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Noninvasive Surface Imaging of Breast Cancer in Humans using a Hand-held Optical Imager

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Abstract

X-ray mammography, the current gold standard for breast cancer detection, has a 20% false-negative rate (cancer is undetected) and increases in younger women with denser breast tissue. Diffuse optical imaging (DOI) is a safe (nonionizing), and relatively inexpensive method for noninvasive imaging of breast cancer in human subjects (including dense breast tissues) by providing physiological information (e.g. oxy- and deoxy- hemoglobin concentration). At the Optical Imaging Laboratory, a hand-held optical imager has been developed which employs a breast contourable probe head to perform simultaneous illumination and detection of large surfaces towards near real-time imaging of human breast cancer. Gen-1 and gen-2 versions of the handheld optical imager have been developed and previously demonstrated imaging in tissue phantoms and healthy human subjects. Herein, the hand-held optical imagers are applied towards *in vivo* imaging of breast cancer subjects in an attempt to determine the ability of the imager to detect breast tumors. Five female human subjects (ages 51–74) diagnosed with breast cancer were imaged with the gen-1 optical imager prior to surgical intervention. One of the subjects was also imaged with the gen-2 optical imager. Both imagers use 785 nm laser diode sources and ICCD camera detectors to generate 2D surface maps of total hemoglobin absorption. The subjects lay in supine position and images were collected at various locations on both the ipsilateral (tumor-containing) and contralateral (non-tumor containing) breasts. The optical images (2D surface maps of optical absorption due to total hemoglobin concentration) show regions of higher intensity at the tumor location, which is indicative of increased vasculature and higher blood content due to the presence of the tumor. Additionally, a preliminary result indicates the potential to image lymphatic spread. This study demonstrates the potential of the hand-held optical devices to noninvasively image breast cancer in human subjects.

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Conflicts of Interest

All authors certify that this manuscript has not been published in whole or in part nor is it being considered for publication elsewhere. The authors have no conflicts of interest to declare.

Introduction

Breast cancer affects 1 in 8 women in the U.S. and is the second leading cause of cancer death, after lung cancer (1). Breast cancer screening by x-ray mammography has been shown to detect cancer at earlier stages leading to more flexibility in treatment options and prolonged life. However, x-ray mammography has an average 20% false-negative rate (meaning cancer is present but is not detected) which increases in younger women (due to denser breast tissue) where the cancers are found to be more aggressive (2). In the past few decades, diffuse optical imaging (DOI) has been explored by several research groups as a safe (nonionizing), and relatively inexpensive method for detecting breast cancer in human subjects (including dense breast tissues) by providing physiological information (i.e. oxy- and deoxy- hemoglobin concentration, etc.) (3–24, selected references). Additionally, DOI has been combined with other imaging modalities to provide complementary information (25–27, selected references). Clinical studies performed by various groups show that breast cancer is characterized by increased levels of total hemoglobin concentration (HbT) which can be detected by increased optical absorption (28, selected reference). Diffuse optical imaging of the breast is carried out using bed-based imagers, where the patient lies prone on a bed with the breast immersed in a cup or between parallel plates (9–14, 17, selected references), or alternatively using portable hand-held imagers (20–23, 29 selected references) that can be used at the bedside (similar to breast ultrasound) or even at the doctor's office. While bed-based methods provide 3D tomographic imaging, most hand-held imagers provide 2D localized spectroscopic information of the breast optical properties.

At the Optical Imaging Laboratory, generation-1 (gen-1) and generation-2 (gen-2) versions of a hand-held optical imager have been developed which employ breast contourable probe heads to perform simultaneous illumination and detection of large surfaces towards near real-time imaging of human breast tissue. The devices have previously been validated in phantom, *in vitro*, and healthy human subject studies with and without external fluorescent contrast (30–39). Our gen-1 hand-held optical imager has demonstrated 2D imaging and 3D tomography using 6 simultaneously illuminated sources in phantoms (30) and normal human breast tissues (31–33). Simultaneous illumination enable rapid data acquisition such that 2D images of large areas ($4 \times 9 \text{ cm}^2$) can be generated in near-real time (~ 2 sec per processed image). Three-dimensional tomographic imaging of human breast tissues using the gen-1 hand-held optical imager was possible due to its unique 3D positional tracking capability during imaging. The positional tracking capability allowed registration of the optical images with respect to the imaged tissue geometry, and hence provided 3D volumetric analysis as well (34). Extensive tissue phantom studies were performed to demonstrate 3D tomographic imaging capabilities of the imager (30, 35) as well as improved target depth detection (36). *In vivo* 3D tomographic breast imaging studies (on healthy subjects containing superficially placed tumor-like targets) were demonstrated using the hand-held optical imager (33). In parallel, a gen-2 version of the hand-held optical imager was also developed and its ability to detect targets was demonstrated in phantoms and human breast tissue (37–38). The gen-2 imager has added features of a two-part probe head that allows bilateral reflectance imaging of both the breast tissues simultaneously, as well as transmittance imaging of a single breast tissue (similar to x-ray mammography set-up) with or without slight compression.

Quantitative validation and comparison of performance between the gen-1 and gen-2 imagers in phantoms are provided in refs 37–39.

Herein, the gen-1 and gen-2 hand-held optical imagers have been applied towards *in vivo* 2D imaging of breast cancer subjects (without external fluorescence) in an attempt to determine the ability of the imager to detect breast tumors qualitatively. The details of the optical imaging instrumentation, experimental *in vivo* breast imaging studies and data analysis are described in the following sections.

Materials and Methods

Instrumentation and data acquisition

The three major components of the bench-top continuous-wave based gen-1 optical imaging system (Fig. 1) are the hand-held probe, the 785 nm laser diode source (HPD1005-9MM, Intense Ltd., North Brunswick, NJ), and the intensified charge-coupled device (ICCD) detector (PI-SCX 7495-0002, Roper Scientific, Trenton, NJ). The hand-held probe head is designed with unique features in that it is flexible to contour to different tissue curvatures and it uses simultaneous illumination and detection for rapid data acquisition of large tissue volumes. The probe head contains 6 source fibers (which split the light from a single laser diode) and 165 detector fibers connected to a $4 \times 9 \text{ cm}^2$ probe face. Each source emits $< 5 \text{ mW}$ of laser light incident on the tissue that is imaged. The 3-piece probe design allows for up to 45 degree curvature of the probe on each side. Extensive details of the imaging system and probe design can be found in reference 31. A gen-2 version of the imager was recently developed in order to overcome several limitations including the bulkiness of the system and the non-uniform laser source intensity distribution. The gen-2 imager is a more compact system that fits on a portable cart designed for clinical translation. The gen-2 probe has a forked design with two probe heads each contain 3 source and 96 detector fibers in a $4 \times 5 \text{ cm}^2$ probe face. The 5-piece probe design allows flexibility to fully contour to the curvature of the breast tissue. Extensive details of the gen-2 imaging system can be found in reference 37. Continuous-wave optical intensity images of total hemoglobin absorption were collected using the gen-1 and gen-2 imagers via reflectance imaging. Each intensity image consisted of a set of 5–10 measurements with a 0.2 second exposure time at a given experimental condition, which was averaged across repeated measurements to obtain average data. Custom-developed MATLAB software was used to post-process the raw data in order to generate a 2D contour plot of optical intensity data corresponding to the probe surface.

Data Analysis

Images were collected from both the left and right breast of each subject. The 2D contour plots represent the absorption intensity corresponding to the detector location (x-y coordinates) in the probe face. The data acquired from the contralateral (non-tumor containing) breast was subtracted from the data acquired from the ipsilateral (tumor containing) breast in order to remove background noise. The quantitative values of the data were reversed in sign (i.e. negative to positive values) such that the signal due to higher absorption appears as a positive (red) value in the images. The absorption intensity images were acquired at 785 nm wavelength. The 785 nm wavelength of light is close to the

isosbestic point, which is the point at which the absorption spectra of oxy and de-oxy hemoglobin intersect (40). Thus the higher absorption regions (due to the presence of tumors) represent the increased total hemoglobin concentrations (HbT), independent of the oxygen saturation content of hemoglobin. The images are presented as 2D contour plots of intensity (HbT concentration) given in arbitrary units for each individual scan (i.e. not normalized across scans).

Experimental studies

Five female subjects (ages 51–74) with breast cancer, as confirmed by needle biopsy, were recruited for the Institutional Review Board (IRB) approved Health Insurance Portability and Accountability Act (HIPAA) compliant studies. Three subjects had invasive ductal carcinoma, one subject had ductal carcinoma in situ, and one subject had metastatic carcinoma. The experimental cases are summarized in Table I.

For each study, the subject lay supine in a chair reclined to 45 degrees (shown in Fig. 2). The probe was placed on the breast with enough pressure to achieve full contact with the tissue without causing discomfort. The gain setting on the image intensifier was independently adjusted for each patient to get the maximum signal, and the absorption intensity was plotted in arbitrary units. For case #1, the gen-1's hand-held probe was placed visually at four different locations starting at the upper 12 o'clock position of the left breast and moving vertically downward (along the same clock position) in order to cover the entire breast. One image (which is an average of 5–10 repeated measurements as described in the Methods section) was collected at each of the four locations. The process was repeated at the same locations on the contralateral (opposite) breast and the corresponding images were used as a reference to remove background noise. For cases #2, 3, and 5 the probe locations were measured (using a ruler or a plastic template) and marked at 1 cm increments from the nipple toward the 12 o'clock and 6 o'clock positions on both the breasts. Since the breast tissue was larger than the area of the probe surface, a single set of vertical scans were not sufficient to image most of the breast. Hence, the tissue was scanned vertically down along the right and left of the central 12 o'clock position (with certain extent of overlap regions between scans). For case #4 the axillary lymph node region was the area of interest. The subject lay on her right side and raised her left hand over her head. The probe was placed visually on the upper region of the breast just under the arm. The probe was moved downward along the breast and images were collected at four discrete locations. A schematic of the probe location placement in each subject is shown along with the acquired optical images in Figs. 3–6 in the following section.

Results

Figs. 3A–3B show x-ray mammography images from the left breast of a 51 year-old breast cancer patient (case #1 in Table I). Fig. 3A is the axial view where the top of the image is toward the left side of the breast and the bottom of the image is toward the right side of the breast. Fig. 3B is the oblique view where the top of the image is toward the patient's head and left side and the bottom of the image is toward the patient's feet and right side. The images show the presence of 3 masses in the outer quadrant of the tissue (indicated by the

yellow arrows and labeled as L1–L3). The medical reports indicated that two of the masses were cancerous (L1 and L2) and the third mass was benign (L3). Figs. 3C–3D show ultrasound images from the left breast of the same subject. The three tumor masses in the outer quadrant of the tissue are indicated by the yellow arrows.

Figs. 3E–3H show the diffuse optical images, in terms of the extent of absorption across the tissue. Since the changes in absorption at the isosbestic point directly relate to changes in HbT, these optical images represent the differences in HbT across the breast tissue. An increased HbT is typically observed at the tumor site due to tumor angiogenesis. The images are shown at their approximate locations on the breast tissue and the estimated tumor locations are indicated by the yellow open circles. In the images, higher (red) signal indicates areas of higher absorption (or increased relative HbT concentrations) due to the tumor vasculature. The detection signal from the tumor is greater when the tumor is close to a strong laser source in the probe face and the signal is lesser when the tumor is located close to a weaker laser source. The six illumination points in the probe do not have uniform or consistent intensity of the laser light, and in this case the strongest source point in the probe was at the bottom right corner. This limitation in the imager has been addressed by the development of a second generation (or gen-2) device which has uniform and consistent source intensity distribution and will be described in more detail at the end of this section. The images show regions of higher absorption in the bottom right corner of the 2D image when the strong source was close to lesion L1 (Fig. 3E) and lesion L2 (Fig. 3G). When the source was located between the two lesions, a contrasted signal was detected, although at a lower intensity (Fig. 3F). The fourth image where the strong source was located below the tumors does not show a high signal since the strong source is located away from the tumor sites.

Figs. 4A–4B show the x-ray mammography and MRI images from a 52 year-old female with ductal carcinoma *in situ* (case #2 in Table I). The yellow arrows indicate the location of the 1.5 cm diameter lesion located at 12 o'clock in the left breast. Fig. 4C and Fig. 4D show the diffuse optical images at two probe locations over the tumor region from the front of the breast tissue while the subject lay supine. The images show areas of greater absorption (red signal) due to higher HbT concentration close to the tumor location, along with artifacts (areas of red signal found away from the tumor location and therefore not associated with higher HbT concentration). The subject was then asked to lie on her side with her arm raised above her head and images were collected from the side of the breast tissue. Fig. 4E shows an image collected from the side with the probe located over the tumor region. The image shows a higher HbT signal from the tumor site and fewer artifacts than the images collected from the front of the tissue.

Figs. 5A–5B show the x-ray mammography and ultrasound images from a 74 year-old female with a 0.7 cm diameter tumor located at 12 o'clock in the left breast (case #3 in Table I). Needle biopsy results indicated the tumor was invasive ductal carcinoma. Figs. 5C–5D show the diffuse optical images at two probe locations from the front of the tissue while the subject lay supine. Both images show regions of greater absorption due to higher HbT concentration (red color) close to the tumor location along with artifacts.

Fig. 6A shows ultrasound images of axillary lymph nodes (which have a typical size of ~ 1 cm (41) from a subject with metastatic carcinoma in the axillary region of the left breast (case #4 in Table I). Fig. 6B shows a diffuse optical image from the axillary lymph node region of the subject who lay on her side with her arm raised over her head. An area of greater absorption due to higher HbT concentration (red signal) is present in the region of the axillary lymph nodes which indicates the potential to image lymphatic spread.

Figs. 7A–7B show the x-ray mammography and ultrasound images from a 69 year-old female subject with invasive ductal carcinoma (case #5 in Table I). The yellow arrows indicate the location of a 0.9 cm diameter tumor at 8 o'clock in the left breast. Figs. 7C–7D show diffuse optical images at two probe locations. A higher signal (i.e. greater absorption due to higher HbT concentration) was detected close to the tumor location along with artifacts.

The subject in case #5 was also imaged using the gen-2 optical imager. Fig. 8 shows diffuse optical images collected using one probe from the gen-2 imager. While the subject lay supine the breast tissue was lifted and the probe placed underneath in order to image the tumor region (Fig. 8A). In Fig. 8B the subject lay at rest and in Fig. 8C the subject raised her arm above her head. The diffuse optical images show areas of greater absorption (red) due to higher HbT concentration close to the tumor location as indicated by the white dotted circles, and fewer artifacts than the gen-1 images.

The gen-2 probe was used to collect images at the different clock positions similar to the ultrasound procedure. Fig. 9 shows diffuse optical images collected with the gen-2 probe at the (A) 9 o'clock and (B) 3 o'clock locations. Area of greater absorption due to higher HbT concentration (red) can be seen at the 9 o'clock position (which is over the tumor location), whereas the image from the 3 o'clock position (which is away from the tumor location) does not show area of high absorption.

Discussion

The results presented here demonstrate the feasibility of imaging human breast cancer *in vivo* using a hand-held optical imager. The imager was used to image five breast cancer patients between ages 51 and 74 and was able to detect invasive ductal carcinoma, ductal carcinoma in situ, and lymphatic spread. In each case, the contralateral (non-tumor containing) breast was imaged to provide a reference (or background). In over 95% of the breast cancer cases, tumors are unilateral (i.e. in a single breast) (42–43) before it spreads to the other breast tissue and/or elsewhere (as in some cases). By performing bilateral imaging of both the breast tissues, where only one of the breasts is diseased, there is a potential to differentiate the diseased vs. normal tissue. Diffuse optical imaging provides functional information (analogous to positron emission tomography) indicating the presence of a tumor within the background rather than individual structural features. The diffuse optical images showed areas of greater absorption due to higher HbT concentration close to the tumor location along with artifacts. The artifacts may be due to several factors including improper contact of the probe with the tissue, the non-uniform source intensity distribution, and differences in the tissue composition of the contralateral breast, which does not allow proper

elimination of the background signal. The first two factors are improved by the second generation optical imager, and ongoing work is performed to further reduce artifacts in the images.

The first generation (gen-1) imager was designed as a bench-top device to demonstrate proof-of-concept. It was used to perform extensive phantom studies and preliminary *in vivo* studies with human subjects. The device is bulky and not conducive to bedside imaging in a clinical setting. Additionally, the laser source system design using a single laser diode split by six optical fibers does not produce a uniform intensity distribution among the six illumination points in the probe head. As a result, a tumor is detected when it is located close to a strong source in the probe and is undetected when it is closer to a weak source. The second generation (gen-2) imager was developed in order to overcome these limitations. The gen-2 probe head is smaller allowing more flexibility during imaging. The uniform intensity distribution of the laser sources allows for tumor detection at any location on the probe face.

For the first subject, the probe locations were determined visually by moving the probe vertically down to cover most of the tissue. A measurement system was later implemented where the probe locations were measured and marked on the tissue using the nipple as a reference point. During repeat visits, the consistency of the probe location is dependent on the operator's measurement. To overcome this limitation, a template with premeasured locations will be placed over the tissue and used to mark the locations and ensure consistency between multiple visits.

This study demonstrates rapid 2D imaging of breast cancer in large tissue volumes using a handheld optical imager, which has not been previously done. Future work will involve 3D localization of the tumors within the breast tissue. A unique feature of the optical image is that it has automated tracking and coregistration facilities in order to enable 3D tomography. Coregistered imaging and tomography have been demonstrated (34) using an acoustic based tracking system. In order to improve the error in coregistration due to tracker instability, an optical based tracking system is currently developed in house. The tracking system will obviate the need to measure and mark the probe locations on the tissue and allow consistent imaging across multiple visits independent of the operator. Additionally, the gen-1 and gen-2 imagers are currently modified to acquire the hemodynamic response in terms of both oxy- (HbO) and deoxy- hemoglobin (HbR) concentrations, apart from the changes in total hemoglobin concentrations (HbT). This is achieved by adding more source wavelengths (e.g. 690 and 830 nm) apart from the 785 nm wavelength source (that corresponds closely to HbT concentration in deep tissues). The HbO and HbR measurements can in turn be used to determine whether a lesion is benign or malignant, as observed by researchers in the past (17, 20, 26, selected references).

Conclusion

A hand-held optical imager has been developed towards real-time imaging of human breast cancer. Results presented herein demonstrate the ability of the gen-1 imager to detect tumors in human breast tissue *in vivo*. A gen-2 version of the imager has been developed to overcome several limitations and is designed to be a more portable system with a smaller

and flexible probe design. A preliminary study with the gen-2 imager in a cancer subject yielded images of the tumor with few or no artifacts. Future work will involve 3D tomographic analysis of the images obtained from breast cancer subjects.

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Figure 1. Schematic of the CW based hand-held optical imaging system. (Inset) The hand-held probe face (figure is not to scale).

Figure 2.
Experimental set-up for *in vivo* breast imaging.

Figure 3.

X-ray, ultrasound, and diffuse optical images of the left breast from a 51 year-old breast cancer patient. (A) X-ray mammography: The top of the image is the left side and the bottom of the image is the right side of the breast tissue. (B) X-ray mammography: Oblique image where the top of the image is toward the patient's head and left side and the bottom of the image is toward the patient's feet and right side. The yellow arrows indicate 3 lesions (labeled as L1, L2, and L3) located in the 1 o'clock to 2 o'clock region of the breast. (C–D) Ultrasound images showing two malignant lesions (L1, and L2), and one benign lesion (L3).

(E–H) Diffuse optical images collected from the breast tissue of a 51 year-old breast cancer patient. The schematics on the left indicate the approximate location of the tumors (red circles) and the probe (blue boxes) on the breast tissue (figure not to scale). The yellow arrows to the left of the probes in the schematic indicate the movement of the probe between each image and the previous. The white dotted circles in the diffuse optical images to the right indicate a rough estimate of the tumor locations. The red color indicates areas of greater absorption due to higher HbT concentration relative to the background.

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Figure 4.

(A) X-ray mammography and (B) MRI images from a 52 year-old female with ductal carcinoma in situ. The images show a 1.5 cm diameter mass at the 12 o'clock region of the left breast. (C–E) Diffuse optical images of ductal carcinoma in situ in the left breast of a 52 year-old female. (C–D) Images collected at two probe locations from the front of the breast while the subject lie in supine position. The schematics on the left indicate the approximate location of the tumors (red circles) and the probe (blue boxes) on the breast tissue (figure not to scale). The yellow arrow to the left of the probe in the schematic indicates the movement of the probe between that image and the previous. (E) Image collected from the left side while the subject lie on her right side with her arm raised above her head. The black open circles represent the approximate tumor location. The red signal indicates areas of greater absorption due to higher HbT concentration.

Figure 5.

(A) X-ray mammography and (B) ultrasound images from a 74 year-old female with invasive ductal carcinoma (case #3). The yellow arrows indicate the location of a 0.7 cm diameter lesion located at 12 o'clock in the left breast. (C–D) Diffuse optical images from two probe locations on the subject from case #3. The schematics on the left indicate the approximate location of the tumors (red circles) and the probe (blue boxes) on the breast tissue (figure not to scale). The yellow arrows to the left of the probes in the schematic indicate the movement of the probe between each image and the previous. The black open circles indicate the approximate tumor location.

Figure 6.

(A) Ultrasound images showing axillary lymph nodes from a subject with metastatic carcinoma in the axillary region of the left breast (case #4). (B) Diffuse optical image of the left axillary region from a female subject with metastatic carcinoma (case #4). The smaller image indicates the location of the probe with respect to the axillary lymph nodes and is zoomed in the larger image. The white dotted circle indicates a region of interest where higher absorption (red) signal corresponding to higher HbT concentration might indicate the lymphatic spread.

Figure 7.

(A) X-ray mammography and (B) ultrasound images from a 69 year-old female subject with invasive ductal carcinoma (case #5). The yellow arrows indicate the location of a 0.9 cm diameter tumor at 8 o'clock in the left breast. (C–D) Diffuse optical images collected at two probe locations (indicated by the schematics to the left). The schematics on the left indicate the approximate location of the tumors (red circles) and the probe (blue boxes) on the breast tissue (figure not to scale). The yellow arrow to the left of the probe in the schematic indicates the movement of the probe between that image and the previous. The white dotted circles indicate a region of interest at the approximate tumor location where higher absorption (red signal) corresponding to higher HbT concentration might indicate the increased vasculature (and therefore blood content) due to the presence of the tumor.

Figure 8.

(A) Schematic showing the approximate tumor location and probe location while imaging a 69 year-old female subject with invasive ductal carcinoma (case #5). (B–C) Diffuse optical images collected from underneath the breast using the gen-2 imager. The white dotted circles indicate a region of interest at the approximate tumor location where higher absorption (red signal) corresponding to higher HbT concentration might indicate the increased vasculature (and therefore blood content) due to the presence of the tumor.

Figure 9.

Diffuse optical images collected with the gen-2 probe at the (A) 9 o'clock and (B) 3 o'clock locations (of the same breast) from a 69 year-old female subject with invasive ductal carcinoma (case #5). Area of higher absorption can be seen at the 9 o'clock position which is over the tumor location, whereas the image from the 3 o'clock location away from the tumor does not show area of high absorption. The white dotted circle indicates a region of interest at the approximate tumor location where higher absorption (red signal) corresponding to higher HbT concentration might indicate the increased vasculature (and therefore blood content) due to the presence of the tumor.

Table 1

Summary of our experimental cases for *in vivo* imaging on breast cancer subjects.

#	Age	Tumor Diameter	Tumor Location	Tumor Type
1	51	3.5 cm, 1.5 cm	2 o'clock, left breast	Invasive ductal carcinoma
2	52	1.5 cm	12 o'clock, left breast	Ductal carcinoma in situ
3	74	0.7 cm	12 o'clock, left breast	Invasive ductal carcinoma
4	64	Lymph node	Axilla, left breast	Metastatic carcinoma
5	69	0.9 cm	8 o'clock, left breast	Invasive ductal carcinoma