The purpose of this analysis has been twofold: first, to suggest that the concepts of nucleation and growth, developed for physical systems, apply to the growth of cell colonies, and second, to show that elementary diffusion equations can be employed to calculate the influence of container geometry upon the critical size of tissue explants. It is further suggested that the spontaneous appearance of cancer may be a process of nucleation in that single cancer cells in normal tissue may be subcritical with respect to malignant growth.

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The Action Potentials Obtained from Venus's-Flytrap

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Venus's-flytrap, *Dionaea muscipula,* is included in a small group of plants designated by botanists as carnivorous because of its reputed ability to digest insects and small bits of living matter caught in the trap structure from which the plant takes its popular name. It is found growing in the wild state only in the eastern part of the Carolinas of the United States.

In 1769, William Young of Philadelphia brought a live specimen to England. Bourdon-Sanderson (*2, 5*) as early as 1873 discovered the electromotive properties of the leaf of *Dionaea* in the excited and unexcited states, using specimens from the laboratory of the Royal Gardens at Kew. Bose (*1*) criticized these earlier results and Bourdon-Sanderson's later results (*3, 4*), published in 1882 and 1888, as being more or less contradictory.

According to Bourdon-Sanderson one should expect, as a result of stimulating the inner surface of *Dionaea* in its prime, to observe a short-lived positive electrical increase, followed by a larger negative rise and fall in potential, i.e., a diphasic action potential. This indicated that the inner surface of the leaf trap first became positive and then negative as the excitatory process passed under the nonpolarizable electrodes placed symmetrically on the two outer surfaces of the trap. When the inner surface was excited, mechanically or electrically, it was found that the electrical disturbance was greater in the neighborhood of the seat of excitation than at a distance from it. The electrical transient lasted about 1 sec and its propagation speed, at 38° C, was about 20 cm/sec. The mean maximum negative potential variations as the result of a strong stimulus was about 20 mv.

The deflections were originally measured with a 5,244-ohm Thomson galvanometer, a 10,000-ohm resistance in series, and nonpolarizable electrodes placed across the leaf. The sensitivity of the instrument was 350 scale divisions for 0.001 Daniell, i.e., 3×10^{-6} v per division. Later measurements of the maximum magnitude of the fluctuations were made with a Lippmann capillary electrometer.

To get a better understanding of the nature of these electrical transients, it is important not only to know that they are local electrical negative potential changes, but also the precise shape of the after potentials, negative and positive, and their temporal variations under controlled conditions. In order to avoid the necessary corrections for the individual characteristics of the electrical deflecting instruments involved in following such rapid fluctuations in potential, one resorts in modern practice to the use of a cathode ray oscillograph.

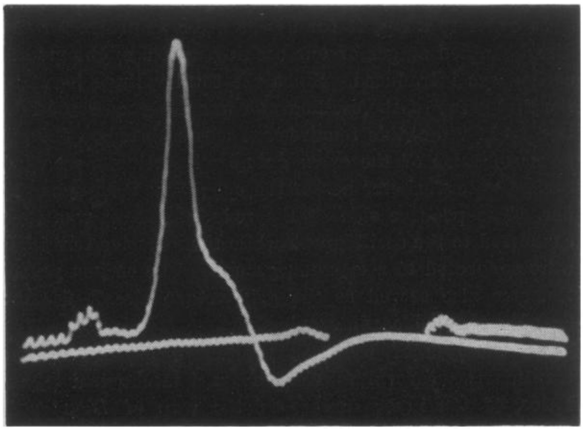

FIG. 1. Oscillograph record of an action potential of Venus's-flytrap, *Dionaea muscipula.* Electrodes taken off under side of trap. Bending of trigger hair used as stimulus. Negative potential up, time scale logarithmic, 60-cycle frequency; stimulating artifact, negative spike potential, negative after potential followed by positive after potential; maximum 0.13 v above resting potential, 24° C.

In neural action potential measurements, the initial negative variation or spike lasts about 2 msec, reaching its crest in about 0.5 msec. In *Dionaea* it was found that the negative variation existed for several tenths of a second. Such a pulse can be highly distorted if amplified by an ordinary resistance-coupled amplifier, such as can be used to amplify the neural spike, unless the coupling circuits have been designed to possess unusually high time constants.

With this in mind, the following instrumentation was devised. A Dumont 208B cathode ray oscillograph was used. The transients on the 5-in. fluorescent screen were compressed to 2 in. and photographed with a Monitor camera equipped with a f/4.5 lens. Lowest frequency X-axis was 2 cycles per sec. Lowest frequency Y-axis was 4 cycles per sec with a 5% distortion at 2 cycles per sec, square wave input.

Since the Y-axis amplifier of the instrument is designed so that the final two stages are directly coupled, and since the deflection factor for a signal amplified only by these two stages is 1.5 d-c volts per inch, a d-c amplifier having a gain of only 100 could be used as a pre-amplifier, to obtain a deflection factor of approximately 15 mv per inch.

A differential d-c amplifier described by Schmitt (*6*) was adapted for this purpose. It was designed to amplify small potential differences between two points, each of which has a high impedance with respect to ground. This is a condition often found in biological systems and is characteristic of the living system in the present investigation, since the approximate resistance between the nonpolarizable electrodes on the plants and the grounded soil was several hundred thousand ohms. The input voltage as a function of output voltage of this amplifier is essentially linear for output, peak to peak up to 100 v, and the differential property is nearly perfect until the mean grid potential deviates by more than 200 mv from normal. The gain of the amplifier was about 160.

In order to adapt it for use as a d-c amplifier for the direct-coupled stages of our oscillograph, a slight modification was introduced. Since the total voltage input to the first stage of the oscillograph was designed to be limited to 11 v d-c, this requirement was satisfied at the expense of some of the extra gain by inserting a megohm carbon potentiometer across the output of the pre-amplifier. The positive side of the reduced output was then connected to the oscillograph while the negative terminals were connected to a common ground. The carbon potentiometer also served as the grid return circuit for the first d-c-coupled oscillograph stage.

An example of the photographic recordings of the transient wave form resulting from a mechanical stimulation of the inner surface of the trap of *Dionaea* is shown in the figure. The connections between the metallic input circuit and the surface of the leaf were bridged by nonpolarizable Ag-AgCl electrodes in 0.7% KCl solution, terminating in glass capillary tubes from which a 0.1-mm brush of asbestos fibers protruded.

The stimulus to excite the electrical activity, but not sufficient to produce mechanical closure of the trap, was applied by suddenly bending one of the trigger hairs with a fine glass hook, until the hair made an angle of 45° with the plane of the supporting irritable inner surface of the leaf.

In accordance with the practice commonly used in representing neural action potentials, positive decreases in potential are graphed upward on the *Y*-axis, with the resting potential set at zero. The record shows the rise and fall of potential between the electrodes placed at two opposite but structurally similar points on the outer surface of the lobes of the trap.

The start of the action potential is gradual—there is no evidence of a sudden or explosive change, although the initial acceleration is very great. Its rising phase is smooth, and its crest is reached in 0.1 sec. The falling phase is much slower, so that the peak is situated well toward the side of the rising phase. Overlapping and immediately following the spike, as in neuron compound action potentials, there occurs a negative after potential which shows marked independence of behavior. Next comes the positive after potential. It attains its maximum value in about 0.07 sec, and then decreases until restoration to its resting potential is completed in about 1.5 sec. As the action potentials are picked off at successively greater distances from the point of stimulation, the transients broaden and become lower.

The action potentials run a course characteristic of mammalian nerve in normal physiological condition. Departures from this normal form are governed by the position of the electrodes, health and age of the plant, temperature and intensity of stimulus. The action potential can run its course without producing closure of Venus's-flytrap.

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Contamination in Orthophosphates Irradiated in a Neutron Pile[1]

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In the course of an investigation with plants grown in sand fertilized with rock phosphate irradiated in a neutron pile, the specific activity of phosphorus in the plant material was as much as three times as great as the specific activity of phosphorus in the fertilizer. The specific activities were determined by precipitation of the phosphorus as magnesium ammonium phosphate according to the method of MacKenzie and Dean (*3*). For no apparent reason a selective absorption of P^{32} seemed to be taking place. One possible explanation was the presence of some radioactive contaminant. An investigation was undertaken to determine the nature of the contaminant, if any, and the extent of its presence in different units of pile-irradiated phosphates.

A sample of $KH_2P^{32}O_4$ obtained by neutron pile bombardment of KH_2PO_4 was dissolved in water and made to volume. Not enough potassium was present to affect the total radioactivity appreciably. An aliquot was removed, the phosphorus was precipitated in acid solution as the phosphomolybdate, and the filtrate was made to volume. The phosphomolybdate was then dissolved in 1*N* NH_4OH and made to volume. A second aliquot, just twice the size of the first, was treated in a like manner. A third

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[1] Since this paper was prepared for publication, E. D. E. Thomas and D. J. D. Nicholas reported similar findings for Na_2HPO_4 placed in the neutron pile at Harwell, England. (*Nature*, Lond., 1949, **719**, 163).