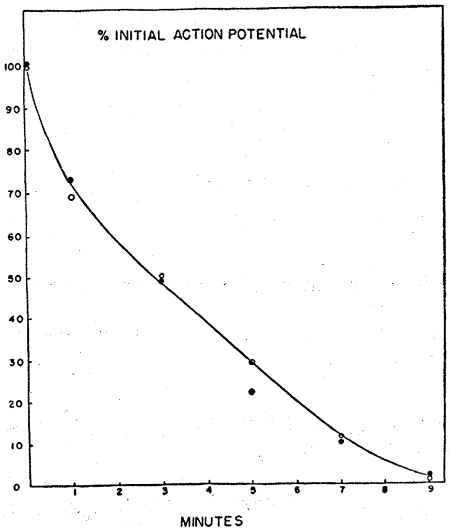

Fig. 1. Amplitude of action potentials obtained for strips immersed in cyanide and α-dinitrophenol.

tials. This mechanism of action implies that cyanide acts physically on the membrane in addition to having an effect on the cytochrome enzymes.

The inhibitors may act on mammalian nerve by blocking energy sources immediately necessary for impulse transmission. This hypothesis assumes that mammalian nerve differs from frog and squid nerve in containing a system in which the action potential is linked to the energy sources. Inhibition of the sodium extrusion mechanism, definitely dependent upon metabolism, appears to be excluded. The action of cyanide and α-dinitrophenol within minutes precludes the possibility that enough internal sodium or potassium ion accumulation occurs to depress the action potential. It is not yet possible, however, to decide whether the inhibitors act by binding physically with a membrane component or whether they act upon a metabolic system within mammalian nerve which generates the action potenial (6).

G. SANT'AMBROGIO
D. T. FRAZIER
L. L. BOYARSKY
Department of Physiology, College of Medicine, University of Kentucky, Lexington

References and Notes

1. A. L. Hodgkin and R. D. Keynes, *J. Physiol. (London)* **128**, 28 (1955).
2. R. Straub, *Helv. Physiol. et Pharmacol. Acta* **14**, 1 (1956).
3. R. Lorente de Nó, "A Study in Nerve Physiology," in *Rockefeller Institute Medical Research Studies* (New York, 1947), vols. 131 and 132.
4. W. D. Dettbarn and R. Stämpfli, *Helv. Physiol. et Pharmacol. Acta* **15**, 25 (1957).
5. G. M. Schoepfle and F. E. Bloom, *Am. J. Physiol.* **197**, 1131 (1959).
6. This work was supported by a U.S. Public Health Service grant B-1176(C).

30 November 1960

Self-Absorption Correction for Isotopes Emitting Weak Beta Rays

Abstract. The shape of the self-absorption correction curve of β-emitters is not universal but depends on the geometrical arrangement of the sample and counting device. This may explain why the correction factor defined by Hendler (*1*) is not always linearly related to the thickness of the sample.

In determining the radioactivity of samples of radioactive isotopes emitting weak β-rays, such as C^{14}, S^{35}, and H^3, one has to take into account that part of the radiation is absorbed in the sample itself. R. W. Hendler has described a method for calculating the self-absorption correction factor for C^{14} assay (*1*). A series of planchets of constant area containing different known quantities of the same radioactive material is prepared , and the weight m (in milligrams) and the specific count rate (in counts per minute per milligram) are recorded for each planchet. A standard weight m_{std} is chosen arbitrarily and the correction factor F (as a function of m) is defined as the ratio of the specific count rate R at weight m_{std} to the specific count rate at weight m. Hendler found experimentally that F was related to m by a linear function from infinite thinness to a weight of several times saturation thickness. This relation was contested by J. Katz (*2*), who calculated F for several sets of experimental data and found that F was not linearly related to m.

The treatment of experimental data on self-absorption is mostly based on the fact that the absorption curves of many β-emitting isotopes are approximately exponential up to a certain thickness of the absorber (*3*). If the exponential equation is applied to the set of data for $BaS^{35}O_4$, published by Katz and Golden (*4*) and quoted by Hendler in his reply (*5*), F is not found to be a linear function of m, although within a limited range of values of m the deviation from a straight line is not great. The experimental data are fitted much better by a function of m which is derived from the exponential absorption equation, provided the absorption coefficient is chosen appropriately. The parameters used for these data were $R/I_\infty = 0.0569$ and $\alpha = 0.0956$ (see below). The reference weight was 12 mg over an area of 3.14 cm^2. Within the range given by Katz and Golden ($m = 4$ to 25 mg), the deviation of the function F was smaller than 1 percent, whereas the straight line, which was suggested by Hendler (*5*), deviates by as much as 2 percent from the curve given by Katz and Golden.

From a mathematical treatment of the exponential absorption equation it follows that the slope of the curve of F plotted against m increases by a factor of two when going from $m = 0$ to $m = \infty$.

The absorption of radiation can be measured by interposing a filter of weight m over a fixed area between a sample containing a certain isotope and the detecting device. It can be seen for many isotopes that

$$I = I_0 \cdot e^{-\alpha m} \qquad (1)$$

where I is the observed count rate in counts per minute, I_0 is the count rate without filter, and α is the absorption coefficient in milligrams^{-1} (*6*). This means that each layer dm of the filter decreases the radiation by the same fraction (Beer's Law). For the self-absorption it follows (*4*) that

$$I = I_\infty (1 - e^{-\alpha m}) \qquad (2)$$

where I_∞ is the observed count rate at infinite thickness. The specific count rate is I/m, and the correction factor F is defined as

$$F = (R/I)m \qquad (3)$$

where R is the specific count rate at weight m_{std}. Substituting Eq. 2 in Eq. 3 we obtain

$$F = \frac{R}{I_\infty} \cdot \frac{1}{1 - e^{-\alpha m}} \cdot m \qquad (4)$$

By definition, F is 1 for $m = m_{std}$, and

$$R = \frac{I_\infty}{m_{std}} (1 - e^{-\alpha m_{std}}) \qquad (5)$$

F can be analyzed for small values of m by expanding the exponential term into a series and by again expanding the resulting fraction into a binomial series, cutting off after the terms of the second degree:

$$F = \frac{R}{I_\infty} \cdot \frac{1}{\alpha m} \left[1 + \frac{\alpha m}{2} + \frac{\alpha^2 m^2}{12} \right] \cdot m$$
$$= \frac{R}{\alpha I_\infty} + \frac{R}{2\,I_\infty} \cdot m + \frac{R \cdot \alpha}{12\,I_\infty} \cdot m^2 \qquad (6)$$

It follows that the intercept of the curve of F against m on the ordinate is $R/(\alpha \cdot I_\infty)$, that the initial slope is $R/(2 \cdot I_\infty)$, and that the curve is bending upwards.

For high values of m (region of infinite thickness) the exponential term in Eq. 4 can be neglected against 1, and we get

$$F = \frac{R}{I_\infty} \cdot m \qquad (7)$$

which is a straight line through the origin with a slope of R/I_∞, that is, two times as high as at $m = 0$.

Whereas the exponential function fits very well the data of Katz and Golden (and many other sets of data published earlier), it is evident that the data cited by Hendler (*1*) are fitted much better by a hyperbolic function than by an exponential one. It seems thus that it depends on the counting arrangement, or possibly on the method of sample preparation, whether or not the apparent absorption coefficient, as obtained from the experimental data, is independent of the sample thickness.

It is obvious that α cannot be compared directly with the absorption coefficient of Beer's law because β-rays interact with matter in a way essentially different from electromagnetic waves. The absorption process of β-rays is a complex one, including, besides the energy losses by excitation and ionization of the absorber atoms, also scattering and backscattering, two processes which change the energy spectrum and its angular distribution (*7*).

It is therefore to be expected that it depends on the geometrical arrangement of the sample and counting device whether or not the self-absorption can be described by an exponential law with a constant α.

P. MASSINI

Philips Research Laboratories,
N.V. Philips' Gloeilampenfabrieken,
Eindhoven, Netherlands

References and Notes

1. R. W. Hendler, *Science* **130**, 772 (1959).
→ J. Katz, *ibid.* **131**, 1886 (1960).
3. This law is discussed already → W. Wilson, *Proc. Roy. Soc. London A* **82**, 612 (1909); see also M. Calvin *et al.*, *Isotopic Carbon* (Wiley, New York, 1949), p. 21.
4. J. Katz and S. B. Golden, *J. Lab. Clin. Med.* **50**, 658 (1959).
→ R. W. Hendler, *Science* **131**, 1887 (1960).
6. α differs in meaning from the absorption coefficient as usually defined, in that it contains the area over which *m* is measured. Usually the area is taken as 1 cm^2 and *m* becomes the thickness in mg/cm^2.
7. G. L. Brownell, *Nucleonics* **10**(6), 30 (1952).

16 November 1960

Action Potential and Contraction of Dionaea muscipula (Venus Flytrap)

Abstract. Observation of the action potential and contraction of the leaf of *Dionaea muscipula* Ellis revealed several interesting phenomena. Two successive stimuli are generally necessary to cause contraction. The first and ineffective stimulus is associated with slow depolarization. The second stimulus has much more rapid depolarization and initiates contraction.

The excitatory and contractile processes of *Dionaea muscipula* Ellis are of interest to biologists because of certain similarities and certain differences from the same phenomena in mammalian organs. *Dionaea muscipula* belongs to a small group of plants of the carnivorous type which are capable of trapping insects and then digesting them. The feature of interest in this study is the leaf or flytrap which contracts upon stimulation of one of the inner sensitive hairs. This plant early attracted the attention of Darwin (*1*). A complete description and pertinent references may be found in Lloyd's classical monograph (*2*). Bourdon-Sanderson (*3*) as early as 1873 recorded its electromotive properties with a capillary electrometer. Relatively little attention seems to have been paid to it until Stuhlman (*4, 5*) recorded its action potential and characteristic of the contractile process in 1948–50 with modern methods. He showed the similarity of the *Dionaea's* action potential to that of mammalian nerve and reported on the variations to be expected from positioning of the electrodes, health and age of the plant, temperature and intensity of the stimulus. It was further shown that the action potential may run its course without producing closure of the flytrap. The present report is directed towards a further delineation of the excitatory and contractile processes.

Fresh adult healthy specimens (*6*) were obtained in early summer. They were kept in a suitable terrarium at 26°C under 12 hours of flourescent light daily. With only minor differences the method of recording the action potential was similar to that described by Stuhlman (*5*). The contraction was simultaneously recorded isometrically with a strain-gauge transducer and amplifier. Stimulation was simply done by a fine cat whisker touching one of the sensitive hairs of the inner leaf. Although the experiment was monitored with a dual-beam oscilloscope, recording was actually done with a Sanborn oscillograph. The events of excitation and contraction proved to be slow enough to be reliably recorded by this method. Only leaves with a diameter of 1.4 cm ±3 mm were used. The leaf opening usually measured 1 cm or more.

In over a hundred trials it was found that the leaf contracted upon the second stimulus. Occasionally contraction required three stimuli at 2-second intervals. Rarely did the leaf contract on the first stimulus. A typical experiment is shown in Fig. 1.

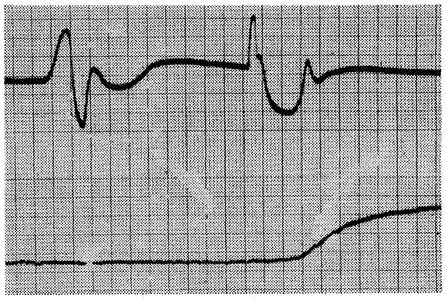

Fig. 1. Action potential and contraction of leaf of *Dionaea muscipula* Ellis. Top, action potential. Bottom, contraction. Abscissa scale (heavy lines) equals 0.2 second. Ordinate scale (heavy lines) equals 5 mv. Note that the first stimulus elicits a slow action potential which is not effective. The second action potential has much faster depolarization and causes contraction of the leaf.

The noteworthy features are the similarity of the general contour of the action potential to that of mammalian tissues such as heart muscle (*7*). There is a rapid negative phase followed by a positve after-potential. Several minor after-oscillations may occur in some leaves. However, the item of note is that the first potential elicited showed comparatively a slower depolarization rate than the following one. This important difference is documented in Table 1, which shows the distinct shortening of the duration of the negative phase (0.13 second) of the second action potential as compared to that of the first action potential (0.24). Apparently, for the excitatory process to initiate contraction, the rate of depolarization must attain a certain velocity.

From Table 1 it may also be ascertained that the positive after-potential does not show the distinct changes upon repeated stimulation as the negative phase. Not infrequently, leaves which failed to develop more rapid deplorization upon repeated stimulation also failed to contract. However, this observation was not constant, for some leaves showing characteristic action potential changes also failed to contract. Other factors which influenced the contractile process were obviously also critically important.

In the 31 leaves it was possible to determine that the mean delay between the second or effective stimulus and the onset of contraction was on the average 0.6 second (standard error, ±0.05). The time which elapsed between the onset of contraction and the development of initial tension was 1.07 seconds (average standard error, ±0.14). The attainment of maximum tension took an additional 6 to 7 seconds. By

Table 1. Comparison of the first action potential (ineffective) and second action potential (effective) of two stimuli on 31 separate leaves of *Dionaea muscipula* Ellis. Figures in the first lines under each phase are means; figures in the second lines are standard errors.

Action potential			
First		Second	
Amplitude (mv)	Duration (msec)	Amplitude (mv)	Duration (msec)
	Negative phase		
11.2	0.24	14.6	0.13
±0.8	±0.1	±0.7	±0.02
	Positive phase		
10.4	0.76	8.4	0.65
±0.8	±0.1	±0.9	±0.07