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Kyoto University
Dielectric Modeling of Biological Cells. Models and Algorithm

Akihiko IRIMAJIRI*, Toshinobu SUZAKI**, Koji ASAMI*** and Tetsuya HANAI***

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Shell models pertinent to the dielectric analysis of biological cells and organelles are described. These include: 1) one-shell, 2) two-abutting-shell, 3) multi-shell, 4) vesicle-inclusion, and 5) composite-shell models, with their spherical or ellipsoidal variations. A systematic procedure for depicting the dielectric behavior of each model is also presented on the basis of the theory of interfacial polarization. As an example, data from a suspension measurement on lymphoid cells are analyzed using model 5.

KEY WORDS: Dielectric dispersion/ Permittivity/ Conductivity/ One-shell model/ Multi-shell model/ Spherical cell/ Ellipsoidal cell/

I. INTRODUCTION

Biological cells, prokaryotic or eukaryotic, are demarcated from their environment by a diffusion barrier of lipidic nature, viz., the plasma membrane. In higher organisms, the cell develops within it a secondary structure of organelles, which are mostly membrane bounded, as is the case with the nucleus, mitochondria and endoplasmic reticulum.

In physical terms, all these membranes can be regarded as a shell dielectric that separates two more conducting aqueous compartments to form an interface on each side of the shell phase. This interface in turn becomes the site of charge accumulation or depletion (i.e., "polarization") depending on the electric field applied. When subjected to a.c. field, the degree of polarization across the interface varies with frequency. Thus, in cells and tissues, interfacial polarization is the major mechanism that is responsible for the dielectric relaxation phenomena we usually observe.

Impedance analysis of cells and tissues has long attracted biophysicists' attention partly because this method allows a noninvasive approach to the electrical properties of living cells. However, some workers, especially those who are electrically oriented, often end up with mere equivalent-circuit analyses, leaving the assignment of dielectric spectroscopic data behind. To some workers with a biomedical background, on the other hand, dielectric modeling and associated theories of interfacial polarization both seem too complicated to have a good command of these. Hence a systematic procedure should be desirable which is easy to apply in extracting the passive electrical prop-
Fig. 1. Zero-shell model: (a) Sphere of radius $R$ and equivalent homogeneous permittivity $\varepsilon_{eq}$; (b) ellipsoid of revolution with semiaxes $a$, $b$ and $c$ (= $b$), an ellipsoidal version of (a).

...erties, such as relative permittivity and conductivity, of the component phases that comprise a cell.

In this article, we summarize the dielectric models of our routine use and present some examples of the models' predicted behavior, followed by a program list with which to execute relevant calculations.

II. SUSPENSION EQUATIONS FOR THE TWO-PHASE SYSTEM

Complex relative permittivity $\varepsilon^*$ for a dilute suspension made up of medium ($\varepsilon_\infty$) interspersed with spherical particles (Fig. 1a) was formulated by Wagner, on the basis of the theory of interfacial polarization, as

$$\varepsilon^* = \varepsilon_\infty + \frac{2(1-\Phi)\varepsilon_\infty + (1+2\Phi)\varepsilon_p}{(2+\Phi)\varepsilon_\infty + (1-\Phi)\varepsilon_p}$$

(1)

where $\Phi$ is volume fraction.

For more concentrated suspensions, Hanai derived an extended version of Eq. (1), which is of the form:

$$\left(\frac{\varepsilon^* - \varepsilon_p^*}{\varepsilon_\infty^* - \varepsilon_p^*}\right) \left(\frac{\varepsilon_\infty^*}{\varepsilon^*}\right)^{1/\gamma} = 1 - \Phi$$

(2)

Combination of Eq. (1) or Eq. (2) with the expression for a particular model to be given in the following section enables calculation of $\varepsilon^*$ for the whole suspension of shelled spheres.

Suspension equations for ellipsoids (Fig. 1b) have been proposed by several authors. Among these, Sillars' extension of the Maxwell-Wagner theory to non-spherical particles makes the starting point for subsequent modifications. (For a didactic derivation of Sillars' equation, see e.g. Takashima.) Pertinent equations have been derived by Asami et al. for dilute systems and by Watanabe et al. for concentrated systems, to name but a few.
Fig. 2. Multi-shell model, spherical. The core of \( \varepsilon_i^* \) is surrounded by \( n \) concentric strata of \( \varepsilon_k^* \)'s and thicknesses \( d_k \)'s (\( k=1, 2, \ldots, n \)).

Fig. 3. One-shell model. (a) Sphere, (b) Ellipsoid of revolution.

III. MODELS FOR THE SUSPENDED PHASE

In the dielectric modeling of spherical cells, a sphere of concentric strata (Fig. 2), the number \( n \) of which is a parameter may be the basic model to start with. Fig. 3 shows the simplest possible versions. Hereafter, the spherical model (Fig. 3a) rather than the spheroidal one (Fig. 3b) will be focused on since mathematics would become too lengthy to give a full description to the latter.

Applying Maxwell's homogenization procedure\(^{8,19} \) one can write down the homogeneous permittivity of the "one-shell" model as

\[
\bar{\varepsilon}_i^* = \varepsilon_m \frac{2(1-v)\varepsilon^*_m + (1+2v)\varepsilon_i^*}{2+v\varepsilon^*_m + (1-v)\varepsilon_i^*}
\]  

with \( v=(1-d/R)^3 \). Likewise, \( \bar{\varepsilon}_i^* \) for a larger number of strata can be readily obtained through repeated applications of Eq. (3).

The model in Fig. 4 illustrates two abutting shells. This model applies to the case
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IMPEDANCE ANALYSIS PROGRAM

<table>
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<tr>
<th>Spherical Models</th>
<th>Ellipsoidal Models</th>
<th>Others</th>
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<tr>
<td>1:0-shell</td>
<td>1:0-shell</td>
<td>E: Cole-Cole</td>
</tr>
<tr>
<td>2:1-shell</td>
<td>2:1-shell</td>
<td>F: Data Display</td>
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<tr>
<td>6:5-shell/vesicles</td>
<td>1:1-shell/vesicles</td>
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<tr>
<td>8:7-shell/vesicles</td>
<td>2:2-shell</td>
<td></td>
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</table>

2-shell model
(Two abutting shells, 4 phases)

\[
\begin{align*}
\varepsilon_{\text{m}} & \quad \varepsilon_{\text{v}} \\
\varepsilon_{\text{mv}} & \quad \varepsilon_{\text{v}} \\
\end{align*}
\]

Fig. 4. Two-abutting shell model (right). The whole picture is a hard copy of "main menu" displayed upon selecting model #3 in the program "IMPEDANCE ANALYSIS mini".

![Fig. 4](image)

Fig. 5. Vesicle-inclusion model.

![Fig. 5](image)

Fig. 6. Triple-shell model with vesicle inclusions.

![Fig. 6](image)

where an overcoat- or an undercoat layer is closely attached to the membrane phase proper.

As previously reported by Irimajiri et al.\textsuperscript{11}, a multi-stratified sphere with \( n \) discrete shells in general gives rise to \( n+1 \) different relaxations, reflecting that the number of emerging relaxations corresponds to the number of interfaces involved.

(424)
Single shells containing membrane-bounded vesicles as a secondary suspension may be modeled as in Fig. 5, which we have named the “vesicle-inclusion” model. Although its theoretical basis is rather weak compared with the concentric shells described above, we attempt to define the $\varepsilon_r^*$ of this model by incorporating the results from a suspension equation (Eq. (1) or (2)) for vesicles into the parameter $\varepsilon_r^*$ in Eq. (3). In this calculation, we prefer Eq. (2) to Eq. (1) because intracellular vesicles are usually resident in a high volume concentration.

Figure 6 depicts a more realistic model for cells, in which the nucleus, demarcated by a double membrane system, resides at the center of cytoplasm that has been simplified to a vesicular suspension.

Besides these, many other models that allow size distribution for the suspended particles as well as cytoplasmic structures have already been developed in our laboratories. For brevity, however, these sophistications are omitted here.

IV. OUTLINE OF THE PROGRAM “IMPEDANCE ANALYSIS mini”

One may start this program by simply choosing an intended model or operation from the “menu” listed in Fig. 4 (left block). For each model, both the calculation conditions (i.e., frequency range, number of points per decade, type of mixture equation, etc.) and parameter values can be easily entered while referring to the symbols displayed as in Fig. 4 (right block). The results of calculations will then be visualized in a colored format of dispersion curves or of Cole-Cole plots. Up to eighth previous calculations are to be stored and hence superimposable on the display for ready comparison. Finally, analyses of experiment by curve fitting are also feasible if the data to be handled are fed through the format “FEK” (cf. “menu”), which means “frequency(F)/relative permittivity(E)/conductivity(K).” The program list is in the

Fig. 7. Dispersion curves predicted from the “multi-shell” model for: $n=1$ (---), 3 (-----), 5 (---), 7 (----), and 99 (--------). Calculations employed the following parameters:

$\varepsilon_1 = \varepsilon_3 = \cdots = \varepsilon_n = \varepsilon_{k}\text{macro}=8, \ k = k_1 = \cdots = k_{n\text{macro}} = 0.1 \text{ nS/cm},$

$\varepsilon_1 = \varepsilon_3 = \cdots = \varepsilon_{k}\text{macro}=78, \ k = k_1 = \cdots = k_{n\text{macro}} = 10 \text{ mS/cm},$

$d_1 = d_2 = \cdots = d_n = 5 \text{ nm}, \text{ and } R = 0.5 \mu\text{m}.
attached Appendix. This program, written in N88-BASIC (NEC), has been confirmed to run on NEC “PC-9801” series computers excepting PC-9801, PC-9801E, and PC-98LT.

V. EXAMPLE OF PREDICTED DIELECTRIC BEHAVIOR

As stated in Section III, the “multi-shell” model may represent a variety of shelled particles composed of concentric strata, so that it is of general interest to depict the model’s behavior predictable upon varying the number of shells involved in it. Figs. 7 and 8 show an example of such calculations where a dielectric shell and a conducting aqueous phase, both being of an identical thickness \( d = 5 \text{ nm} \), alternately build up to increase the number of strata towards filling up the core phase. Clearly, the “one-shell” model \( (n=1) \) traced a semicircle in the Cole-Cole plots (Fig. 8), while the final concentric structure \( (n=99) \) gave rise to a skewed-arc-like pattern indicative of the involvement of many relaxation times. The results of calculations for intermediate numbers of strata such as the “double-shell” \( (n=3) \) and “triple-shell” \( (n=5) \) models are also displayed in Fig. 8.

Figure 9 shows the behavior of the “vesicle-inclusion” model. In this calculation, we have chosen a thin-walled particle of \( 8 \mu \text{m} \) in diameter and changed the vesicle size \( R_v \), as a parameter, from \( 3 \mu \text{m} \) down to \( 0.1 \mu \text{m} \) with the intraparticulate volume fraction of those vesicles \( \phi_v \) fixed at 0.4234. Under such constraints, the case with \( R_v = 3 \mu \text{m} \) corresponds to the “double-shell” model, which showed a flattened arc in the complex permittivity plane plot (Fig. 9, top). With a decrease in the vesicle size (i.e., with an increase in the vesicle number), separation between two major relaxation frequencies, one being due to the outer shell and the other due to the vesicle membrane,

![Fig. 8. Cole-Cole plots for curves in Fig. 7. Line specifications, same as in Fig. 7.](image-url)
Dielectric Modeling of Biological Cells

Fig. 9 Cole-Cole plots for the dielectric behavior predictable from model 6 (one-shell/vesicles) or the "vesicle-inclusion model" (Fig. 5). Calculations employed the following parameters: $\varepsilon_r = \varepsilon_m = 8$, $\varepsilon_i = \varepsilon_i = 78$, $x_m = x_m = 0.1$ nS/cm, $x_i = x_i = 10$ mS/cm, $d_m = d_i = 5$ nm, $R = 4$ µm, $\phi_v$ (volume fraction of vesicles) = 0.4234, and $R_0 = 3$ (---), 1 (-----), 0.5 (---), 0.2 (--------), and 0.1 (............) µm.

Fig. 10. Dielectric behavior of cultured lymphoma cells (L5178Y) in suspension. Circles, observed; lines A-D, best-fit theoretical curves calculated using Eq. (1) and models A-D with appropriate parameter values.

became dominant, as shown in Fig. 9 (bottom).

The last example (Fig. 10) deals with simulation of the dispersion curves obtained from measurements with cultured lymphoma cells in suspension\(^{14}\). Here, model A refers to the "one-shell" or the simplest available model for living cells. Model B is the
"triple-shell", a version of the "multiple shells", whose inner double-shell is meant for
the nucleus. Models C and D are special versions of the "triple-shell with vesicle
inclusions", or the "composite cell" model. The "vesicles" in model C represent
mitochondria whose size (~0.6 \(\mu m\)) and cytoplasmic volume fraction \(\phi_c \approx 0.05\) were
both determined stereologically. In model D, the presence of vesicles smaller than the
mitochondria was also considered. The most sophisticated model has thus been shown
to mimic the dielectric behavior observed.

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Appendix

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