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Improving treatment efficacy with biological or biophysical feedback

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Optical measurements of tissue changes during photodynamic and radiation-based cancer treatments help to develop and improve these therapies.

Novel optical technologies can provide detailed microscopic information about tissue structure and physiology in a non-invasive manner, minimizing patient discomfort. This otherwise unobtainable information can help physicians select the best treatments, thus greatly benefiting patients. When used for early disease detection, it results in more effective curative therapies. It also allows inspection of the progress of various therapies to ensure they are working, enabling improvements to be made. We have investigated this last aspect of the benefits of optical methods, showing how they can be used to monitor tissue changes during X-ray and photodynamic therapy-based (PDT) cancer treatments.

Many of the current clinical cancer treatments are 'blind,' in that the success or failure of a treatment is often not known for weeks or months afterwards. For example, efficacy of radiation therapy, a treatment modality used in >50% of all cancers, is often assessed by a computed tomography (CT) or a magnetic resonance (MR) scan several months after treatment completion. A sensitive method reporting on treatment-induced tissue changes earlier—-during the course of treatment, or shortly thereafter—would clearly be beneficial in terms of optimizing therapy for the patient. Some initial work on radiation therapy monitoring using high-frequency ultrasound and positron emission tomography imaging has been reported.[1,2] However, the results are preliminary and inconclusive. Biophotonic approaches, in comparison, yield excellent tissue contrast, high sensitivity, and specificity. Their ability to detect subtle tissue changes and draw on mature technological embodiments (such as advances in photonics and fiber optics) makes them well-suited for sensitive assessment of treatment-induced tissue changes. We have used optical coherence tomography (OCT) to image tissue microstructure and microvasculature in X-ray irradiated or PDT-treated normal and cancerous tissues in mice.

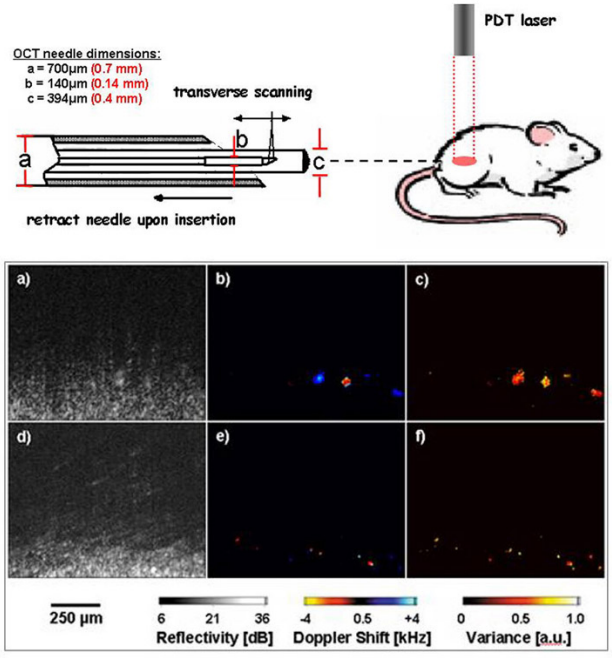

Figure 1. *Interstitial (within-tissue) optical coherence tomography (OCT) needle probe for monitoring microstructural and microvascular tissue responses. Shown on top is a cartoon of the needle probe design, emphasizing the sub-millimeter diameter of the probe. We monitored Photofrin-based photodynamic therapy (PDT) treatments in real-time in a rat with an implanted Dunning prostate tumor. Lower panels: (a) structural, (b) color-Doppler, and (c) variance images of several large detected vessels; (d) structural, (e) color-Doppler, and (f) variance images of many small detected blood vessels in the rat tumor.*[4]

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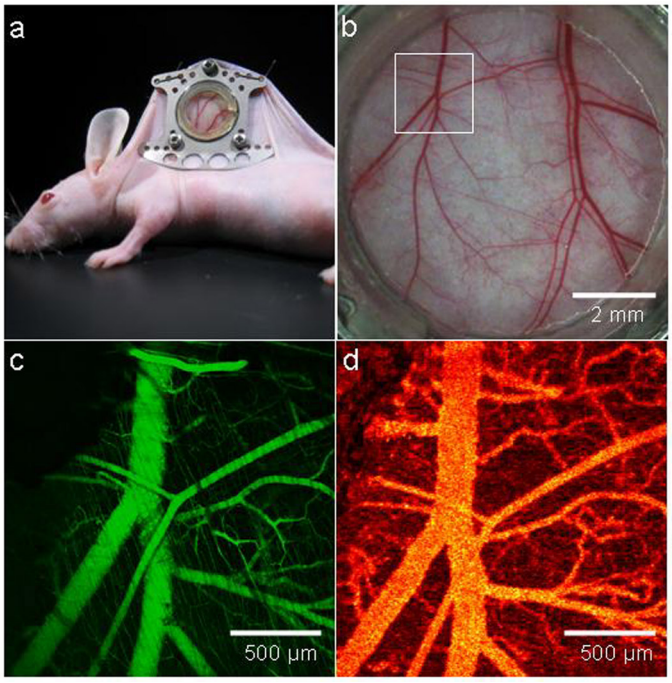

Figure 2. *Speckle variance OCT (svOCT) platform for sensitive detection of the microvasculature in animals. (a) Doppler angle and flow details make this method suitable for detection of microvasculature at the capillary level, as shown with this mouse window chamber. (b) White light microscopy of entire window. The white box represents confocal fluorescence and svOCT imaging locations. (c) Maximum intensity projection image of a fluorescence confocal z stack obtained using fluorescein Dextran (a fluorescence vascular contrast agent). (d) svOCT en face projection image of 3D vasculature without use of any external contrast agents.*[5]

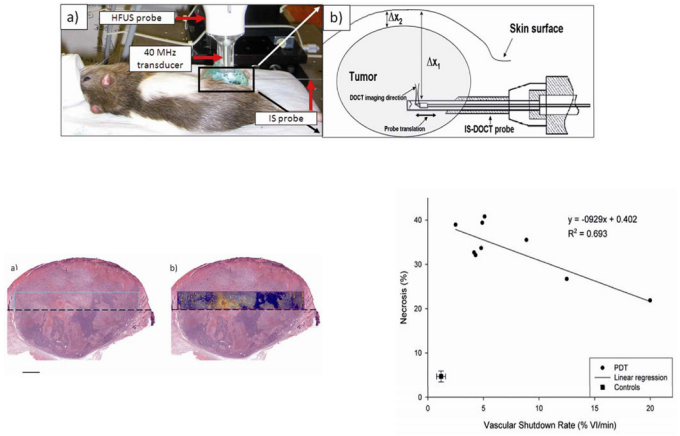

Figure 3. *Real-time PDT-treatment monitoring of Dunning prostate tumors in rats, using the interstitial (within-tissue) Doppler OCT system in Figure 1. Probe insertion under high-frequency ultrasound guidance and geometry of the probe/tumor locations are shown on top. Location of the Doppler OCT imaging plane is known from ultrasound data and can be correlated with biological end point of tumor necrosis, as shown on bottom left. Here, two histological sections of an excised tumor, stained with haemotoxylin and eosin, have been analyzed to assess the percent of tumor necrosis induced by PDT. The graph on the bottom right summarizes the correlation between the Doppler OCT in vivo metric measured during PDT delivery (x-axis) and eventual biological outcome of tumor necrosis (y-axis).*[6]

OCT is an emerging non-invasive cross-sectional imaging modality for visualizing subsurface tissue details in vivo at resolutions approaching histology.[3] Depth-resolved optical signals of subsurface tissue reflectivity are generated using the principles of optical interferometry and coherence gating. This results in high-resolution subsurface microstructural and functional (e.g. blood microvasculature) in vivo OCT images of biological tissues in a noninvasive tomographic manner. We use both structural and vascular tissue data to develop metrics that display changes as a result of radiation and PDT treatments. We developed technical pre-clinical platforms that enable us to track these changes over time in a longitudinal manner. This is important given the subtle nature of most treatment changes and the biological heterogeneity or variability of tissue features within tumors. Using OCT, we aim to address the following: what is the best image feature/metric? How sensitive is it to a given treatment? What is its specificity? (Tissue swelling may be detectable, but perhaps not important in the eventual patient response.) How predictive are the selected metrics of the overall treatment outcome?

The reported experiments make use of two OCT imaging interfaces: an interstitial (within-tissue) needle probe (<1mm diameter) that can be inserted into an animal tumor under radiological guidance to report on deep structural and functional tissue changes, and a larger OCT probe that has been optimized to image tumors implanted in a dorsal skin-fold window chamber in immuno-compromised mice. While the former offers the advantages of realistic 3D tumor milieu, the issues of lower signal-to-noise ratios, tissue motion, and detection of subtle microvascular features present formidable challenges. Conversely, the window chamber platform offers a more manageable and controlled—though 'artificial'—environment to develop OCT tissue structural and vascular metrics in vivo, and to validate results (e.g. with fluorescence confocal microscopy).

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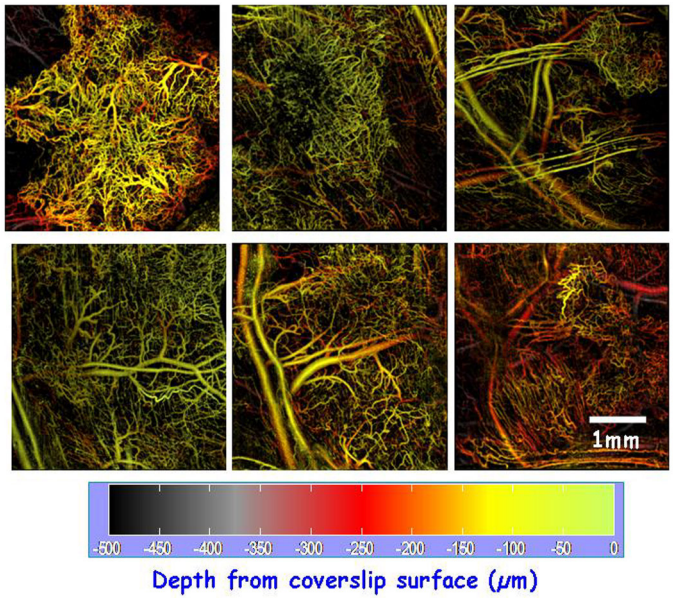

Figure 4. *Depth-encoded speckle variance optical coherence tomography images of six window chambers seeded with 9L gliosarcoma tumors, nine days after implantation. A significant amount of tumor microvascular variability is evident both between and within individual tumors. This inherent biological variability, effectively a shifting baseline, presents a significant challenge in deriving appropriate microvascular metrics that are meaningful and robust (e.g. quantitative measures of tumor response to therapies). Work is ongoing to develop such metrics that track tissue response assessment.*

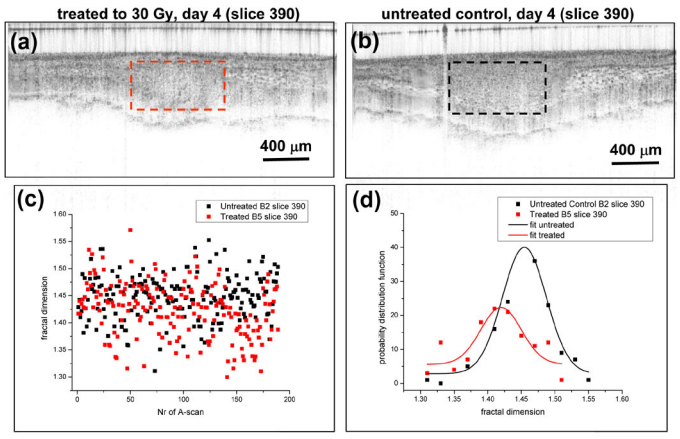

Figure 5. *Preliminary in vivo OCT spatial speckle (fractal) analysis of irradiated tissues. Direct structural OCT images of tumor-bearing mouse tissue in vivo in a window chamber model (ME-180 human cervical carcinoma tumors). (a) 4 days after 30Gy irradiation, and (b) corresponding untreated control at the same t=4 days time point. The top transparent band in both images is the 200μm-thick glass coverslip. Tumors are visible in the central area of both images, where the tissue bulges downwards slightly. (c) Box-counting fractal analysis of the spatial speckle pattern of the two OCT images, yielding fractal dimensions of the 190 A-scans in the regions of interest indicated by dashed boxes centered on the tumors in (a) and (b). (d) Data reduction of the results in (c), showing the histograms of the fractal dimension distributions and the corresponding Gaussian fits. Clear differences in the fractal dimension distributions are evident (average fractal dimension, distribution shape, Gaussian area).*

Figure 1 shows the OCT imaging needle and typical Doppler OCT blood flow data obtained interstitially;[4] Figure 2 is an example of a window chamber study using the speckle-variance approach to OCT microvascular analysis and validation by fluorescence confocal microscopy.[5] (Speckle is a superposition of dark and light spots common to wave-based examination methods, including ultrasound and laser imaging.) In Figure 1, several representative vessels are seen in the six Doppler images (a)–(f) (top row with larger vessels, bottom row with smaller ones), but the tissue motion artifacts and challenging signal-to-noise-ratio environment mask some of the finer capillaries. Some of these shortcomings are addressed with speckle-variance OCT in the mouse window chamber model (see Figure 2 and Figure 4). SvOCT in Figure 2 uses temporal speckle variations between consecutive structural (B-mode) OCT images and yields vascular contrast based on viscosity differences between more solid and more liquid compartments of tissues. Nevertheless, the interstitial Doppler OCT needle arrangement permits real-time detection of biologically validated PDT treatment effects (see Figure 3).

PDT is a promising treatment modality for several oncologic and non-oncologic pathologies, but it is both dynamic and complex owing to the interplay of light, photosensitizer, and molecular oxygen. As such, the need for treatment monitoring/guidance is crucial, and the sensitive microvascular detection afforded by OCT may be suitable for this purpose. Figure 3 summarizes the results of an interstitial Doppler-OCT derived microvascular metric measured during PDT treatment and its correlation with eventual biological end-point of tumor necrosis. The correlation suggests a possible role for this quantifiable real-time OCT metric for dynamic PDT treatment feedback.[6] However, exploring the microvascular imaging theme further, Figure 4 shows the speckle-variance OCT results in a mouse window chamber, demonstrating the inherent variability of tumor

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vascular patterns even prior to treatment. This inherent biological 'noise' must be accounted for in developing robust treatment-specific metrics, and it underscores the importance of a longitudinal imaging approach to elucidate the important relative changes induced by PDT or radiation therapy.

OCT images also carry useful information on tissue microstructure and organization, including speckle. Speckle analysis may provide unique signatures, related to tissue type, cellularity, and response to therapeutic insult, even though the specific tissue characteristics giving rise to the speckle pattern are not spatially resolved. Figure 5 shows the results of spatial speckle analyses[7] of OCT structural images in irradiated and control mouse tumors in the window chamber. We studied spatial speckle characteristics by analyzing the fractal dimension of individual depth scans within a region of interest encompassing the tumor. Clear differences were evident in the obtained fractal statistics of the spatial speckle pattern in a 30Gy-irradiated tumor, as compared to an unirradiated control tumor four days after irradiation. Many outstanding questions related to the robustness of these initial results, including detectable dose levels, normal versus tumor sensitivities, pros and cons relative to other OCT metrics of radiation effects, temporal dynamics of the response, and underlying radiobiological mechanisms, remain.

In the context of therapeutic monitoring, feedback, optimization, and understanding, the various OCT techniques discussed here may have a significant impact on improving treatments. As shown through illustrative examples, tissue microstructure and microvasculature exhibits sensitive, dynamic, and crucial details modulated by a variety of non-invasive therapies such as PDT and radiation therapy. The outstanding challenges are many: to improve structural and functional feature detectability, to develop and quantify the various tissue signal metrics, to test whether detected changes are specific to a therapy, and to determine if these detected changes are indicative of the eventual treatment outcome. Some of these challenges have been addressed, as summarized above, while work continues on others.

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References

1. I. Madani, W. Duthoy, C. Derie, *et al.*, *PET-guided, focal dose escalation using intensity modulated radiotherapy for head and neck cancers*, **Int'l. J. of Radiation Oncology, Biol. and Phys. 68**, pp. 126–35, 2007.
2. M. C. Kolios and G. J. Czarnota, *Potential of ultrasound for the detection of cell changes in cancer treatment*, **Future Oncol. 5**, pp. 1527–32, 2009.
3. A. F. Fercher, W. Drexler, C. K. Hitzenberger, and T. Lasser, *Optical coherence tomography: principles and applications*, **Rep. Prog. Phys. 66**, pp. 239–303, 2003.
4. X. B. A. Standish, J. Smolen, A. Mariampillai, N. R. Munce, B. C. Wilson, I. A. Vitkin, and V. X. D. Yang, *Interstitial Doppler optical coherence tomography monitors microvascular changes during photodynamic therapy in a Dunning prostate model under varying treatment conditions*, **J. Biomed. Opt. 12**, pp. 034022–8, 2007.
5. A. Mariampillai, B. A. Standish, E. H. Moriyama, M. Khurana, N. R. Munce, M. K. K. Leung, J. Jiang, A. Cable, B. C. Wilson, I. A. Vitkin, and Y. X. D. Yang, *Speckle variance detection of microvasculature using swept source optical coherence tomography*, **Opt. Lett. 33**, pp. 1530–2, 2008.
6. B. A. Standish, K. K. C. Lee, X. Jin, J. Smolen, A. Mariampillai, N. R. Munce, B. C. Wilson, I. A. Vitkin, and V. X. D. Yang, *Interstitial Doppler optical coherence tomography as a local tumour necrosis predictor in photodynamic therapy of prostatic carcinoma: an in-vivo study*, **Cancer Res. 68**, pp. 9987–95, 2008.
7. C. Flueraru, D. P. Popescu, Y. Mao, S. Chang, and M. Sowa, *Added soft tissue contrast using signal attenuation and the fractal dimension in OCT images of porcine arterial tissue*, **Phys. Med. Biol. 55**, pp. 2317–31, 2010.