 2.5 SUV or approximately 40% of the $SUV_{max}$.[4] Using 50% of the FDG $SUV_{max}$ to automatically delineate primary tumors of the H&N region led to volumes that were larger than those delineated with CT in 25% of the cases.[13] However, results from this study have to be taken with caution since the use of a single threshold appears questionable. Another study showed indeed that the threshold required to match macroscopic laryngectomy specimens used as a "gold standard" varied from one specimen to another between 36% and 73% of the $SUV_{max}$.[14] Such data and the lack of validation studies illustrate that fixed thresholds achieve poor delineation accuracy.

Adaptive thresholding addresses some of the above limitations. The uptake threshold depends then on both the maximum uptake in the tumor and the average uptake in the surrounding background. This method has been shown to be accurate for segmenting PET images in a series of pharyngolaryngeal tumors.[15] Although validated as a reliable

delineation method, it still suffers from some limitations, like the necessity of calibrating carefully its parameters for each PET system and reconstruction protocol. Other adaptive thresholding methods have been developed. Some of them subtract the background uptake from the $SUV_{max}$ instead of computing the ratio $SUV_{max}/SUV_{background}$. Iterative thresholding proceeds with successive refinements of the threshold, based on direct modelling of the camera resolution.[16] Threshold-based delineation can lose specificity in images with low signal-to-background ratios, as with undifferentiated tumors or peritumoral inflammation induced by radiotherapy.[9]

Many methods based on more complicated image segmentation techniques have been described and applied to PET data.[4,11,12] They involve, for instance, probabilistic thresholds[17] or (fuzzy) clustering techniques,[18] which attempt to address the issues of low resolution and partial volume effect. In this respect, resolution blur has long prevented the application to PET of widespread segmentation techniques that associate object edges with ridges in the magnitude of the uptake gradient. Image restoration tools, like edge-preserving noise filters and deblurring algorithms, partly overcome the problems of blur and noise and make gradient-based segmentation applicable to PET. A method using these tools and segmenting the tumors with a watershed transform and a hierarchical cluster analysis has been successfully validated by comparison with surgical specimens in both HNSCC and NSCLC[9,19] (Fig. 1). This approach has the advantage of accounting explicitly for all imperfections of PET images. Therefore, it does not require any calibration of abstract model parameters. Only the knowledge of the PET image resolution is necessary. Such methods relying on gradient crest detection appear to be more robust than thresholding. The former can distinguish uptake levels that are relatively close, like the tumor and surrounding inflammation, whereas the latter usually assumes that the background uptake is uniformly low.

To date, the difficulty of delineating tumors accurately in PET images with rather low resolution has given rise to many different delineation methods, validated for specific tumor sites and to various extents (with different figures of merit and versus phantoms, synthetic images, other imaging modalities like CT, or ground truth).[11,12,20] Ongoing efforts attempt however to standardize image quality (not specifically for delineation purposes), survey existing or emerging developments in the field, emit recommendations, and provide benchmarking tools.[12,21]

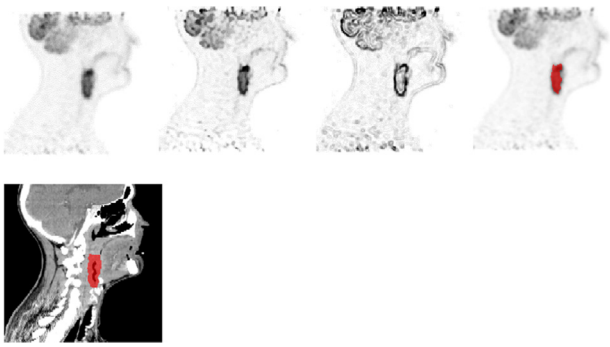

**Figure 1** Example of target delineation with a gradient-based segmentation method. On the top row, from left to right, the raw PET image, the PET image after denoising and deblurring, the raw PET image again with the GTV PET in red; on the bottom row, the CT image with the GTV PET. (Color version of the figure available online.)

A few recent studies have proposed to base automatic tumor delineation on combined information from PET, MRI, and CT.[21-23] The basic idea of such cosegmentation approaches is to compensate for the low-spatial resolution of PET with high-resolution MRI or CT to improve the accuracy of automatic methods. However, segmentation algorithms, which are based on different imaging modalities require advanced algorithmic tools such as machine learning to combine multisource information.[21,23] Especially the availability of combined PET/MRI scanners promotes a cosegmentation using PET and MRI data at the same time.[24,25] Note that the usage of integrated PET/MRI for radiotherapy target volume delineation requires a number of technical considerations. PET quantification acquired with PET/MRI relies on MR-based attenuation correction, which may introduce slight uncertainties.[26] To guarantee a minimum of geometric uncertainties, PET/MRI acquisition for radiotherapy planning purposes is recommended in treatment position. Consequently, additional hardware tools for patient fixation and positioning need to be considered during PET/MRI reconstruction.[27] Furthermore, only the use of dedicated MRI acquisition protocols optimized for geometric fidelity and image contrast should be used for high quality radiotherapy target definition on the basis of anatomic and functional MRI.[28,29]

Diffusion-weighted-MRI detects differences in tissue microenvironment due to random displacement of water molecules. This movement between pairs of opposing magnetic field gradients is detectable as a signal loss proportional to the amount of movement and the strength of the gradient. These differences are quantified as apparent diffusion coefficients (ADC), which are inversely correlated with tissue cellularity. However, DW-MRI is extremely prone to distortion artifacts which is crucial for integration of geometric volume-based data into the radiotherapy planning process.[29] Only a few studies have investigated so far if DW-MRI may be suitable for target volume delineation in clinical practice.[30,31] In those studies, DW-MRI was compared to FDG PET/CT imaging information and no direct correlation between target volumes delineated based on FDG PET compared to ADC measured with DW-MRI was found. A recent study explored overlap volumes as well as voxel-based image information from FDG PET and ADC maps acquired using integrated PET/MRI.[32] Also here, no consistent correlation was found between FDG PET and DW-MRI, confirming similar findings of earlier studies.[24,33]

## FDG-PET and DW-MRI for the Selection of Neck Node Target Volume

In H&N radiotherapy, the first step in the treatment planning chain is the selection of the appropriate target volume in the neck based on all available diagnostic information and the

knowledge of the disease physiopathology, that is, the probability of local and nodal infiltration for a given tumor stage. This relies partly on various imaging modalities, which reveal more or less accurately the true tumor extent.

More than 10 years ago, proof of principle studies conducted in a limited number of patients with HNSCC concluded that FDG-PET/CT could have significantly altered the treatment plan, by changing either the target volume selection or the dose prescription.[34,35] However, these studies were overenthusiastic, as the challenge with using imaging modalities is that none of them has sensitivity or specificity of 100% (no false positive and no false-negative, respectively).

Thus, when incorporating PET or multiparametric MRI into treatment planning, their sensitivity and specificity have to be compared to similar data obtained with CT or anatomical MRI, and confronted to the pathologic ground truth, if it is available. Table 1 summarizes the available data on specificity and sensitivity of FDG-PET, CT, anatomical MRI, and DW-MRI for lymph node staging in H&N cancer in comparison with the surgical lymph node specimen as the gold standard.[36-38] All these modalities led to comparable diagnostic performance for nodal staging, that is, a range of sensitivity and specificity below 90% in all cases. A limited number of small studies indicated that DW-MRI might be superior to conventional imaging for nodal staging of HNSCC, particularly in the detection of subcentrimeter nodal metastases. In a study of surgically treated patients, DW-MRI showed higher sensitivity and specificity to detect nodal metastasis in the neck, leading to 91% accuracy, compared to 83% for 1.5 T turbo-spin echo MRI.[39] None of the MRI protocols could detect lymph node metastases smaller than 4 mm.

A potentially practice-changing use of molecular imaging consists in the staging of clinically node-negative patients where the issue could be to avoid treating the neck nodes if molecular imaging examination also turns out to be negative. The meta-analysis by Liao, which concentrated on clinically neck node negative patients unfortunately did not report a high enough value for FDG-PET sensitivity, thus ruling out the possibility of watchful policy for neck treatment in those patients.[37] In the meta-analysis by Kyzas, when only the cN0 patients were analyzed, the pooled sensitivity of FDG-PET did not exceed 50% (95% CI: 37%-63%), thus confirming the previous conclusion.[36] These data are not surprising, considering that in node negative patients who underwent prophylactic neck node dissection, microscopic nodal infiltration could be observed in up to 30% of cases.[40] Thus, the rather low signal-to-background ratio of FDG and the limited spatial resolution of the images currently preclude the detection of microscopic disease with PET, and therefore, compared to anatomic imaging modalities such as CT and MRI, it is unlikely that FDG-PET will be of any added value in selecting prophylactic nodal target volumes in the neck. Similar conclusions appear true for the use of DW-MRI.

Another way of looking at the data presented in Table 1 is to concentrate on staging specificity of FDG-PET, which could demonstrate nodal infiltration and justify a higher radiation dose prescription, if sufficiently high. Unfortunately, the range of specificity, typically around 85%, cannot justify such practices.

## Target Volume Delineation With FDG-PET and DW-MRI

FDG-PET has been shown to be of value for delineation of the primary tumor gross tumor volume (GTV) by comparing 3D registration of CT, MRI, and FDG-PET images of oropharyngeal, hypopharyngeal, and laryngeal SCCs in several studies (Table 2).[15,41-45] In all these studies, although the methods of defining the FDG-PET GTV were different (ie, manual delineation by nuclear medicine physicians, fixed SUV, automatic user-independent thresholding), the primary tumor GTV defined on the basis of FDG-PET was on average smaller than when defined by contrast-enhanced CT. Such finding was however not observed for delineation of the neck nodes, where no volume change was reported between CT and FDG-PET.[41,42]

**Table 1** Comparison of meta-analyses of the value of MRI, CT, FDG-PET, and DW-MRI for nodal staging in Patients With Head and Neck Squamous Cell Carcinoma

| | Kyzas et al[36] | Liao et al[37] | Wu et al[38] |
|---|---|---|---|
| Number of patients or studies | 1236 | 21 studies | 878 |
| Time frame | 1994-2007 | 1985-2010 | 1990-2011 |
| Sensitivity (%)* | | | |
| MRI | 78 (54-92) | 65 (34-87) | 76 (70-82) |
| CT | 74 (61-83) | 52 (39-65) | 64 (61-68) |
| FDG-PET | 79 (72-85) | 66 (47-88) | 66 (62-68) |
| DW-MRI | n.a. | n.a. | 86 (78-92) |
| Specificity (%)* | | | |
| MRI | 80 (67-88) | 81 (64-91) | 86 (73-93) |
| CT | 76 (68-83) | 93 (87-97) | 75 (63-80) |
| FDG-PET | 86 (83-89) | 87 (77-93) | 81 (77-87) |
| DW-MRI | n.a. | n.a. | 95 (93-97) |

n.a., nonavailable.

*Average value with 95% CI into parentheses.

**Table 2** Comparison Between CT, MRI, and FDG-PET for the Delineation of the Primary Tumor Gross Tumor Volume (GTV) in Pharyngolaryngeal Squamous Cell Carcinoma

| Author | *n* | Site | T-stage | $GTV_{CT}$ (mL) | $GTV_{MRI}$ (mL) | $GTV_{PET}$ (mL) |
|---|---|---|---|---|---|---|
| Daisne et al[15] | 10 | Oro | T2-T4 | 32 (5.1-137.7)* | 27.9 (0–92.8) | 20.3 (5.1-88.9) |
| | 19 | Lar/Hyp | T2-T4 | 21.4 (1.9-55.6) | 21.4 (1.4–58.4) | 13.4 (1.2-34.2) |
| Delouya et al[41] | 25 | Oro, Lar, NPC | T1-T4 | 24 (1-53) | n.a. | 18 (1-48) |
| Chatterjee et al[42] | 20 | Oro | T1-T4 | 36.6 (3.4-184) | n.a. | 25.2 (1.6-166) |
| Caldas-Magalhaes et al[43] | 10 | Lar, Hyp | T3-T4 | 14.9 ± 5.3# | 18.3 ± 10.5 | 9.8 ± 4.1 |
| Ligtenberg et al[45] | 25 | Lar | T3-T4 | 17.5 (5.9-88.7) | 15.5 (4.9-66.3) | 14.5 (6.1-82.7) |

Oro, oropharynx; Lar, larynx; Hyp, hypopharynx; NPC, nasopharyngeal; n.a., nonavailable.
*Range.
#Standard deviation.

Of interest, one study reported that the advantage of using FDG-PET was not observed in small T1 or T2 tumors, likely as a result of the intrinsic limitations of PET, namely, low resolution and partial volume effect, when imaging small volumes.[42] The use of FDG-PET for GTV delineation has also been shown spatio-temporally stable within a few days before the start of radiotherapy; the little difference in GTV delineation observed between the 2 scans obtained few days apart did not have any influence on dose distribution.[46] MRI did not provide any added value to CT, either for GTV delineation or ifor reduced interobserver variability.[47]

But the real question is how these imaging modalities perform in comparison with the real tumor volume assessed on a pathologic specimen. In two of the studies presented in Table 2, for a subset of laryngeal or hypopharyngeal tumors scheduled for total laryngectomy, the imaging modalities were also registered with the actual surgical specimen taken as a "gold standard."[15,43] In both studies, FDG-PET demonstrated higher accuracy in delineating the primary tumor GTV with a statistically significant reduction in the target volumes compared to CT or MRI. But it should be stressed, however, that in both studies, CT, MRI, and FDG-PET failed to visualize the superficial tumor extent, illustrating their known limitation in spatial resolution.

Now that FDG-PET has been shown to be an optimal imaging modality for the delineation of the primary tumor GTV, at least for locally advanced SCC, the next questions are, first, whether its use will effect on delineation of the clinical target volume (CTV) and of the planning target volume (PTV) and, second whether a more conformed dose distribution can be achieved in treatment planning. Regarding the target volume issue, interestingly, the differences observed between CT and FDG-PET for the GTV delineation translated into significant differences in CTV and PTV delineation.[48] A similar finding was also found in another group of patients treated for pharyngolaryngeal tumors.[45] Regarding the dose distribution, when comparative 3D conformal radiotherapy plans were made, FDG-PET-based plans were more conformal than the CT-based plans, and reduction in the isodose volumes with subsequent reduction in the dose to the surrounding normal tissues were observed with FDG-PET-based plans.[48]

Such promising findings were validated in a prospective multicentric phase II study that enrolled 40 patients with locally advanced oropharyngeal, laryngeal and hypopharyngeal SCC.[49] First, as anticipated, the use of FDG-PET for delineation of the primary tumor GTV translated into smaller CTV and PTV; second, the dose planning translated into more conformal dose distributions with improved parotid sparing in the FDG-PET-based plans compared to CT-based plans. In that study, tumor recurrence was systematically observed within the FDG-PET GTV and could not be explained by geographical miss, thus ruling out the idea that a conventional CT-based plan would have prevented recurrence.

Next to PET, the use of DW-MRI for target volume delineation has only been investigated in a few studies. Primary tumor GTV delineated on DW-MRI were significantly smaller than the GTV delineated on contrast-enhanced CT, and less interobserver variability was observed.[50,51] In one of these studies, over a median follow-up of 30.7 months, 7 patients had recurrent disease; recurrence was always located within the area of overlap between the $GTV_{CT}$, $GTV_{DW\text{-}MRI}$, and $GTV_{FDG\text{-}PET}$.[50] Only a few studies have investigated so far if DW-MRI may be suitable for target volume delineation in clinical practice.[30,31] In those studies, DW-MRI was compared to FDG PET/CT imaging information and no direct correlation between target volumes delineated based on FDG PET compared to ADC measured with DW-MRI was found. A recent study explored overlap volumes as well as voxel-based image information from FDG PET and ADC maps acquired using integrated PET/MRI.[32] Also here, no consistent correlation was found between FDG PET and DW-MRI, confirming similar findings of earlier studies.[24,33] For lymph nodes in the neck, a planning study showed a better correlation between the nodal GTV and the pathology findings, when contouring was performed based on DW-MRI findings in comparison with contouring based on CT or conventional MRI findings.[52] Although such data are promising, further research and development is needed before DW-MRI could be routinely used for contouring patients with HNSCC.

In radiotherapy for H&N tumors, anatomic variations affecting normal tissues and target volumes have been reported. Such variations may also impact on dose distribution, thus calling for adaptive radiotherapy, not only as a way to better conform the dose to the organs at risk and target volumes, but also as an option to escalate the prescribed dose to maximize tumor control.[53] Very few studies have systematically studied the evolution of the GTV during treatment with molecular imaging. In a study of 10 patients with locally advanced pharyngolaryngeal SCC, Geets et al systematically performed contrast-enhanced CT, MRI, and FDG-PET on a

weekly basis during the first 5 weeks of concomitant chemoradiotherapy. Provided a sufficiently specific method is used to automatically contour the FDG-PET GTV, progressive shrinkage of the GTV was observed, translating into progressive reduction of the CTV, PTV, and high-isodose volumes.[54,9] Similar studies of patients with locally advanced H&N tumors failed to report similar trends, but all these studies relied on suboptimal automatic segmentation methods of the PET images, which could not discriminate between residual tumor FDG activity and peritumoral inflammation induced by radiotherapy.[55,56]

Data on the systematic use of DW-MRI during treatment are even scarcer. A prospective study on 10 patients with locally advanced oropharyngeal SCC indicated that the ADC value in the neck nodes progressively increased and then stabilized during treatment, but how this could be used to adapt the radiotherapy treatment is not known yet.[57] In a study including 8 patients, an increase in ADC value during treatment was also observed, and such variation already appeared at fraction #11.[58] Further studies are, however, needed before routine use of FDG-PET or DW-MRI becomes recommended during treatment to help reduce the target volumes.

## Dose Escalation and the Concept of "Dose Painting"

Dose escalation in H&N tumors has been investigated in many different ways, assuming that it could lead to increased local tumor control and, consequently, improved overall survival. It was first tested many years ago in the context of accelerated or hyperfractionated radiotherapy, which by increasing the biological effective dose, has been shown to significantly increase the locoregional control rate with a small improvement in overall survival.[59] With the refinement in target volumes delineation and the improvement in the conformality of dose delivery, homogeneous radiation dose escalation phase I-II studies have been reported.[60,61] The increase in physical dose was, however, limited by the dose delivered to the surrounding normal tissues, which could be associated with increased late toxicity.

These data raised the following question: could refined radiation dose escalation strategies be more likely to provide some therapeutic gain? Imaging-based dose painting, namely, the prescription and delivery of a nonuniform dose to the GTV is a different paradigm for prescribing radiation therapy.[62,63] The basic idea is to replace, completely or in part, the morphologically or anatomically defined target volumes with the spatial distribution of a specific tumor phenotype, which is acquired through molecular imaging and related to local tumor control after radiotherapy (or at least hypothesized to be so). A dose prescription function is then used to convert this distribution into a map of prescribed doses that can be fed into an inverse planning optimizer.

Two prototypical strategies have been considered in the literature. On one hand, subvolume boosting, also known as dose painting by volume, involves some discrete volume that is defined in the image and given an additional "boost" of radiation dose. On the other hand, dose painting by numbers defines the dose prescription at the voxel level.[64] In the latter case, the prescription function maps a range of image intensities onto a range of doses. Hybrids between these 2 strategies use a series of nested volumes, often about 5, with a prescribed dose assigned to each of them.

The dose painting paradigm is supported by several clinical or biological hypotheses: (1) local recurrence arises from cellular or microenvironmental niches that are (relatively) resistant at the radiation dose level that can safely be routinely delivered using a uniform dose distribution[65]; (2) molecular imaging will allow spatio-temporal mapping of these regions of relative radioresistance[66-68]; and (3) advances in radiation therapy planning and delivery technologies facilitates delivery of a graded boost dose to such regions, which in turn should lead to improved local tumor control without increasing morbidity.[69,70] Support for the dose-painting hypothesis comes in part from mathematical modeling studies. It has been shown that for a fixed integral dose to a tumor with a uniform spatial radiosensitivity distribution, delivering a uniform dose of radiation will maximize the tumor control probability; but for a nonuniform radiosensitivity distribution, a uniform dose distribution is inferior to a distribution that delivers a relatively higher proportion of the integral dose to the more resistant regions of the tumor, that is, by dose painting.[71] However, a recent preclinical study testing this hypothesis based on FDG uptake in a rat rhabdomyosarcoma model failed to demonstrate the benefit of a dose redistribution approach and argued that dose intensity could not go below a standard dose level.[72] Per se, this observation does not kill the concept of dose redistribution, but simply exemplifies the need of an appropriate marker of radioresistance, which FDG may not be.

The current interest in dose painting focuses mainly on 3 evidence-based causes of radiation therapy failure in the clinic: tumor burden (ie, tumor cell density), tumor cell proliferation, and tumor hypoxia.

Regarding tumor burden, FDG uptake is commonly considered as a good surrogate for tumor cell density, although various parameters influence its accumulation, such as the rate of glycolysis and tumor perfusion, proliferation, inflammation, and hypoxia.[73] As already discussed earlier, FDG uptake has been shown to correlate spatially with tumor extent as assessed on pathology specimens. There are few "proof-of-concept" planning studies that have demonstrated the feasibility of selective dose escalation based on FDG distribution.[54,70,74-77] All these studies demonstrated the possibility to deliver more radiation dose (typically by 15%-20%) to the GTV without increasing the dose delivered to organs at risks like the parotid glands, the spinal cord, and, to some extent, the constrictor muscles. The use of DW-MRI is an appealing alternative imaging modality to define targets for dose painting, but no clinical study has been reported yet.

Only 1 dose painting phase-I study has been reported in patients with locally advanced H&N SCC. In a trial enrolling 21 patients, it was shown that an increase in dose per fraction (to 2.5 and 3 Gy per fraction) up to a median dose of 80.9 and 85.9 Gy, respectively, could be safely delivered to the FDG-PET-based GTV during part of the treatment.[78] However, in

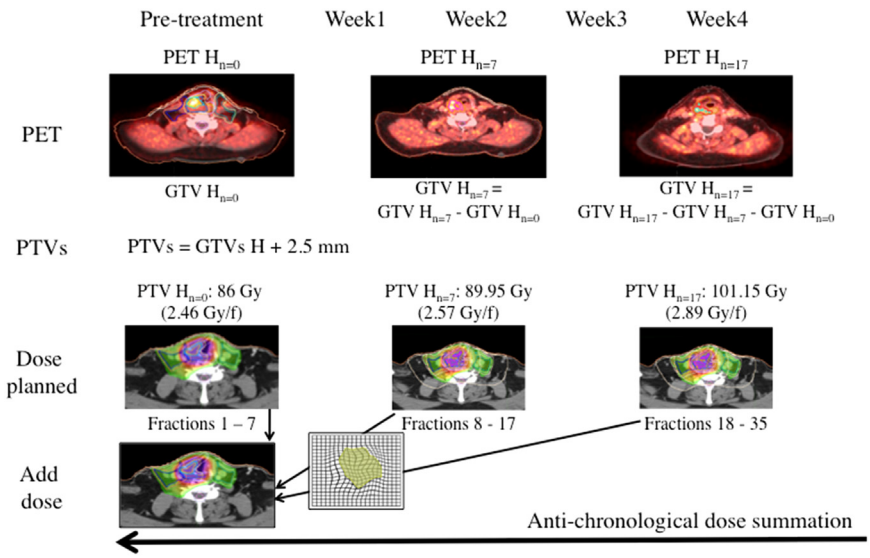


**Figure 2** Design of the adaptive dose escalation procedure. For each planning phase, separate image sets (ie, PET CT $H_0$ [before start of RT], PET CT $H_7$ [at fraction #7] and PET CT $H_{17}$ [at fraction #17]) were acquired. Using deformable image coregistration, regions of interest were deformed from one PET-CT to the next and manually adjusted when needed. For each phase, a new treatment plan was made only on the newborn voxel of that phase (ie, at the seventh and seventeenth fraction). The doses were summed antichronologically on the pretreatment PET-CT. (Adapted with permission from Ref 88.)

that study, late mucosal ulcers were observed in the highest radiation dose group, and thus the maximal tolerated median dose of 80.9 Gy was defined.[79] According to the NCI ClinicalTrials.gov website, at least 2 other phase I trials are ongoing or have been completed, but no data are yet available.

Regarding tumor cell proliferation, the use of the radiofluorinated thymidine analogue 3′-deoxy-3′-$^{18}$F-fluorothymidine (FLT) has been investigated. It is a terminator of the growing DNA chain, which is therefore only incorporated into DNA during synthesis to a very limited extent, but it is retained in cells after phosphorylation by the thymidine kinase 1 enzyme.[80] FLT has been shown to be an imaging surrogate for tumor cell growth, with enough sensitivity to detect various growth inhibition response.[81] FLT-PET scans at baseline and 2 weeks into fractionated radiotherapy for patients with HNSCC have been used to define targets for sub-volume boosting in a recent radiation therapy planning study.[82] However, in the absence of direct clinical evidence for an association between these regions and a subsequent local treatment failure, the biological rationale for this boost strategy is not completely clear, and further data are thus needed before using dose painting based on pretreatment FLT distribution. Dose boosting based on residual FLT uptake is another possibility, but again further data are needed before recommending on such strategy.

Tumor hypoxia has been observed in a wide variety of solid human tumors and has been shown to be a strong factor of radio-resistance and tumor failure after radiotherapy.[83] Various noninvasive indirect methods have been developed to detect tumor hypoxia, and among them, the use of positron-labeled tracers has been used in conjunction with PET systems. Typically, these tracers detect the presence of hypoxia when $pO_2$ drops below 10 mmHg. Among the various PET tracers that can be easily synthesized, $^{18}$F-misonidazole (MISO) is the most commonly used, but more recently, other PET tracers such as $^{18}$F-FAZA, $^{18}$F-FETNIM, $^{18}$F-EF3, $^{18}$F-EF5, and $^{18}$F-HX4 have also been introduced in the clinic.[84] $^{18}$F-FAZA has several advantages, including an easy production with high specific activity, chemical stability after injection, a specific metabolism in hypoxic cells, and rapid clearance of unbound tracer from non-hypoxic tissues, leading to higher tumor-to-background ratios compared to other tracers.[85] Using FAZA, tumor hypoxia has been identified in 0%-51% of HNSCC cases.[86,87] In a feasibility dose planning study in patients with locally advanced SCC, it has been shown that doses up to 86 Gy could be delivered to hypoxic voxels, without significantly increasing the dose delivered to the surrounding normal tissues.[88] However, the magnitude of the required dose to control disease in PET hypoxic regions is not clear. Simplistic back-of-an-envelope estimates based on in vitro oxygen-enhancement ratios are likely to be gross overestimates of the dose required to increase cell kill in human tumors. In proof-of-concept planning studies using F-MISO to redistribute the radiation dose to the hypoxic subvolumes, it has been calculated that a dose increase to tumor hypoxic areas by 15%-20% could substantially increase the control probability without affecting normal tissue toxicity.[89-92] Tumor hypoxia varies both in intensity and location throughout the course of treatment, thus calling for hypoxia-directed adaptive radiotherapy (Fig. 2).[87] Based on the dynamic of hypoxia during radiotherapy, it has also been suggested that a dose escalation protocol using assessment early on during treatment might be optimal.[93] Along this line, it has been demonstrated that in HPV-positive patients with oropharyngeal SCC, a reduction of the prescribed radiation dose of 10 Gy to the PTV associated

**Table 3** Summary of the ongoing phase-III trials in radiotherapy "dose painting" for Head and Neck squamous cell carcinoma.

| Acronym/investigator (NCT #) | Tumor location | HPV status | Tumor stage | Molecular imaging | Phase | Study design | | Completion date |
|---|---|---|---|---|---|---|---|---|
| | | | | | | Standard arm | Experimental arm | |
| Xuzhou Medical College, China (NCT# 02089204) | NPC | Not relevant | III-IVa | FDG-PET FMISO-PET | III | IMRT + cddp + docetaxel | (1) IMRT + cddp + docetaxel + boost dose on FDG<br>(2) IMRT + cddp + docetaxel + boost dose on FMISO | December 2015? |
| De Neve (NCT# 01341535) | Oro, Hyp, Cav, Lar | HPV− | III-IV | FDG-PET | rand. II | 69.12/56 Gy in 6.5w + weekly cddp | 84/40 Gy in 6w + weekly cddp | Q1 2018 |
| Eisbruch (NCT# 02031250) | Oro, Hyp, Lar, Cav, NPC | HPV−HPV+ high risk | III-IV | DCE-MRI | rand. II | 70 Gy in 7w + cddp/carbo | 80 Gy in 7w+ cddp/carbo | December 2020 |
| INTELHOPE (NCT# 0275722) | Oro, Hyp, Lar | n.a. | III-IV | FDG-PET | rand. II | 66/54 Gy in 6w + concomitant cddp | 73.5/63/54 Gy in 6 weeks + cddp | December 2020 |
| Zips (NCT# 02352792) | Oro, Hyp, Cav, Lar | n.a. | III-IV | FMISO-PET | Rand. II | 70 Gy in 7w + 5Fu + mitomycin C or cddp | 77 Gy in 7w + 5Fu + mitomycin C or cddp | December 2022 |
| ESCALOX (NCT # 01212354) | Oro, Hyp, Cav | n.a. | n.a. | FMISO-PET | III | 70/56 Gy in 7w (SIB-IMRT) + concomitant cddp | 80.5/70/56 Gy in 7w (SIB-IMRT) + concomitant cddp | January 2025 |

DCE, dynamic contrast-enhanced; Oro, oropharynx; Hyp, hypopharynx; Cav, oral cavity; Lar, larynx; NPC, nasopharynx; CH, chemotherapy; n.a., non available; rand., randomized.

with the lymph nodes in those patients showing a resolution of hypoxia after 1 week of treatment was a safe approach.[94]

Supported by the promising data presented above, and according to the ClinicalTrials.gov website, 6 randomized phase II or phase III studies are ongoing to validate the concept of dose painting using FDG-PET, F-MISO-PET or dynamic contrast-enhanced-MRI (Table 3). Although 1 study has supposedly closed its accrual, no data has been published or reported yet. Another study in which the dose will be increased based on FDG-PET distribution is about to start in Dresden (Germany). Preliminary data have been recently released from the study of Tubigen.[95] Patients with baseline hypoxic tumor had a lower locoregional control probability, which was somehow overcome with the delivery of a higher radiation dose.

What should be emphasized is that all the data collected so far were obtained from patients with HPV-negative SCC, and that a different treatment paradigm will likely have to be implemented for patient with HPV-positive tumors, at least in the oropharynx.

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