RECTIFICATION AND INDUCTANCE IN THE SQUID GIANT AXON

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The function of nerve has been extensively investigated for many years and careful measurements have been made of the initiation and propagation of the impulse under a wide variety of conditions. It was only natural that there should be numerous attempts to correlate and explain these experimental data. The general phenomenological theories of nerve activity have avoided the physical characteristics of nerve structures and the specific theories have had to consider these properties as undetermined parameters. It has long been assumed, for example, that the axon membrane could be represented electrically by capacity and resistance elements, yet these characteristics have only been measured directly in the past few years. Excitation phenomena have also led to a few suggestions that the membrane might act as an electrical rectifier and to at least one mention of the possibility of an inductive element in a nerve.

The preceding papers have presented experimental evidence, independent of excitation, for the rectification and inductance elements in the membrane of the squid giant axon (Cole and Baker, 1941 *a* and *b*; Cole and Curtis, 1941). The various aspects of the evidence should now be correlated and the implications as to both the physical structure of the membrane and its physiological function should be investigated as far as possible.

The electrical properties of the membrane will for several reasons be considered in terms of an equivalent circuit. Probably the most important of these is that the measurements have for the most part been made by a direct comparison of the nerve with electrical circuit elements. However, information in this form is quite usable because the techniques for analyzing the behavior of circuits are versatile and powerful and have been widely used. The numerical values and the configuration of the circuit elements may then be used to describe molecular structure on the one hand and physiological function on the other. But it must be emphasized again that any particular circuit is not necessarily unique and should not be interpreted intuitively (Cole, 1928, 1933 *a*, 1937). To avoid as much complexity as possible at the present time, we shall only consider the simplest circuit, shown in Fig. 1, which has a reasonable correspondence to the known membrane properties (Cole and

Baker, 1941 *b*). This circuit makes many concessions to simplicity and several of the obvious defects will be mentioned later (p. 48). Before turning to the physiological applications of the circuit, we shall consider the physical characteristics and implications of the circuit elements themselves.

MEMBRANE STRUCTURE

Capacity and Resistance

The membrane capacity, C (Fig. 1) of the squid axon is about 1.1 μf/cm.^{2} and it has a constant phase angle of about 75°. These values are in the range of those found for a variety of other cells, and their significance has been discussed recently (Cole, 1940). From physiological evidence, this capacity is probably a characteristic of the ion impermeable portion of the membrane and the phase angle a measure of the dielectric loss in it. For physical reasons, it was suggested that the membrane is a highly organized and coherent structure

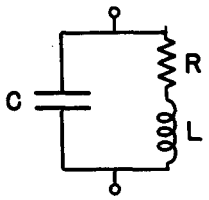

FIG. 1. An approximate equivalent circuit for the membrane of the squid giant axon with capacity, C, inductance, L, and resistance, R.

having a high dielectric constant and a considerable thickness on a molecular scale.

The membrane resistance, R (Fig. 1) of the squid axon is quite variable, but it is usually of the order of a few hundred ohm cm.^{2} for small currents, and similar values have been found for several other cells. This resistance is assumed to be a measure of the ion permeability on the basis of physiological and physical evidence recently summarized (Cole, 1940).

Rectification

However, as the current is increased, it is no longer found that the resistance, R, may be considered as a linear circuit element. Measurements of impedance (Cole and Baker, 1941 *a*) and membrane potential (Cole and Curtis, 1941) show that the membrane is a rectifier with a limiting resistance for an outward current flow about one-hundredth of that for an inward flow.

It is very instructive to compare the rectification characteristic of the membrane with those of widely used commercial barrier layer rectifiers, such as copper-copper oxide and selenium. It is found for Cu-CuO that the limiting resistance in forward direction ranges from one-hundredth to one-thousandth of that in reverse direction and for selenium that this ratio is about one-

hundredth. Thus the ratio of one-hundredth found for the axon membrane is quite comparable to that for these rectifiers. However, the similarity probably ends at this point. Although there has been considerable uncertainty as to the exact mechanism of the barrier layer rectifier action, there seems to be little doubt that it is an electronic phenomenon. It seems rather improbable at the present time that there is an electronic conduction in the cell membrane, and we are much more inclined to look for an explanation in terms of an ionic conduction. Such an example has been found by Blinks (1930) in a dried collodion membrane separating different electrolytes, where the rectification was accounted for by the mobility of the permeating ion forced into the membrane by the applied field.

One might also consider the possibility of a conduction by potassium ions alone. It seems reasonable to suppose in such a case that both the concentration and the mobility of this ion are quite small in the membrane structure itself. If the concentration is governed by partition coefficients at the two interfaces we may expect a concentration gradient within the membrane comparable to the external concentration gradient. When there is an inward current flow, a steady state will be set up in which the initial low outside concentration of potassium ions will be decreased by the ions crossing into the membrane. At the inside the potassium ions are readily taken up as they emerge from the membrane and the concentration at each point in the membrane will be reduced. The overall effect will then be an effective increase of the membrane resistance. For outward current flow, the large supply of these ions available at the inner face of the membrane is augmented by the current flow, and at the outer face only a relatively low equilibrium concentration of the ions is possible. As a consequence of this large supply and small delivery the concentration may be expected to increase at every point and so to decrease the resistance.

This picture has not been put into quantitative terms, and it may be simpler to consider it in terms of potential barriers at each interface which are then alternative expressions of the partition coefficients. One may then anticipate a resistance-current relationship of the form $R = R_0 + R_1 \tanh i/i_0$ and the observed rectification curves are of this general form. However, it seems premature to compare the experimental data with such an analytical expression until each has established on a better basis than at present. It is obvious, however, that the rectification characteristics will at least be an important guide in the formulation and verification of the nature of the mechanism.

Inductance

The idea of a membrane capacity is easy to rationalize and it has not been particularly surprising to find that the axon membrane acts as a rectifier, although the efficiency may be rather unexpected. It is quite another matter

to find that the membrane has the electrical characteristics of an inductance. In ordinary experience an inductance is a characteristic associated only with the storage of energy in a magnetic field, and it is singularly difficult to imagine a membrane structure which would allow an electromagnetic field corresponding to more than a few microhenries. This could be ignored but the actual membrane inductance of one-fifth henry for a cm.^{2} cannot.

Although the concept of inductance was originated to express electromagnetic phenomena and our standards of inductance have long been electromagnetic, it is not necessary to restrict the use of the term to this field. An inductance may be defined by means of the relation of the current, I, and the voltage, V, in a manner analogous to the operational definition of a resistance, R, by Ohm's law, $V = RI$. Then anything in which it is found that the potential difference is proportional to the rate of change of current is an inductance, electrically speaking, and it is measured as the factor of proportionality, L, in $V = L\,dI/dt$. Similarly, the definition of capacity, C, is $I = C\,dV/dt$. Turning to the energy, we find that $T = 1/2\,LI^2$ for the inductance, $U = 1/2\,CV^2$ for the capacity, and the rate of dissipation $F = RI^2$ for a resistance, completely irrespective of the ultimate mechanisms by which the storage or dissipation of energy may take place.

However, to point out that an inductance can be defined without reference to a magnetic field is hardly a satisfactory explanation of the very large membrane inductance. And indeed it will not be possible to give a complete explanation except in terms of the membrane structure. On the other hand, it may be possible to make the existence of a membrane inductance seem less unreasonable by examples of purely physical structures having inductive characteristics without a corresponding magnetic field.

One example is the carbon filament incandescent lamp. In this instance the explanation of the inductance is relatively simple, and is easily understood by a consideration of the current flow after the application of a constant voltage to the lamp. At the first instant a current will flow corresponding to the cold resistance of the filament. The filament then begins to heat as a result of the current flow at a rate depending upon its heat capacity and to an extent determined by the heat losses. However, a carbon filament has a negative temperature coefficient of resistance and consequently the resistance becomes less than it was initially. Since a constant potential was applied the current rises from the initial value as an exponential function of time and finally reaches a constant value. These electrical characteristics are, however, identical with those of a resistance and inductance network, and it is possible to calculate the thermal characteristics of the lamp from the electrically equivalent structure. When measured in a Wheatstone bridge it is found that such a lamp has an inductive reactance at all frequencies and that the equivalent network of a 60 watt lamp requires an inductance of 30 henries. A much more dramatic

example of an inductance of thermal origin is found in the recently developed Western Electric 1-A Thermistor (Pearson, 1940). In this device a bead of uranium oxide is mounted on two platinum wires and sealed in vacuum. This material also has a negative temperature coefficient which with the heat capacity and losses gives the Thermistor an inductance of many henries. It is not impossible of course that the cell membrane has similar characteristics, but this particular inductance mechanism probably has little value to us except as an example of a non-magnetic inductance.

There is another and more common class of inductances arising from mechanical motions, and the most familiar example of these is the piezoelectric crystal, such as quartz or Rochelle salt. A potential difference applied to two electrodes on one of these crystals tends to change its shape, and conversely if its shape is changed mechanically a potential difference will appear between the electrodes. When the crystal is not constrained, its distortion after the application of a constant potential to the electrodes depends upon the mechanical characteristics of mass, elasticity and damping. As this distortion takes place the electrical charge generated modifies the current flow from the outside circuit in just the same manner as an electrical circuit containing resistance, capacity, and inductance. In this electro-mechanical system the inductance is determined by the mass of the crystal, the capacity by its elasticity, and the resistance by the internal and external frictional losses. When the alternating current has a frequency about that of a natural mechanical frequency of the crystal, the mechanical vibrations may have large amplitude and the electrical reaction on the circuit is particularly powerful. The crystal may then control the frequency of the associated circuit with a high degree of accuracy and serve as a standard of frequency and time. A common type of quartz crystal about 1 cm. square and 1 mm. thick with a natural frequency of 2 megacycles is found to have an inductance of about one-tenth henry or approximately half the inductance of a similar area of axon membrane.

It may seem quite unreasonable to suppose that the axon membrane may be piezoelectric with a natural frequency of a few hundred cycles, but in the present state of our information this possibility cannot be excluded. There is a potential difference across the membrane and probably a very high electrical field strength giving rise to considerable mechanical forces. Any alteration of the field strength by an externally applied potential may be expected to alter the mechanical stress. A resultant deformation reacting on the membrane potential difference might then be measured electrically. However, for a crystal as thin as the axon membrane presumably is to have a natural frequency of a few hundred cycles requires a very high density and very small coefficient of elasticity. But this may not be impossible if the membrane has the laminar structure proposed for the myelin sheath on the basis of x-ray evidence (Schmitt, 1936). This picture of parallel lipo-protein sheets separated by

aqueous layers is highly suggestive of the colloid coacervates such as bentonite, in which the micro-structure, as in the tobacco mosaic virus, is maintained by the interaction of the ion atmospheres (Langmuir, 1938). It is possible that the forces holding a particular particle in its equilibrium position may be small compared to the mass of the particle, and the natural frequency of oscillation about the equilibrium may be of the order required by the membrane measurements. There is of course no direct evidence to support this suggestion, but observations on the double refraction of bentonite sols in an alternating current field (Mueller, 1939; Norton, 1939) are at least helpful. Here it is found that the sign of the double refraction at a frequency of the applied field above about 200 cycles is opposite to that obtained at lower frequencies. We may then expect that impedance measurements of these sols will also show a change of sign of their reactance in this same frequency range although the measurements may be somewhat obscured by surface conductance effects.

If we consider structures containing sources of energy, the possible interpretations of an inductive reactance are almost unlimited. In general we can only say with certainty that the membrane structure is such that it gives rise to a potential difference during current flow which is proportional to the rate of change of the current.

In this discussion of the possible sources of inductance it has been emphasized that the common association of an inductance with a magnetic field may be misleading, and it is equally apparent that the common association of a capacity with a dielectric may be equally misleading. There has been little reason to question the applicability of this concept because it gave so satisfactory a picture of a thin membrane. It has already been mentioned that the elasticity of a piezoelectric crystal gives rise to a capacity. This capacity element is separate and distinct from the dielectric capacity of the crystal which may be obtained when the crystal is constrained and cannot react mechanically to the electric field. Also we find that there are capacities of a thermal origin and as may be expected they arise from a positive temperature coefficient of resistance. A 3 watt tungsten lamp, for example, has been found to have a capacity of about one thousand microfarads. As a consequence we must also be prepared to discard the simple and obvious interpretation of the membrane capacity measurements.

Until there is more complete evidence as to the nature of the membrane structure and function we have no reason to assume that these physical measurements must be explained in terms of physical structures analogous to those which have been used for illustrative purposes. On the contrary, it is probably safest to consider the membrane as far as possible from a physical-chemical viewpoint in which no sharp division can be made between physical and chemical phenomena as has often been done. For example, let us assume that the

variables of the general two factor formulations of excitation (p. 43) can actually be identified chemically, as ions for example, and that the changes of the difference in these ion concentrations correspond to changes of the membrane potential difference. As will be shown, this is electrically equivalent to an inductance, capacity, and resistance, in equations (7) and (8), and the measurements of these elements then constitute an analysis of the chemical kinetics of the membrane system. If we wish to describe the membrane phenomena in purely chemical terms, as is perfectly reasonable and possible, it then seems quite probable that the electrical measurements as they stand contain by far the most accurate and detailed chemical information available at the present time for the axon membrane.

MEMBRANE RESPONSE

We shall turn now to the performance of the membrane as a whole, both by itself and in relation to the rest of the axon. It is convenient to divide the field into the two regions of (1) passive or subthreshold phenomena, and (2) active or threshold phenomena. These regions probably should be separated according to whether or not potential energy is released, but for the present the terms will be used without careful definition.

Subthreshold Potential

The subthreshold phenomena will be considered first in terms of calculated and measured electrical potentials. When a current flows through any part of an axon having an appreciable resistance or impedance, it is obvious that there will be a change of potential. These potentials may be roughly classified as linear and non-linear. For sufficiently small currents, the potential difference is proportional to the current and the factor of proportionality is the resistance for direct current and the impedance for alternating current. When the alternating current characteristics are known it then becomes possible to calculate the linear subthreshold potential difference for the application of any small current. The equivalent circuit of Fig. 1 is, however, an approximate representation of the alternating current characteristics of the membrane and may be used directly.

For a square centimeter of membrane, the capacity, C, in this circuit is about 1 microfarad, and for simplicity the dielectric loss will be ignored. The resistance, R, may have a resting value in the neighborhood of 300 ohms per cm.^{2} for small currents, and the inductance, L, is about one-fifth henry. If now v_m is the potential difference across the membrane at one point and i_m is the current density at this point, the relation between these two quantities at any time is given by the ordinary differential equation:

$$LC\frac{d^2v_m}{dt^2} + RC\frac{dv_m}{dt} + v_m = L\frac{di_m}{dt} + Ri_m. \qquad (1)$$

This is a familiar second order differential equation with constant coefficients and the methods of solution are well known and direct. However, as the current is increased beyond perhaps 10 per cent of rheobase, it has been found that the resistance, R, is a rectifier element having a lower resistance for an outward flow of current and a higher resistance for inward flow. This property may be expressed in the form $v/i = R(i)$ where v is the potential difference across and i the current through the element, and we have:

$$LC\frac{d^2v_m}{dt^2} + R(i)C\frac{dv_m}{dt} + v_m = L\frac{di_m}{dt} + R(i)i_m, \tag{2}$$

for the non-linear subthreshold potential. The solution of this equation with the variable coefficients has been found for some analytical forms of $R(i)$, but the solution must be obtained by numerical, graphical, or mechanical integration when $R(i)$ is given in numerical or graphical form only. If we turn now from the consideration of a single point on the membrane to a treatment of the axon as a whole we must allow the potential and current to vary along the length of the axon as well as with the time. Equation (2) is then to be replaced by the partial differential equation:

$$LC\frac{\partial^2v_m}{\partial t^2} + R(i)C\frac{\partial v_m}{\partial t} + v_m = L\frac{\partial i_m}{\partial t} + R(i)i_m. \tag{3}$$

As yet there has been no indication that the exterior and interior of an axon are anything but electrolytic conductors and these may be expressed as the resistances r_1 and r_2 for a unit length of axon. Then with the ordinary assumptions of a negligible radial potential difference inside and outside the membrane and a negligible longitudinal current flow in the membrane we obtain:

$$\frac{\partial^2v_m}{\partial x^2} = (r_1 + r_2)i_m, \tag{4}$$

where i_m is now the current per unit length. This becomes the usual cable equation if the membrane is a pure resistance, r_m for a unit length, and the relation between v_m and i_m is given by $v_m = r_m i_m$. In the present case, however, this relation is given by equation (3) and the axon behavior is completely determined by the two equations, (3) and (4). In general the membrane current, i_m, cannot be measured directly under any particular experimental conditions and only the externally applied current, $I_0(t)$, is known. The part of this current which crosses the membrane, $I(t)$, aptly termed the "penetrating fraction" by Rushton (1937 *b*), is readily found and we have the further relation

$$I(t) = \int i_m(x, t)\,dx \tag{5}$$

to be satisfied for each electrode. In the case of no applied current, $I(t) = 0$. The behavior of the nerve model under any set of conditions is then to be found by solving the three equations (3), (4), and (5). The complete solution of these partial differential equations is a difficult and laborious procedure which has not yet been carried out for even a single problem, and there are at least as many possible problems as there have been experiments on nerve. The effect of the presence of inductance and rectification elements in the membrane may, however, be illustrated by a few special cases and approximate calculations.

Constant Current

When a small constant current is applied uniformly to a unit area of membrane, the subthreshold potential may be calculated by equation (1) or from the circuit of Fig. 1. This potential will be over-damped, critically damped, or under-damped and oscillatory according as the resistance R is greater than, equal to, or less than $2\sqrt{L/C}$. Also the potential will have the same form for both directions of current flow. For larger currents, the non-linearity of the resistance, R, must be taken into account. It is obvious that under an anode where the membrane resistance is increased, the circuit will be more and more highly damped as the current is made larger and that the damping will be progressively decreased under the cathode. A rigorous calculation of these potentials from equations (3), (4), and (5) has not been attempted, but the graphical integrations of equation (2) have been carried out assuming a uniform density, as shown in Fig. 2. To compensate in some degree for not solving the partial differential equations, the apparent rectification curve for the applied current (Fig. 4, Cole and Curtis, 1941) was used rather than the otherwise more appropriate curve for a uniform current density (Fig. 5 of that paper).

The curves of Fig. 2 exhibit the same characteristics as the squid axon membrane potentials (Cole and Curtis, 1941) and *Sepia* axon potentials (Arvanitaki, 1939). During current flow at the anode, the damping and consequently the effective time constant during the rising phase of the potentials increase with increasing current, and the final steady levels of potential increase more rapidly than the applied currents. The cathode potentials show progressively less damping until they are under-damped and oscillatory with the steady levels of potential rising less rapidly than the applied currents. The qualitative agreement between these rather crude calculations and the experimental results is sufficiently good to be considered as additional evidence that the electrical picture of the membrane given in Fig. 1 is essentially correct. It is certainly indicated that the *Sepia* axon membrane is a rectifier. In view of the direct evidence for the squid axon and the similarity of the behavior of the *Sepia* it is difficult to believe that the *Sepia* membrane does not have a similar inductance element. There is, however, a discrepancy in the frequency

of the oscillations. For a zero membrane resistance the potentials will have a maximum frequency given by $\nu_0 = 1/2\pi \sqrt{LC}$. Using the values of one microfarad/cm.^{2} obtained from the transverse impedance measurements and the one-fifth henry cm.^{2} from the longitudinal measurements, we find a frequency, ν_0 = 330 cycles/second. The *Sepia* data give somewhat lower frequencies, as would be expected because of the damping, but the data of Fig. 2 (Cole and Curtis, 1941) give nearly 500 cycles. Until the reasons for this difference are clear or until all of the necessary measurements can be made on a single axon, it will be assumed that this lack of agreement is within the range of variability of the axons and their properties under experimental conditions.

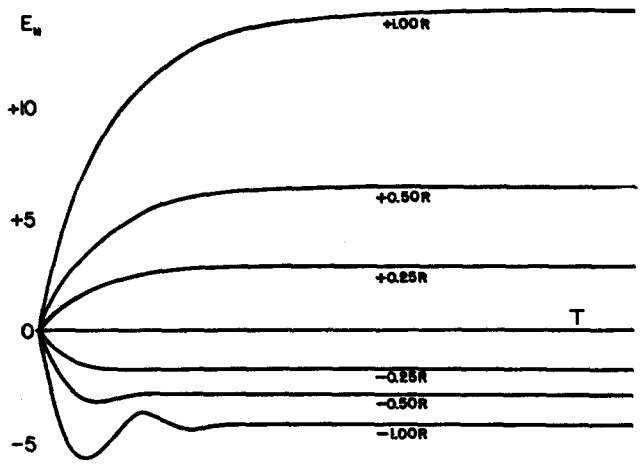

FIG. 2. Approximate graphical calculation of subthreshold potentials from measured membrane characteristics. Changes of membrane potential, E_M at anode and cathode, *vs.* time, T, after the application of constant currents of 0.25, 0.5, and 1.0 rheobase. Increase of potential at anode is upward.

The most striking oscillations of potential have been obtained by Arvanitaki (1939) in partially decalcified preparations. With decreasing calcium, the damping with an applied current is decreased. In the extreme case the damping has become zero or slightly negative, with no applied current. The oscillations of potential are then spontaneous and they increase in amplitude until excitation occurs. It will be particularly interesting to know whether the entire form of the rectification curve of the membrane is changed or only the point on the curve corresponding to zero current flow is altered under these conditions.

Short Pulse

This formulation of the electrical properties of the membrane should be equally applicable to the measurements of Hodgkin (1938) on the subthreshold

potential changes in the isolated single crab axon with the application of a short shock, and those of Pumphrey, Schmitt and Young (1940) on the squid axon. However this is apparently not the case since the approximate integration of equation (2) used above does not give the characteristic responses found by Hodgkin. This is evidence that the equivalent circuit of Fig. 1 is not adequate for this preparation and experiment. If the circuit also fails to represent the behavior of the squid axon after a short shock, it follows immediately that the actual and calculated membrane potentials do not agree during the initial phases of the application of a constant current. However, as has been pointed out, the amplifier characteristics have obscured the actual behavior of the membrane potential at short times (Cole and Curtis, 1941), and this prediction cannot be verified from the present records. But Hodgkin's records make it rather probable that the simplifications of either the circuit, or the calculations, or both, are not always justified.

Electrotonic Potential

Suppose that an axon is in a moist chamber, or in a medium restricted by a close-fitting insulating trough, with two very narrow electrodes applied some distance apart. At the first instant that a direct current is applied, the entire membrane current will flow through the capacity because the rectifier and particularly the inductance very effectively oppose a sudden charge of current in that branch. The membrane current density at the first instant, although theoretically infinite under an electrode will decrease symmetrically on each side of the electrode and the distribution at the other electrode will be identical except for sign. As time goes on, the current is increasingly transferred from the condenser to the inductance-rectifier branch until, when a steady state is reached, the current is entirely governed by the rectifier. The current flow near the anode is entirely inward and consequently the membrane resistance is increased in this region. Since the current and voltage are no longer changing with time, equation (3) reduces to $v_m = R(i_m)i_m$, and now we may calculate the membrane current density at different distances from each electrode by the data of Fig. 4 (Cole and Curtis, 1941) in a manner analogous to that used in determining the membrane potential as a function of the current density. This leads to the results shown in Fig. 3 for three values of anode and cathode current. Quite apart from the analysis, the underlying development of these current distributions is easily seen qualitatively. A small current from the anode increases the membrane resistance immediately under the electrode and so tends to divert the current from this region. As the current is increased it must spread farther along the axon before it can penetrate with reasonable ease. At the cathode the situation is reversed, because the decreased resistance under the electrode reduces the necessity for the current spread that would otherwise take place, and the current concentration becomes relatively larger the higher the current. Then the characteristic length, $\lambda = \sqrt{R/(r_1 + r_2)}$

may not be a fixed and invariable property of any axon in a steady state as has usually been assumed in theories of excitation and propagation.

It has been known since the time of Pflüger that anelectrotonus had a somewhat wider spatial distribution than catalectrotonus, and if the electrotonus is directly related to the membrane current flow, this is evidence to indicate that other axon membranes also have rectification properties.

Propagated Impulse

Turning now to the propagated impulse, we may reconsider the difficulties in correlating the phenomena during the rising phase. As has been pointed out (Cole and Baker, 1941*a*), the outward current flow during the foot of the action potential should produce an impedance decrease if the common assump-

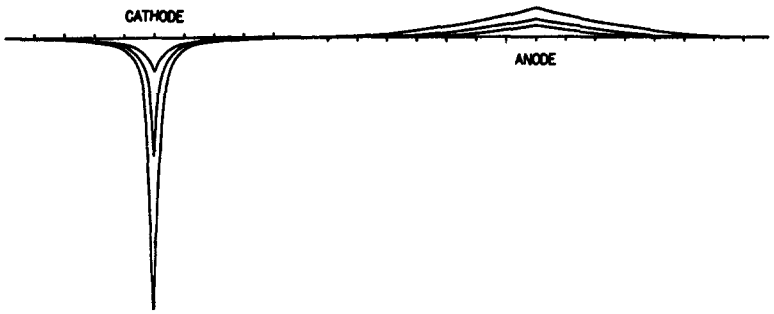

FIG. 3. Steady state membrane current density, ordinate, for narrow electrodes and currents of 0.25, 0.5, and 1.0 rheobase, calculated from measured membrane characteristics. The unit of distance, abscissa, is the characteristic length for small current.

tion of an increased ion permeability of a membrane under a cathode is valid. However, the impedance change was negligible before the point of inflection (Cole and Curtis, 1939) although the steady state effect of current flow was as predicted (Cole and Baker, 1941*a*). The contradiction then remains on the basis of a capacity-resistance membrane structure and the effect of the inductance element should be considered.

The foot of the action potential has been found to be accurately exponential in form in *Nitella* (Cole and Curtis, 1938) and in the squid axon (unpublished experiments), and this form is to be expected, in a capacity or capacity-resistance membrane, ahead of a sudden partial short circuit of the membrane moving with a constant velocity along the axon (Cole and Curtis, 1938). We may now extend this calculation to include any form of membrane impedance so long as it is linear; *i.e.*, if the effect of the rectification may be ignored. Thus the observed exponential form agrees with the impedance observations in requiring that the membrane rectification is not involved in the foot of the

action potential. But this requirement can only be met if both the potential difference across, and the current flow through, the rectifier element are so small that its non-linear characteristics are negligible. Subject to verification, we may assume the resistance of Fig. 1 to be linear and to have a value of 300 ohm cm.^{2} and calculate the potential and current during the foot of the action potential. For the membrane capacity, we have the current $I_c = C\ dV/dt$, and if $V = V_0\ e^{t/\tau}$ during this period, $I_c = CV/\tau$ where τ is the time constant of the action potential foot. For the inductance-resistance arm of the circuit, $V = RI_R + L\ dI_R/dt$, from which $I_R = V/(R + L/\tau)$. Then when $\tau = 10^{-4}$ sec., $\tau/C = 100$ ohms and $L/\tau = 2000$ ohms. Thus we see that only 5 per cent of the total current flows through the resistance and inductance arm and that only about 15 per cent of the action potential is across the resistance. If the action potential is 50 mv. and the point of inflection is at 25 mv., the capacity current at this time will be 250 μ amp./cm.^{2} The current density through the rectifier will be about 12 μ amp./cm.^{2} at this time and the potential across it will be about 3.6 mv. From Fig. 5 (Cole and Curtis, 1941) it is seen that these values of current and potential difference lie so near the origin that the potential difference is practically proportional to the current. Since the membrane has not entered the non-linear range it may be considered as a simple resistance. With the current flow in this arm and potential difference across the resistance small, we must conclude that the inductive element effectively removes this arm from participation in the foot of the action potential. For slow changes, on the contrary, we may expect the inductance to become less effective and allow a larger potential difference and current flow for the rectifier element. These conclusions find considerable confirmation in unpublished records of the form of the action potential during passage through a polarized region. It has been found that the time constant of the foot of the potential remains the same for anodic and cathodic current flow as the unpolarized axon. Also, the recovery of polarized nerve is slower under the anode and faster under the cathode than normal, but the effect of change of velocity of propagation has not been investigated.

After the application of a constant current, it would be expected that the paths of the current would vary in a similar manner. The current flow through the inductance and rectifier should be negligible at the start and pass through a point of inflection as it builds up to the steady state value. The impedance of the membrane should then show similar characteristics as the current flow modifies the apparent resistance of the rectifier, and a few unpublished records of the impedance change during current flow show the expected characteristics.

Thus we may conclude that, on the basis of the evidence available, there is good reason to believe that the equivalent circuit of Fig. 1 is an adequate correlation and representation of the observed characteristics of the squid axon with the subthreshold potentials at intermediate and long times. At short times

the computations are certainly unsatisfactory, and it is also quite probable that we have here additional evidence that the simple circuit is not a complete representation of the membrane.

Threshold and Excitability

So far, attention has been centered on subthreshold potentials during current flow and the foot of the action potential as measured independent of excitation, and a considerable degree of consistency has been found, along with a number of unsatisfactory aspects. The temptation, however, is very strong to ignore the difficulties for the present and turn to excitation. This work is only a preliminary step in the description of nerve function until it can be extended to include excitation and propagation phenomena. The qualitative description of these phenomena in terms of a membrane breakdown has been so satisfactory that it is certainly not expedient to desert this picture until it can be shown necessary to do so. We may then expect that the two crucial steps in a quantitative description of the phenomena are (1) the conditions for this breakdown to occur, and (2) the nature of the breakdown. But it must be admitted at the present time that there is no satisfactory description of either of these two steps. As a consequence it has been necessary, in order to formulate the initiation and propagation of the nerve impulse mathematically, to make largely unsupported assumptions as to the nature of these two steps. It is possible to assume a variety of membrane circuits and assume that a critical charge, potential difference, or current flow, etc., in any of the elements will result in a change in one or more of the elements of capacity, resistance, inductance and E.M.F. It has been shown that several combinations of these *a priori* possibilities can indeed describe various nerve phenomena rather well, but it seems certain that many other—although perhaps intuitively less satisfactory—permutations of assumptions may do equally well.

We should, therefore, not consider these comparatively specific theories in detail but go back to the experimental data which they attempt, more or less successfully, to correlate. Almost without exception these data have been data of excitation and it appears to have been too optimistic to hope that it would have been possible to determine both the structural and the functional characteristics of nerve from these functional data alone.

These functional data, however, are the backbone of classical electrophysiology of nerve, and they form a self-consistent picture as pointed out qualitatively by Cole (1933 *b*). The quantitative correlations were carried out by Rashevsky, 1933, Monnier, 1934 and Hill, 1936 as two factor theories in which one of the factors, U, tended towards the excitation and the other, V, away from it. All the formulations may be considered as special cases of general equations set up by Young (1937)

$$\frac{dU}{dt} = k_{11}U + k_{12}V + a_1 I$$
$$\frac{dV}{dt} = k_{21}U + k_{22}V + a_2 I, \tag{6}$$

where I is the stimulating current, a_1 and a_2 are constants, and the k's are velocity coefficients, from which the two time constants may be derived. These equations have been considered from a general conventional mathematical viewpoint by Young, but we may consider them in a more informal manner since it has usually been assumed that excitation is governed only by the difference $U - V = \theta$, which we may call the excitability.

The two simultaneous first order equations (6) may now be replaced by a single second order equation (Forsythe, 1921[1]) as was mentioned by Rushton (1937*a*) for the form considered by Hill. Then equations (6) may be put in the form (Parrack, 1940)

$$\frac{d^2\theta}{dt^2} - (k_{11} + k_{22})\frac{d\theta}{dt} + (k_{11}k_{22} - k_{12}k_{21})\theta = (a_1 - a_2)\frac{dI}{dt} - [(k_{21} + k_{22})a_1 - (k_{12} + k_{11})a_2]I \tag{7}$$

This equation correlates a vast amount of data as reviewed by Katz (1939) for various forms of electrical stimuli, square pulse, condenser discharge and charge, and alternating current. However, a difficulty arose when an oscillatory excitability was found for a single frog fiber by Erlanger and Blair (1936). This phenomenon was investigated in more detail and particularly in relation to nerve calcium and alternating current threshold by Monnier and Coppée (1939) and it was concluded that one of the two time constants in Monnier's formulation would have to be a complex quantity.

It can be shown quite generally that under certain conditions U and V may be oscillatory (Forsythe, 1921[2]), and these conditions have been applied to population problems by Lotka (1925[3]) and repetitive discharge by Householder (1939). Considering only the excitability, it is seen that equation (7) has an oscillatory solution when k_{12} and k_{21} are of opposite sign and $(k_{11} - k_{22})^2 < 4k_{12}k_{21}$. These are then the conditions for complex time constants in the sense of Monnier and Coppée and in other cases the time constants are real. Thus we see that equation (7) for the excitability of a nerve can represent a very considerable amount of the available data.

[1] Page 350.
[2] Page 342.
[3] Page 78.

If we now may represent the axon membrane by a capacity, inductance, and constant resistance, the relation between the potential difference across it, V, and the current, I, given by equation (1), is

$$\frac{d^2V}{dt^2} + \frac{R}{L}\frac{dV}{dt} + \frac{1}{LC}V = \frac{1}{C}\cdot\frac{dI}{dt} + \frac{R}{LC}I. \tag{8}$$

This equation also has an oscillatory solution when $R < 2\sqrt{L/C}$, and two real time constants for $R > 2\sqrt{L/C}$. The similarity of equations (7) and (8) is highly suggestive, and one is tempted at this point to identify the excitability θ with the membrane potential, V, and combinations of the k and a coefficients with R, L, and C. It is obviously possible to replace the various quantities and coefficients defined only by excitation measurements with constants which are physically defined and measured without reference to excitation. Then all data which are represented by equation (7) may be represented equally well by equation (8) if the numerical values of R, L, and C will permit.

It has long been a favorite assumption that excitation took place when a threshold change of the membrane potential occurred. Qualitative support for this assumption is seen in the records of the potential following the application of a constant stimulus. As the threshold is reached the action potential appears to begin at the time of the first minimum of the subthreshold potential (Cole and Curtis, 1941). The recent investigation by Pumphrey, Schmitt, and Young (1940) of the excitability and local potential following short shocks also indicates a close correlation between these two quantities. Consequently we are entirely justified in assuming a close relation between the two, but for the present it is not certain that a simple proportionality applies for all types of stimulation. Furthermore in repetitive response to a constant stimulus the successive excitations take place at approximately the times when the successive maxima of the subthreshold potential would be expected (Arvanitaki, 1939). However, sufficient data are not available in such experiments to satisfactorily define the time at which excitation takes place and determine the membrane potential at that point.

On the other hand, in the propagated impulse it does seem very plausible that excitation takes place at about the point of inflection of the rising phase of the action potential, and it has often been assumed that a threshold change of membrane potential has occurred at this point. From unpublished records of the membrane potential during the passage of an impulse through a polarized region there is evidence that the potential at the point of inflection depends upon the polarizing current and also that the difference between this potential and the steady state potential depends upon the current. In other words, it has not yet been possible to find any membrane potential or change of membrane potential having a value at the point of inflection which is independent of the polarizing current.

Although equation (8) may be solved for all possible forms of stimulating current, the general features are most easily seen for alternating current stimulation. The current necessary for a constant value of the potential difference may be computed directly as a function of frequency from equation (8). It is also proportional to the reciprocal of the absolute value of the impedance of the equivalent circuit of Fig. 1. The membrane impedance has a maximum for all values of the damping, $\eta = R\sqrt{C/L}$, less than 0.7 and at a frequency below the natural undamped frequency $1/2\ \pi\sqrt{LC}$ as seen in Figs. 3 *b*, 5, and 7 of the previous paper (Cole and Baker, 1941 *b*). An optimum frequency for excitation is then to be expected somewhat below 330 cycles for the squid axon membrane. Adequate data are not available, but preliminary results between 150 cycles and 200 cycles at least indicate that the orders of magnitude of the membrane capacity, C, and inductance, L, are correct.

The excitability oscillations at a frequency of 200 cycles (Erlanger and Blair, 1936; Monnier and Coppée, 1939) and optimum frequency for alternating current excitation from 100 to 250 cycles (Katz, 1939; Monnier and Coppée, 1939) for the frog sciatic nerve are too near the region of the undamped natural frequency of the squid membrane to be ignored. This approximate agreement may certainly be taken as an indication that the electrical structure of the two nerves may be similar.

It has usually been found that the threshold alternating current stimulus, as a function of the logarithm of the frequency, was symmetrical with respect to the optimum frequency. This characteristic is dependent upon the presence of complete accommodation, which means that the coefficients of the current, I, in equations (7) and (8) must be zero (Parrack, 1940). Since no evidence of incomplete accommodation was found in the two squid axons tested, it is again apparent that the simple circuit of Fig. 1 is not adequate on the basis of the present assumptions.

However interesting as this comparison may be, it cannot be taken too seriously. From a rigorous point of view it probably must be regarded at present as coincidental and fortuitous. In the first place the excitability equation (7) takes into account distributed effects and the variation of membrane current flow at and near the electrodes, while the potential equation (8) applies only to a uniform current density. In the second place, the potential equation (8) does not take into account the rectifier characteristic of the membrane. Thirdly, a variety of observations indicate that a liminal change of membrane potential is not the necessary and sufficient condition for excitation. When it becomes possible to make calculations from equations (4), (5), and (6) and more complete information on excitation is available, we shall be in a better position to judge the extent to which the physical data may explain the phenomenological formulation of excitability.

It is interesting to note that the two factor theories have been created for the sole purpose of explaining the behavior of nerve at relatively long times as compared with chronaxie or excitation time, and that a linear relation between cause and effect has been assumed here as well as for short times. The electrical measurements, however, indicate a non-linear element which probably becomes a controlling factor at long times but is relatively unimportant, as we have seen, at short times. However, the excitability data may not be symmetrical for anode and cathode (Parrack, 1940) although this asymmetry is much less than is found for the membrane potential or calculated by integration of the rectifier equation (2). Mention has already been made of the relative spread of steady state electrotonus at the anode and cathode (p. 39).

As a basis for calculation, Rushton (1937 *b*) and Offner, Weinberg, and Young (1940) have each explicitly assumed a membrane structure, and a condition and nature of excitation which are equivalent. On the basis of these assumptions, Rushton found it necessary to have a minimal length of the axon fulfill this excitation condition before an impulse could be propagated. As soon as it is recognized that there can be a departure from the resting or linear properties of the axon membrane before excitation takes place, the minimal length becomes difficult to specify exactly. It is quite possible that this length is an equivalent of the region over which the requisite non-linear change has taken place without excitation. Similarly, we may expect to find that the non-linear responses of Pumphrey, Schmitt, and Young (1940), which they referred to as physiological, are to be related to the electrical non-linearity of the membrane. A considerable amount of experiment and careful theory, taking into account the capacity, rectification, and inductance, will be necessary to verify such suggestions.

Until reasonably detailed calculations have been carried out, it is unsafe to predict the performance of the equivalent circuit for high frequency currents at and near the threshold for excitation. It may, however, be expected, optimistically perhaps, that, as has been suggested, the membrane rectification plays a considerable rôle in these various phenomena (Gildemeister, 1930; Katz, 1937; Rosenbleuth, Reboul, and Grass, 1940). For example, the considerable delay in the appearance of excitation at high frequencies would be explained by a rectification which allowed an average cathode direct current to flow across the membrane under the electrode. This current might be expected to build up at a rate determined largely by the natural frequency and damping of the membrane until the threshold was reached. It is obvious, however, that we cannot expect this characteristic at one electrode and not at the other. Consequently, this postulated average cathode current is a local rather than an external current and there must be nearby anode regions and perhaps even more distant alternating cathode and anode regions. Under some conditions the anode regions may have so high a current density as to give

the alternating current blocks of conduction which have often been observed, and it is then completely consistent to have the well known "break" excitation from this region when the alternating current is stopped.

DISCUSSION

In the first place it is interesting to consider the lack of earlier direct evidence for rectification and inductance elements in nerve. If these elements are present in other nerves, the usual small size of the axons is probably the most important obstacle to their measurement. The measurements must be made at low frequencies or long times, relatively speaking, to avoid having the membrane capacity carry too large a fraction of the membrane current. The membrane then presents so high an impedance that the current will flow almost entirely in the extra-cellular fluids unless a large area of membrane is exposed and the alternative paths are reduced to a minimum. These requirements are most easily approached by a large single axon such as Young's squid giant axon preparation. However, transverse impedance measurements on this axon failed to give any indication of an inductance or even a finite membrane resistance because there was so much fluid surrounding the axon that the membrane current at the low frequencies was negligible with the measuring sensitivity available. This difficulty is greatly reduced in the longitudinal measurements because a relatively large proportion of the electrode current crosses the membrane. However, rectification was not found in the investigation of membrane resistance by direct current longitudinal measurements (Cole and Hodgkin, 1939) where one might have expected a change of resistance with current strength. The decrease of resistance at the cathode certainly compensated for the increase at the anode to some extent in these experiments, and the net result of these two effects may be calculated from the data of Fig. 5 (Cole and Curtis, 1941). The total current flow through the membrane is given by $i_2 = [r_1/(r_1 + r_2)]i_0$, where i_0 is the electrode current, and r_1 and r_2 are the external and internal resistances per unit length. The apparent resistance of the membrane for one electrode, $r_m = V_m/i_2$, where V_m is the change of membrane potential, is calculated as the function of the cathode current given in Fig. 4. The membrane current flow for the second electrode is in the opposite direction, giving the anode curve. For the two membranes in series we then have the sum of the two individual resistances as shown by the third curve. The change of resistance with current for the membranes alone is about 10 per cent, but when the internal and external resistances are included the maximum variation of the measured resistance predicted for a 1 cm. interpolar distance is about 1 per cent. Not only is this within the limits of measurement but the currents considered here are much larger than those used by Cole and Hodgkin. Furthermore, in preliminary experiments on the squid axon with one killed end, the variation of the longitudinal direct current re-

sistance with the magnitude and direction of the current has been measured (Guttman, unpublished).

Since the membrane circuit discussed is admittedly inadequate, a few of the more obvious defects may be pointed out. The membrane electromotive force has been assumed constant and is therefore omitted except insofar as it appears in the rectifier element. The capacity, for example, has been adequately demonstrated to have considerable dielectric loss, which is ignored for simplicity. The inductance is treated as independent of frequency but this cannot be verified until the complicated effect of dielectric loss is adequately

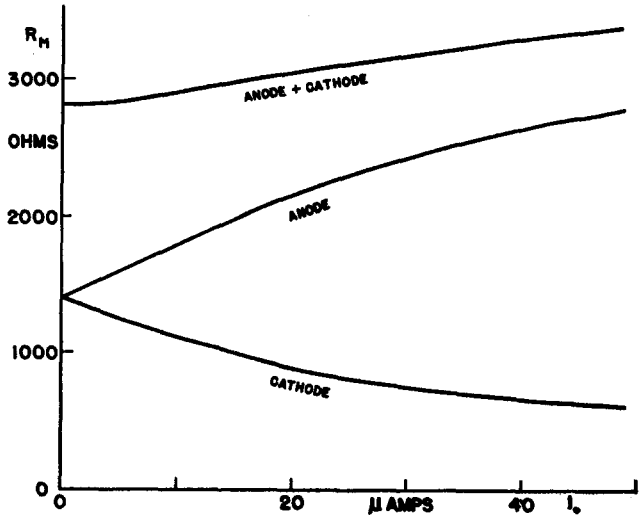

FIG. 4. Effective resistance of axon membrane, R^M, *vs.* total measuring current, I_0, for anode and cathode separately and in series.

calculated. Also, both the capacity and inductance are assumed to be quite invariable. This seems to be justified for the capacity, but there is little evidence as yet concerning the stability of the inductance. Turning to the rectifier, it has been noted that the theoretical impedance loci for the circuit shown in Fig. 1 are only in qualitative agreement with the measurements. A decrease of the resistance should decrease the damping and so increase the inductive reactances at low frequencies. But it was found that the inductive reactance decreased as the direct current resistance of the axon membranes decreased when they deteriorated and lost excitability.

The measurements of the changes of transverse impedance during current flow and the membrane potential during current flow have given similar rectification characteristics for the membrane. The membrane potential was

considered under the direct current steady state conditions in which the inductance would have no effect, but the transverse impedances were measured at frequencies of 2 kilocycles and higher. Under these conditions, the effect of a rectifier placed as indicated in Fig. 1 would be considerably reduced by the inductance and consequently dependent upon the frequency, but the measurements were interpreted on the basis of a frequency independent rectifier element in parallel with the capacity. The addition of such an element in the circuit of Fig. 1 will lessen or alleviate some of these inconsistencies, but more adequate experimental data are needed to specify such an additional factor definitely and justify the increased complexity of analysis.

The corollaries of a piezoelectric membrane structure must not be overlooked. If an applied potential difference can deform the membrane, the action potential should have a similar effect. In either case a change of the membrane double refraction is to be expected, but careful experiments by Schmitt and Schmitt (1940) failed to show such an effect during the passage of an impulse in the squid axon. It is of course possible that the axon membrane is too thin for such observations, but this can only be conjecture at present. On the other hand, if a potential difference can deform the membrane, a deformation can change the potential difference. If there is a threshold potential difference for excitation there should also be a threshold mechanical deformation with similar temporal characteristics. Mechanical excitation of nerve has long been known, but it has been difficult to measure experimentally. None the less the temporal characteristics as exhibited by the strength-duration curve (Blair, 1936) are at least in the same general range as for electrical excitation.

It has already been mentioned that simple calculations of the potential after a short pulse of current applied to the circuit of Fig. 1 do not agree with the observations on crab and squid axons. Also the excitation characteristics to be expected from this circuit do not include the case of complete accommodation which has usually been treated theoretically and investigated experimentally. These defects may well be results of the over-simplification of the membrane properties which has been made. Preliminary measurements and calculations indicate that the modifications of the membrane circuit which are needed to explain the impedance measurements more satisfactorily are in the proper direction to reduce the discrepancies between the observed and predicted potentials and excitabilities.

Measurements of a range of forms have revealed a considerable consistency in the value of the membrane capacity and in a few cases it has been found also that this capacity is quite independent of the physiological function and condition of the cell. The reasonable conclusion is then that the capacity represents a highly constant and inert structure of the membrane. It has been found that a number of under- and over-damped oscillatory nerve phenomena occur

in the range from 100 to 500 cycles and an inductance has been shown to be an essential element in squid axon structure. If this element also appears in frog and *Sepia* axons, the order of magnitude of the inductance will be the same as for the squid, for a variety of conditions and experiments. It would then seem very likely that all of these and probably other nerve phenomena are dependent upon a membrane inductance and that this inductance is at least as constant and inactive as the membrane capacity. It is much too early to predict that an inductance is a characteristic of a nerve membrane, much less of all cell membranes, for it could conceivably be the distinguishing characteristic of membranes having a propagated activity.

SUMMARY

Previous measurements have shown that the electrical properties of the squid axon membrane are approximately equivalent to those of a circuit containing a capacity shunted by an inductance and a rectifier in series. Selective ion permeability of a membrane separating two electrolytes may be expected to give rise to the rectification. A quasi-crystalline piezoelectric structure of the membrane is a plausible explanation of the inductance. Some approximate calculations of behavior of an axon with these membrane characteristics have been made. Fair agreement is obtained with the observed constant current subthreshold potential and impedance during the foot of the action potential.

In a simple case a formal analogy is found between the calculated membrane potential and the excitability defined by the two factor formulations of excitation. Several excitation phenomena may then be explained semi-quantitatively by further assuming the excitability proportional to the membrane potential.

Some previous measurements and subthreshold potential and excitability observations are not consistent with the circuit considered and indicate that this circuit is only approximately equivalent to the membrane.

REFERENCES

Arvanitaki, A., 1939, *Arch. int. physiol.*, **49**, 209.

Blair, H. A., 1936, *Am. J. Physiol.*, **114**, 586.

Blinks, L. R., 1930, *J. Gen. Physiol.*, **14**, 127.

Cole, K. S., 1928, *J. Gen. Physiol.*, **12**, 29; 1933*a*, Cold Spring Harbor symposia on quantitative biology, Cold Spring Harbor, Long Island Biological Association, **1**, 107; 1933*b*, Cold Spring Harbor symposia on quantitative biology, Cold Spring Harbor, Long Island Biological Association, **1**, 131; 1937, *Tr. Faraday Soc.*, **33**, 966; 1940, Cold Spring Harbor symposia on quantitative biology, Cold Spring Harbor, Long Island Biological Association, **8**, 110.

Cole, K. S., and Baker, R. F., 1941*a*, *J. Gen. Physiol.*, **24**, 535; 1941*b*, *J. Gen. Physiol.*, **24**, 771.

Cole, K. S., and Curtis, H. J., 1938, *J. Gen. Physiol.*, **22,** 37; 1939, **22,** 649; 1941, J. Gen. Physiol., **24,** 551.
Cole, K. S., and Hodgkin, A. L., 1939, *J. Gen. Physiol.*, **22,** 671.
Erlanger, J., and Blair, E. A., 1936, *Am. J. Physiol.*, **114,** 328.
Forsyth, A. R., 1921, A treatise on differential equations, London, Macmillan.
Gildemeister, M., 1930, *Ber. Verhandl. sächs. Akad. Wissensch. Leipzig*, **81,** 303.
Hill, A. V., 1936, *Proc. Roy. Soc. London, Series B*, **119,** 305.
Hodgkin, A. L., 1938, *Proc. Roy. Soc. London, Series B*, **126,** 87.
Householder, A. S., 1939, *Bull. Math. Biophysics*, **1,** 129.
Katz, B., 1937, *Proc. Roy. Soc. London, Series B*, **124,** 244; 1939, Electric excitation of nerve, London, Oxford University Press.
Langmuir, I., 1938, *J. Chem. Physics*, **6,** 873.
Lotka, A. J., 1925, Elements of physical biology, Baltimore, The Williams & Wilkins Co.
Monnier, A. M., 1934, L'excitation électrique des tissus, Paris, Hermann.
Monnier, A. M., and Coppee, G., 1939, *Arch. int. physiol.*, **48,** 129.
Mueller, H., 1939, *Physic. Rev.*, **55,** 792.
Norton, F. J., 1939, *Physic. Rev.*, **55,** 668.
Offner, F., Weinberg, A., and Young, G., 1940, *Bull. Math. Biophysics*, **2,** 89.
Parrack, H. O., 1940, *Am. J. Physiol.*, **130,** 481.
Pearson, G. L., 1940, *Bell. Lab. Rec.*, **19,** 106.
Pumphrey, R. J., Schmitt, O. H., and Young, J. Z., 1940, *J. Physiol.*, **98,** 47.
Rashevsky, N., 1933, *Protoplasma*, **20,** 42.
Rosenbleuth, A., Reboul, J., and Grass, A. M., 1940, *Am. J. Physiol.*, **130,** 525.
Rushton, W. A. H., 1937*a*, *Proc. Roy. Soc., London, Series B*, **123,** 382; 1937*b*, *Proc. Roy. Soc. London, Series B*, **124,** 201.
Schmitt, F. O., 1936, Cold Spring Harbor symposia on quantitative biology, Cold Spring Harbor, Long Island Biological Association, **4,** 7.
Schmitt, F. O., and Schmitt, O. H., 1940, *J. Physiol.*, **98,** 26.
Young, G., 1937, *Psychometrika*, **2,** 103.