Inspired gas-induced vascular change in tumors with magnetic-resonance-guided near-infrared imaging: human breast pilot study

Abstract. This study investigates differences in the response of breast tumor tissue versus healthy fibroglandular tissue to inspired gases. Cycles of carbogen and oxygen gas are administered while measuring the changes with magnetic-resonance-guided near-infrared imaging in a pilot study of breast cancers. For two patients, analyses are performed with cross-correlation techniques, which measure the strength of hemodynamic modulation. The results show that the overall vasoreponse, indicated by total hemoglobin, of healthy tissue has approximately a 72% and 41% greater correlation to the gas stimulus than the tumor region, in two patients respectively, when background physiological changes are controlled. These data support the hypothesis that tumor vasculature has a poorly functioning vasodilatory mechanism, most likely caused by dysfunctional smooth muscle cells lining the vasculature. This study presents a methodology to quantitatively analyze inspired gas changes in human breast tumors, and demonstrates this technique in a pilot patient population.

Keywords: breast tumor; inspired gas change; magnetic resonance; near-infrared imaging.

1 Introduction

Magnetic resonance-guided near infrared (MRg-NIR) imaging leverages knowledge of the location of a suspect lesion (provided by MR) to provide hemodynamic information such as total hemoglobin and oxygenation. This study reports on the potential of contrast derived from measuring the vascular compliance between normal and diseased tissue, which may have promising utility in increasing specificity.

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to-air ratio (GAR), shown in Eq. (1), is calculated as a quantitative metric to determine how large the modulation due to gas is compared to the background physiological modulation during air breathing.

$$\text{GAR} = \frac{\text{max}(\text{corr}_{\text{gas}})}{\text{max}(\text{corr}_{\text{air}})}.$$  

In Eq. (1), the GAR is computed by finding the ratio of the maximum correlation of the gas to the stimulus, to the maximum correlation of the all air control.

All three subjects in this study provided informed consent, following the procedures approved by the Institutional Review Board at Stanford Medical Center. Of the three subjects, two subjects were analyzed. One subject was dropped because of lack of compliance to the breathing protocol (subject was not breathing through the mouth piece), which was apparent when examining the oxycapnometer data. Patient 2 was aged 43 years, premenopausal, and had a 1.8-cm grade-2 invasive ductal carcinoma of the left breast. Patient 3 was aged 47 years, and had a 1.2-cm grade-1 invasive ductal carcinoma of the left breast. Imaging was performed before treatment.

3 Results and Discussion

An example of the vascular response to the gas stimulus for both the healthy fibroglandular and tumor tissue is shown in Figs. 2(b) and 2(d), respectively. The response during an air/air control for healthy and tumor vasculature is shown in Figs. 2(a) and 2(c), respectively. MRG optical data points are plotted as open circles. Overlaid in black (with calculated points labeled as closed circles) are the best-fit sinusoids that match the data. These sinusoids are phase shifted to match the time lag of the tissue response, and normalized to the amplitude of the response. These curves allow a visual comparison of the cross correlation between the tissue response and the modeled gas stimulus. Clearly, the response during air/air breathing, shown in Figs. 2(a) and 2(c) has lesser correlation to the sinusoidal stimulus than that of oxygen/carbogen breathing, shown in Figs. 2(b) and 2(d) (0.70 versus 0.26 in healthy tissue, and 0.65 versus 0.34 in tumor tissue).

Since the vascular changes induced during gas breathing can be insignificant in some subjects, the GAR ratio was computed to compare healthy versus tumor tissue. This metric provides a means to gauge the relative magnitude of the change with respect to normal air variations caused by biological noise. The tumor regions of interest for these cases were identified by CE-MRI. These images are shown in Figs. 3(a) and 3(c). The sequences used to collect CE-MRI data were 3-D T1-weighted gradient echo images (TR/TE/flip angle = 30/8/40 deg) collected on a 1.5 T MR system (Signa Excite, GE Medical Systems, Waukesha, Wisconsin). These images show regions of contrast enhancement that were histologically confirmed invasive ductal carcinomas. The optical data were collected in the plane indicated by the vertical lines in the CE-MRI images. Data from these planes were used to form images of the maximum correlation of the breast tissue to the respiratory stimulus compared to the air control.

The GAR for total hemoglobin of the tumor tissue compared to the normal surrounding fibroglandular tissue is shown in Figs. 3(b) and 3(d). There is a substantial separation in the average GAR between the normal and tumor tissue, which is on average 1.8 in tumor tissue to 2.8 on average in normal tissue, for these two patients. This GAR change is due to a weaker correlation in the tissue response to the gas stimu-
lus in these cases. These results could be explained by a lack of adequate smooth muscle cells to allow proper vasodilatory response. A similar lack of response has been observed in rodent R3230 mammary tumors by Hull et al., where a switch from air to carbogen caused a small and variable change in total hemoglobin (possibly indicating a decreased blood flow).

These results are predicted based on histological studies in tumors that reveal inadequate smooth muscle cells lining the vasculature. Without properly functioning smooth muscle cells to control vascular tone, blood volume changes are expected to be smaller. For example, in healthy tissue, the breathing of carbogen normally leads to vascular dilation due to the presence of CO2, a byproduct of cellular respiration. Excess CO2 typically indicates that the cells are consuming more oxygen than the vasculature is transporting. The vasculature normally responds by dilating, bringing in freshly oxygenated blood. The lack of smooth muscle cells in tumor tissue prevents the vasculature from dilating, resulting in a smaller or inconsistent change to the carbogen stimulus. Conversely, the switch to oxygen gas from carbogen would lead to vascular constriction in normal tissue (in this case, due to the lack of CO2). The lack of properly functioning vasocontrol in a tumor would result in less vascular constriction. These changes are better determined with the cross-correlation magnitude, which provides a quantification of the agreement of the measured tissue response to the gas stimulus.

It is interesting to note that the correlation in the tumor tissue for Ox/Cb of patient 2 is less than the air/air correlation for patient 3 (see Table 1). Based on the results from a previous study, we do not believe that comparing interpatient gas correlations has any real meaning, because there is such a large variance in air/air correlations in healthy subjects; these differences underscore the need for the GAR metric, which accounts for background physiology, and might be a way to quantitatively compare patients. As this is a pilot study, certainly a larger population needs to be measured to determine the utility of this technique.

Interestingly, the phase of the vascular change in the tumor tissue, shown in Fig. 2(d), is different than that of the healthy tissue, shown in Fig. 2(b). The phase delay (in units of π) is displayed in more detail in Table 2. In both cases, there is a time lag between the healthy fibroglandular tissue response and the tumor tissue response. This indicates differences in blood delivery between the healthy and diseased tissue. This lag could be explained by a delay in the blood arriving in the tumor due to poor tissue perfusion, a well-documented characteristic of tumors.

### Table 1 Cross-correlation in the fibroglandular (FG) and tumor tissue for the oxygen/carbogen (Ox/Cb) stimulus. GAR are the maximum correlations of the Ox/Cb stimulus versus the maximum correlation of the all-air control.

<table>
<thead>
<tr>
<th>Max correlation in FG, Ox/Cb</th>
<th>Max correlation in FG, air/air</th>
<th>GAR in FG</th>
<th>Max correlation in tumor, Ox/Cb</th>
<th>Max correlation in tumor, air/air</th>
<th>GAR in tumor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patient 2</td>
<td>0.37</td>
<td>0.13</td>
<td>2.85</td>
<td>0.25</td>
<td>0.15</td>
</tr>
<tr>
<td>Patient 3</td>
<td>0.70</td>
<td>0.26</td>
<td>2.69</td>
<td>0.65</td>
<td>0.34</td>
</tr>
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### 4 Conclusions

The search for increased specificity in breast cancer continues in the radiology community due to inadequate biomarkers of malignancy. Vascular functional deficiency has been noted as one of the key hallmarks of breast cancer, but tools to measure it have been inadequate. Breathing oxygenated gases has been used to investigate bladder, prostate, head and neck, and other tissues using BOLD MR imaging. However, breast BOLD has not been well studied, presumably because of the difficulty measuring changes in hemodynamics with adequate SNR. To date, only one case has been reported of changes in a human breast tumor during hyperoxygenated gas breathing, where the mean signal change was 62%.

Here, a pilot study is carried out using an oxygen/carbogen breathing protocol to produce hemodynamic changes. The functional changes in the tumor regions, measured by MRG optical imaging, are compared to the surrounding healthy tissue by cross correlating the measured response with the modeled breathing stimulus. This robust method provides self-referenced maps of the magnitude and time delay of vascular response of a region of interest detected during breast MR mammography.

The results show that relative total hemoglobin variation is higher in healthy tissue than tumor tissue. Additionally, the cross-correlation magnitude between the tumor tissue during inspired gas compared to an air-air control is less than that of the fibroglandular tissue compared to the control. Although this is a preliminary pilot study, this study shows the potential of using hemodynamic modulation as a tissue biomarker for diagnostic potential. Alternately, this metric could be used as a means to measure vascular function to determine sensitivity to therapy.

### Table 2 Time lag at maximum correlation between the measured total hemoglobin response and the Ox/Cb stimulus. Data are presented for the healthy surrounding fibroglandular tissue and the tumor tissue for patients 2 and 3.

<table>
<thead>
<tr>
<th></th>
<th>Phase delay for FG tissue (units of π)</th>
<th>Phase delay for tumor tissue (units of π)</th>
<th>Phase difference (units of π)</th>
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<tbody>
<tr>
<td>Patient 2</td>
<td>1.75</td>
<td>1.5</td>
<td>0.25</td>
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<tr>
<td>Patient 3</td>
<td>0</td>
<td>1</td>
<td>1</td>
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References