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Utility of functional imaging in prediction or assessment of treatment response and prognosis following thermotherapy

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Abstract

The purpose of this review is to examine the roles that functional imaging may play in prediction of treatment response and determination of overall prognosis in patients who are enrolled in thermotherapy trials, either in combination with radiotherapy, chemotherapy or both. Most of the historical work that has been done in this field has focused on MRI/MRS methods, so the emphasis will be there, although some discussion of the role that PET might play will also be examined. New optical technologies also hold promise for obtaining low cost, yet valuable physiologic data from optically accessible sites. The review is organized by traditional outcome parameters: local response, local control and progression free or overall survival. Included in the review is a discussion of future directions for this type of translational work.

Introduction

In this era of personalized medicine, much effort is being directed to the development of methods that can assist in the selection of the best therapeutic options for individual cancer patients. The drive toward individualization of treatment has led to the publication of literally thousands of papers on this subject and the generation of journals that are specifically dedicated toward this goal. Examples include: The Open Biomarkers Journal - <http://www.bentham.org/open/tobiomj/>, Biomarker Insights - <http://www.lapress.com/biomarker-insights-journal-j4>), and Disease Markers - <http://www.iospress.nl/loadtop/load.php?isbn=02780240>.

One of the most intensely investigated methods for individualizing cancer treatment has been in the characterization of genomic markers of biologic diversity. A myriad of host and tumor specific variations in gene expression occurs in cancer, which provides a rationale for believing that individualization of treatment is possible (1,2). Indeed, genomic profiling in particular holds great promise for prediction of chemotherapy response in numerous tumor types, including breast cancer (3,4), prostate cancer (5) and non-small cell lung cancer (6,7). In the field of hyperthermia, there has been limited activity in this area with only one study being reported thus far, wherein a genomic profile was derived for prediction of persistence of positive lymph nodes following neoadjuvant thermochemotherapy in locally advanced breast cancer (8).

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In spite of the enthusiasm for genomic profiling or other types of molecular and cellular biomarkers, one cannot ignore the physiologic microenvironment in tumors, particularly when considering use of hyperthermia. With therapeutic hyperthermia, perhaps more than any other cancer treatment modality, the physiologic microenvironment can affect treatment delivery, can influence treatment outcome, and can be altered by the treatment both positively and negatively. For example, tumor perfusion will be inversely related to the ability to heat a tumor using external methods, heat-induced vasculopathy may lead to upregulation of oxygen-sensitive proteins that could increase the biologic aggressiveness of the tumor, hyperthermia might increase tumor oxygenation leading to enhanced radiosensitivity, and acute changes in pH could increase the chance of thermal cytotoxicity.

In this review, we focus on examining how functional imaging parameters that reflect the tumor microenvironment can be used to predict treatment outcome and overall survival in thermotherapy trials. Endpoints that have been assessed using functional imaging include tumor response, duration of local tumor control and progression free and overall survival.

Overview of molecular imaging methods

Magnetic Resonance Imaging (MRI), Dynamic Contrast Enhanced MRI (DCE-MRI) and Magnetic Resonance Spectroscopy (MRS)

MRI is based on the principle of nuclear magnetic resonance, where nuclei in an external magnetic field selectively absorb and then release energy unique to those nuclei. The released energy can be used to create an anatomic image that is characterized by outstanding contrast resolution. The outstanding contrast resolution and lack of a need for ionizing radiation have led to MRI being widely used for tumor detection and staging and for assessing response to treatment. Though the signals used to create a magnetic resonance image can be affected by the microenvironment, MRI is essentially an anatomic imaging modality. However, modifications of basic MRI techniques can be used to gain more functional information.

In DCE-MRI, the kinetics of intratumoral water soluble contrast medium concentration, i.e. signal, as a function of time after intravenous injection, allow physiologic parameters related to tissue perfusion/permeability to be evaluated. Methods of evaluation include visual inspection of data in movie format, inspection of graphs of signal intensity vs. time, empirical institution dependent measurements and pharmacokinetic modeling using multi-compartment analysis (9). There are advantages and disadvantages to each of these analytical methods, but overall the lack of a universally-accepted analytical method has likely impeded understanding the full potential of this noninvasive imaging method for staging patients and predicting outcome (10). Nevertheless, the noninvasive nature of DCE-MRI and the feasibility of follow-up studies are attributes that make DCE-MRI attractive for assessing tumor perfusion in clinical trials.

MRS differs from MRI in that in the latter frequency is used for spatial localization, while in the former frequency is used for both for spatial localization and chemical identification (11). Thus, in MRS, specific chemicals can be detected based on their frequency signature. Protons are usually used for spectroscopy due to their abundance and high magnetic sensitivity, though special techniques allow other nuclei, such as phosphorus or fluorine, to be evaluated as well. Due to the pH sensitivity of the frequency signature of certain molecules, MRS can also be used for noninvasive measurement of intracellular pH (12). Noninvasive assessment of chemical spectra can provide unique information regarding the evaluation of tumor behavior and possibly response to treatment.

Positron Emission Tomography (PET)

PET imaging can provide qualitative and quantitative metabolic information using molecular probes labeled with positron-emitting radionuclides, such as ^{18}F , ^{11}C , ^{15}O , $^{60,62,64}\text{Cu}$, ^{124}I . A positron emitted by a radiotracer travels only a small distance before it encounters an electron, at which point an annihilation reaction occurs where matter is converted to energy, and two 511 keV photons are emitted in opposite directions. The PET scanner detects both these photons using a cylindrical array of detectors. The fact that the two photons are produced simultaneously and travel at 180° to one another is used to further localize each event. Most PET scans are now performed on hybrid PET/computed tomography (CT) scanners. This allows the anatomic information from CT and metabolic information from PET to be obtained in a single imaging session. The CT scan is also used to provide attenuation-correction of the PET images, enabling activity within tumors to be accurately measured. Using PET, radiotracer uptake within tumors is quantified routinely by measuring standardized uptake value (SUV), which normalizes the radiotracer activity to body weight and administered radiopharmaceutical dose.

At present, the most commonly used PET radiotracer is 2-deoxy-2- ^{18}F fluoro-D-glucose (FDG). FDG is a glucose analog that is actively transported into cells where it is trapped and not further metabolized. Malignant cells use more glucose than normal cells (Warburg effect), and often have higher glycolytic metabolism related to tumor hypoxia. In general, aggressive malignancies exhibit higher accumulation of FDG compared to lower-grade tumors, and the degree of FDG activity may provide prognostic information. Malignant tumors responding to chemotherapy are typically characterized by a measurable reduction in FDG uptake on PET scans before becoming measurably smaller on CT or MRI scans, and therefore may be predictive of early response to therapy (13).

While FDG is currently the dominant radiotracer used in PET imaging, other tracers are being developed which may be useful in determining response to therapy but are not yet available for routine clinical use; these are summarized in a recent review by Larson (14). Examples include ^{18}F -fluorothymidine (FLT) as a marker for cell proliferation and hormonal markers such as ^{18}F -fluoroestradiol (FES). Several PET tracers are being studied as surrogate markers for hypoxia. These include 2-nitroimidazole agents such as ^{18}F -fluoromisonidazole (FMISO), ^{18}F -fluoroetanidazole (FETA), ^{18}F -fluoroazomycin-arabinofuranoside (FAZA), and ^{18}F -fluoroalkyl acetamide derivatives (EF1, EF3, and EF5) (15,16). These agents diffuse freely throughout the body after injection so that delivery to target areas is independent of blood flow. Under conditions of hypoxia, the tracer is reduced and irreversibly trapped intracellularly. Copper-labeled dithiosemicarbazones are also reduced and retained in hypoxic tissues. Cu-ATSM (diacetyl-bis(N^4 -methylthiosemicarbazone)) can be labeled with several positron-emitting copper isotopes, including ^{60}Cu ($t_{1/2} = 23$ min), ^{62}Cu ($t_{1/2} = 9.7$ min), and ^{64}Cu ($t_{1/2} = 12.7$ hr). Cu-ATSM accumulates more rapidly in hypoxic tissues compared to FMISO, but is more sensitive to blood flow effects and tumor type (15) (17). In the future, PET imaging with hypoxia markers may be useful for characterizing tumors, and for evaluating response to chemoradiation and thermal therapy.

Optical Imaging

Optical spectroscopy involves the interrogation of tissue optical properties to gain insight into a variety of functional and structural features. There are a wide range of potential sources of optical contrast in tissue, but the properties focused on in this review involve the absorption, scattering, and fluorescence properties of tissue. Such measurements have a variety of advantages, including safety (non-ionizing), low-cost, quantitative, non-destructive, and wide sensitivity to a range of functional parameters. Optical measurements are commonly made using a fiber optic probe placed in contact with the tissue of interest, or using a non-contact imaging system (18). Light enters the tissue and undergoes absorption, scattering, and or

fluorescence. Exiting photons can then be collected and recorded with an optical detector. The penetration depth of light ranges from a few hundred microns in the UV, up to several cm in the near infrared, making this technique applicable to superficial tumors as well as sites such as the intact breast using NIR wavelengths (19). Measurement via a biopsy needle or endoscope enable measurement of more deeply seated tumors (18).

Two types of measurements possible using this type of setup will be discussed: diffuse reflectance and fluorescence spectroscopy. Diffuse reflectance involves illuminating the tissue and recording the backscattered light as a function of wavelength. This is dependent on the wavelength-dependent absorption and scattering properties of the tissue. Fluorescence spectra are recorded by illuminating the tissue with a given wavelength of light, and recording the intensity emitted at a longer wavelength. This relies on tissue fluorophores to absorb the incident photon and emit a fluorescent photon at a longer wavelength. This measurement is sensitive to the absorption, scattering, and fluorescence properties of the tissue. Tissue is highly scattering, with scattering tending to decrease at longer wavelengths, and emitted photons are typically multiply scattered. Absorption in tissue in the visible wavelength range is dominated by hemoglobin, and tends to increase towards the ultraviolet wavelength range. This turbidity makes determination of the underlying optical properties non-trivial due to the complex interactions of light with tissue, but a variety of quantitative models of light-tissue interaction have been developed that are capable of extracting the absorption, scattering, and fluorescence properties from measured tissue spectra (20).

The functional parameters to which optical spectroscopy is sensitive include: 1) total hemoglobin content of tissue which is related to the vascular fraction of the tissue, 2) the hemoglobin oxygen saturation, which is related to vascular oxygenation, 3) scattering properties, which are affected by tissue morphology including cellular and extracellular matrix density, and 4) intrinsic sources of fluorescence, including the electron carriers NADH and FAD (18). This enables optical spectroscopy to be intrinsically sensitive to a wide range of functional and morphological properties, many of which are known to be affected by thermotherapy.

The turbidity of tissue to optical radiation serves to limit the effective penetration depth and resolution at which tissue can be characterized at depth. The effective penetration depth ranges from several hundred microns or less in the UV, up to several cm in the near infrared (18). This limits the types of measurements that can be made in some clinical settings, where the site of interest is not superficial. Several solutions to this problem have been used, including limiting the measurement to NIR wavelengths, where the penetration depth is greater but fewer sources of contrast are available, or using endoscopic or needle-based measurement systems to access deep tissue (18).

Prediction and Assessment of Tumor Response

The change in ATP content within tumors has been used to predict response rates. Ohtsubo described a linear relationship between decreases in tumor cell ATP content and decreases in cell survival when cells were heated in vitro using a range of temperatures (21). This provides a rationale for considering change in tumor ATP content as a surrogate for cell viability. Glickson (13,14) and Vaupel (22) used 31-P MRS to measure changes in 31-P metabolites in response to hyperthermia. Vaupel used graded thermal doses by changing the time of heating at 43.5C and found that inorganic phosphate (Pi) increased with heating time, whereas all ATP peaks declined (22).

Sijens et al., found that the ATP/Pi ratio in a murine mammary carcinoma increased slightly 18hr following 42C heating for 15 min (23). For higher temperatures, however, there was a temperature-dependent decline in the ATP/Pi ratio. At 45C the ratio decreased by more than

a factor of 10 (Figure 1A). In a thermoradiotherapy trial in spontaneous canine soft tissue sarcomas, 31-P MRS was performed prior to and 24h after the first hyperthermia treatment (24). When ATP/Pi was plotted as a function of the median temperature achieved during hyperthermia, the temperature dependence of the ATP/Pi ratio was strikingly similar to that found in the murine mammary adenocarcinoma (Figure 1B). In a parallel trial in human soft tissue sarcomas in which thermoradiotherapy was administered prior to surgical resection, a decrease in adenosine triphosphate/phosphomonoester (ATP/PME) after the first hyperthermia treatment was associated with more necrosis in resected tumors (24) (Figure 1C). Thus, in all of these models, the decrease in ATP observed at higher temperatures was consistent with increased thermal cytotoxicity and this quantification of ATP using MRS may have value as a measure of thermal treatment efficacy.

DCE-MRI has been used to both predict and assess effects of thermal therapy on tumor response. In human soft tissue sarcomas treated neoadjuvantly with thermochemotherapy, there was an association between loss of contrast medium uptake and extent of necrosis (25). Craciunescu et al., developed a semi-quantitative scoring scheme to describe the pattern and kinetics of contrast enhancement prior to the onset of treatment in 20 patients with locally advanced breast cancer (26). The scoring system has three components: the first is related to morphology and comes from whether the shape of the enhancement pattern as described by the MR parametric maps is centrifugal or centripetal. The second and third components are physiologic, one related to tumor vascularity/permeability and the other to tumor extracellular extravascular space, as defined by the washin and washout parameters (Figure 2). Tumors with a higher score were more likely to respond clinically compared to tumors with a lower score. Low scores were in tumors that likely had relatively poor perfusion. Although based on a very low number of patients, the specificity and sensitivity were 78 and 91%, respectively ($p=0.002$).

Assessing the effect of hyperthermia on tumor metabolism through PET imaging of glucose analogues is a powerful way to evaluate efficacy. Progressive loss of uptake of fluoro-deoxyglucose (FDG) as assessed with PET imaging has been found to be related to the amount of necrosis found in excised high grade soft tissue sarcomas that had been treated preoperatively with thermoradiotherapy (27,28). Westerterp et al., performed serial FDG PET studies in 17 patients with esophageal cancer who were treated preoperatively with paclitaxel, cisplatin, radiotherapy and hyperthermia (29). Patients with greater decreases in FDG uptake after two weeks of treatment had more necrosis in the resected tumor specimen. The positive and negative predictive values were both 75% in this limited series (Figure 3). In an additional report, reduction in FDG PET uptake was a good indicator of pathologic response in 20 rectal cancer patients treated with radiotherapy, 5FU and hyperthermia (30). In this study, the second PET imaging study was done after therapy was completed, so it was not tested as a predictor of response.

In summary, there are several methods that appear to reflect the degree of direct cytotoxicity elicited from thermotherapy trials that integrate radiation and/or chemotherapy. Early changes in these parameters may allow accurate prediction of the response at the end of therapy, or at the time of surgical resection. In one study, a single measurement of pretreatment tumor perfusion characteristics was predictive of response (26). In most other studies, a pretreatment determination was compared to a second observation at some point early in the treatment course. Though multiple imaging sessions is a strength of noninvasive tumor imaging, the need for two measurements can occasionally be problematic, as discussed later.

Prediction of Local Tumor Control

Local tumor control is difficult to quantify because of the issue of competing risks. In assessing local control as the primary endpoint, there is also usually a risk for development of metastases

or death, prior to local failure (31). These events occurring prior to local failure, the competing risks, can be accounted for by censoring in the statistical analysis, but censoring can lead to inaccurate and misleading estimates of the time to local failure (31). Nevertheless, there are parameters that have allowed prediction of local tumor control, particularly when assessing treatment regimens that include radiation therapy. A prime example is the degree of tumor hypoxia. Tumor hypoxia has been measured most often using invasive oxygen electrodes, but recently non-invasive measurement of tumor hypoxia using PET imaging has been investigated and, similar to invasive measurements, results from PET imaging suggest that hypoxic tumors are more likely to undergo local tumor recurrence after radiotherapy (32,33). However, using the same PET imaging approach, others have not found an association between hypoxia and local tumor control following chemoradiation therapy (34). In soft tissue sarcomas, tumor hypoxia assessed using invasive oxygen electrodes was predictive of metastasis free survival but not local tumor control following pre-operative thermoradiotherapy and surgical resection (35). Thus far, PET imaging of hypoxia has not been assessed in a thermoradiotherapy trial where local control was the endpoint.

In most studies where invasive measures of tumor hypoxia have been used, only pretreatment measurements were made, or a small number of measurements were made early in the treatment course. In a study of tumor oxygenation in canine sarcomas where measurements were made throughout a course of thermoradiotherapy, it was apparent that fluctuations in oxygen throughout treatment were more pronounced than expected (36). This emphasizes the value of noninvasive measurement of tumor oxygenation at multiple times in thermoradiotherapy trials so that a more complete understanding of the kinetics of this important microenvironmental factor can be achieved.

MRS has also been used to assess local tumor control in thermoradiotherapy trials. In 29 canine soft tissue sarcomas, the ratio of phosphodiester to phosphocreatine (PDE/PCr) was associated with probability of local tumor control following thermoradiotherapy (37). The biological significance of the PDE/PCr ratio is difficult to interpret and this observation may have been spurious since a relatively large number of potentially predictive factors were assessed. In this same canine study, however, there was a positive association between the area under the time-intensity contrast enhancement curve and local tumor control. This suggests that the probability of local control was better in more highly perfused tumors. This is similar to that observed in the locally advanced breast cancer study in humans, where more poorly perfused tumors were less likely to respond to thermochemotherapy (26).

Progression Free and Overall Survival

In several relatively small clinical trials, tumor lactate was quantified using bioluminescence imaging of tumor biopsies and high lactate levels were associated with decreased metastasis free survival and decreased overall survival. These trials included patients with head and neck (38), cervix (39) and colorectal cancer (40). There is biologic rationale for this observation, as the primary lactate transporters are chaperoned to the cell surface by EMMPRIN (CD147), a protein that is linked to breakdown of extracellular matrix and invasion (41). MCT-1 and EMMPRIN expression levels and trafficking to the cell membrane are upregulated upon exposure to elevated levels of lactate (42). Upon reaching the cell surface, EMMPRIN is shed in vesicles into the extracellular space, where it activates matrix metalloproteinases (43). Thus, the link between tissue lactate concentrations and more aggressive disease may occur as a result of the concomitant recruitment of EMMPRIN and MCT-1 to the cell surface in response to elevated lactate.

Lactate can be measured using MRS and associations between tumor lactate and survival after radiotherapy of malignant gliomas (44) and non-small cell lung cancer (45) have been

identified. However, there is no information on the importance of lactate in predicting survival in thermotherapy trials.

Extracellular pH (pHe) has been measured using invasive needle electrodes in canine sarcomas treated with thermoradiotherapy (37). Metastasis free survival was longer in animals with tumors that had pretreatment pHe values greater than 7.0 than if the tumor pHe was less than 7.0 (Figure 4). These less acidic tumors may have had better perfusion. This is also supported by the finding of longer metastasis free survival in animals with tumors having rapid contrast medium washin and washout as assessed using DCE-MRI (46) (Figure 5). The results of these two studies provide evidence of a link between pHe and perfusion in influencing treatment outcome in thermo-radiotherapy trials. Further work is needed to assess this important concept.

Phosphorus MRS has been used to predict survival. In humans with high grade soft tissue sarcomas treated pre-operatively with thermoradiotherapy, the PME/PDE ratio was predictive of progression free and overall survival (47). Differences in this ratio may likely reflect the rate of membrane turnover, which has been hypothesized to be related to tumor cell motility (48). In a canine trial, the PDE/ATP ratio was found to be of significance with regard to metastasis free survival. In that trial, subjects with tumors characterized by a high PDE/ATP ratio had significantly longer metastasis free survival (37). Given the relationship of elevated PDE with regard membrane degradation, tumors with a low PDE/ATP ratio may have had increased susceptibility to metastasis.

In the canine and human sarcomas trials, other ³¹P MRS parameters were examined for an association with metastasis-free and overall survival, including those that predicted for response, such as ATP/Pi. No significant correlation was between these parameters and outcome was identified in either trial (37,47).

Optical Spectroscopy – a new promising new molecular imaging tool

To date, optical spectroscopy has been used primarily for tumor diagnosis, as opposed to assessing prognosis. However, the inherent optical properties that can be derived may be related to prognosis as well. Relevant parameters include total hemoglobin, which is related to vascular volume, hemoglobin saturation and the redox ratio, which are related to hypoxia, and lipid absorption spectra, which are related to lipid content and type. In a small clinical trial in breast cancer, near infrared spectral analysis allowed identification of malignant tumors with 100% specificity and sensitivity (49). Diffuse optical spectroscopy has also been combined with diffuse correlation spectroscopy, which measures perfusion, to show that these spectroscopic parameters change within a few days after neoadjuvant chemotherapy treatment. It is possible that changes in these parameters early in the course of therapy may be useful in assessing tumor response (50). Similarly, Brown et al., have reported a significant decrease in the total hemoglobin saturation in malignant breast tumors, as compared with normal or benign lesions (18). They also report a significant positive correlation between hemoglobin saturation and total hemoglobin content. Finally, they found a significant positive correlation between HER2/neu status and both hemoglobin saturation and total hemoglobin content.

At the preclinical level, we have begun to perform studies to validate optical spectroscopic data against other validated physiologic methods. For example, we found an excellent correlation between changes in hemoglobin saturation and invasive oxygen measurements in flank tumors of mice that are switched from breathing room air to carbogen (51). We also identified a positive correlation between deoxyhemoglobin concentration measured with optical spectroscopy and hypoxic fraction assessed with pimonidazole hypoxia marker binding (52). Additionally, we found a positive correlation between an optical parameter derived from Monte Carlo modeling of diffuse optical reflectance, the backscatter factor (μ_s), and the histologic estimate of percent necrosis. The primary advantage of optical spectroscopy is that

it enables longitudinal monitoring of physiologic information, making it ideal to identify transient changes induced by therapy. Fig. 6 shows such an example in animals treated with MTD doxorubicin (10 mg/kg i.v.). It can be seen that there is a transient increase in hemoglobin saturation peaking 10 days post-treatment, possibly due to a reduction in oxygen consumption due to tumor cell killing, that may be difficult to pick up using a more limited selection of time points available using immunohistochemistry or other modalities. These results suggest strongly that optical spectroscopy may be a useful tool for monitoring treatment responses that involve thermotherapy. To date, however, optical spectroscopic methods have not been reported in conjunction with thermotherapy studies, either at the preclinical or clinical levels.

Discussion

We have summarized the current state of the art with respect to functional imaging methods that have been applied successfully to predict tumor response, duration of local control and overall survival in thermotherapy trials. We have also summarized methods that have potential in this regard, but have not yet been applied in this setting. A significant association has been found between several functional imaging methods and progression-free and overall survival. With the exception of DCE-MRI, however, there is little overlap between which parameters are predictive of which endpoint. Although changes in ATP/Pi appear to reflect direct changes in cell viability, such changes are not related to the ultimate outcome of the patient.

One of the challenges in functional imaging is the complexity and cost of MRI/MRS and PET methods. This becomes particularly problematic when one considers doing repeated measurements on the same subject over time. In our clinical series, the overall success of being able to obtain only two serial measurements was around 50% in both human and canine trials (37,47). Measurements made before treatment are relatively straightforward, since there is typically ample time to schedule the measurement before treatment starts. However, once treatment begins, obtaining a follow-up measurement becomes more difficult. Scheduling issues and patient compliance can all contribute to difficulties in obtaining serial imaging data. The rare occurrence of imaging equipment malfunction can also contribute to lost data.

One solution to this problem would be to consider use of optical methods, which are relatively cost effective and rapid to perform. They can be done in the clinic with a portable system at the bedside. Thus, they may provide the most robust tool to obtain serial data, at least for those tumors that are amenable to being studied with this modality.

With regard to molecular markers of outcome, there are important questions regarding whether physiologic parameters, such as those discussed in this paper, can be predicted by genomics. Fundamentally, is the physiology of a tumor predetermined by its genomics, or is the physiology independent of this? This relationship has been addressed in multiple myeloma where in a study of over 200 patients the loss of FDG PET signal after induction chemotherapy was a positive predictive factor, and independent of genomic profile (53). Tzike examined both 1-H MRS parameters and genomic data to classify primary brain tumors (54) and although there were associations between the 1-H MRS parameters and genomics whether these markers were related to outcome was not assessed. To date, there is no information on the relationship of tumor physiology vs genomics in hyperthermia trials. This is potentially important since a genomic biomarker would be much easier to implement compared with functional imaging.

Expansion of the role of hyperthermia for human cancer treatment will ultimately rely on identification of tumor sites and treatment prescriptions that lead to improved outcome compared to current treatments. Given the time requirements and expense of conducting phase III trials, and the technical effort required to initiate hyperthermia treatment programs, information that can facilitate identification of patients likely to succeed, or more efficacious

treatment prescriptions, could help streamline the definition of optimal hyperthermia treatment settings. Functional imaging has great potential to play an important role in this regard and its potential to modify current hyperthermia treatment practice is only beginning to be tapped.

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Figure 1.

Changes in ATP/Pi, as measured from ^{31}P MRS, are temperature dependent. (A) Data from mouse mammary carcinoma grown in the flank measured 18 h after local heating for 15 min at the designated temperature. Figure reproduced with permission from the author and publisher (23). (B) Data from canine soft tissue sarcomas. Dogs were enrolled on a clinical trial in which hyperthermia was combined with radiation therapy. ^{31}P MRS studies were performed prior to and 24hr after the first hyperthermia treatment. The change is the difference between these two time points. Data reproduced with permission from the author and publisher (24). T50 = median temperature. (C) Relationship between change in ATP/PME (pre vs. post first heat treatment) and probability for pathologic CR in human high grade soft tissue sarcomas, based on change in signal between the pre-treatment study and a study performed 24hr after the first hyperthermia treatment (24). Reproduced with permission from the author and publisher. These data are consistent with the concept that a reduction in ATP after hyperthermia treatment is related to cell killing by hyperthermia.

Figure 2.

DCE-MRI perfusion patterns in locally advanced breast cancer (26). Left panel shows a centrifugal pattern, with central contrast medium filling seen in the washin and washout parameters (top vs. bottom). Right panel is a tumor exhibiting a centripetal pattern. Data reproduced with permission from the publisher and author.

Figure 3.

Receiver Operating Curve for prediction of pathologic CR rate following thermochemoradiotherapy, using change in FDG PET uptake after two weeks of treatment (29). The receiver operating curve assesses the true positive rate (Y axis) as a function of the false positive rate (X-axis). An ideal test would yield a true positive rate of 1.0, and a false positive rate of 0. Data reproduced with permission from the author and publisher.

Figure 4.
(A). Probability of metastasis free survival as a function of time for dogs with tumors having extracellular pH>7 vs dogs with tumors having extracellular pH<7 (37). Dogs with tumors having a more alkaline extracellular pH had longer metastasis free survival.

Figure 5.

Metastasis free survival in dogs with tumors dichotomized by contrast medium washin rate (A) and contrast medium washout rate (B), as determined from DCE-MRI (46). Metastasis free survival was longer in dogs with higher washin and washout values, likely reflecting tumors with better perfusion. The Kaplan Meier curves shown compare the survival for animals, grouped above and below the median for the whole population. Washin value is a rate constant derived from the initial slope of the dynamic contrast enhanced image set and is influenced most strongly by the perfusion rate in the tumor (10). The washout value is derived from the terminal slope of the image set and reflects the rate of contrast clearance from the tumor. This is also influenced by perfusion, but is also reflective of the extracellular volume and permeability of the microvasculature.

Figure 6.

Hemoglobin saturation is plotted as a function of time after treatment with MTD doxorubicin. The bars represent the mean and standard deviations of the treated (blue) and control (red) groups. The dashed lines show linear regression lines for each group. A significant difference in the longitudinal trend associated with treatment was found using a linear mixed effects model ($p < 0.001$).