Non invasive microwave dielectric spectroscopy for biological characterization and healthcare applications : Importance of a differential approach

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***Abstract*— This paper aims to highlight the importance of applying a differential approach to electrically characterize biological samples, such as biomolecules or cells in suspension, up to microtissues and organs, and to enhance measurement sensitivity and consequent discrimination. Whenever S parameters, capacitive or conductive signals, or dielectric characteristics are considered, the introduction of contrasts therefore extends the sensing applicability of microwave dielectric spectroscopy and opens interesting applicative fields in biology and healthcare instrumentations.**

Keywords*— *microwave, dielectric spectroscopy, cells, living materials, characterization, RF measurements

I. Introduction

Microwave dielectric spectroscopy constitutes an interesting analytic technique to characterize biological samples and tissues as it presents several advantages. The use of electromagnetic waves to sense living organisms is indeed intrinsically non-destructive and non-invasive at low power levels [1-6]. In the microwave range, it is also very sensitive to water molecules and enables intracellular investigations [1].

For long, this technique has been applied to cumbersome samples with also quite large equipment. Electrical signals such as S parameters or impedance were looked at. However, with the advent of microtechnology with RF circuits, a new scale of bio-samples has been made accessible. In parallel, high sensitivity levels in detection were also requested. However, given the large γ dispersion phenomenon occurring in the microwave range due to the water molecules polarization and considering also the intrinsic variability of biological samples, important variations in dielectric response are obtained, which leads to a detection loss between samples. To circumvent this drawback, a differential approach may be used.

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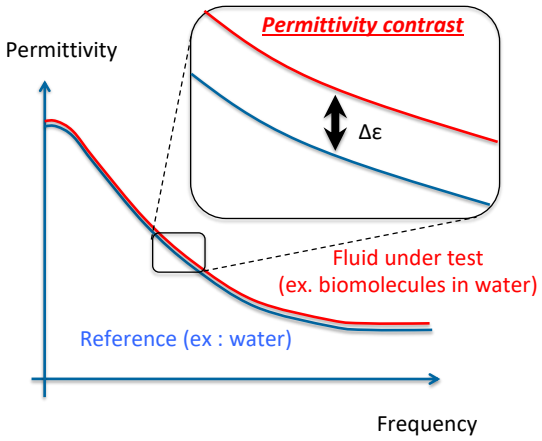

Fig. 1. Large relaxation phenomenon of biological liquids and samples

In this paper, the importance of using such a differential approach is therefore highlighted to re-allow a possible discrimination between similar biological samples at different scales, while considering molecules in solution, individual cells measured in their host culture medium, as well as with cells in suspension and even particular biological liquids such as egg constituents.

II. Sensitivity Issue while Measuring Biological Samples in the Microwave Range

For long, those using or developing dielectric spectroscopy have been considering absolute values of permittivity or other electrical parameters. This is well adapted in the case of the characterization of very different materials. However, while considering biological samples, this habit leads rapidly in a strong limitation in detection with RF signals located in the microwave range. Biological materials are indeed mainly composed of water molecules, which present a relaxation close to 17 GHz at room temperature, i.e. strong losses in the frequency range as well as a large variation of permittivity values, as illustrated in Fig. 1. In consequence, the water molecules of the investigated

element and those usually present in the host media of the biological solutions or suspensions screen the targeted bio-information. The discrimination between two similar

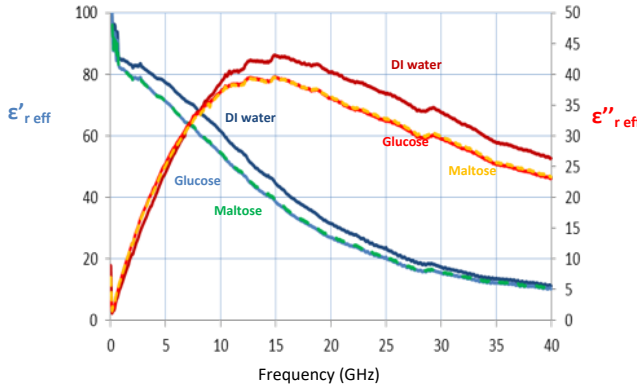

Fig. 2. Real and imaginary parts of the permittivity of Di-Ionized water, glucose and maltose in aqueous solution. The real parts are presented in blue and green, whereas the imaginary parts are in yellow, orange and red.

biological configurations or constituted solutions becomes impossible.

To overcome this electrical situation and to open new perspectives of applications of the dielectric spectroscopy to biological materials, a solution is not to consider absolute parameters any longer but relative ones. A differential approach should therefore be preferred and well defined according to the investigated bio-element.

III. Differential Approach

It consists in subtracting the impact of the screening host medium, which is chosen as a reference during the measurements. This may be done with the permittivity characteristics as given with Eq. 1 or with S parameters as with Eq. 2, or also with the electrical parameters such as capacitor (cf. Eq. 3) and conductance.

$$\Delta\varepsilon = \varepsilon_{\,fluid\ under\ test} - \varepsilon_{\,ref} \tag{1}$$

$$\Delta\,|S21| = S21_{\,fluid\ under\ test} - S21_{\,ref} \tag{2}$$

$$\Delta C = C_{\,fluid\ under\ test} - C_{\,ref} \tag{3}$$

Where ε corresponds to a permittivity characteristic, the one of the complete fluid under test, $\varepsilon_{\text{fluid under test}}$, or the one of the host medium taken as the reference for the calculation, ε_{ref}. S21 is a transmission coefficient, whereas C is a capacitance. It allows to focus then on the dielectric or electrical contrasts induced by the bio-element itself and eliminates the contribution of the dominant constituent, as indicated in the inset of Fig. 1.

To illustrate this approach, different examples are then given, starting from biomolecules in aqueous solution, single cell measurements, cells suspensions, and bio-liquids such as albumen and yolk of eggs.

IV. Illustrations of the Differential Approach with Various Biological Samples

A. Biomolecules in solution

As a first example, biomolecules in aqueous solution are considered. In this case, two similar oses, glucose and maltose, are measured within a miniature coplanar waveguide [7]. Fig. 2 presents the real part and the imaginary part of the permittivity for the two oses in aqueous solution with a concentration of 100 g/L as well as the ones of De-Ionized

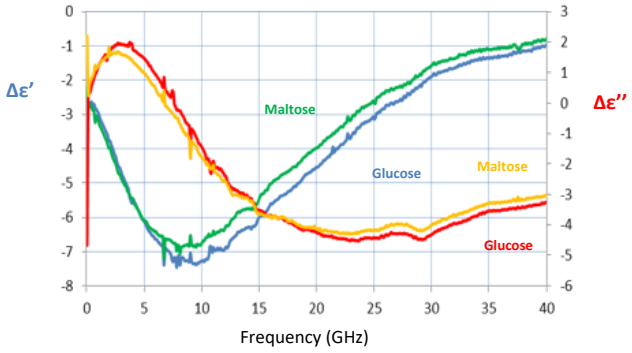

Fig. 3. Dielectric contrasts obtained for maltose and glucose in aqueous solution at a concentration of 100 g/l. The contrats based the real parts of permittivity are presented in blue and green, whereas the ones of the imaginary parts of the permittivity are given in yellow and red.

(DI) water versus frequency, from 40 MHz to 40 GHz. One may first notice the similarity of the dielectric characteristics of all these solutions under test. Moreover, the water ones present the highest values of permittivity, indicating its prevalent role in the molecular relaxation phenomenon. Finally, the two carbohydrates dielectric curves may not be distinguished from each other.

By applying the differential approach, i.e. by removing the contribution of the host water medium, which is then considered as a reference (the zero level on the ordinate axis), the discrimination of the glucose and the maltose in solution becomes possible. This is presented in Fig. 3 with the contrasts versus frequency of both permittivity parts, the real and imaginary ones.

B. Cells in suspension in their culture medium

The next example is related to cells in suspension in a traditional culture medium, which enables the cells to continue to live normally. This host medium contains salt as well as numerous nutrients.

Here, RF lymphoma cells at different concentrations in a Roswell Park Memorial Institute (RPMI) medium are investigated. Once again, if absolute values of permittivity are

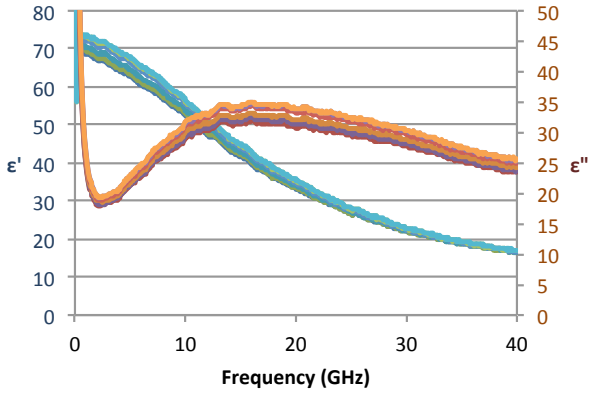

Fig. 4. Real and imaginary parts of the permittivity of RL lymphoma cells in their culture medium at several concentrations, 4, 8, 17, 35, 70 and 108 millions of cells per milliliter. The real parts are presented in blue and green, whereas the imaginary parts are in orange and brown.

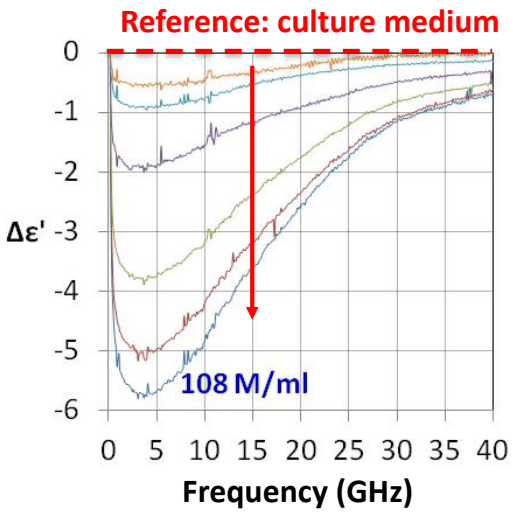

Fig. 5. Contrasts of the real part of the permittivity of RL lymphoma cells in their culture medium at several concentrations, 4, 8, 17, 35, 70 and 108 millions of cells per milliliter.

considered, the discrimination of the cells solutions is made difficult, as illustrated in Fig. 4.

On the other side, when the host medium contribution is removed, the different concentrations of cells are identifiable, as given in Fig. 5.

C. *Individual cells in their culture medium*

The dielectric evaluation of individual cells in their culture medium is also of interest. In that case, capacitive and conductive contrasts may be defined [5]. Fig. 6 presents the example of the spectrum of a single cell of the human monocytic cell line THP1 in RPMI culture medium supplemented by 10% of Fetal Calf Serum. The required sensitivity is very high for such a measurement, as a capacitive contrast of the order of tenths femtoFarad is requested. Such a dielectric characterization would not have been possible by considering only absolute electrical parameters. This example therefore demonstrates the power of employing a differential approach to enable the measurement of cells as small as 10 µm in diameter and with so weak electrical variation compared to its host medium.

A similar result is obtained if considering the conductive contribution of the cell. Fig. 7 presents the spectrum of the conductive contrast of the same single cell. A conductive contrast of the order of tens of microSiemens compare to the host liquid is extracted, which is also remarkable. One may

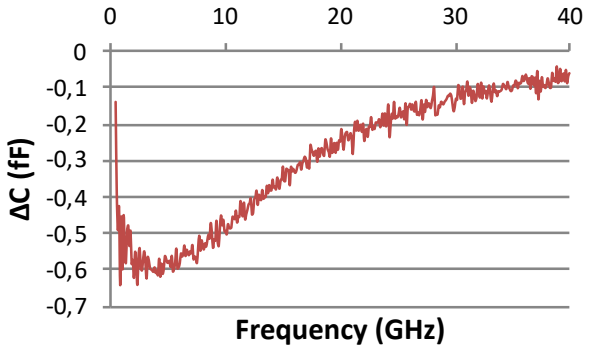

Fig. 6. Capacitive contrast of a single THP1 cells in its culture medium from 400 MHz to 40 GHz.

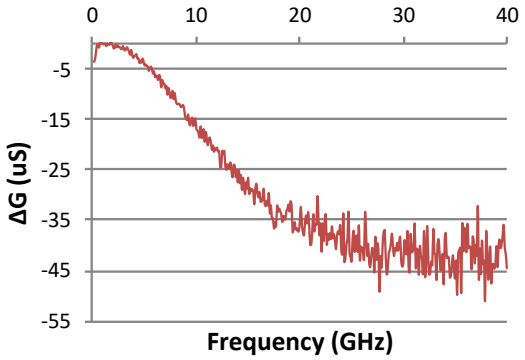

Fig. 7. Conductive contrast of a single THP1 cells in its culture medium from 400 MHz to 40 GHz.

also notice that even if the culture medium is ionic due to salt content, the microwave sensing is not screened by the presence of these ions and it does not prevent from extracting very low conductive contrast levels.

D. *Particular biological liquids*

As a last selected example, there may be also an interest to analyze the RF data based on the original S measured parameters of different liquids. Here, two constitutive liquids of eggs are studied, the albumen and the yolk. They are very sticky and viscous, which makes their characterization very tricky and complicated. Equation (2) is used to extract the contrast of the transmission coefficient, which is indicated in Fig. 7. Even if such liquids are delicate to handle in the millifluidic sensor [6], standard deviations obtained after ten successive measurements are reasonable.

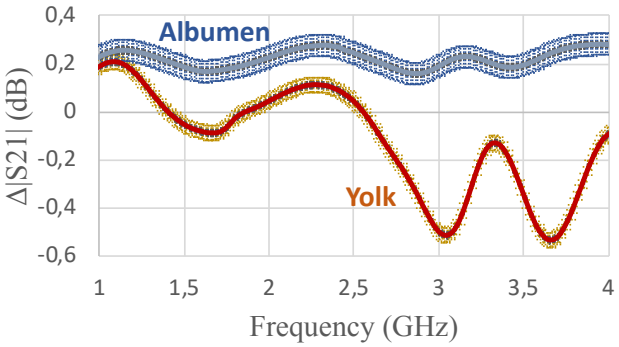

Fig. 8. Average values and standard deviations of the differences of modulus contrasts of measured S_{21} for albumen and yolk of a fertilized

V. Conclusions

In this paper, the use of a differential approach to remove the impact of the host medium while considering a biological liquid or element under test is presented and illustrated with different examples, from biomolecules in aqueous solution to cells in suspension or at the individual scale as well as for particular biological liquids (albumen and yolk of fertilized eggs). This enables to reach a sensitivity, which is not possible while only considering absolute electrical parameters. This approach is demonstrated with a calculation, which incorporates the suppression of the surrounding liquid compare to the bio-element under investigation. This imposes two measurements steps, the one of the bio-material of interest and the one of its host medium. It may also be performed directly by using differential sensors. This facilitates the

experimental procedure and the time of measurement. It however involves the requirement of more expensive and cumbersome equipment such as a multiport Vector Network Analyzer. The dielectric analyses in the microwave range of biological materials down to the scale of cells and even single cells is consequently possible, providing applications in molecular and cellular studies.

Finally, this approach may also be extended to organs and applied to in vivo measurements, leading to new perspectives of the dielectric spectroscopy to healthcare applications.

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