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On the Dielectric Properties of Poly(glutamic acid) in Aqueous Solution in the Frequency Range 10^7–10^{10} Hz

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ABSTRACT: The permittivity spectra of aqueous poly(glutamic acid) and adenosine triphosphate solutions have been measured by the dielectric time domain spectroscopy technique in the frequency range of 20 MHz to 5 GHz. No evidence of a reported dispersion in the 100-MHz region due to side-group motions in poly(glutamic acid) was found. It is demonstrated experimentally and shown theoretically that the premature truncation of time domain line shapes may introduce seeming dispersions, the frequency region of which will depend on the time window used.

1. Introduction

Dielectric spectroscopy is well established as a tool for studying proteins in solution.[1,2] The β-dispersion observed in the megahertz range has been attributed to molecular reorientation, and the dipole moment can be calculated from the corresponding dielectric increment. In addition, the dielectric relaxation times give information on the structure, the physical dimensions of the proteins being related to this relaxation time.

As model system for proteins, synthetic polypeptides have been intensively studied by using different physical techniques. The dielectric dispersion due to side-chain motion in polypeptides in dioxane solution has been investigated by the dielectric time domain spectroscopy (TDS) method.[3,4] Relaxation times in the range 0.02–0.5 ns were found and attributed to the rotation of C–O bonds in the side group. This line of work has recently been extended to a study by the TDS technique of the dielectric properties at high frequencies of poly(L-glutamic acid) (PGA) in aqueous solutions.[5] In this work two dispersions were reported, a dominant high-frequency dispersion due to water and a smaller dispersion in the region around 100 MHz. This latter dispersion was attributed to the motion of side-chain dipoles, the relaxation time being of order 2–3 ns.

The dielectric spectra of the PGA solutions reported by Mashimo et al.[5] fall in the frequency range of 10 MHz to 10 GHz. Aqueous solutions of poly(glutamic acid) have earlier been investigated at lower frequencies by bridge techniques. Takashima[6] studied the dielectric properties and their pH dependence of PGA solutions in the frequency range of 1 kHz to 1 MHz. The large dielectric increment observed was found to be larger for the coiled state than for the helical state, the transition from helix to coil state occurring around pH 6. Müller et al.[7] extended the frequency range to 100 MHz and found that the dielectric spectrum was characterized by two dispersions. The slower relaxation process had a relaxation time on the order of 10 μs, and the faster relaxation was on the order of 100 ns. The high-frequency relaxation was attributed to the diffusion process of counterions along subunits of the macromolecule. The low-frequency relaxation depended on the molecular weight and was explained as due to either counterion diffusion along the complete macromolecule or overall reorientation or possibly both effects. Nakamura and Wada[8] using a Fourier synthesized pseudorandom noise method studied aqueous PGA solutions in the frequency range 0.2–10 kHz. The large dielectric increment in the random coil state was confirmed by the specific increment near the value predicted by the counterion fluctuation mechanism. No distinct relaxation curves were observed in the helical state.

In the work by Müller et al.[7] the permittivity spectrum was measured at densely falling frequencies up to 100 MHz. It was found that the high-frequency limit of the permittivity was always equal to the static permittivity of water as is also seen from the spectrum presented in the paper. No dispersion was thus observed in the region between the water relaxation with a relaxation time 10 ps and the counterion relaxation of order 100 ns. This is in contrast to the results obtained in the recent time domain measurements,[5] where a clear dispersion is reported to fall in the range $\lesssim$100 MHz. In order to clarify the existence of such a dispersion and its possible use as a tool in studying the side-chain dynamics, we have investigated the high-frequency dielectric properties of some aqueous poly(glutamic acid) solutions of different pH values.

In this paper we report on the results from TDS measurements, which do not corroborate the existence of a side-group dispersion in the 100-MHz region. However, a seeming dispersion in this region may be introduced on truncation of time domain line shapes, and the possibility of such a false dispersion in a TDS spectrum is analyzed.

2. Materials

The sodium salt of poly(L-glutamic acid) (NaPGA) with an average molecular weight of 74 000 was obtained from Sigma.

A total of 66 mg of NaPGA was dissolved in 5 mL of water and dialyzed against double-distilled water for 7 days at 4 °C to remove the sodium ion. The solution was concentrated and the solvent changed to 0.1 M NaOH with repeated runs in a Centricon microconcentrator.

The concentrate was divided into three aliquots and the pH set to 4, 7, and 9, respectively, by adding 0.1 M NaOH and 0.1 M HCl to a final volume of 4.5 mL, giving a concentration of approximately 5 mg/mL of poly(glutamic acid) per sample.

3. TDS Measurements

In a dielectric time domain spectroscopy measurement the permittivity spectrum is determined by the study of the influence of the dielectric sample on the shape of a

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pulse propagating in a transmission line. Several TDS methods have been developed, which differ from each other in the arrangement of the sample. In one reflection method the sample terminates the open-ended transmission line, and the dielectric spectrum is obtained from the reflected pulse shapes.[9,10] An alternative is given by the total transmission method, in which the pulse transmitted through the sample is compared to the pulse transmitted through the empty sample cell.[11,12] In the present study both methods were employed; the spectra presented below were obtained by using the total transmission method.

In the total transmission method a Fourier transformation $F(\omega) = \int_{-\infty}^{\infty} f(t)\, e^{-i\omega t}\, \mathrm{d}t$ of the pulse $\nu(t)$ transmitted through the empty cell and the pulse $r(t)$ transmitted through the sample gives the transmission coefficient at a chosen frequency $T(\omega) = R(\omega)/V(\omega)$. The complex permittivity, ϵ^*, of the sample is then obtained from $T(\omega)$ by numerical solution of the transmission line equation

$$T(\omega) = \frac{(1-\rho^2)e^{-(i\omega l/c)(\sqrt{\epsilon^*}-1)}}{1-\rho^2 e^{-(i2\omega l/c)\sqrt{\epsilon^*}}} \tag{1}$$

Here $\rho = (1 - \sqrt{\epsilon^*})/(1 + \sqrt{\epsilon^*})$, c is the speed of light in free space, and l is the sample cell length.

The ideal transmission coefficient of eq 1 will be disturbed by deviations from ideal transmission line conditions, e.g., in cell design and connector mismatches. The influence of these sources of error can be reduced by measuring relative to some standard dielectric with a known permittivity spectrum. Preferably this spectrum should lie close to that of the unknown dielectric. The transmission coefficient $T_{\mathrm{ref}}(\omega)$ for the reference liquid can then be calculated from eq 1, and the unknown spectrum is obtained by solution of the equation

$$\frac{R(\omega)}{R_{\mathrm{ref}}(\omega)} = \frac{T(\omega)}{T_{\mathrm{ref}}(\omega)} \tag{2}$$

For a dielectric showing dc conductivity the transmitted pulse will not reach the final level of the incident step pulse $\nu(t)$. Instead, the asymptotic value of the time domain transmission coefficient is given by

$$t_\infty = \lim_{t\to\infty} \frac{r(t)}{\nu(t)} = \frac{1}{1 + \sigma l/2\epsilon_0 c} \tag{3}$$

Provided a sufficiently long time window is used to allow the complete relaxation of all dielectric processes, the time domain data can thus be used to directly determine σ.

The aqueous poly(glutamic acid) solutions show a high conductivity, distorting the pulse shapes too much to allow an accurate measurement relative to the empty cell, using eq 1. However, an appropriate reference liquid is easily prepared by adding potassium chloride to water and carefully adjusting the electrolyte concentration to a conductivity close to that of the poly(glutamic acid) solution, as judged from the final level of the transmitted pulse. At low salt concentrations the dielectric parameters of water will not be significantly influenced[13,14] and the dielectric spectrum of the reference electrolyte can be calculated from

$$\epsilon^*_{\mathrm{ref}} = \epsilon_{\infty\mathrm{w}} + \frac{\epsilon_{\mathrm{sw}} - \epsilon_{\infty\mathrm{w}}}{1 + i\omega\tau_{\mathrm{w}}} - \frac{i\sigma}{\omega\epsilon_0} \tag{4}$$

where ϵ_0 is the permittivity of a vacuum = 8.854×10^{-12} (A s)/(V m). The water parameters[15] at 20 °C are ϵ_{sw} = 80.2, $\epsilon_{\infty\mathrm{w}}$ = 5.6, and τ_{w} = 9.4 ps. Insertion of eq 4 into eq 1 will give the transmission coefficient $T_{\mathrm{ref}}(\omega)$ to be used in the solution of eq 2.

In order to ensure the applicability of the TDS measurement relative to an aqueous electrolyte of high conductivity, solutions of the sodium salt of adenosine triphosphate (ATP) were prepared and set to the same conductivity as the PGA solutions. ATP is known to show a dispersion in the 10^8–10^9-Hz region;[2,16] i.e., the resolution of such a dispersion serves as a critical test of the experimental method.

A necessary criterion for the characterization of a dispersion in TDS is that the pulse shapes are monitored during a time that is longer than the relaxation time studied. This may be checked already in the time domain, since the transmitted pulse will not have reached a steady final level if too short an observation time is used. As further discussed below, the difference $d(t)$ between the pulses transmitted through the sample and the reference, respectively, will show an exponential decay. The use of an ATP solution, with its short decay time, is then valuable as a check already in the time domain. An observation of an incomplete decay of $d(t)$ for an unknown of the same conductivity must then be due to dielectric decay processes and not to an experimental artifact inherent in the TDS measurement.

In the total reflection method the reflection coefficient S of the dielectric is analogously determined from the reflected line shape. The reflection coefficient for an open-ended cell of effective length l is given by

$$S(\omega) = \frac{R(\omega)}{V(\omega)} = \frac{(\rho + e^{-2i\omega l\sqrt{\epsilon^*}/c})e^{2i\omega l/c}}{1 + \rho e^{-2i\omega l\sqrt{\epsilon^*}/c}} \tag{5}$$

Here $R(\omega)$ and $V(\omega)$ are the transforms of the pulses reflected from the dielectric filled and empty cell, respectively, and the remaining symbols have the same meaning as those in eq 1. In analogy with the total transmission method, a conducting dielectric will give a reflected pulse which does not reach the final level of the incident pulse, the offset giving the conductivity σ. Also in this case a reduction of errors in the permittivity spectrum is achieved by using a potassium chloride solution as the reference liquid, with the unknown permittivity obtained by solving the equation

$$\frac{R(\omega)}{R_{\mathrm{ref}}(\omega)} = \frac{S(\omega)}{S_{\mathrm{ref}}(\omega)} \tag{6}$$

Here $R(\omega)$ and $R_{\mathrm{ref}}(\omega)$ are the Fourier transforms of the pulse shapes reflected from the unknown and reference samples, respectively.

With creation of the difference $V(\omega) - R(\omega)$ and the sum $V(\omega) + R(\omega)$, the solution of eq 5 can be reformulated,[9,10,17,18] giving ϵ^* expressed in these quantities. However, since all information lies in the original pulse transforms $R(\omega)$, it was preferred to directly obtain the spectrum from the numerical solution of eq 5.

A comparison of the two discussed TDS methods shows that the integrated pulse difference $\int d(t)\, \mathrm{d}t$, which is proportional to l, is larger for the reflection method compared to the transmission method for equal l. However, the reflection method requires small sample lengths since the solution of eq 5 is very sensitive to experimental errors in $S(\omega)$ and may diverge unless $|\omega l\sqrt{\epsilon^*}/c| < 1$. This is in contrast to the total transmission method where no such restraint exists, the length of the sample rather being limited by the requirement that the modulus of the transmission coefficient does not become too small. In the present measurements 5- and 10-mm cell lengths were used in transmission measurements, and a 2.25-mm cell

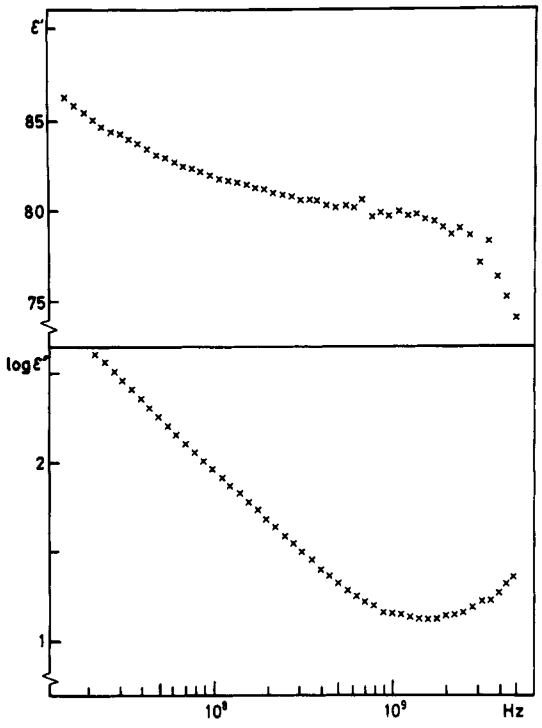

Figure 1. Permittivity spectrum of an aqueous poly(glutamic acid) solution (5 mg/mL) at pH 7.

length was used for the reflection measurements. This can be compared with an effective length 0.28 mm for the reflection measurements in ref 5.

4. Results

Figure 1 shows the permittivity spectrum for a poly(glutamic acid) solution at pH 7. The spectrum is an average of 10 independent spectra obtained by alternating filling of the 10-mm sample cell with the reference electrolyte solution and the poly(glutamic acid) solution. The observation time window for the pulse shapes was 50 ns. The Fourier transforms were calculated at 50 frequencies between 20 MHz and 5 GHz using the Samulon formula.[19] The observation of the difference in pulse shape $r_{ref}(t) - r(t)$ clearly showed an incomplete decay, indicating the presence of dielectric relaxation processes, which were not completed within the 50-ns time window. Under these circumstances the lower frequency limit was set to 20 MHz, i.e., the inverse of the observation time window. The upper frequency limit is determined by the strong attenuation of the transmitted signal at frequencies above a few gigahertz for the long sample cell length used. The spectrum in Figure 1 shows, besides the high-frequency water dispersion, the trailing permittivity of a strong dispersion at lower frequencies than the available range.

Figure 2 shows the permittivity spectrum of an ATP solution set to the same conductivity as the poly(glutamic acid) solution and obtained under identical experimental conditions. In contrast to the poly(glutamic acid) solution, the permittivity here levels off at low frequencies. A strong dispersion can be discerned in the ϵ' spectrum centered around 400 MHz. Subtraction of the dc conductivity contribution brings out the corresponding peak also in the plot of the dielectric loss factor ϵ''. The successful resolution of this dispersion demonstrates the applicability of the total transmission TDS method also on these highly conducting solutions. The ATP solutions were also studied by using the total reflection method, giving the same results, however, with a lower high-frequency limit in this case as discussed above. The full drawn line in Figure 2 shows the fit to a theoretical spectrum calculated as a sum of two Debye dispersions

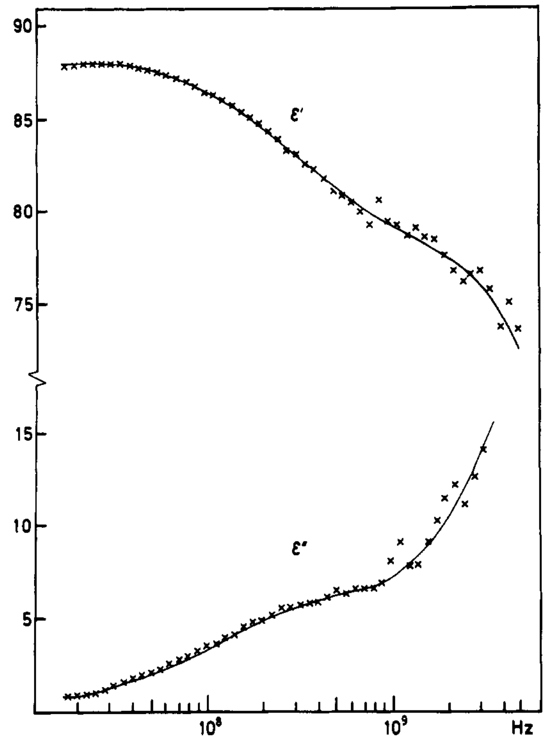

Figure 2. Permittivity spectrum of an ATP solution set to the same conductivity as in Figure 1. The conductivity contribution to ϵ'' has been subtracted. The full line shows the theoretical spectrum using a two relaxation time model function.

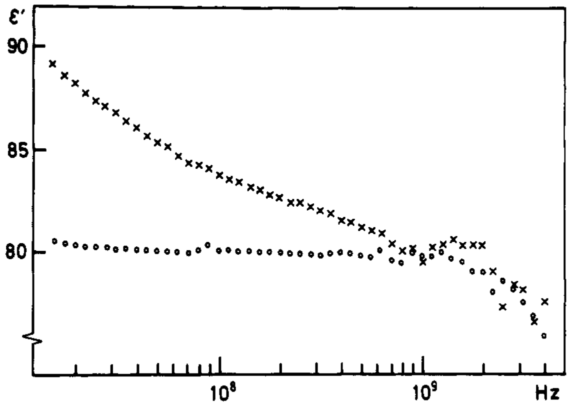

Figure 3. Permittivity spectrum ϵ' for poly(glutamic acid) solutions at pH 4 (○) and pH 9 (×).

$$\epsilon^* = \frac{\epsilon_s - \epsilon_1}{1 + i\omega\tau_1} + \frac{\epsilon_1 - \epsilon_\infty}{1 + i\omega\tau_2} + \epsilon_\infty \tag{7}$$

with the parameters $\epsilon_s = 88.3$, $\epsilon_1 = 77.9$, $\epsilon_\infty = 5.6$, $\tau_1 = 0.51$ ns, and $\tau_2 = 9.4$ ps.

Figure 3 shows the real part of the permittivity ϵ^* of poly(glutamic acid) solutions of pH 9 and pH 4, respectively, also obtained from measurements with the total transmission method. The onset of the high-frequency dispersion > 1 GHz is found also in these solutions; however, the lower frequency dispersion features have virtually disappeared in the pH 4 solution. This result is also obtained by measurements using the total reflection method.

Unfortunately, not sufficient information is given on the experimental conditions used in ref 5 to ascertain the cause of the discrepancy with the results in Figures 1 and 3. The lower limit of the permittivity spectrum presented

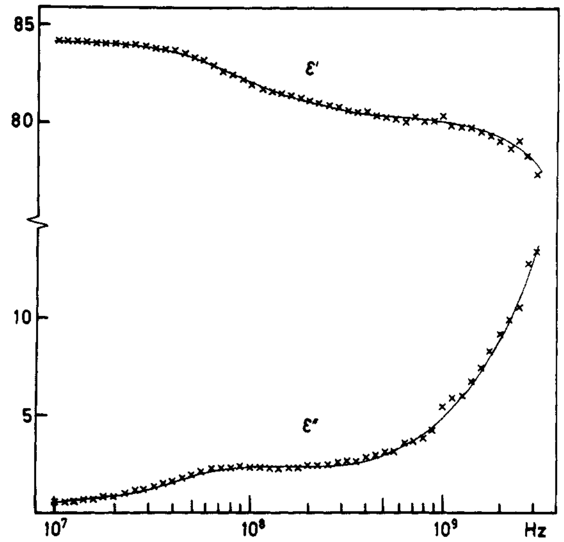

Figure 4. Permittivity spectrum from the PGA solution in Figure 1 on truncation of time domain pulse shapes after 10 ns. A conductivity contribution with $\sigma = 0.49$ S/m has been subtracted. The full line shows a model spectrum with two relaxation times $\tau_1 = 9.4$ ps and $\tau_2 = 1.7$ ns.

in ref 5 is 10 MHz; i.e., a time window of order 100 ns is required in the measurement. No time window is given. However, in a previous paper on time domain methods for aqueous solutions, similar poly(glutamic acid) spectra were presented showing a dispersion in the 100-MHz range, these spectra being obtained with a time window <20 ns.[10]

In order to check the influence of changing the time window T_w, the poly(glutamic acid) solutions of pH > 4 were studied with both the transmission and reflection methods, with T_w ranging from 10 to 100 ns, the spectra being evaluated from frequencies below T_w^{-1}. In each case a seeming dispersion was obtained, the position of which was dependent upon the time window used. For longer time windows this dispersion falls at lower frequencies, shifting to higher frequencies with the shortening of the observation time T_w.

The result of the premature truncation of the time domain data underlying Figure 1 is shown in Figure 4. Here the Fourier transforms were evaluated from the same registered pulse shapes, but truncated after 10 ns, using the same frequency range of 20 MHz to 5 GHz. A seeming dispersion in ϵ' is introduced, falling at frequencies of around T_w^{-1}. At higher frequencies the spectra of Figures 1 and 4 agree as expected. In Figure 4 is also shown the ϵ'' spectrum corrected for a dc contribution $\sigma/\omega\epsilon_0$. A seeming loss peak is introduced in the same frequency range around T_w^{-1}.

The full line shows a theoretical spectrum calculated from a two-Debye relaxation time model function with $\tau_1 = 9.4$ ps and $\tau_2 = 1.7$ ns, respectively. The same results as in Figure 4 are obtained also on the premature truncation of the pulse shapes from the reflection measurements.

The spectra of Figures 1 and 4 give an experimental demonstration of the introduction of a seeming dispersion in conductive dielectrics on the premature truncation of TDS line shapes from a low-frequency relaxation process. This dispersion can easily be mistaken as due to a dipolar relaxation process, as further theoretically analyzed in the following section.

5. Theoretical Analysis

For a total transmission measurement with a thin sample, i.e., frequencies for which $|\omega l\sqrt{\epsilon^*}/c| \ll 1$, the transmission coefficient, eq 1, can be expanded to

$$T_{\text{thin}}(\omega) \simeq 1 + \frac{i\omega l}{2c} - \frac{i\omega l}{2c}\epsilon^* \qquad (8)$$

If we observe the difference $d(t)$ between the pulse transmitted through the empty cell $\nu(t)$ and the pulse transmitted through the sample $r(t)$ and solve for the permittivity, we obtain

$$\epsilon^*(\omega) = \frac{2cD(\omega)}{i\omega l V(\omega)} + 1 \qquad (9)$$

A series expansion of the reflection coefficient $S(\omega)$ under the thin sample condition similarly gives

$$\epsilon^*(\omega) = \frac{cD(\omega)}{2i\omega l V(\omega)} + 1 \qquad (10)$$

That is, the transform of the pulse difference $D(\omega)$ in both cases gives the permittivity. The following analysis is then valid for both TDS methods.

The decay function $\phi(t)$ of the dielectric gives the normalized decay of the polarization when a steady macroscopic electric field is removed from the medium. The permittivity is related to $\phi(t)$ by the Fourier transform[20]

$$\frac{d\phi}{dt} = -\frac{1}{2\pi}\int_{-\infty}^{\infty} \frac{\epsilon^* - \epsilon_\infty}{\epsilon_s - \epsilon_\infty} e^{i\omega t}\, d\omega \qquad (11)$$

with the inverse transform

$$\frac{\epsilon^* - \epsilon_\infty}{\epsilon_s - \epsilon_\infty} = \int_0^{\infty} -\dot{\phi} e^{-i\omega t}\, dt \qquad (12)$$

Insertion of eq 9 into eq 11 and deletion of δ functions gives

$$\frac{d\phi}{dt} = -\frac{1}{2\pi}\int_{-\infty}^{\infty} \frac{2cD(\omega)e^{i\omega t}\, d\omega}{(\epsilon_s - \epsilon_\infty)i\omega l V(\omega)} \qquad (13)$$

If the incident pulse $\nu(t)$ is a perfect step pulse $\nu_0 U(t)$ ($U(t) = 0$ for $t < 0$, $U(t) = 1$ for $t > 0$), its transform is given by $\nu_0/i\omega$. Insertion into eq 13 gives

$$\frac{d\phi}{dt} = -\frac{2cd(t)}{(\epsilon_s - \epsilon_\infty)l\nu_0} \qquad (14)$$

That is, the difference in the time domain between the two pulses, $d(t)$, will directly give information on the decay function.

In a case where the total permittivity is given by a sum of two Debye dispersions and a dc conductivity contribution, the total permittivity can be written

$$\epsilon^* - \epsilon_\infty = \frac{\Delta\epsilon_1}{1 + i\omega\tau_1} + \frac{\Delta\epsilon_2}{1 + i\omega\tau_2} - \frac{i\sigma}{\omega\epsilon_0} \qquad (15)$$

Here $\Delta\epsilon_i$ is the dielectric increment of the dispersion and τ_i the corresponding relaxation time. Insertion into eq 11 gives

$$-\frac{d\phi}{dt} = \frac{\Delta\epsilon_1 e^{-t/\tau_1}}{(\epsilon_s - \epsilon_\infty)\tau_1} + \frac{\Delta\epsilon_2 e^{-t/\tau_2}}{(\epsilon_s - \epsilon_\infty)\tau_2} + \frac{\sigma}{\epsilon_0(\epsilon_s - \epsilon_\infty)}U(t) \qquad (16)$$

for $t > 0$.

A comparison with eq 14 gives for $d(t)$

$$d(t) = \frac{l\nu_0\Delta\epsilon_1 e^{-t/\tau_1}}{2c\tau_1} + \frac{l\nu_0\Delta\epsilon_2 e^{-t/\tau_2}}{2c\tau_2} + \frac{\sigma l\nu_0}{2c\epsilon_0}U(t) \qquad (17)$$

The pulse difference $d(t) = \nu(t) - r(t)$ will thus consist of two exponential terms, characterized by the relaxation times, plus a constant that depends on the conductivity. For an aqueous solution where τ_1 is the water relaxation time of order 10 ps the pulse shapes will after a sufficiently long time window be determined by the low-frequency dispersion and the conductivity.

If in a measurement the pulse shapes are truncated at time T_w, before the slow relaxation process τ_2 has reached an end, the experimentally observed pulse shape will be

$$d_0(t) = d(t) + \frac{l\nu_0\Delta\epsilon_2}{2c\tau_2}(e^{-T_w/\tau_2} - e^{-t/\tau_2})U(t - T_w) \quad (18)$$

That is, a tail is added to the correct pulse shape. A Fourier transformation of $d_0(t)$ into the frequency domain according to eqs 12 and 14 then leads to

$$\epsilon^* = \int_0^\infty \frac{2cd_0(t)\, e^{-i\omega t}}{l\nu_0}\, dt + \epsilon_\infty = \frac{\Delta\epsilon_1}{1 + i\omega\tau_1} + \frac{\Delta\epsilon_2}{1 + i\omega\tau_2} + \epsilon_\infty - \frac{i\sigma}{\omega\epsilon_0} + \frac{\Delta\epsilon_2 e^{-T_w/\tau_2}e^{-i\omega T_w}}{i\omega\tau_2(1 + \omega\tau_2)} \quad (19)$$

The permittivity spectrum calculated from the truncated pulse shapes will thus include an extra frequency-dependent term besides the two true dispersions.

If the conductivity is deduced from the pulse levels at the end of the observation time window, a premature truncation of the pulse shapes will lead to an apparent conductivity σ' that is higher than the true dc conductivity σ. A truncation of line shapes at time T_w gives an apparent conductivity

$$\sigma' = \sigma + \frac{\Delta\epsilon_2\epsilon_0 e^{-T_w/\tau_2}}{\tau_2} \quad (20)$$

A subtraction of this conductivity contribution from the total permittivity given in eq 19 finally leads to the following apparent dipolar permittivity spectrum

$$\epsilon^*_{\mathrm{app}} = \frac{\Delta\epsilon_1}{1 + i\omega\tau_1} + \frac{\Delta\epsilon_2}{1 + i\omega\tau_2} + \epsilon_\infty - \frac{i(\sigma - \sigma')}{\omega\epsilon_0} + \frac{\Delta\epsilon_2 e^{-T_w/\tau_2}e^{-i\omega T_w}}{i\omega\tau_2(1 + i\omega\tau_2)} \quad (21)$$

Equation 21 is readily programmed to calculate the spectrum for different values of the parameters.

Figure 5 shows the theoretical spectrum calculated from eq 21 for a model aqueous solution with a truncated low-frequency dispersion. In this model calculation the dielectric parameters were set to $\epsilon_\infty = 5$, $\Delta\epsilon_1 = 75$, $\tau_1 = 10$ ps, $\Delta\epsilon_2 = 400$, $\tau_2 = 50$ ns, and $\sigma = 0.5$ S/m. The influence of changing the time window T_w is seen by comparing parts a and b of Figure 5 in the former $T_w = 10$ ns and in the latter $T_w = 5$ ns. In both spectra a seeming dispersion is introduced falling in the frequency region of the inverse of the time window T_w^{-1}.

Equation 21 and the spectra in Figure 5 are based on the assumption of a slow relaxation with a single relaxation time. A distribution of relaxation times in this low-frequency dispersion will broaden the features also for the seeming dispersion around T_w^{-1}.

6. Conclusions

Dielectric time domain spectroscopy is a fast and accurate method of measuring permittivity spectra over a wide frequency range. A useful feature of the method is the ability to study also liquids with a high dc conductivity. However, the establishment of the completion of the pulse shapes in these circumstances becomes more difficult, and it is necessary that great care is exercised in extension of the calculated spectra to lower frequencies. In the presence of low-frequency dispersions the premature truncation of line shapes will introduce a false dispersion, the apparent relaxation time of which will depend on the truncating time window used.

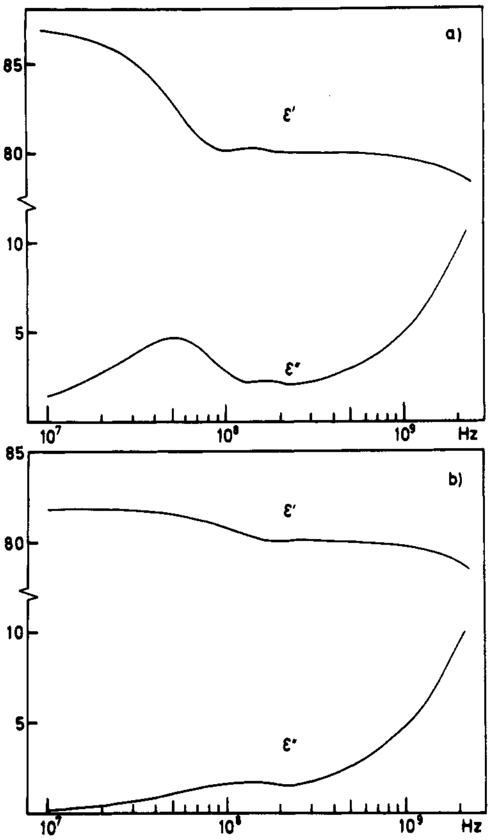

Figure 5. Model permittivity spectra calculated from eq 21 with parameters $\epsilon_\infty = 5$, $\Delta\epsilon_1 = 75$, $\tau_1 = 10$ ps, $\Delta\epsilon_2 = 400$, $\tau_2 = 50$ ns, and $\sigma = 0.5$ S/m. The time window is (a) $T_w = 10$ and (b) $T_w = 5$ ns, respectively.

For the poly(glutamic acid) solution studied at pH 7 and pH 9, i.e., in the coil state, the high-frequency tail of the dispersion due to counterion diffusion processes extends into the frequency range > 20 MHz. These dispersion features virtually disappear at pH 4, i.e., on the helical state of the macromolecule, in agreement with earlier observations at lower frequencies. The premature truncation of the time domain line shapes introduced a seeming dispersion, the position of which was dependent upon the time window used. This effect may well explain the dielectric relaxation processes with a relaxation time $\tau = 2$–3 ns recently reported, which have been attributed to side-group motions in the macromolecule.

References and Notes

(1) Oncley, J. L. In *Protein, Amino Acids and Peptides*; Cohn, E. J., Edsall, J. T., Eds.; Reinhold: New York, 1943.
(2) Grant, E. H.; Sheppard, R. J.; South, G. P. *Dielectric Behaviour of Biological Molecules in Solution*; Oxford University Press: Oxford, 1978.
(3) Nakamura, H.; Mashimo, S.; Wada, A. *Macromolecules* **1981**, *14*, 1698.
(4) Yagihara, S.; Nozaki, R.; Mashimo, S.; Hikichi, K. *Macromolecules* **1984**, *17*, 2700.
(5) Mashimo, S.; Ota, T.; Shinyashiki, N.; Tanaka, S.; Yagihara, S. *Macromolecules* **1989**, *22*, 1285.
(6) Takashima, S. *Biopolymers* **1963**, *1*, 171.
(7) Müller, G.; van der Touw, F.; Zwolle, S.; Mandel, M. *Biophys. Chem.* **1974**, *2*, 242.
(8) Nakamura, H.; Wada, H. *Biopolymers* **1981**, *20*, 2567.

(9) Cole, R. H.; Berberian, J. G.; Mashimo, S.; Chryssikos, G.; Burns, A.; Tombari, E. *J. Appl. Phys.* **1989**, *66*, 793.
(10) Mashimo, S.; Umehara, T.; Ota, T.; Kuwabara, S.; Shinyashiki, N.; Yagihara, S. *J. Mol. Liq.* **1987**, *36*, 135.
(11) Gestblom, B.; Noreland, E. *J. Phys. Chem.* **1977**, *81*, 781.
(12) Gestblom, B.; Elmgren, H. *Chem. Phys. Lett.* **1982**, *90*, 412.
(13) Winsor, P.; Cole, R. H. *J. Phys. Chem.* **1985**, *89*, 3775.
(14) Kaatze, U. *J. Phys. Chem.* **1987**, *91*, 3113.
(15) Kaatze, U.; Uhlendorf, V. *Z. Phys. Chem. (Frankfurt am Main)* **1981**, *126*, 151.
(16) Sheppard, R. J. *Annu. Rep. Conf. Insul. Dielectr. Phenom.* **1978**, 278.
(17) Cole, R. H. *J. Phys. Chem.* **1975**, *79*, 1469.
(18) Cole, R. H.; Mashimo, S.; Winsor, P. *J. Phys. Chem.* **1980**, *84*, 786.
(19) Samulon, H. A. *Proc. IRE* **1951**, *39*, 175.
(20) Williams, G. *Chem. Rev.* **1972**, *72*, 55.

Registry No. PGA (homopolymer), 25513-46-6; PGA (SRU), 24991-23-9; ATP, 56-65-5.