# PASSIVE ELECTRICAL PROPERTIES OF MICROORGANISMS

## I. CONDUCTIVITY OF *Escherichia coli* AND *Micrococcus lysodeikticus*

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ABSTRACT Effective conductivities are reported for the bacteria *Escherichia coli* and *Micrococcus lysodeikticus* over a range of environmental conductivity. The apparent conductivities of the organisms can be explained in terms of the properties of the cell wall. At low conductivities of the environment, the conductivtiy of the cell appears to be dominated by the counterions of the fixed charge of the cell wall. At higher conductivities of the suspending medium, evidence suggests that ions from the environment invade the cell wall causing an increase in the effective conductivity of the cell so that it takes on values roughly proportional to that of the environment. The model points to the usefulness of dielectric techniques in studies of the properties of intact cell walls.

A variety of electrical measurements has been used in the study of microorganisms. Microscope electrophoresis has yielded information about the outer surface of the cell (Brinton, 1959, James, 1957). Studies in which adsorption of ions is monitored by electrophoretic mobility of cells have assisted in speculations about the electrical and chemical nature of the cell surface (Davies, 1956, Haydon, 1962). Measurements of dielectric dispersion (1 to 100 megacycles/sec.) in suspensions of bacteria have provided information about the cell membrane and the conductivity of the cytoplasm of intact cells (Fricke, 1956, Pauly, 1962). In each case, the electrical technique provides a non-destructive test of the intact, living cell.

It is here proposed that information about the region defined as the cell wall can be obtained from measurements of the low frequency (10 to 10,000 CPS) conductivity of microorganisms. The first evidence of the importance of the electrical properties of the cell wall is found in studies by Curtis and Cole (1937) of the

conductivity of the algal cell, *Nitella*. Their work demonstrated that the apparent high conductivity of this organism could be traced to the cell wall. In this report the possibilities and problems are illustrated by two bacteria, *E. coli* and *M. lysodeikticus*. Both have effective conductivities which are high enough to be measured under a variety of environmental conditions. Differences in the conductivities of the two organisms suggest different properties for their cell walls.

## EXPERIMENTAL PROCEDURE

Values for the conductivity of the bacteria were determined from measurements of suspensions of the cells in aqueous solutions of electrolytes. To compute the effective, homogeneous conductivity of the bacteria it is necessary to know the conductivity of the suspension and the suspending fluid and the volume fraction of the suspension occupied by the bacteria.

*Growth and Preparation of Samples.* The organisms studied were *E. coli* Strain B and *M. lysodeikticus*.[1] *E. coli* was grown on a pilot-plant scale in nutrient broth at 37°C. The axis ratio of the cells obtained in this manner averaged about 1.5:1. The resulting product was harvested and stored at −20°C until used for measurements. Special studies demonstrated that neither cell volume nor conductivity were effected by a single freezing and thawing. *M. lysodeikticus* was grown in shaken flasks of trypticase soy broth at 33°C, harvested at 48 hours and used fresh. These cells are roughly spherical. The maximum dimension of both organisms is slightly in excess of 1 $\mu$.

The bacteria were washed twice in distilled water before use. Viability of the material was not affected by procedures used in the preparation of the samples.

*Bacterial Cell Volume.* The method for determination of bacterial cell volume was essentially that described by Conway and Downey (1950) and Black and Gerhardt (1962). In these measurements advantage is taken of the observation (Gerhardt, 1961, 1964) that very large molecules are excluded by the entire cell including the cell wall. Hence the dextran (mol. w., 150,000) inaccessible volume is roughly equivalent to the total cell volume. On the other hand inorganic phosphate (Mitchell, 1956) and sucrose (Gerhardt, 1961) are excluded by the cytoplasmic membrane but mix freely with the water of the cell wall. The difference between the dextran inaccessible volume and the phosphate or sucrose inaccessible volume is a measure of the water space of the cell wall. For this work sucrose and dextran concentrations were determined gravimetrically or from measurements of index of refraction in a differential refractometer. Analysis for phosphate was by the Friske-Subba row method as modified by Boltz and Mellon (1947). Precautions were taken to avoid errors in the analysis which might have been introduced by the presence in the samples of leakage products from the cells.[2]

Volume data for the two organisms as a function of environmental conductivity (or osmolality) are summarized in Fig. 1. Volumes are given relative to the dry weight of the cells. The reference weights were determined by drying washed samples of cells for

[1] Extensive measurements were also carried out on *Serratia mercescens* Strain 8 UK. The results were similar to those obtained for *E. coli* and to save space will not be reported in this paper.

[2] An internal report describing in somewhat greater detail these and other procedures used in this work and including tabular data needed for the solution of equation (1) can be supplied to the interested reader.

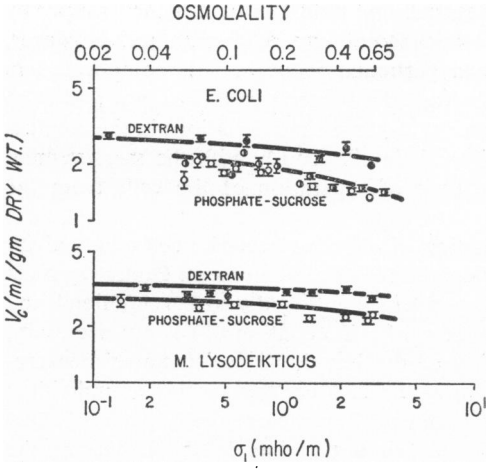


FIGURE 1 Volume of *E. coli* and *M. lysodeikticus* as a function of osmolality and conductivity of the suspending fluid. Closed circles (●) are dextran as a tracer, half closed circles (◑) are sucrose as a tracer, open circles (○) are phosphate as a tracer.

24 hours at 100°C. For *E. coli* the phosphate and sucrose data are plotted separately. In neither organism was there an experimentally significant difference between the phosphate and sucrose inaccessible volume. The agreement between volumes available to these two solutes combined with the previously referenced observations of Mitchell and Gerhardt supports the conclusion that these tracers enter the water space of the cell wall but do not penetrate the membrane. The phosphate and sucrose data for *M. lysodeikticus* have been averaged together.

*Electric Conductivity.* The conductivity $\sigma$ of a suspension of randomly oriented speroids with conductivity $\sigma_2$ in a suspending medium with a conductivity $\sigma_1$ was given by Fricke (1924) as

$$\frac{\sigma - \sigma_1}{\sigma + x\sigma_1} = p\,\frac{\sigma_2 - \sigma_1}{\sigma_2 + x\sigma_1} \tag{1}$$

where $p$ is the volume concentration of suspended particles and $x$ is a number very nearly equal to 2 for the conditions of this investigation.[3]

To measure the effective conductivity of bacteria as a function of conductivity of environment, suspensions of cells were prepared by adding NaCl solutions to soft pellets of bacteria which had been obtained under standard centrifuging conditions and for which information on cell volume had been obtained by the methods described above. Samples of the suspension were placed in conductivity cells[4] and their conductance measured at a frequency of 1592 CPS with a Wayne Kerr universal bridge (Wayne Kerr Lab., Ltd., Chessington, Surrey, England).[5] The measured conductance is proportional

[3] The parameter $x$ is a slowly varying function of the ratio $\sigma_2/\sigma_1$ and the axis ratio of the spheroid. For the case of *E. coli* it takes on values between 1.90 and 1.95 while for *M. lysodeikticus* it is always 2.00.

[4] A set of four conductivity cells were mounted together in a lucite block. Each cell consists of a pair of parallel, platinized, platinum electrodes approximately 4 mm in diameter mounted at the ends of a cylindrical, lucite tube. Separation of the electrodes was adjusted to give a cell constant of $10^3$ $m^{-1}$. All measurements were made at 29°C.

[5] The Wayne Kerr bridge is a transformer ratio arm bridge for the frequency range of 20 to 20,000 CPS with an accuracy of ±0.2 per cent and a discrimination of the order of 0.02 per cent.

*(Footnote continued on following page.)*

to $\sigma$. In principle, the conductivity $\sigma_1$ of the suspending fluid can be obtained simply by centrifuging the suspension to remove cells and sampling the supernatant. However, obtaining a value of $\sigma_1$ that corresponds to a particular value of $\sigma$ is complicated in certain cases by the fact that leakage of ions from the bacteria causes both $\sigma$ and $\sigma_1$ to change with time. To minimize errors connected with these time variations in the conductivity, a sample of the suspension was centrifuged simultaneously with the measurement of $\sigma$. The value of $\sigma$ corresponding to the time of separation of the cells from the supernatant was used in the calculations.

The accuracy of this method for determination of effective bacterial cell conductivity depends to a great extent upon the particular conditions of measurement. Under the most favorable conditions, *i.e.*, when the leakage problem mentioned above is minimal and when $\sigma_1$ approaches $\sigma_2$ so that a precise knowledge of $p$ in equation (1) is not necessary, the accuracy depends simply on the precision of the individual conductance measurements. In this case, the $\sigma_2$ values are accurate to within $\pm$ 5 per cent. Although individual conductance measurements are accurate to within $\pm$ 2 per cent, the leakage of electrolytes at the extremes of the range of $\sigma_1$ adds an uncertainty in the choice of the proper $\sigma$, $\sigma_1$ pair. This may increase the effective error in these values to the order of $\pm$5 per cent. From the nature of equation (1), when the ratio $\sigma_1/\sigma_2$ is of the order of 10, a 5 per cent error in either $\sigma$, $\sigma_1$, or $p$ is magnified to an error of approximately 30 per cent in $\sigma_2$. Errors of this magnitude may be found in the *E. coli* data at the highest values of $\sigma_1$.

Some day to day variations in the electrical properties of the bacteria themselves have been observed. This was more evident with *M. lysodeikticus*, which was grown on a small batch basis for each experiment, than with *E. coli* which was prepared on a pilot-plant scale and small fractions of the large batch removed from frozen storage for each series of measurements. Values of $\sigma_2$ at low $\sigma_1$ for *M. lysodeikticus* range from 0.15 mho/m to as high as 0.30 mho/m in rare cases. However, the lower value is considered more nearly characteristic of the organism.

Electrophoretic mobilities of the bacteria were measured by the microscope electrophoresis method which has been described by James (1957) and Brinton and Lauffer (1959) and many others.

## EXPERIMENTAL RESULTS

From observed values of $p$, $\sigma$, and $\sigma_1$, equation (1) gives the effective, homogeneous conductivity $\sigma_2$ of the suspended particle. Values of $\sigma_2$ for *E. coli* and *M. lysodeikticus* are presented in Fig. 2 for a range of values of the conductivity of the environment. There is no intent by this presentation to imply that bacteria are in fact homogeneous conductors. On the contrary, it appears more probable that the observed, effective conductivity is controlled entirely by the outer shell (cell wall). However, to go beyond values of $\sigma_2$ requires some speculation about the mechanisms involved. For the moment the effective homogeneous conductivity is a convenient

*(Footnote concluded.)*
This bridge has proven sensitive enough to provide crude estimates of the dielectric constant of suspensions of these bacteria in the range 200 to 20,000 cps. In the dielectric constant measurements, a large cell was used to minimize stray field errors and the variable electrode spacing technique (Schwan, 1961) was used to eliminate the effect of electrode polarization.

quantity to use for some preliminary observations and comparisons. First, it will be noted that the difference in conductivity between the two bacteria is striking—particularly at low $\sigma_1$. This difference is a qualitative one in the sense that after washing and resuspension in water, *M. lysodeikticus* invariably has a higher conductivity than its environment; whereas, the conductivity of *E. coli* is always less than that of the surrounding medium even though it is washed as many as 6 to 10 times in an attempt to reduce the environmental conductivity. Second, the conductivity of both organisms is high by comparison with that reported for non-biological,

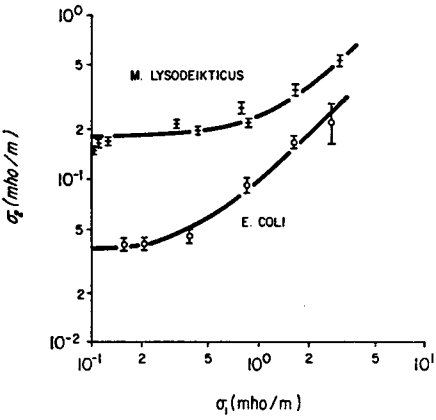


FIGURE 2 Effective, homogeneous conductivity of *E. coli* and *M. lysodeikticus* as a function of the conductivity of the suspending fluid. Solid curves are equation (9) with the constant $A$ chosen for best fit of the observed data.

colloidal particles of similar size. Table I compares glass and polystryrene spheres with the two bacteria under roughly the same environmental conditions. The results for glass and polystyrene can be explained as surface conductance phenomena, *i.e.*, the observed, effective, homogeneous conductivity can be predicted from the surface charge density as determined by electrophoresis measurements. To explore this as a possible explanation for the observations with bacteria, the electrophoretic mobility

TABLE I

EFFECTIVE CONDUCTIVITY OF COLLOIDAL PARTICLES

The Conductivity of the suspending medium is approximately 0.10 mho/m in each case.

| | Diameter | Effective conductivity | Reference |
|---|---|---|---|
| | $\mu$ | *mho/m* | |
| Glass spheres | 1.7 | 0.0042 | (Fricke, 1936) |
| Polystyrene spheres | 0.55 | 0.0150 | (Schwan, 1962) |
| *E. coli* | 1.0 | 0.0300 | |
| *M. lysodeikticus* | 1.2 | 0.1500 | |

of the two organisms was measured and the results are presented in Fig. 3. The failure of this model is qualitatively apparent. Whereas the conductivity of *M. lysodeikticus* is five times that of *E. coli* at low $\sigma_1$, the electrophoretic mobility of *M. lysodeikticus* is actually lower than that found for *E. coli*. Furthermore, the quantitative prediction (Cole, 1933, Fricke, 1936) of effective, homogeneous conductivity, which is obtained from surface charge density as measured electrophoretically, is lower than that observed by a factor of three in the case of *E. coli* and by a factor of twenty in the case of *M. lysodeikticus*.

Since the surface properties of the bacteria do not appear to explain their con-

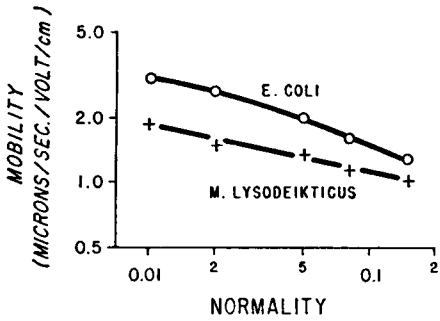


FIGURE 3 Electrophoretic mobility of *E. coli* and *M. lysodeikticus* as a function of salt concentration in the suspending medium.

ductivity, consider instead the membrane which surrounds the cytoplasm with its high concentration of ions and ask what properties would be required of the cell membrane if it were to explain the observations. The effective, homogeneous conductivity $\sigma_2$ of a conducting sphere of conductivity $\sigma_i$ surrounded by a thin poorly conducting shell was given by Cole (1928) as[6]

$$\frac{1}{\sigma_2} = \frac{1}{\sigma_i} + \frac{R_m}{a} \tag{2}$$

where $R_m$ is the membrane resistance and $a$ is the radius of the sphere. From the high internal osmolality of the cells (Mitchell, 1956) and direct observations of internal conductivity of these and other cells (Pauly 1959, 1962) it is reasonable to assume that the internal conductivity $\sigma_i$ of *E. coli* is of the order of magnitude of 0.3 to 0.5 mho/m and for *M. lysodeikticus* roughly 1 mho/m. Since $\sigma_i \gg \sigma_2$, the value assumed for $\sigma_i$ is not critical in the calculation of $R_m$. Thus, to explain the data of Fig. 2 in terms of the properties of the cell membrane, values of $R_m$ would have to be less than $20 \times 10^{-6}$ ohm m$^2$ for *E. coli* and $3 \times 10^{-6}$ ohm m$^2$ for *M. lysodeikticus*. Cole and Moore (1960) report values as low as $10^{-3}$ ohm m$^2$ for

[6] This equation follows directly from equation (7) under the assumption that the thickness of the shell is much less than the radius of the sphere and that the conductivity of the shell material is comparable to or less than that of the interior.

active nerve membrane but values of the order of 0.1 ohm $m^2$ or higher would be more reasonable for these resting cells.

## DISCUSSION

The effective, homogeneous conductivities $\sigma_2$ of bacteria are high enough at low frequencies to be measured with fair accuracy over a range of a decade and one half in the environmental conductivity $\sigma_1$. Within this range of measurement, the electrical behavior of the bacteria undergoes a transition from roughly constant values of $\sigma_2$ at low $\sigma_1$ to an approximately linear dependence upon $\sigma_1$ at high $\sigma_1$. Although direct measurements of the membrane resistance of these organisms are not available, it appears highly unlikely that the membranes have properties which can explain the high conductivity which is observed at low frequencies. Similarly, surface conductance, as determined *via* measurements of the electrophoretic mobility of the bacteria, can neither qualitatively nor quantitatively explain the observations.

Recognizing that these organisms are not homogeneous, one can formulate a model which describes the electrical properties of the bacterial cell. Bacteria can be described anatomically as having cytoplasm enclosed by a membrane which, in turn, is surrounded by a cell wall (Salton, 1964). Although the cytoplasm contains a high concentration of ions, the membrane in all probability has such a high resistance that the entire region which it encloses may be considered non-conducting at low frequencies (below 100,000 CPS). The cell wall, on the other hand, is readily permeable to small molecules and ions. In fact, many of the structural molecules of the wall are ionized so that even when the environmental conductivity is low the region called the cell wall may be highly conducting by virtue of the presence of mobile counterions. In this respect the material composing the cell wall has electrical properties which are similar to an ion exchange resin.[7] If the cell wall region is in equilibrium with the environment and furthermore if the concentration of fixed charge within the cell wall is $c_f^w$, then the requirement of electroneutrality is satisfied (Overbeek, 1956) if

$$c_+^w - c_-^w + c_f^w = 0, \tag{3}$$

where $c_+^w$ and $c_-^w$ are the co- and counterion concentrations within the wall. If the chemical potential is equal for both regions,

$$c_+^w c_-^w = (c^o)^2, \tag{4}$$

where $c^o$ is the concentration of ions in the environment.[8] The conductivity of the

[7] The cation exchange properties of the cell wall of the algae *Chara* has been noted by Gaffey and Mullins (1958) and of bacteria by McCalla (1940).

[8] Two qualifications in this use of the Donnan Theory in the present application must be made. First, in a region such as the cell wall where a sizeable fraction of the total space is occupied by solids, the concentrations $c_+^w$ and $c_-^w$ refer to the water space rather than the total space of the wall. Thus concentrations are in moles per liter of solution not moles per liter of cell wall space. Second,

*(Footnote continued on following page.)*

cell wall should be proportional to the sum of the products of the concentrations of mobile ions in this region and their mobilities. If $u_+^w$ and $u_-^w$ are the mobilities of co- and counterions, it is found from equations (3) and (4) that

$$u_+^w c_+^w + u_-^w c_-^w = \tfrac{1}{2} c_f^w \left[(u_-^w - u_+^w) + (u_-^w + u_+^w)\sqrt{1 + (2c^o/c_f^w)^2}\right] \tag{5}$$

and if the mobilities of the two ions are comparable this can be written approximately as

$$\approx c_f^w u^w [1 + (2c^o/c_f^w)^2]^{1/2} \tag{6}$$

Thus for small conductivities of the environment, the conductivity of the wall will approach a constant value which will depend upon the concentration of the fixed charges in the region. For large conductivities of the environment, the wall conductivity will approach a linear dependence upon $c^o$. If it were possible to determine the transition from constant to linear dependence (*i.e.*, where $2c^o/c_f^w = 1$), it should be possible to estimate the fixed charge concentration of the wall. It appears from the experimental data that this transition occurs within the range of observation.

For purposes of this discussion the bacterial cell will be assumed to consist of a central non-conducting core, corresponding anatomically to the region bounded by the cytoplasmic membrane, surrounded by a conducting shell which corresponds to the cell wall (Fig. 4). Maxwell gave an expression for the effective, homogeneous conductivity $\sigma_2$ of a shelled sphere which is formally identical with equation (1) (Maxwell, 1881, Fricke, 1955), namely

$$\frac{\sigma_2 - \sigma_w}{\sigma_2 + 2\sigma_w} = p \frac{\sigma_i - \sigma_w}{\sigma_i + 2\sigma_w}, \tag{7}$$

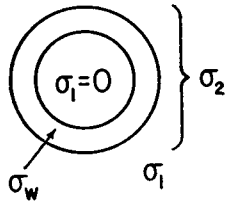


FIGURE 4 Model for the bacterial cell. The inner, non-conducting core corresponds to the region enclosed by the cytoplasmic membrane. The outer shell represents the cell wall.

where $\sigma_i$ and $\sigma_w$ are the conductivities of the interior and wall respectively and $p$ is the fraction of the total cell volume occupied by the core. If $\sigma_i \ll \sigma_w$, this can be written

$$\sigma_2 = \sigma_w \frac{1 - p}{1 + \frac{1}{2}p}. \tag{8}$$

If $\sigma_w$ is assumed to have the form of equation (6) and if $p$ is independent of $\sigma_1$,

*(Footnote concluded.)*
it should be recognized that in writing equation (4) it has been assumed that the activity coefficients for the ions are unity. This assumption is not strictly valid particularly for the ions within the cell wall. However, even without information on the activity coefficients, the analysis will be useful for rough quantitative estimates of $c_f^w$ from the experimental data.

$$\sigma_2 = A[1 + (2c^\circ/c_f^{\prime\prime})^2]^{1/2}, \quad (9)$$

where $A$ is a constant which depends upon the effective mobility of the ions, the magnitude of the fixed charge concentration and the anatomy of the cell. From the solid curves in Fig. 2, it is apparent that one can fit curves of the form of equation (9) to the observed values of $\sigma_2$. From the values of $c^\circ$ corresponding to the condition that $2c^\circ/c_f^{\prime\prime} = 1$, it appears that the concentration of fixed charge in the cell wall of *E. coli* is of the order of 0.07 equivalent per liter of cell wall water, whereas, for *M. lysodeikticus* the corresponding value is roughly 0.2 equivalent per liter. These values would be 10 to 40 per cent lower if expressed in terms of total cell wall volume rather than wall water volume. The correction depends upon a knowledge of the volume fraction of the wall which the solids occupy. Britt and Gerhardt (1958) report a value of 89 per cent for the water content of isolated walls of *M. lysodeikticus*. Thus, although knowledge of the specific volume and hydration of the solids is lacking, it appears that the correction would not be substantial for this organism.

The nature of the data and curve-fitting process illustrated in Fig. 2 gives rise to uncertainties in the estimated values of $c_f^{\prime\prime}$ which are of the order of $\pm 20$ per cent in the case of *M. lysodeikticus* and about 15 per cent for *E. coli*. In addition, the analysis has been carried out with the realization that $p$ is not strictly independent of $\sigma_1$, that the activity coefficients of the mobile, cell wall ions are probably less than unity and that the mobilities of the co- and counterions may not be equal. Furthermore, it has been assumed that the fixed charge concentration in the wall is independent of $\sigma_1$. Although there is little reason to believe that monovalent cations will adsorb to the negatively charged cell wall, the constancy of $c_f^{\prime\prime}$ remains to be demonstrated. The analysis, therefore, is presented not as a precision technique for determination of cell wall, fixed charge but as a rough test of the feasibility of the model proposed for the bacterial cell.

It remains now to determine whether the estimates of $c_f^{\prime\prime}$ obtained simply from the position of the curves relative to the abscissa of Fig. 2 are consistent with the magnitude of the effective, homogeneous conductivity observed for the bacteria at low $\sigma_1$. Table II gives the data used in estimating $c_f^{\prime\prime}$ and the prediction of $\sigma_2$ based on these values. An equivalent conductivity of 11 mho/m/equivalent per liter is used in converting $\sigma_1$ to $c^\circ$. To obtain the conductivity of the cell wall water at low $\sigma_1$ from $c_f^{\prime\prime}$ the equivalent conductivity of the counterion is taken as 4.5 mho/m/equivalent per liter, which is about the value to be expected for the sodium ion at this concentration and 29°C. The volume fraction $p$ is taken as the ratio of the sucrose-phosphate volume inaccessible to the dextran inaccessible volume. This is equivalent to saying that the cell wall volume is equal to the water space of the wall. Of course, a larger volume for the wall with correspondingly lower effective conductivity could be taken but the result would be the same. In light of the assumptions and experimental error the predicted and observed values of $\sigma_2$ may be considered to be in

TABLE II

EFFECTIVE, HOMOGENEOUS CONDUCTIVITY AT LOW $\sigma_1$ FOR *E. COLI* AND *M. LYSODEIKTICUS*

To test the internal consistency of the proposed model for the electrical properties of the bacterial cell, the effective, homogeneous conductivity is computed from estimates of concentration of fixed charge in the cell wall. (see text)

| | *E. coli* | *M. lysodeikticus* |
|---|---|---|
| $\sigma_1$(for $2c^o/c_f^w = 1$), *mho/m* | 0.37 | 1.1 |
| $c^o$(for $2c^o/c_f^w = 1$), *M* | 0.034 | 0.10 |
| $c_f^w$, *equivalents/liter* | 0.07 | 0.2 |
| $\sigma_w$(for low $\sigma_1$), *mho/m* | 0.3 | 0.9 |
| $p = V_{sucrose}/_{dextran}$ | 0.80 | 0.83 |
| $\sigma_2$ From equation (8), *mho/m* | 0.04 | 0.11 |
| $\sigma_2$ (observed), *mho/m* | 0.04 | 0.17 |

agreement. Actually, the mobility of the ion within the wall might have been expected to be lower than in free solution. Hence it would not have been surprising to find the observed values lower than predicted. On the other hand, there is no reason to believe that at low $\sigma_1$ the counterion is $Na^+$. In fact, under these environmental conditions the leakage ions from the cells are apt to be dominant. Even a small contribution from hydrogen ions could cause a large increase in the effective conductivity of the wall. The observations, therefore, are consistent with the postulate that the effective conductivity of the bacterial cell at low $\sigma_1$ can be attributed to the counterions distributed within the cell wall.

This model bears a close resemblance to the model used by Schwartz (1962) to explain the complex conductivity of suspensions of polystyrene particles. In his problem, the particle itself is truly non-conducting and has a low dielectric constant by comparison with water. The counterions, which explain the high dielectric constants observed for these particles, are confined to the very thin, diffuse layer just outside the charged surface of the particle. The low frequency limit of the dielectric constant in the Schwartz theory is directly proportional to the surface charge density of the particle and is not a function of the mobility of the counterion. In the case of bacteria, the fixed charges and counterions are distributed throughout a shell of finite thickness (the cell wall). The core is non-conducting but may have an effective dielectric constant of the order of 500. However, the similarity of the two models leads one to predict that if the counterions within the wall participate in the Schwartz effect, very high, low frequency, dielectric constants should be observed for these suspensions and the magnitude should be proportional to the estimated values of $c_f^w$ in Table II. Preliminary exploratory measurements of the dielectric constant of suspensions of the two bacteria were extended as low as 200 CPS.[5] For 50 per cent cell suspensions the 200 CPS dielectric constants were $3 \cdot 10^4$ for *E. coli* and $9 \cdot 10^4$ for *M. lysodeikticus*. These values are in the same ratio as the $c_f^w$ values of Table II.

This points to the use of dielectric constant measurements as another, independent method for the determination of fixed charge in the cell wall, a method which would not suffer from uncertainties in the choice of ion mobility but which would in fact, contribute information as to the nature of the counterions involved. To thoroughly exploit this technique would require a more sensitive bridge than was available for these studies. It appears, however, that with improved theory and experimental procedure, low frequency, dielectric measurements may give the kind of information about the cell walls of bacteria that microscope electrophoresis studies have provided for the outer surface of the cell.

## SUMMARY

The passive electrical properties which are reported for the bacteria *E. coli* and *M. lysodeikticus* can be explained by an anatomical model consisting of a central, non-conducting core, the region enclosed by the cytoplasmic membrane, surrounded by a conducting shell, the cell wall. The cell wall itself appears to be similar to an ion exchange resin in the sense that fixed, charged sites are distributed throughout the porous structure. The conductivity of this region is proportional to the environmental conductivity for high, environmental salt concentrations and dependent only on counterion concentration when the environmental salt concentration is low. This relationship leads to an estimate of 0.07 equivalents per liter for the fixed charge concentration in the wall of *E. coli* and 0.2 equivalents per liter for *M. lysodeikticus*. These values are in agreement with the magnitude of the conductivity observed for the two organisms. The study points to the possible use of these and more advanced dielectric techniques to provide the kind of information about the cell walls of bacteria that microscope electrophoresis has given for the outer surface of the cells.

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