Assessment of Water Content in Biological Samples by Terahertz Time-Domain Spectroscopy

Mariia Borovkova*,1,2, Mikhail Khodzitsky2, Alexey Popov1, Alexander Bykov1, Igor Meglinski1
1University of Oulu, Optoelectronics and Measurement Techniques Unit, Erkki Koiso-Kanttilankatu 3, 90570 Oulu, Finland
2ITMO University, Terahertz Biomedicine Laboratory, 3 b Kadetskaya Line, Vasilievskiy Island, 199034 St. Petersburg, Russia
* Correspondence: mariia.borovkova@oulu.fi

Abstract: The noninvasive measurement of water content in biological samples utilizing the terahertz waves is a promising tool for various biomedical applications. We introduce the method of water concentration measurement in biological samples by terahertz time-domain spectroscopy.

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1. Introduction
In recent years, different techniques using terahertz (THz) radiation have been applied in a variety of scopes due to unique properties of this type of emission. One of the most significant areas of interest is the diagnosis of different diseases or stresses through examination of biological tissues and objects. Sensitive and precise tool for noninvasive water concentration measurement would help in such areas as cancer diagnosis, products quality control or the plants’ stress responses monitoring [1-3]. THz radiation is characterized by the strong water absorption, which allows this frequency range to be used as a sensitive noninvasive hydration probe.

The study is devoted to the development and testing of the methods for noninvasive measurement of water concentrations in biological tissues and samples by THz TDS. Leaves and fruits from different plants along with samples of pork meat were used as the objects of investigation. The measurement of water concentration in botanic tissues and objects is significant due to the steadily worsening ecological situation in the world. Giving that plants and trees are essential for humanity living, there is a need for noninvasive tools to measure and evaluate stress responses of different types of plants, including response for drought. The investigation of humidity of pork meat is a step towards development of the methods for diagnostics of human skin and organspathologies.

Generally, water content in plants can be monitored using such methods as gravimetric or pressure chamber, which lead to the destruction of the sample. Besides, there also are some non-destructive methods which are used for water content measurement (chlorophyll fluorescence, visual estimate), which are usually not very precise. Moreover, there have already been reported the methods for noninvasive water concentration measurement using THz TDS or continuous THz setups, based on correlation between water content level and attenuation of transmitted through the sample THz signal [3]. However, these methods work in transmission mode, which significantly limits their application. Due to the strong water absorption, only thin tissues can be used for the investigation.

In order to examine thicker biological objects (fruit skins and pork meat samples), the reflection geometry was applied. Previous studies demonstrated that water content of human or animal tissues of skin or organs differs in zones with pathologies and healthy zones [4,5]. It should be noted, that the THz radiation is non-ionizing, therefore in general it is harmless for biological tissues. A water content sensor based on the reflection geometry would be a helpful tool for distinguishing healthy and injured tissues when visual assessment is difficult.

2. Theoretical background
The method of water concentrations determination is based on the extended Landau-Looyenga-Lifshitz model [6]. Equation (1) shows the relation between effective permittivity of a water-containing sample and the dielectric function of its components [6]:

\[ \hat{\varepsilon}_{mix}^e(f) = a_w \hat{\varepsilon}_w^e(f) + a_1 \hat{\varepsilon}_1^e(f) + a_2 \hat{\varepsilon}_2^e(f), \]

where \( \hat{\varepsilon}_{mix} \), \( \hat{\varepsilon}_w \), \( \hat{\varepsilon}_1 \) and \( \hat{\varepsilon}_2 \) are permittivities of the whole sample, water, the 1st and 2nd components, respectively; \( a_w \), \( a_1 \) and \( a_2 \) are volume fractions of water, 1st and 2nd component in the sample.

3. Experiments description
For this investigation, a series of experiments were performed both in transmission and in reflection modes. In transmission mode, the leaves from different plants were examined by the mini-Z spectrometer by Zomega corp. For the experiments in reflection mode, some thicker biological objects were used: skin of orange, pieces of carrot, slices...
of pork. The principal scheme of the THz TD spectrometer used for the experiments is presented in the Fig. 1.

![Fig. 1. Principal scheme of the THz TDS setup (reflection mode). Filter – set of teflon filters cutting the IR frequencies, BS - beam splitter, M – mirror, L – lens, BD – balanced detector, PC – personal computer, PM – parabolic mirror, ADC – analog-to-digital converter, DAC – digital-to-analog converter.](image)

The first stage of the experimental series was calculation of the dispersion of the water dielectric function in the frequency region of 0.1-0.9 THz. The data obtained experimentally was consistent to what was published before.

The second stage was performance of an experiment with a number of stages: at each stage, the sample was being dried up using a special heater for a certain time. Therefore, a number of different water content levels were obtained within each experiment in order to test the methods sensitivity and precision. By the end of each experiment, the sample was completely dried up and used as a solid component in the calculations. Simultaneously, the water concentrations were measured gravimetrically at each stage in order to verify the THz data.

For the convenient water concentrations calculation in transmission and reflection modes, two programs in Matlab with Graphical User Interface (GUI) were developed. In the programs, the user has to upload the waveforms of dehydrated sample and investigated sample obtained experimentally. We suggest that in order to control the water status in the leaves or fruits from the same plant, it is enough to use the same dehydrated sample as a solid component for testing other samples from the same species.

4. Results

During the series of experiments, optical properties of different samples in relation to their water concentration were studied. The experiments demonstrated how the optical properties of the samples move from near water dispersion towards the dispersion of absolutely dry tissue.

The Fig.2 shows dispersion of complex refractive indices of the Carpinus caroliniana tree leaf in 5 different stages of dryness obtained in the experiment using the mini-Z spectrometer by Zomega corp. in transmission mode.

![Fig.2. Dispersion of complex refractive index of a leaf (Carpinus caroliniana) in 5 different stages of dryness in the experiment in transmission mode.](image)
The Fig. 2 demonstrates that while the leaf was still hydrated, its optical properties were similar to water dispersion. By the 3rd, 4th and final stages of dryness the refractive index dispersion of the sample was completely flat.

Fig. 3 shows the values of water concentration obtained in different samples in the experiments both in transmission (a) and reflection (b) geometries at 5 stages of dryness. The results acquired by THz method are compared to the gravimetrical data.

![Fig. 3. Water concentration values by THz TDS and gravimetric methods at 5 stages of dryness for a) a leaf of *Carpinus caroliniana* obtained in transmission mode, b) pork meat sample obtained in reflection mode.](image)

The values of water concentration acquired by THz TD spectrometer in transmission geometry (Fig. 3 (a)) conform with the gravimetrical data quite well. This proves that a THz TDS system can be used as a precise sensor of hydration in biological samples. Though values of water concentration obtained gravimetrically and by THz TDS in reflection mode (Fig. 3 (b)) do not completely coincide (which is explained by the fact that gravimetric method allows estimation of the average water concentration in a thick (2-3 mm) sample, whereas measurement by THz TDS corresponds to the surface tissue water level), the experimental curves have quite similar behavior.

Finally, the results obtained with the THz time-domain spectroscopy systems have been compared and cross-validated with the results of convention optical/NIR spectroscopy and tissue polarimetry.

5. Conclusions

It is shown that the THz time-domain spectroscopy systems can be used as a tool for *in vivo* measurements of water content in the biological tissues or other objects of interests, both in the transmission in reflection modes. Presented experimental approach allows to perform non-destructive measurements that potentially can be used for indication of anomalies of human or animal skin or organs, in food quality control, plants stress respond estimation, and cosmetic industry.

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7. References