Study of wound healing by terahertz spectroscopy

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ABSTRACT

Terahertz medical imaging is an expanding field of research in the diagnosis of several medical conditions, including wound healing. Study was carried out in rat models where a THz imaging system was used to mark the difference in absorption spectra between a wound and a healthy tissue. Absorption spectra of the wound and healthy tissues have been measured in the 0.3-1.5 THz range.

Keywords: wound healing, THz Time-Domain Spectroscopy, absorption, medical imaging

1. INTRODUCTION

Wound healing is an essential physiological process consisting of the collaboration of many cell strains and their products.[1] It has three main phases, which do not mutually exclude themselves: inflammation, proliferation, and remodeling.[2]

Traditionally, visual observation is the most common tool for wound assessment.[3] Its appearance can then be documented and categorized based on wound assessment charts, such as the Bates-Jensen Wound Assessment Chart (BJWAT), which contains thirteen items to assess the wound: size, depth, edges, undermining, amount of necrotic, granulation, necrotic tissue type and epithelialization tissue, exudate type and amount, skin color, edema and induration.[4]

Currently, the noninvasive optical imaging assesses wound severity, healing potential, objective, and noninvasive manner by collecting the information through light and tissue interaction. Additionally, optical microscopic techniques evaluate cell regeneration, metabolic activity, collagen remodeling, inflammation, blood flow, and vascular structure.[5] For example, optical coherence tomography (OCT) is one of the techniques for 3D in vivo tissue imaging. That uses reflected light from tissue to construct cross-sectional images from a deeper part of the skin.[6] Multiphoton microscopy (MFM) is a promising method of diagnosis and visualization of wound healing. Multiphoton microscopy techniques, specifically autofluorescence (AF), second harmonic generation (SHG) and fluorescence lifetime imaging (FLIM) are effective assessment methods of wound healing because of their high-resolution, penetration depth, and determining real-time cellular metabolic activity.[5]

Terahertz spectroscopy is a noninvasive and high-contrast optical imaging method for medical imaging. Terahertz (THz) refers to the band of electromagnetic waves with frequencies ranging from 0.1 THz to 10 THz. Recently, had developed terahertz techniques in various fields and specifically in medical applications.[7,8] In addition to being biologically safe, THz has been shown to have a strong interaction with water in tissues.[9] Where in the terahertz range, the water in the biological tissue is the main absorber.[10] Biological tissues differently have water content, so the terahertz absorption also different, which limits the penetration depth of terahertz radiation into biological tissue to the order of a few millimeters.[11] For example, the penetration depth for skin tissues is 500 μm at most for a system with a signal-to-noise ratio (SNR) of 1000 at a frequency of 1 THz.[12] The high sensitivity to water makes terahertz radiation of such interest in the medical field as it can be used to detect subtle changes in the tissue composition, in cancer imaging and wound healing.[13, 14]

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Also, terahertz imaging has a future in wound assessment; It depends on the severity and degree of the wound. When a wound is fresh, the water is present in a liquid form, the tissue saturated with plasma.[15] As the wound heals, macromolecules of collagen form to seal the wound. It traps many of the water molecules that are present, thus altering the Terahertz signature from normal tissue.[16]

On the other side, water absorption is a strong limitation of THz spectroscopy usefulness in medical applications, where the range of water absorption is much higher compared to most tissue molecular components. It creates difficulties in measuring any other chemical components in fresh or frozen tissues.[14] However, the difference between healthy and diseased tissues can be studied by THz regardless of the water content. It could be due to the difference in tissue structure, the density of affected tissues, and other factors.[14,17]

In this paper, the difference in terahertz images of the wound, surrounded tissues, and healthy tissues is studied, using their absorption spectra in the 0.3–1.5 THz range by the laser THz spectroscopy.

2. MATERIALS AND METHODS

2.1 Animal model and wound protocol

Experiments have been done with Wistar male rats, weight 400-450 g. Before surgery, rats were anesthetized by intramuscular injection of zoletil (1-4 mg / 100g), then depilation of the surgical field was performed (Veet cream, France), the surgical field with chlorhexidine (0.05%) were processed and formed wound in each rat. The wound was obtained by cutting a 0.5 cm x 0.5 cm skin flap at the back of the head (between the ears) to the full thickness of the skin in the interscapular region. Formalin-fixed, alcohol dehydrated biopsies samples will be utilized in this work. The example of wound biopsy is shown in figure 1.

Figure 1. The wound biopsy after surgery.

2.2 Terahertz spectroscopy technique

Time-domain THz spectrometer (EKSPLA, Estonia) was used to obtain absorption spectra of normal and wound tissues with tuning range 0.3-1.5 THz. A spatial 2D-scanning was carried out: from 12 to 14 mm in increments of 0.1 mm vertically, from 12 to 14 mm in increments of 0.1 mm horizontally. Averaging over time (by 1024 scan) was carried out for each signal using the software supplied with the spectrometer. The measurements were performed at room temperature. Images were analyzed using OriginPro.

3. RESULTS AND DISCUSSION

Time-domain wavefronts of the electric field of THz radiation transmitted through the air, the wound, and normal tissues are shown in figure 2. The difference in the waves was noticed.

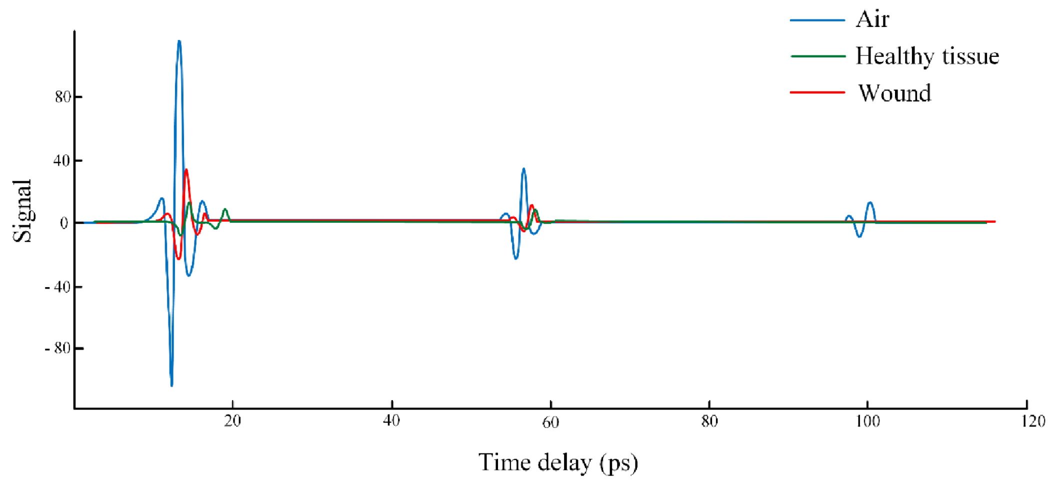

Figure 2. An example of a THz radiation wavefront through the air, wound, and healthy tissue.

Next, the Fourier transform of the temporal THz radiation signal transmitted through the sample was performed to obtain the absorption spectrum. The absorbance spectra for the healthy and wound tissues on the range of 0.3-1.5 THz are shown in figure 3.

Here we show THz spectra in term of absorbance, which was calculated by a standard way:

$$\text{Absorbance} = -\lg\left({}^{I}/_{I_{ref}}\right),$$

where I is the output THz wave intensity, when a sample was placed in the device optical pathway; I_{ref} is the output THz wave intensity without a sample.

The wound was shown to have a lower absorption spectrum than the healthy tissue.

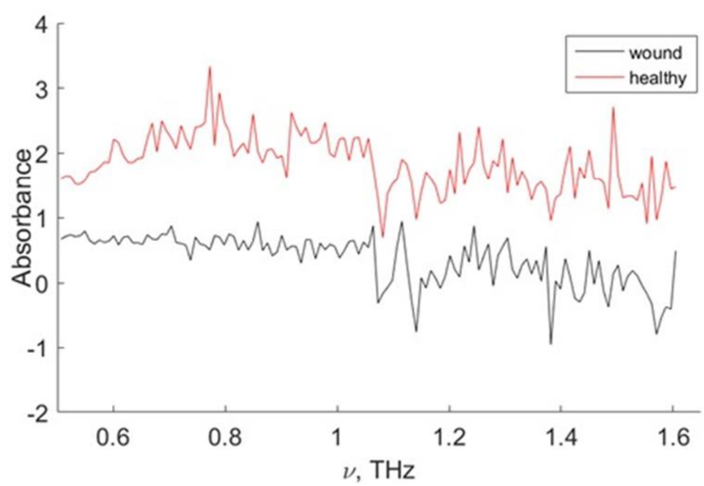

Figure 3. The THz absorption spectra for healthy tissue and wound.

Significant changes in the terahertz signals and absorption spectra configure 2D images, which acquires the THz signal passed through the sample for every position. Absorption spectra of healthy tissue and wound were measured using the

EKSPLA Time-domain THz spectrometer in the spectral range of 0.3-1.5 THz. Spatial 2D scanning of the healthy and wound tissue was carried out with a step of 0.1mm in vertical and horizontal directions and with averaging over scans at every spatial point (1024 scans). The comparison of the absorption spectra at different spatial points of the sample: wound, healthy tissue, and the surrounded tissues are shown in Fig.4. This image corresponds to 0.9 THz frequency.

In total, 100 scans for each wound and healthy tissues were analyzed. Color changes indicate a change in the absorption value. Terahertz images demonstrate an essential contrast between the wound and healthy tissues.

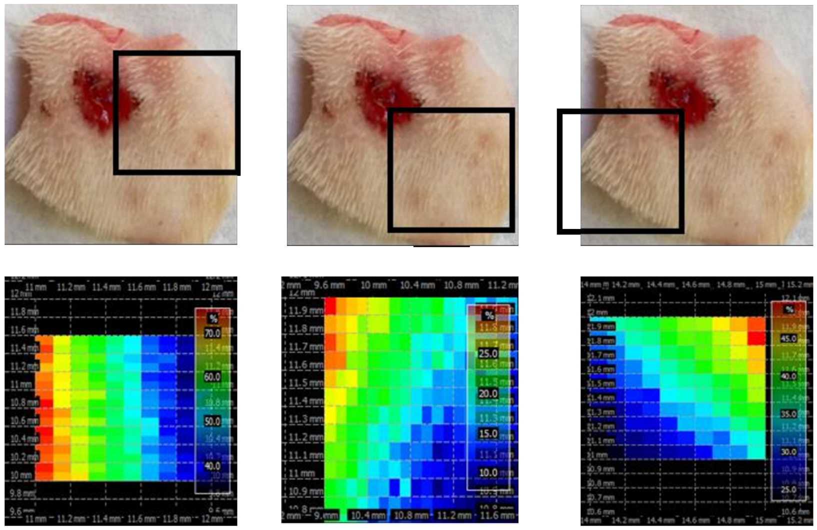

Figure 4. 2D images of the spatial distribution of THz radiation absorption in health, in the wound, and in the surrounding tissues.

4. CONCLUSION

The ability to distinguish between healthy and wound tissue has presented by Time-domain THz spectroscopy. A comparison of absorption spectra shows an essential difference between samples. Therefore, THz imaging will be able to monitor wound healing.

5. ACKNOWLEDGMENTS

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