

# PHILOSOPHICAL TRANSACTIONS.

---

## I. *On the Electromotive Properties of the Leaf of Dionæa in the Excited and Unexcited States.*

*By* J. BURDON-SANDERSON, M.D., F.R.S., *Professor of Physiology in University College, London.*

Received October 27,--Read December 15, 1881.

IN 1873 I communicated to the Society the results of experiments showing that in the leaf of *Dionæa*, as in the excitable organs of animals, the change of form which is the visible effect of mechanical or electrical excitation, is preceded by an electrical change of a nature similar to that which occurs, under corresponding conditions, in animal muscle. In a second paper, which was read on the 14th December, 1876, I submitted to the Society, in conjunction with Mr. PAGE, an account of a number of experiments, made, for the most part in the Laboratory of the Royal Gardens, Kew, with the aid of LIPPMANN'S electrometer, for the purpose of elucidating the relation between the electrical phenomena and the physiological process which they accompany. In that paper we confined ourselves to the mere relation of our observations, recognising their incompleteness, but hoping that the work of future seasons might yield better fruits. Although each year since 1876 some additions have been made to our knowledge, it has not seemed desirable to publish anything until the present occasion. If the accumulated products of so many years appear insignificant, I must ask that it may be remembered that the investigation is one of great difficulty, and that the time during each summer which is available for experiment is very short.

As almost all the experiments on which the conclusions of the present paper are founded have been of such a nature as to require the co-operation of two persons, it is obvious that the work could not have been accomplished without the aid of a practised

MDCCCLXXXII.

B

observer. This aid I have received from Mr. PAGE, to whose technical skill and ingenuity in overcoming difficulties, whatever success may have been attained in the experimental part of the enquiry, is largely due.

I have to express my great indebtedness to the Director of Kew Gardens, Sir JOSEPH HOOKER, and to the Assistant Director, Professor THISELTON DYER, for the invariable kindness with which they have placed the resources of the Royal Gardens at my disposal.

Since 1873 several researches have been published in Germany relating to the excitatory process in *Dionæa*, or more generally to plant excitability. In 1876 an elaborate work appeared in Berlin by Professor MUNK, of which the aim was to acquire "a more exact knowledge of the leaf of *Dionæa* in its electrical relations;" and in addition two important experimental papers by Dr. KUNKEL relating chiefly to *Mimosa* have been lately published in vol. xxv. of PFLÜGER'S Archiv, under the title "Electrical Researches Relating to Animal and Vegetable Structures." For the purpose of deriving from them valuable aid, no less than for that of criticising the observations and conclusions of their authors, it will be necessary to examine these papers carefully. I therefore propose to devote an introductory section to this object.

#### PART I.

#### EXAMINATION OF THE RECENT RESEARCHES OF PROFESSOR MUNK AND OF DR. KUNKEL ON PLANT ELECTRICITY.

1. *Summary of Professor MUNK's Paper*.\*—The introduction (§ 1) begins by referring to the discovery, made by myself in 1873, that the leaf of *Dionæa* is endowed with electromotive activity resembling that of muscle, and that when the leaf contracts a negative variation occurs. He states that his attention was first directed to *Dionæa* in November, 1873, and that, in consequence, he made experiments in 1874 and 1875, using about 100 leaves, none of which were detached from the plants to which they belonged.

§ 2 relates to the distribution of electrical tension on the surface of the leaf. After confirming the observations recorded in my first communication to the Royal Society, that in general when the *Dionæa* leaf is led off by the opposite ends of the outer surface of the midrib, the end furthest from the leaf stalk (distal) is found to be positive to the other end†, he states (1) that the most positive point is not the distal end of the midrib, but the point corresponding to the junction of its middle with its distal third, (p. 37) to which point, though both ends are negative, the petiole end is twice

\* Die elektrischen und Bewegungs-Erscheinungen am Blatte der *Dionæa Muscipula*, von Dr. HERMANN MUNK, a. o. Professor an der Universität zu Berlin. Leipzig, 1876. Aus REICHERT'S und DU BOIS-REYMOND'S Archiv für Anatomie, &c., 1876, besonders abgedruckt.

† "Herrn SANDERSON'S Angabe ist leicht zu bestätigen: man findet in der That regelmässig zwischen jenen Blattenden, nach unserer Bezeichnungsweise, einen aufsteigenden Strom."

as much so as the distal end. (2) That in any line drawn across the midrib on the outer (under) surface of the leaf from side to side, a point can be found on either side which is negative to all other points, the negative point being situated nearer to the border of the leaf than to the midrib (p. 38), and finally that the distribution of tension is the same on the under surface as the upper (p. 42) and that opposite points are always equipotential.\* In § 3 the difference of potential between the most negative and the most positive points of the outer (under) surface of the leaf is found to be 0.04 to 0.05 DANIELL. In § 4 the question of the seat of the electromotive forces on which this and the other observed differences are dependent is discussed.

From preliminary considerations he concludes that inasmuch as it is out of the question "das Blatt zu zerstückeln" so as to investigate the electromotive properties of its parts separately, the only way is to start from the well grounded assumption that wherever the same organisation exists there will be similar arrangement of electromotive forces, and accordingly that in the individual cell the electromotive force will show a distribution which has a definite relation to its long axis. Hence, inasmuch as the form of the cell admits of but few possibilities, we may, by ascertaining which of these possibilities when taken in relation with the known arrangement of the cells in the leaf, affords the best explanation of what is actually observed in the distribution of electrical tension on the surface of the leaf, select that which accords best with fact. In order to get at this result Professor MUNK employs a method of investigation which I shall not endeavour to describe in detail, but content myself with stating that it is founded on the assumption that it is possible to represent the living cell, as regards its electromotive properties, by a zinc cylinder having a zone of copper and surrounded by a uniform layer of a moist conductor;† and that a schema or model made up of a number of such cylinders arranged like the cells of any living part of a plant would exhibit electromotive properties similar to those of the organ it represented. Stated as shortly as possible, the actual arrangement of the parenchyma cells of the leaf of *Dionæa* is as follows: in the lobes the cells run all in one direction, namely, parallel to the veining and at right angles to the midrib. In the midrib the cells of the ridge (under surface) run longitudinally, but those of the trough (upper surface) are continuous with those of the cells of the lobe. For reasons which need not be here entered on (see p. 86) Professor MUNK considers that the arrangement which determines the electromotive properties of the leaf, is that of the cells of the lobe-parenchyma of which the long axes are parallel to the surface of the lobe and at right

\* "An der oberen Blattfläche die gleiche Vertheilung der Spannungen herrscht, wie an der unteren Blattfläche" (p. 42). "Dass die absolute Grösse der Spannungen an der oberen und an der unteren Blattfläche die gleiche ist" (p. ).

† "Die einzelne Parenchymzelle entspricht hinsichts ihrer Kräfte dem einzelnen Metallcylinder" (p. 96).

Du Bois-REYMOND's schema of an electromotive element of muscle is a copper cylinder with a zinc zone surrounded by a uniform layer of a moist conductor. For the plant electromotive element, according to MUNK, the sign must be reversed. The cylinder must be zinc and the zone copper.

angles to the midrib. Accordingly, the leaf ought, on the assumption that the cells have the properties assigned to them (*i.e.*, that the middle of each cell is negative to its ends), to be represented by a bilateral schema consisting of two symmetrical parts, resembling in form the two lobes of the leaf, and each made up of metallic cylinders arranged like the cells in the leaf, as above described. A full account of the properties actually demonstrable in such an arrangement is given in the paper (pp. 54 and 55). It corresponds in the most important respects, as regards the distribution of electrical tension, with Professor MUNK's account of the electrical properties of the resting leaf.

§ 5 relates to the "Mechanism of the Excitatory Motion." On this subject it will be sufficient to state that the account given by Professor MUNK of the way in which the leaf closes when touched, agrees with the explanation originally given by BRÜCKE of the mechanism of the motion of the sensitive plant; namely, that the immediate cause of the change of form is the sudden passing of certain cells (in the case of *Dionæa* those of the upper (inner) layer of the lamina and midrib) from the state of distension into that of flaccidity; but, in addition to this, there is, he says, an active lengthening and extension of the cells of the lower or external layer.\* From the context it appears that all that is here meant is that the water discharged in the sudden relaxation of the cells of the inner (upper) stratum, finds its way at once to the outer (lower). At all events, the only evidence given in support of active extension is the fact, proved by measurement, that two marks on the external surface of the expanded leaf, in a line parallel to the veining and at a measured distance from each other, are found to be further apart after the leaf has closed (p. 117).

In § 6 (p. 123) the author approaches the question which more immediately concerns our present purpose, *viz.*: the electrical phenomena which result from excitation. I will endeavour to give an account, first of the experiments, and secondly of the theoretical explanations—premising that, in this as in the section on the electromotive properties of the unexcited leaf, observations and theories are so mixed that it requires much patience to separate them.

The mode of experiment was always the same. The leaf was led off, either by different points on the under surface of the midrib, as in my original experiments in 1873, or by the midrib and by a spot on the external surface opposite to it. The leaf was not fixed or restrained, so that only a very few excitations could be observed before closure.

All of the experimental results are stated in the following table, in the first column of which I have entered the leading off contacts, distinguishing them by the letters *A* and *B*, in the second column whatever information is given as to the difference of potential between them before excitation. The third column gives the character of the excitatory effect as observed by the galvanometer; and the fourth, the reference to the page where the experiment is recorded.

\* "Die Bewegung kommt dadurch zu Stande dass das reizbare Parenchym erschlaft und kürzer wird, das nicht reizbare sich activ verlängert" (p. 119).



TABULAR Summary of the Results of Professor MUNK's Experiments.

	Contacts.		Electrical condition before excitation.	Excitatory effect.	Reference.	Remarks.
	A.	B.				
I.	Distal end of midrib.	Proximal end	Aufsteigender Strom, <i>i.e.</i> $A$ 30 scale negative to $B$ .	"Negativer Vorschlag," <i>i.e.</i> diminution of previous deflection, followed by "positive Schwankung," <i>i.e.</i> , increase of previous deflection.	P. 128 . . .	Electromotive force of the "positive Schwankung" said to be about equal to that of "aufsteiger der Strom."
II.	Distal half .	Proximal end	Not stated; in general, the difference of potential said to be twice as great as in the previous case. (P. 38, line 7.)	"Neg. Vorschlag" so strong as to reverse the previous deflection, and to exceed the Schwankung several times in extent.	P. 131 . . .	Relation of Vorschlag and Schwankung (Doppelschwankung) to previous deflection inconstant.
III.	Distal half .	Proximal half	Contacts isoelectrical .	First effect "aufsteigend," <i>i.e.</i> in direction of the Schwankung, followed by slow swing in opposite direction, and return to zero.	Pp. 131, 132.	
IV.	Under surface of midrib at junction of middle and distal third.	Outer concave surface of lobe opposite $A$ , and at most negative point.	$B$ negative to $A$ . Difference amounts to 0.05 DANIELL.	Vorschlag of 2 to 3 scale, in which $B$ becomes less negative, followed by very large deflection in the opposite direction.	P. 145 . . .	In this experiment the leaf is led off by the two points of which the potentials differ most.

Professor MUNK's explanation of the *excitatory variation* is, of course, in conformity with his doctrine of the electromotive activity of the individual cell, which has been already explained ; namely, that each parenchyma cell has the endowment of a cylindrical electromotive element of which the poles are positive to the middle,\* and the excitatory change produced in each cell consists in increase or diminution of the electromotive force of each cell. If the variation consisted of a single phase, it might be accounted for by supposing that all the cells of the parenchyma of the lobes are affected by excitation in the same way. As, however, it presents the characters of a "*Doppelschwankung*"—*i.e.*, consists of two phases succeeding each other, of opposite signs, this must be accounted for either by supposing (1) that all the electromotive elements in the structure are similarly affected, but not at the same time, the excitatory change being propagated from the seat of excitation in such a way that it first affects the electromotive elements at one contact, then those at the other ; (2) that all the electromotive elements are similarly affected at the same time, changes of opposite signs following each other in each element simultaneously ; (3) that there exist in the structure two sets of electromotive elements, which are affected in opposite directions by excitation, the disturbance reaching its maximum in the one set later than in the other set.

Of these three alternatives only the third appears to Professor MUNK admissible. The first he rejects on the ground that if it were true the character of the excitatory variation would be affected by the seat of excitation. This he finds not to be the case. Thus in Exp. II. (see Table) the effect should be different according as the leaf is excited by touching the proximal or distal hairs. According to Professor MUNK there is no difference (p. 136) either in this or other similar cases. He therefore concludes that the "*Doppelschwankung*" is not due to propagation of the excitatory effect from the seat of excitation. "Danach kann weder die complicirte Schwankung in irgend einer Beziehung zum Orte der Reizung stehen, noch kann überhaupt von diesem Orte der Erfolg der Reizung, innerhalb der Genauigkeitsgrenzen unserer Untersuchung, irgendwie abhängig sein" (p. 138). The second alternative is rejected on the ground that in IV. (see Table) the Vorschlag, if it occurs at all is very inconsiderable, and is very frequently absent, consisting of one phase only, which could not be the case if each cell underwent first a diminution then an increase of its electromotive force, and all acted simultaneously. The third alternative, thus adopted by exclusion, *viz.* : that the two sets of electromotive elements are oppositely affected by excitation, serves, Professor MUNK thinks, to explain the characters of the excitatory effect in all the cases observed by him.

\* "Die ohngefähr cylindrischen Zellen des Blattflügel-Parenchyms und der beiden Mittelrippen-Parenchyme sind mit Kräften ausgestattet der Art, dass die positive Elektrizität von der Mitte der Zelle nach jedem der beiden Pole hingetrieben wird, die Pole positiv sind gegen die Mitte" (p. 97).

Professor MUNK's paper does not contain any statement of his conclusions. For further reference I have arranged them as follows, in the order in which they have been referred to in the preceding sketch :—

1. Each individual parenchyma cell of the leaf corresponds, in respect of its electromotive properties, with a zinc cylinder zoned with copper and surrounded by a uniform layer of moist conductor. In other words, the nearly cylindrical cells are endowed with electromotive forces such, that positive electricity is driven from the middle of each cell towards each of its poles, *i.e.*, the middle is negative to the poles (p. 97).

2. The electromotive properties of the whole leaf (pp. 47–51) in the unexcited state are represented by those of a schema (see diagram, fig. 20, *a*), consisting of such cylinders arranged in the same order as its cells (p. 87).

3. On excitation, the cells of the upper layer of the leaf undergo a diminution of electromotive force, those of the under layer an increase, *i.e.*, the middles of the cells become less negative than before in the upper layer, more negative in the lower layer (p. 152).

4. The diphasic character of the variation is not due to the fact that the individual cells undergo opposite changes in succession, but to the interference of the opposite electromotive actions of the upper and under cells, of which the time relations are different (see diagrams 29 and 30 and p. 142).

5. The effect of an excitation is in no way dependent on its seat (p. 138).

6. The electromotive activity of the cells has no relation to their water-content (p. 158).\*

\* The author's criticisms of my methods and conclusions will be best considered in connexion with the several subjects to which they refer. His paper contains some misconceptions which it will be convenient to correct in this place. One of the most important, as having led other writers into error, is founded on a casual expression of mine contained in a lecture at the Royal Institution in 1874, to the effect that the excitatory motion of the leaf is identical with muscular motion. No such expression is contained either in my communication to the Royal Society, or in the translation of it which appeared shortly afterwards in the 'Centralblatt.' ("Ueber elektrische Vorgänge im Blatte der *Dionaea muscipula*." 'Centralblatt f. d. Med. Wissensch.,' 1873, No. 53.) If Professor MUNK had considered the context, he would have seen that in my lecture I meant nothing more than that both motions are expressions of the same elementary endowment of protoplasm—that of changing its form on excitation. I held then, and hold now, that, as regards what happens in the living protoplasm, the mechanical effects of excitation in the contractile parts of plants and animals are closely related. As to the mechanism by which the changes of form of the excitable organs of plants are brought about, nothing was said in my paper. I took for granted the explanation usually given, founded on BRÜCKE's experiments. I had not become acquainted with the then recently-published researches of PFEFFER on the excitability of plants ('Physiologische Untersuchungen,' von Dr. W. PFEFFER. Leipzig: 1873).

As regards the original experiments by which I demonstrated the fact that the leaf of *Dionaea* responds to excitation by an electrical disturbance analogous to the excitatory variation of muscle, Professor MUNK's

The researches of KUNKEL,\* like those of MUNK, have been directed to two questions, viz. : to that of the differences of electric tension which present themselves in the uninjured leaf, and to the electromotive phenomena which follow excitation in the excitable organs of plants. As regards the first question, he has found that in leaves generally, when they are led off by non-polarizable electrodes in the usual way, differences of tension present themselves, these being always of such a nature that if one electrode is in contact with a vein, the other with the green surface between the veins, the former is positive to the latter (p. 359). But he considers this due, not to any real difference of tension between the surfaces, but exclusively to inhibition changes which begin at the moment that contact is made. As evidence that the relations of the two surfaces to water are different, he adduces the following observations.

That diffusion actually takes place at the uninjured surfaces of leaves which are in contact with water, he proves by the observation that potassium salts may be detected

observations are, happily, in accordance with mine, as may be seen by comparing his account of the phenomena with mine:—

SANDERSON, 1873.

Wenn das Blatt so auf die Elektroden aufgelegt wird dass der normale Strom des Blattes durch eine Ablenkung der Nadel nach links angezeigt wird und die sensitiven Haare der oberen Fläche berührt werden, so schwingt die Nadel nach rechts. . . . In jedem Falle kommt die Nadel nach der negativen Schwankung in einer Stellung zur Ruhe, die weiter nach links gelegen ist als zuvor, und nimmt dann allmählig ihre frühere Stellung wieder ein. (Centralblatt, 1873, p. 835.)

MUNK, 1876.

Reizen wir unser Blatt bei Ableitung von den beiden Enden der Mittelrippe, so beobachten wir Folgendes: (Es sei z. B. eine Ablenkung von 30 Sc. vorhanden.) Zuerst nach der Reizung nimmt die Ablenkung sehr rasch auf 25–30 Sc. ab, und darauf nimmt sie sogleich etwas langsamer, doch immer sehr rasch auf 40–60 Sc. zu. Nun verweilt der Spiegel nur momentan auf dem Maximum der Ablenkung und kehrt dann langsam, in etwa einer Minute, zu seiner Anfangsstellung zurück (pp. 127, 128).

Notwithstanding this close correspondence, Professor MUNK speaks of my description as grossly defective, “not merely in details, but in what is essential and important” (p. 124); and proceeds, that for aught I knew to the contrary, my “*negative Schwankung*” might be nothing more than a result of shifting of contacts. The serious defect, however, was not so much incompetence as an observer as inability to understand what I saw; for, referring to the very same observation further on, he says that “Herr S. den Erfolg der Reizung richtig beobachtet hat und nur seine Beobachtung durchaus missverstanden hat, indem er . . . eine negative Schwankung da zu sehen vermeinte, wo in der Wirklichkeit eine positive Schwankung mit negativem Vorschlage sich zeigte.” On another of my observations (see p. 152), which he had not time to repeat, but which he thinks may be relied on though it was made “*natürlich ohne von seiner Bedeutung eine Ahnung zu haben*,” he finds an additional prop for his own theories. In conclusion, Professor MUNK congratulates himself on having substituted “an die Stelle der Spurweisen und vielfach unzutreffenden ersten Wahrnehmungen von Herrn Sanderson eine genauere Kenntniss des *Dionæa*-Blattes in elektrischer Beziehung.”

\* KUNKEL. “Electrische Untersuchungen an pflanzlichen und thierischen Gebilden,” PFLÜGER'S Archiv, Bd. xxv.

in water in which such leaves have been immersed even after a few minutes' immersion. He further finds that if a freshly cut off leaf is immersed in water, the whole of the vein surface is wetted at once. The green surface of the lamina exhibits a beautiful silvery lustre indicating that air adheres to it, and that water comes very slowly and incompletely into contact with it. When the leaf is removed from the water, the veins remain permanently and completely wet, but the water runs off the rest of the surface, leaving it dry. Consequently he concludes that when moist electrodes are brought into contact with surfaces which thus differ, the diffusion processes which result from the contact must go on at different rates.

By other experiments, physical rather than physiological, he has shown that in all bodies capable of imbibition with water, electrical changes are associated with the process of imbibition. His first experiments on the subject were made with the ordinary porous cells of batteries. Similar results were obtained by the investigation of non-living vegetable structures, as *e.g.*, laminaria stems. By comparing these facts with those already stated as to the electrical phenomena observed in the investigation of leaves, he derived the conclusion that whatever differences present themselves between the different parts of leaves are due to "*Unterschied in der Benetzbarkeit*" of the surfaces led off. What the direction of this effect is, KUNKEL had determined by experiment. If a drop of water is placed on the green surface of a leaf between the veins and allowed to remain there some time and the leaf is now led off by another similar surface and by the drop, it is found that the surface first wetted is always positive to the other (p. 368). If after one electrode has been brought into contact with an ordinary surface, the other is applied to a vein, the direction of the difference is opposed to the normal. The vein is at first negative but the difference quickly diminishes and is soon reversed (p. 360). In other words, when a leaf or other similar structure is led off by two surfaces of which one is more "wetable" than the other, the former is positive to the latter, provided that both contacts have been made simultaneously.

From these observations, of which the value and interest must be recognised as showing what sort of results may arise from imbibition processes taking place at surfaces of contact, the author proceeds to draw inferences as to the nature of excitatory electromotive changes in the organs of plants, confining his attention to *Mimosa*. The excitatory effect in the swelling at the base of the leaf stalk of *Mimosa*, is described by KUNKEL, on the basis of observations made with the capillary electrometer, the organ being led off by thread electrodes applied to its opposite ends as follows. It is said to consist of a "*Vorschlag*," in which the base becomes negative, followed by a larger excursion in the opposite direction. KUNKEL thinks that this observation is in harmony with those of MUNK. Three fragmentary experiments are given in confirmation of this, on which the theory is founded, that inasmuch as in *Mimosa* the active motions of the leaf are caused by a sudden alteration in the distribution of water in the different parts of the swelling at the base of the leaf stalk, that these "*Wasser-*

*verschiebungen*” are the causes of the electrical changes observed, the proof being, that in general “*Wasserverschiebungen in imbibirten Körpern*” are attended with electrical phenomena. He confesses himself unable to connect the particular phenomena observed with “*einzelne Phasen prevalirender Wasserverschiebungen.*” He thinks, however, that the first “*Vorschlag*” is not due to *Wasserverschiebung*, but to the fact that the diffusion processes which are called into existence by the application of the electrodes are disturbed by alterations in the protoplasm. It is to be noticed that these explanations, which are contained in the concluding paragraph of the paper, are not in accordance with the more general statement on p. 371. In one place it is stated that “all electrical phenomena are conditioned by *Wasserverschiebung*,” in the other, that the *Vorschlag* in *Mimosa* “cannot be referred to *Wasserverschiebung*, but to the alteration of protoplasm which precedes it.” The difference is a fundamental one.

From the preceding pages it will be seen that the two observers who have investigated the electromotive phenomena which are associated with the excitatory process, viz. : Professor MUNK and Professor KUNKEL, have arrived at opposite conclusions as to their nature. According to MUNK, the electromotive activity of the plant cell stands “in no direct relation to the water it contains” (p. 158). It is KUNKEL’s main conclusion that they are determined by diffusion processes. MUNK gives as the result of his observations a complicated map of the distribution of electrical tension on the surface of the leaf of *Dionæa* (p. 40, figs. 14 and 15). KUNKEL starts from the proposition (which he does not stop to demonstrate) that electrical differences do not exist at all in uninjured and untouched leaves (p. 372). According to MUNK the electromotive properties of the leaf are referable to electromotors which resemble the electromotive molecules of DU BOIS-REYMOND, with the exception that their signs are reversed (p. 51). This hypothesis KUNKEL only mentions for the purpose of rejecting it (p. 371). There is, in short, but one respect in which they agree. Their descriptions of the phenomena are sufficiently explicit to make it pretty certain that they have both witnessed the same process. As regards the physiological nature of the phenomena the contradiction is complete.

## PART II.

### DESCRIPTION OF THE METHODS AND INSTRUMENTS EMPLOYED.

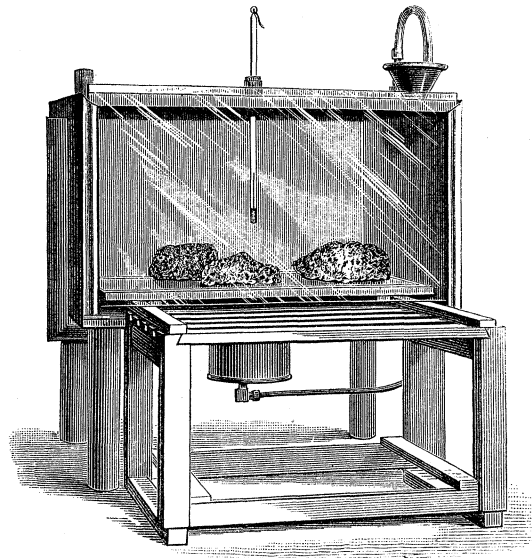
The methods used in the study of the electromotive properties of an excitable living structure, whether of plant or animal, may be grouped under four heads according as they are adapted (A) for maintaining, during the period of observation, the external conditions of temperature, moisture, &c., which are most conducive to vigorous life ; (B) for the investigation of the differences of tension (electrical potential) which exist at one and the same time between different parts of the surface of the organ ; (C) for

investigating the *successive* states (tension differences) which present themselves after excitation, with reference to their duration and their time-relations with other phenomena or processes which precede or accompany them ; or (D) for exciting, or otherwise temporarily modifying the physiological conditions of the organ.

The appliances used for these several purposes in the case of the leaf of *Dionæa* are as follows :—

A. *Means of maintaining favourable conditions of temperature, &c., during the period of observation.*—By preliminary experiments it was ascertained that the conditions most favourable to the activity of the leaf are, that the temperature of the leaf should be from 32° C. to 35° C. and that the air should be nearly saturated. For the maintenance of these conditions the plant is placed in a chamber of the following construction (fig. 1).

Fig. 1.



The Warm Chamber.

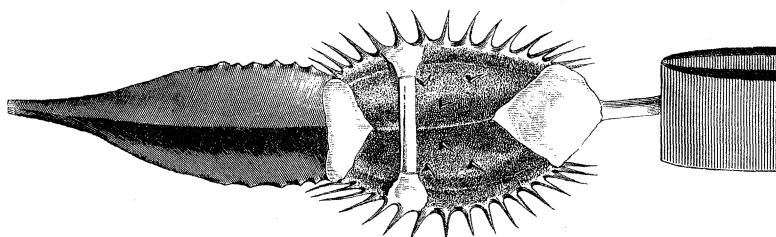
The sponges which serve to keep the air saturated rest on the glass plate which covers the sliding floor. On this glass plate the plant, electrode holders, &c., rest. To withdraw them, the floor is made to slide out along the rails. The upper part of the tube of the regulator is seen on the right side. (From a photograph.)

The chamber measures 47 centims. wide and 40 centims. from back to front, and is 26 centims. high. It is made of japped tin plate. Its top, bottom, and sides are double, and separated from each other by a closed cavity of  $1\frac{1}{4}$  centims. in width, which is filled with water. Its front and back are closed by panes of glass which slide up and down in grooves. The water contained in the bottom, sides, and top is heated by an ordinary gas burner, 3 inches below the bottom, along which, and up one side a chimney,  $2\frac{1}{2}$  centims. by  $7\frac{1}{4}$ , leads. The temperature of the water is governed by an automatic regulator of the form described by Mr. PAGE in the Journal

of the Chemical Society.\* The chamber is provided with a sliding floor of wood, covered with glass, which serves to support the apparatus. The wires leading to the electrodes, &c., enter by openings left on either side of the floor for the purpose.

*Mode of preparing the leaf for observation.*—Whether the leaf is on the plant or not, it is necessary to fix it in such a way as to render it motionless. In our earlier experiments glass forceps were used, ingeniously contrived and constructed by Mr. PAGE, of which the blades were of such form that while one of them applied itself to the upper surface of the leaf when in the fully expanded state, the lower supported

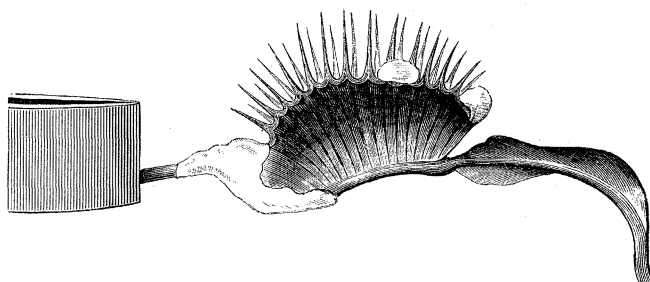
Fig. 2.



The Leaf, seen from above.

The two end plugs and the cross-bar, which stretches from margin to margin, serve to prevent it from closing.

Fig. 3.



The same Leaf, seen from the side. (Both figures are copied from photographs.)

the midrib, for the reception of which it was suitably grooved. A figure of this instrument was given in our former paper.† We have now adopted a much better mode of immobilization which will be understood from the figure. Before otherwise interfering with the leaf, plaster of Paris, which has just been mixed with water, is introduced between the lobes at either end of the midrib, so as to bridge the space between them. A splinter of dry wood is laid across from edge to edge of the lobes, the ends of which are cemented by plaster to the marginal hairs. The leaf so prepared is unable to close (figs. 2, 3).

B. *The galvanometer* used is a THOMSON of 5244 ohms resistance. The degree of sensibility best adapted for the ordinary purposes of the investigation is such that with a thousandth of a DANIELL and a resistance of 10,000 ohms, added to that of the

\* Vol. i., 1876, p. 24.

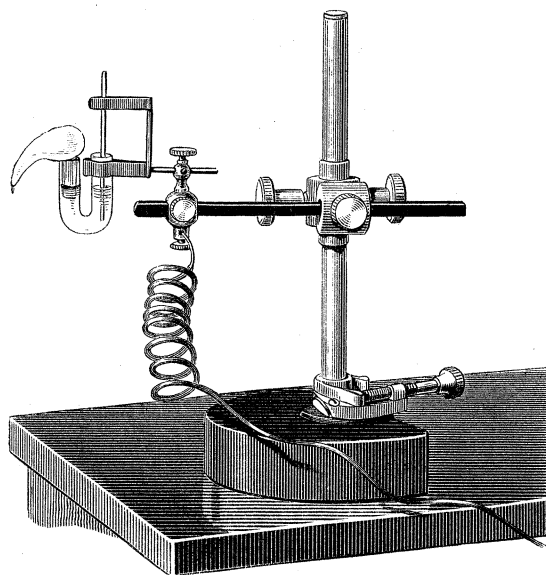
† Proceedings, 1873, p. 413.



galvanometer, we have a deflection of 350 scale ; but for some rheotome observations, a much higher sensibility is required. With a view to more accurate reading of small deflections, a concave mirror, 10 centims. wide, is placed in front of and below the scale, by means of which that part of the graduation nearest to zero can be seen enlarged.

*The electrodes.*—The most important part of each electrode is a U-shaped tube containing saturated zinc sulphate, which is supported by a holder of the form shown in the figure (fig. 4). Into the end of the U contained in the clamp, sinks a zinc rod,

Fig. 4.



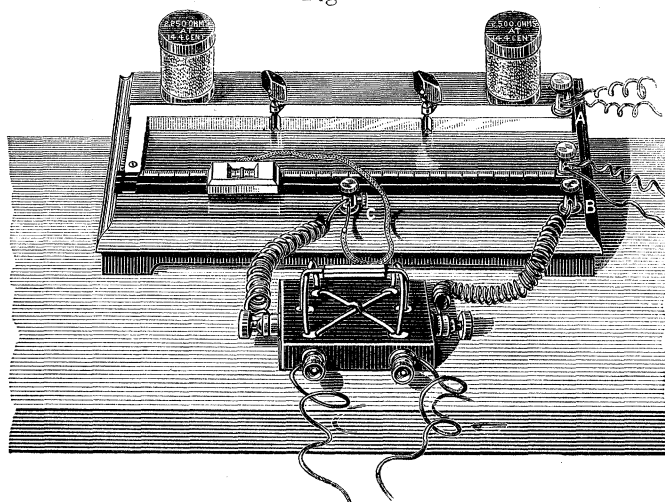
A Non-polarizable Electrode. (From a photograph.)

which is held by the spring clip, and is thus brought into metallic connexion with the coiled wire. This rod is hammered smooth at the end which is intended to be immersed, polished, and amalgamated by treating it sparingly with very weak solution of mercury in nitrohydrochloric acid. Into the opposite limb of the U slips a narrow straight tube 1 centim. in length and about 5 millims. in internal width, which is charged with paste of kaolin and solution of zinc sulphate. The tube has a rim of sealing wax at the top, which prevents it from sinking too far, and is surrounded by a conical beak, shaped with the fingers out of modelling clay (kaolin made into a paste with 0.75 per cent. solution of chloride of sodium). In order to bring the modelling clay to a pencil-like point, a length of several strands of ligature thread steeped in the solution is incorporated with it. These electrodes have a resistance of about 6000 ohms and keep in order for any required time if the atmosphere is saturated or nearly so. The construction of the supports for the electrodes can be understood from the figure. It is sufficient to notice that the collar slides on the horizontal rod of ebonite. This rod can be raised or lowered, or made to rotate round the

vertical axis, by working screws, of which the milled heads are conveniently placed for the purpose.

*The compensator.*—As is well known to every reader of physiological books, Professor DU BOIS-REYMOND has applied the term to an instrument which consists essentially of a long thin wire connecting the poles of a standard cell. The wire being of uniform thickness, the difference of potential between any two points in its length is to the whole difference between the ends, as the distance between these points is to the total length. In a compensator so constructed the total difference has to be determined by a previous operation. The compensator we employ differs in no respect in principle from the potentiometer of Mr. LATIMER CLARKE\* in which the total difference is equal to the whole electromotive force of the standard cell, and not, as in DU BOIS' compensator, to a certain portion of it. The form we have given to the instrument is very different from that of Mr. CLARKE'S instrument, and far more convenient for our purpose (fig. 5). The compensator wire, or rheochord, has

Fig. 5.



Compensator and Reverser. (From a photograph.)

a total resistance of 5 ohms. On a board, 27 centims. long, is fixed a rail of vulcanite 1 centim. broad and as much high, and 25 centims. long. Along the upper margin of this rail, which is divided into millimeters, runs a wire of platinum, of which the resistance is equal to a  $\frac{1}{4}$  ohm. Each end terminates in a metal block. A second rail or bar of brass runs along the board parallel to the first. It is broken at two points for connexion with the end of the coils 1 and 2, of which the resistances are severally  $2\frac{1}{4}$  and  $2\frac{1}{2}$  ohms, so that when the plugs are out the total resistance between the ends of the bar is 4.75 ohms. The bar is united at one end with one of the terminals of the graduated wire. At the other it has a binding screw with two holes. The opposite terminal of the graduated wire has two binding screws, one with two, the

\* "On a Voltaic Standard of Electromotive Force," Proceedings, 1872, p. 444.

other with one hole. On the vulcanite rail runs a brass sliding block or rider, the form of which is seen in fig. 5. It rests on the surface of the graduated wire by its cross wire, and is connected with the binding screw by a flexible wire. From the construction it is obvious that the difference of potential between the binding screws B and C, is to the total difference between A and B, as the length of wire, in millims. between B and the point at which the cross wire rests on the graduated wire is to 5000 millims. In other words, that each millim. of the graduation corresponds to  $\frac{1}{5000}$ th of the total difference. In use, A and B are connected by one pair of wires with the standard cell through a galvanometer, by the other pair with an "auxiliary battery" through a rheostat as described in Mr. LATIMER CLARKE's paper.

*Measurement of electrical resistance.*—It is of great importance to have the means of becoming promptly aware of the changes of resistance which occur in the course of a long observation. For this purpose we adopt the plan of from time to time measuring the deflection due to 10 millims. of wire =  $\frac{1}{500}$  DANIELL. In investigating plant structures that method is practically of great value, for the resistances we have to encounter vary, roughly speaking, between 50,000 and 100,000 ohms. As, therefore, we know approximately that our circuit, when the electrodes are in contact, is about 12,000 ohms, viz.: 6000 for the galvanometer and about as much for the electrodes, such changes as often occur are obvious enough. In the mode of leading off used in the fundamental experiment (see Part IV., p. 25) we obtain, if the contact is successfully made, with an over compensation of  $\frac{1}{500}$  DANIELL a deflection of about a tenth of the deflection due to the same electromotive force when the electrodes are in contact with each other. If it is less than that, we regard the leading off as unsatisfactory. For actual measurement, we have at hand on an adjoining table a WHEATSTONE's bridge arrangement, which can be brought into connexion with our circuit without loss of time when required, by means of a switch which is substituted for the key which otherwise receives the wires from the leading off electrodes.\*

\* This corresponds to K<sup>5</sup> in the diagram. Here L represents the leaf, *f* and *m* the leading off contacts, *x* and *x'* the exciting contacts, I and II the primary and secondary coils, R the rheotome, C the com-

Fig. 6.

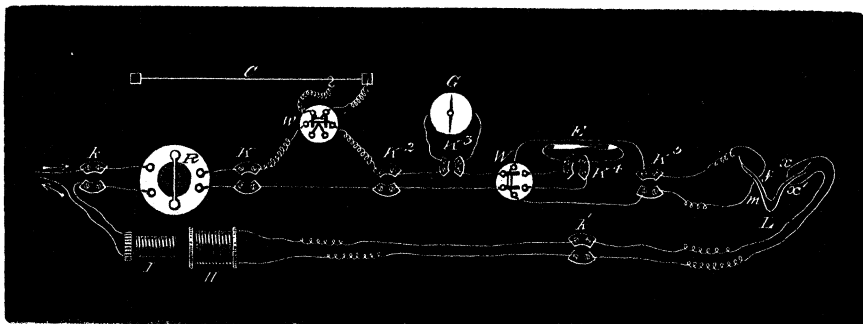


Diagram of Connexions of Compensator.

C. For the preliminary investigation of the time relations of the electromotive changes which follow excitation the most valuable instrument is the capillary electrometer. Two important improvements have been recently made in the construction of this instrument by my friend Professor LOVÉN,\* of Stockholm. The first of these consists in constructing the tube which contains the dilute sulphuric acid, of extremely thin glass so that the capillary tube which is in contact with its internal surface can be observed with a magnifying power of at least 300 or 400 diameters (No. 10 HARTNACK). The other consists in cementing the sulphuric acid tube to the mercury tube, so that no evaporation takes place. By the first, the instrument is rendered much more sensitive, by the second, variations in the strength of the sulphuric acid which arise from evaporation are avoided. When used for the purpose of observing a rapid succession of changes in the electrical relations of two led off surfaces, its superiority to the galvanometer is very striking. The movements of the mercurial column not only correspond closely in time to the actual changes which they represent, but express with very great accuracy the differences of potential which actually exist in each successive phase of a variation. In the beginning of an investigation this is invaluable, for the summary information of the duration, character, and progress of the electrical disturbance produced by excitation, which the electrometer affords, renders it possible to avoid loss of time in the subsequent application of more accurate modes of measurement. For the rougher kinds of time measurement, the use of the electrometer may be combined with that of the electro-magnetic signal, in the manner explained in our former paper. By this means the occurrence of an event can be determined within a tenth of a second by a single observation, and, of course, much more accurately by repetition. As a means of measuring electromotive force, the electrometer may be used with great advantage when the electrical states to be investigated are transitory, and at the same time so irregular that the rheotome could not be applied. For this purpose it is much better to measure the electromotive value of each excursion with the aid of the compensator than to deduce it, as has been done by FLEISCHL,† from the "compensation pressure." With this exception the measurements of electromotive force can be made much more accurately with the galvanometer. If it were possible to photograph the motions of the mercurial column of the electrometer it would be an addition to its usefulness.

*Use of the Pendulum Myograph as a Rheotome.*—The pendulum myograph (fig. 7) is essentially a seconds pendulum, the weight of which carries a glass plate so fixed

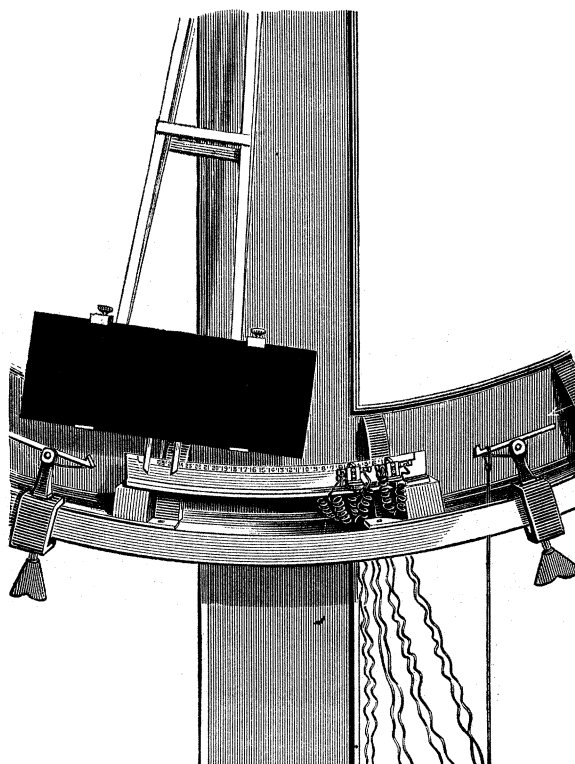
pensator, G the galvanometer, E the capillary electrometer. The wires marked with arrows lead to a battery. K, K<sup>1</sup>, &c., are keys. W is a switch by which the wires from the electrodes may be connected either with the galvanometer (as shown in the diagram), or with the electrometer (as when the bridge is turned to the right). w is the reverser connected with the compensator shown in fig. 5.

\* LOVÉN, "Om kapillarelektrometern och kviksilfvertelefonen," Nordiskt medic. Arkiv. vol. xi., No. 14.

† v. FLEISCHL, "Ueber die Construction und Verwendung des Capillar-Electrometers, für electrophysiologische Zwecke." Archiv. f. Anat. u. Physiol., 1879, p. 269.

that the surface oscillates in its own plane. If this surface is blackened, and a tuning fork is allowed to inscribe its vibrations on it during its swing, it is easy to determine the time occupied by the pendulum in accomplishing any given distance taken at any part of its course from the tracing. I have provided my pendulum with a fixed circular scale, on which each division is at such a distance from the next that the trigger in passing from one to the other occupies one hundredth of a second. Against this scale slide four keys, as seen in the drawing. Each key is so constructed

Fig. 7.



The Pendulum-Rheotome (represented as if in motion).

The motion of the pendulum is from right to left. The detent (on the right) is provided with a wire, by means of which the pendulum can be "let off" by the observer at the galvanometer, which must necessarily be at a distance. (From a photograph.)

that the trigger in passing, opens it. By means of these four keys two circuits can be independently closed and opened one after the other, and the time of closing and of opening can be determined with great exactitude, according to the following plan (fig. 8).

Provided that the events of which the time relations are to be investigated occur within a third of a second, this is an excellent form of rheotome. In opening a key of the kind employed no time is lost; consequently, the time-interval of closure of the circuit corresponds exactly to the distance between I. and II., or between III.

and IV., as the case may be. When longer periods have to be investigated the rheotome previously described by me, in relation to my experiments on the time relations of the excitatory process in the heart, answers perfectly.

D. *Methods of Excitation.*—For electrical excitation (whether by the voltaic or induction current) it is necessary to use non-polarizable electrodes. As the space between the upper surfaces of the lobes is insufficient to allow of the introduction of two electrodes of the form used for leading off, it is convenient to use a simpler form which, although not so perfect, answers the purpose completely. It is merely a zinc rod of which the amalgamated end is sheathed in wash-leather steeped in solution of zinc sulphate. The sheathed end is enclosed in a larger sheath of the same material, steeped in solution of salt, which ends in a point convenient for contact. The zinc rods are held by supports which allow of their points being brought into any required position with accuracy.

Fig. 8.

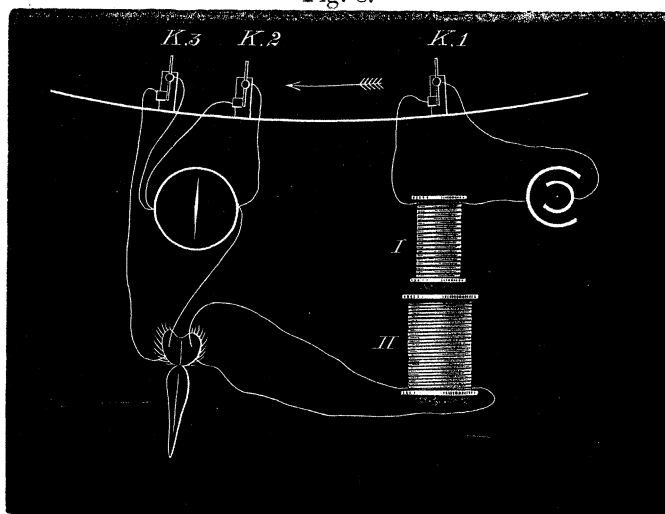


Diagram of the connexions of the Pendulum Rheotome as used for determining time of commencement of first phase, rate of propagation, &c.

### PART III.

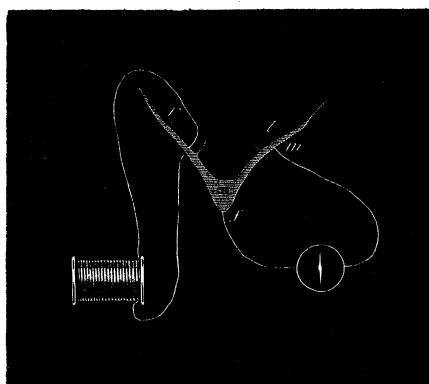
#### ELECTROMOTIVE PROPERTIES OF THE UNINJURED LEAF.

The preliminary examination which was made in 1876 of the electromotive properties of the uninjured leaf appeared to show, first, that the external surface of each lobe is always positive to the internal surface, and, secondly, that in general the part of the midrib which is nearest to the sensitive hairs is positive to other parts of the external surface of the leaf. The more complete knowledge which has been acquired by work done since, justifies the statement that these two propositions express the truth.

With reference to the second and less important of them, I will refer to the following observations made in the Laboratory of the Royal Gardens at Kew in the summer of 1878.

*Negativity of lobe to midrib.*—A leaf having been cut off and placed in the warm chamber, of which the temperature varied from  $29^{\circ}$  C. to  $33^{\circ}$  C. with the leaf-stalk in water, leading off contacts were made on the external surface of one lobe ( $m$ ) and on the midrib  $f$  (see fig. 9). The difference\* of potential was at first  $-0.006$  D. The leaf was then excited by touching the right central hair at intervals of  $5''$  for  $70'$ . Each excitation produced a normal diphasic variation, indicating that  $m$  became first negative, then positive to  $f$ . After each excitation the difference was compensated. After the first two it fell to  $-0.0026$  D. and thereafter to  $-0.0006$  D. In other words, the after-effect of excitation (see Part V.) was opposed in direction to the original difference. A second leaf was prepared in the same way

Fig. 9.



Leaf led off from under surface of right lobe and under surface of midrib, excited from left lobe.

and left in the chamber, of which the air was nearly saturated, all night, the temperature being  $29^{\circ}$  C. At 11.40 next morning the difference was  $-0.004$  D., having been  $-0.009$  D. the evening before. After excitation of the hairs of the opposite lobe it fell to  $-0.0026$  D., and subsequently to  $-0.001$  D. A third leaf, similarly led off and placed in a chamber as before, showed a difference of  $-0.0214$  D., the lobes and marginal hairs being fully expanded. After the first mechanical excitation of the hairs of the opposite lobe, the marginal hairs became incurved without any further change of form of the leaf; the difference fell to  $-0.019$  D. After a second excitation it was  $-0.015$  D. and then sank, after repeated touches of the hairs, until, 75 minutes after the first observation, the leaf escaped from the holder without further disturbance of the leading off contacts, the last reading being  $-0.012$  D. It remained

\* To save unnecessary repetition in the following pages the word difference, printed in leaded type, will be used for "difference of potential." It need scarcely be added that the letter D. stands for DANIELL.

in the chamber until next morning, when a fine glass rod was introduced between the marginal hairs for the purpose of excitation; the difference was now  $-0.007$  D. The leaf was subjected to a series of excitations, each of which was followed by a normal diphasic variation. A fourth leaf was led off in the same way, but it was not, like the preceding ones, restrained by the glass holder. It was placed in the warm chamber at 1.30 P.M. and observed at intervals until 4.30 P.M. The difference varied from  $-0.032$  D. to  $-0.029$  D. On returning next morning it had diminished to  $-0.018$  D. It was at once excited by gently touching the distal sensitive hair of the led off lobe, the leaf responding by a normal variation but not closing. At once the difference sunk to  $-0.054$  D. After a second excitation of the same kind the leaf closed smartly, the subsequent difference being  $-0.045$  D. Another leaf (not of the same series) was led off by three contacts, one ( $f$ ) at the midrib, the other two at the middle of the external surface of each lobe ( $m$  and  $m'$ ), so that these surfaces could be severally compared with  $f$ . Both  $m$  and  $m'$  were negative to  $f$ , the difference being  $m -0.018$  D., and  $m' -0.024$  D. In another similar observation the corresponding differences were  $m -0.038$  D., and  $m' -0.033$  D. After a few excitations these diminished to  $-0.011$  D. and  $-0.004$  D.

The above observations, with the exception of the two last, were made for the purpose of ascertaining the truth of Professor MUNK's statement, that the external surface of the lobe is negative to the under surface of the midrib. They tend to show that the difference in question is one of frequent (perhaps constant) occurrence in unexcited leaves, and that it diminishes in consequence of excitation.

We now proceed to the study of the second, and much more important, of the two statements made in our previous paper relating to the electromotive properties of the resting leaf. The fact that the external surface of the lobe is positive to the internal surface opposite to it, we propose to designate by the term "cross difference," which sufficiently expresses its own meaning. It will be remembered that the existence of any such difference is denied by Professor MUNK, who holds that the opposite surfaces of the lobe are isoelectrical.

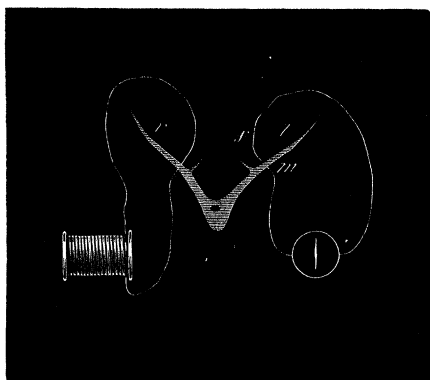
In a preliminary series of observations made in the Laboratory of the Royal Gardens at the same time with those already referred to no exceptions were met with. No importance, however, could be attached to the bare fact of the negativity of the internal surface unless it could be shown to be physiological in its nature—that is, not a result of the merely chemical or physical differences of the two opposite surfaces of contact. The method which is employed for determining whether a phenomenon is physiological or not, must always be the same. It consists in observing whether its development is so associated with the functions of the living part or organism in which it has its seat as to indicate that it is dependent upon it or forms part of it. It was therefore the first step towards determining the physiological meaning of the cross difference, to ascertain whether it is modified by changes in the vital activity of the leaf, and particularly whether it is the same in unexcited and in excited leaves, and



whether the change produced by excitation, if any, is in relation with the excitatory process itself.

The first observation on this subject was made in September, 1879, in the course of experiments made for other purposes. A leaf was led off as shown in the diagram (fig. 9)—that is, by opposite contacts, of which the internal corresponded to the space between the distal and proximal sensitive hairs, the surfaces of contact being as large as possible. The leaf was placed in the chamber at  $38^{\circ}\text{C}.$ , and was kept there for an hour, at the end of which the two surfaces were nearly equipotential, there having been previously a difference of  $-0.007\text{ D.}$  During a period of 40 minutes the leaf was subjected to a series of 16 excitations, in the course of which the difference rose

Fig. 10.



Leading off electrodes on right lobe. Exciting electrodes on left.

from  $+0.001\text{ D.}$  to  $+0.0114\text{ D.}$  After a second series of 15 excitations it rose to  $+0.032\text{ D.}$  In another leaf, observed the following day by the same method, the difference increased during 14 excitations from  $+0.0084\text{ D.}$  to  $+0.046\text{ D.}$  In a third the initial difference was  $+0.0036\text{ D.}$  During the following two hours the leaf was excited 28 times at nearly equal intervals. At the end of the first half hour the difference was  $+0.0082\text{ D.}$ ; at the end of the second,  $+0.118\text{ D.}$ ; of the third,  $+0.0178\text{ D.}$ ; of the fourth,  $+0.024\text{ D.}$  In the first of these three observations the leaf was excited mechanically—that is, by touching the sensitive hairs of the opposite lobe. In the second and third, single opening induction currents were led through the opposite lobe by pointed electrodes which penetrated its external surface.

It was not until the next year that the opportunity offered of further investigating this effect. A leaf was led off for the purpose, as in the previous experiment, by opposite contacts, the electrode being applied to the internal surface between the distal and proximal sensitive hairs. To ensure a more satisfactory initial determination of the cross difference in the unexcited leaf, it was introduced into the chamber (of which the temperature was  $20^{\circ}\text{C.}$ ) the day before, in order that any accidental difference due to initial changes at the surface of contact might be got rid of. The

observations were as follows:—Before excitation the cross difference was  $+0.024$  D.; immediately after excitation, by exciting mechanically the sensitive hairs of the opposite lobe, it was  $+0.031$  D., but soon diminished to  $+0.026$  D. A second excitation had a similar effect—it rose to  $+0.029$  D. After an hour's rest in the chamber the experiment was repeated. Before excitation the cross difference was  $+0.025$  D.; on compensating as quickly as possible it was found to have risen (about half a minute after excitation) to  $+0.035$  D.; in five minutes it had fallen to  $+0.028$  D. A second excitation similarly observed raised the cross difference to  $+0.031$  D., after which it fell to  $+0.028$  D. Four similar observations gave corresponding results, but the effect produced (increase of the cross difference), which in the first experiment of the series amounted to one-hundredth of a DANIELL, did not in the later ones exceed one-five hundredth. The indication that these phenomena afford that the electrical difference which exists in the unexcited leaf of *Dionæa* between the opposite surfaces is in physiological relation with the excitatory properties of the leaf will receive confirmation when, in the next part of this paper, we have before us the immediate effects of excitation.

In the short communication I made to the Royal Society in 1873, in which I announced the discovery of the excitatory electrical change in the leaf of *Dionæa*, I stated the existence of a "leaf current" directed from the proximal to the distal end of the midrib in the uninjured leaf. I had observed that, in general, the attached end of the midrib was negative to the further end, and expressed this fact in the language commonly used in physiological writings. I had also found that the under surface of the leaf stalk in the neighbourhood of the joint by which the leaf is attached to it, was negative to the rest of the leaf stalk, and designated this negativity by the term "stalk current." I had further observed that if the current from a battery (a small DANIELL'S cell) was directed through the leaf stalk at the same time that the two ends of the midrib were led off to the galvanometer, the difference previously existing between the ends of the midrib was increased if the current led through the leaf stalk were in the same direction with the leaf current, diminished if it were in the opposite direction. I had finally observed that in leaves severed from the plant, the "leaf current" was curiously dependent on the length of the petiole,\* so that the shorter the petiole the greater was the negativity of the proximal end as compared with the distal. Of these four observations relating to the electromotive properties of the unexcited leaf, all may be readily demonstrated in most leaves. The more complete knowledge of the subject which I now possess leads me to regard the first two as of little importance. The third possesses a greater interest, for it affords evidence that in the plant a voltaic current conducted through a living part not only influences the condition of the part through which it flows, but extends that influence to extra-

\* The experiment was related as follows:—"In a leaf with a petiole an inch long I observed a deflection of 40 scale. I then cut off half, then half the remainder, and so on. After these successive amputations, the deflections were respectively 50, 65, 90, 120." (Proceedings, vol. 21, p. 495.)

polar parts, affecting them in the same way as in nerve, so that whatever may be the nature of the action in the two cases, the phenomena scarcely admit of being distinguished from each other, as may be seen from the following experiments made in the Kew Laboratory in July, 1878.

A leaf was led off at the points  $f$  and  $m$  and  $f'$   $m'$  in the diagram (fig. 11). The negativity of the attached to the free end of the leaf required 0·01 D. to compensate it, which was expressed by a deflection of 27 scale of the galvanometer. Between the two led off surfaces of the petiole there was a difference of 0·006 D., the leaf end being negative to the other. These measurements having been made, the contacts

Fig. 11.

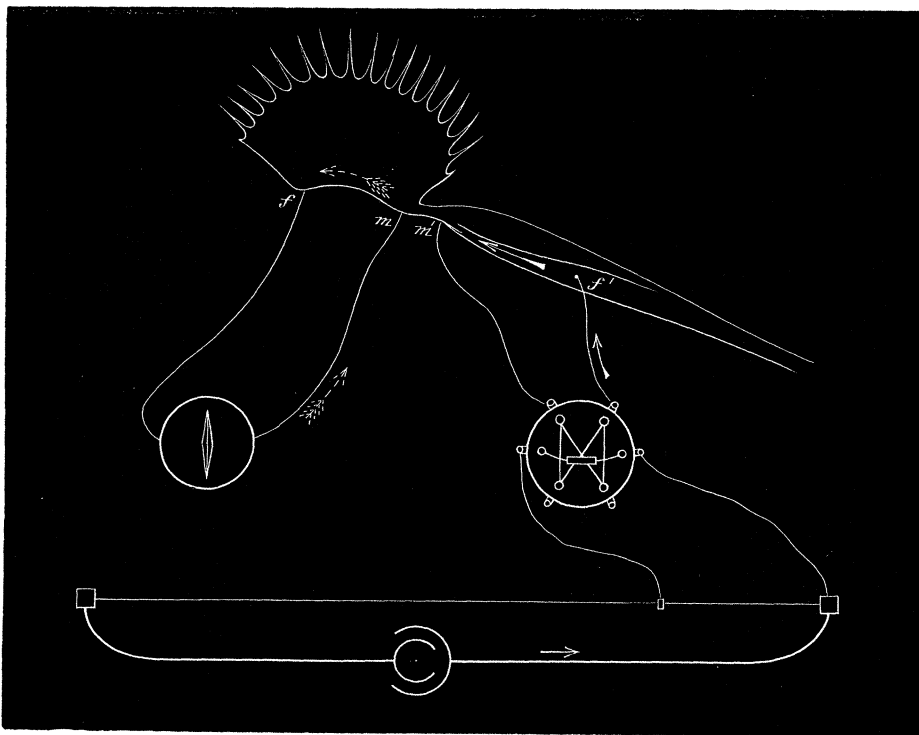


Diagram of experiment as to extra polar-influence of voltaic currents. The strong arrows indicate the direction of the derived current; the weak arrows that of the current determined by it in the leaf. N.B.—The reverser should be turned the other way.

$f$  and  $m$  were led off to the galvanometer, while  $f'$  and  $m'$  were connected with the compensator. The compensator was then reversed so as to conduct a current into the petiole opposite to that which would have been required to compensate it—in the same direction, therefore, as the natural difference—and the slide was pushed along until the needle of the galvanometer was brought to zero. The slide was then at a position indicating +0·024 D.

I have since endeavoured to determine the relation between the remarkable extra

polar effect (which so strikingly resembles the electrotonic variation of nerve) and the strength of the current which produces it, more accurately. For this purpose currents of various strengths, derived from a rheochord, were conducted through the leaf stalk, the leaf being led off as in the experiment last described. The effects observed were as follows:—With 25 centims. of rheochord wire,\* corresponding to a difference of potential of about a quarter of a DANIELL, the difference between the two leaf electrodes was diminished when the derived current was directed from the leaf, from  $-0.007$  to  $-0.0058$ , and increased to about  $-0.008$  when it was reversed. With twice the length of wire (about half a DANIELL) it was diminished from  $-0.006$  to  $-0.004$  when the derived current was directed from the leaf. Finally, with three times the length of wire the difference, which had now fallen to  $-0.005$ , was abolished; so that when the galvanometer was uncompensated, the needle was brought to zero by the derived current through the petiole.

It was ascertained by careful experiments that the electromotive change in the leaf ceased with the current which induced it. When the experiment was so arranged that the galvanometer circuit having been open during the passage of the “polarizing” current was closed at the moment that it ceased, the needle remained motionless.

In the above experiment it is seen that the induced difference of potential between the leading off contacts was very small indeed as compared with that of the contacts by which the derived current entered and left the petiole. But it is to be borne in mind that this relation does not express the true relation between the electromotive forces, for this reason: We have already assumed that all electrical changes dependent on physiological action, have their seat in the parenchyma, and this assumption is as applicable to the leaf-stalk as to the leaf itself. Consequently the “polarizing” effect of a voltaic current led from one point to another of the surface of the leaf-stalk depends, not on the electrical difference thereby determined between the surfaces of contact, but on the differences between the end cells of the interpolar stratum of parenchyma. Now it is known experimentally, first, that the resistance encountered by a current led as above described through a leaf-stalk is enormous, and secondly, that this resistance has its seat at the surface, whereas the longitudinal resistance of the middle stratum is relatively small. The result is that the physiological or “polarizing” effect of the current on the parenchyma which is the immediate effect of the electrical difference determined by the current between the most and the least positive (or negative, as the case may be) cells of the parenchyma, is a very inconsiderable fraction indeed of what it would be if the polarizing electrodes could be applied directly to the parenchyma itself. These considerations lead me to believe that although in the experiment related on the previous page the currents used were relatively considerable, there is no reason to suppose that the electrical changes in the parenchyma exceeded those (to be described in the next part of this paper) which, in

\* With reference to this mode of using the rheochord, see p. 42 (Excitatory influence of the voltaic current).

the excitable parenchyma of the leaf are associated with the excitatory state, and consequently I am led to regard it as possible that these changes may not be inadequate to produce secondary changes of the same kind in the petiole—in other words, that if the petiole were led off during the excitatory variation, it would show a variation in the same direction. Want of material has rendered it impossible to settle this important question experimentally.

The statement contained in my preliminary note in 1873, that the amputation of the petiole always increases and often doubles or trebles the electromotive force of the "leaf current," in other words, increases the negativity or diminishes the positivity of the end of the leaf which is next to it, has been confirmed by further and more exact observations. In illustration of it the following experiment may be given. A leaf was led off in the way described in my note by the two ends of the under surface of the midrib, with the fixed electrode at the distal end, it being still on the plant. The difference between the two surfaces was found to be  $-0.011$  D. On excitation by touching a hair a normal variation was observed, consisting of a positive deflection followed by a prolonged negative after effect, that is, the same phenomena as were described in 1873. On cutting off the leaf with as long a petiole as possible, no effect could be observed on the difference. Half the petiole having been removed without effect, the difference being now  $-0.0112$ , half the remainder was removed, the result of which was that the difference increased (the observation being made as soon as compensation could be effected, that is, within a minute after the amputation) to  $-0.0198$ . In the course of a few minutes the difference sank to  $-0.01$ , that is, a little below its original amount. Amputating half of the remainder of the petiole still attached raised it at once to  $-0.023$ , but it soon sank to  $-0.017$ . Clipping off the stump of the petiole which still remained, produced only a slight increase to  $-0.02$ . Throughout these experiments the excitatory variation exhibited the same character. We have thus before us two sets of facts relating to the electrical influence of the petiole on the leaf, viz.: (1) that a current through the petiole, if directed towards the leaf, renders the attached and proximal end negative to the distal, and (2) that removal of the petiole produces the same effect. I am unable at present to explain either of these phenomena. The nature of the electrical relation between the two organs which underlies both must remain undetermined until it is possible to make further experiments.

#### PART IV.

#### THE ELECTRICAL EFFECTS OF EXCITATION.

1. *The fundamental experiment.*—Under this term I propose to designate the observation of the electrical effects which present themselves when the leaf is led off by opposite surfaces of one lobe, while the other is excited by any of the methods

described in Part II. It is to be understood that the electrode  $f^*$  is always applied to the internal surface of the leaf between the three hairs (in the trigone) and the electrode  $m$  to the outside surface exactly opposite  $f$ . What happens under this condition is always the same. After a delay—of which the duration varies with the temperature—the external surface becomes first negative, then positive, to the internal. For the purpose of giving a general view of the time-relation of the first phase of the variation, I submit the following table, which gives the results of three successive rheotome observations† :—

	Time after excitation at which galvanometer circuit opened.							
	0·2"	0·4"	0·6"	0·8"	1"·0.	1"·2.	1"·4.	1"·6.
Obs. I.—Deflection . . . (100 sc.=0·063 D.)	0 0	3 4	8 10	22 28	28 35	25 31	29 38	16 20
Obs. II.—Deflection . . . (100 sc.=0·045 D.)	1 1	19 17	30 27	45 40	44 39	29 26	8 7	2 2
Obs. III.—Deflection . . . (100 sc.=0·0353 D.)	0 0	5 3	36 25	55 39	42 30	17 13	21 15	7 5
Mean result . . .	0·3	8	20	36	35	23	20	9

Each of the leaves employed in the experiments of which the results are given was fixed in the glass holder described in Part III., and led off, as above explained, by opposite contacts. The three observations were made consecutively; the opposite lobe to the one led off was excited by the rheotome (see fig. 6), the electrodes used being steel needles, of which the points (2 millims. apart) were inserted into the external surface of the lobe. It was consequently necessary to move them three or four times during the period of observation, care being taken that each new insertion should be at the same distance from the middle line of the leaf as its predecessor. Every excitation was repeated twice, and, if the deflections obtained differed in extent, a third time. As it was of much more importance to complete the observation while the leaf remained in a normal condition than to obtain great accuracy in

\* In using the letters  $f$  and  $m$  to designate the leading off electrodes, it is assumed, as in former papers, for convenience of description and for no other purpose, that any difference or current is to be called negative when it is of such a nature that  $m$  becomes negative to  $f$ , and *vice versa*. The words "negative" and "positive" therefore have only a relative meaning.

† The numbers in ordinary figures represent the actual deflection observed; those in black type, what they would have been had the resistance been equal in all the experiments and such that a difference of 0·05 D. would have been expressed by 100 scale of the galvanometer. The sensibility of the galvanometer was purposely reduced for this preliminary experiment, it being desirable to lengthen the periods of closure to 0·2".

the readings, they were not repeated more frequently than appeared necessary. In each experiment the leaf was introduced into the warm chamber at 32° C. some time before the observation began. The observations were made at intervals of two minutes and a half, and the number of excitations necessary for each was from 20 to 25. Consequently, each lasted nearly an hour.

The observations given in the table serve to show that the variation lasts during the whole of the first and the greater part of the second second, and that it culminates at from six to eight-tenths of a second after excitation. For these purposes it was thought best to employ long periods of closure (using the galvanometer in a very insensible state) and to make the end of one period coincide with the beginning of the next in such a way as to cover the whole period to be investigated. But such a method was quite inadequate to give information as to the question of greatest importance in relation to the excitatory variation—that of the time at which the effect commences. For this purpose it was necessary to shorten the periods of closure, and consequently to increase the sensibility of the galvanometer. We therefore substituted for the revolving rheotome the “pendulum” rheotome, of which the description is given in Part III. The instrument is so arranged for the experiment that on swinging from right to left it opens three keys in succession—K 1, K 2, K 3 (see fig. 8). Of these, the first occupies a fixed position, so that it is opened about 0.1” after the pendulum is liberated. This position corresponds to zero of the pendulum scale. The other two keys, K 2 and K 3, can be slid along to any required distance from K 1. The binding screws of K 1 are connected with the primary circuit of the induction coil; those of K 2 with the shunt of the galvanometer, so that when this key is closed the galvanometer is “short circuited;” those of K 3 are interpolated in the galvanometer circuit between the leading-off electrodes and the shunt. Consequently, when the three keys (being previously closed) are opened in succession by the pendulum, the primary circuit is first opened by K 1, the galvanometer circuit is unbridged—that is, closed—by K 2, and again broken by K 3. As the scale of the pendulum is divided into hundredths of a second and extends over a period of a quarter of a second, this arrangement affords the means of investigating with great exactitude the events of the beginning of the variation.

The galvanometer is brought to such a degree of sensibility that with a resistance of 10,000 ohms, in addition to that of the galvanometer in circuit, 0.0001 D. gives a deflection of 97 scale. To facilitate accurate reading a slightly concave mirror is placed in front of the scale, by means of which an enlarged image of the part of the scale most used can be seen by the observer without fatigue. The leaf is led off and excited as in the fundamental experiment. For excitation, non-polarizable electrodes of the form described at D, p. 18, are used, the contacts being opposite, and corresponding in position to those of the leading off electrodes applied to the opposite lobe. The plant of which it forms part is placed in the chamber, the air of which is saturated, at 26° C.

It having been first ascertained that when K 2 was at 0.2'' of the scale of the pendulum, and K 3 at 0.04'', there was no variation, successive observations were taken with K 3 at positions further and further removed from K 2 (which remained stationary at 0.02'') with the following results:—With K 3 at 0.06'' there was a trace of movement of the needle; with K 3 at 0.08'' it had increased to —4 scale; at 0.1'' to 12; at 0.15'' to —39 sc. To complete the observation K 2 was moved to 0.1'' and K 3 to 0.2'', so that the period of closure now corresponded to the second tenth of a second after excitation; the galvanometer reading was —161 sc.; finally, between 0.2'' and 0.3'' we had —248 sc.

In four other experiments the results (stated shortly) were as follows:—

Experiment I. Temperature 32–35° C. 0.02''–0.4'', no deflection; 0.02''–0.05'', a trace; 0.02''–0.06'', —5 sc.; 0.02''–0.1'', —35 sc., —31 sc.

Experiment II. Temperature 34° C. 0.02''–0.04'', no deflection; 0.02''–0.05'', —3 sc.; 0.02''–0.06'', —6 sc.; 0.02''–0.15'', —70 sc.

Experiment III. Temperature 32° C. 0.02''–0.06'', no deflection; 0.02''–0.08'', no deflection; 0.02''–0.10'', —13 sc., 22 sc., 23 sc.; 0.02''–0.9'', —10 sc., 7 sc., after which previous negative results were verified by repetition.

Experiment IV. Temperature 36° C. 0.02''–0.04'', trace; 0.02''–0.05'', —4 sc., 6 sc.; 0.02''–0.6'', —8 sc., 10 sc.; 0.02''–0.8'', —12 sc.; 0.02''–0.1'', —16 sc.

The experiments were made consecutively, that is, on successive days, and, with the exception of Experiment IV., under precisely similar conditions. In Experiment IV. the temperature was inadvertently allowed to rise to 36°,\* a fact which sufficiently accounts for the somewhat earlier beginning of the variation.

The experimental results which have been given justify the statement (1) that the first phase of the variation is entirely negative; (2) that it begins (at the temperature of 32° C.) about one-twentieth of a second after the excitation; (3) that its electromotive force rises rapidly at first and subsequently very gradually; (4) that it culminates about the end of the first half-second after excitation and subsides gradually towards the end of the third half-second.

In the paper which I communicated to the Royal Society with Mr. PAGE in 1876, I stated that the variation of the leaf did not begin until two-tenths of a second after excitation, and gave the grounds on which the statement was based. The experiments which led us to conclude that what we then called the "electrical delay" (the interval of apparent electrical inactivity) were correctly made. We repeated them under the most favourable circumstances in the Kew Laboratory in 1878,† with perfectly similar results, but nevertheless our interpretation of them was erroneous.

\* It is probably also worth noting that the day on which this experiment was made was the hottest of last summer, Friday, July 16.

† The observations made at Kew, in 1878, gave 0.12'' as the shortest period of delay when the



In 1876 we used an arrangement by which (1) the act of excitation (if mechanical, the depressing of a lever which brought a camel-hair pencil into contact with one of the sensitive hairs; if electrical, the breaking of the primary circuit of the induction apparatus) recorded itself on a blackened cylinder revolving at a rapid rate by clock-work; and (2) the record of the response was inscribed on the same cylinder.

In the first record, the inscription of which was effected with the aid of DEPRez' electro-magnetic "signal," or chronograph, the loss of time was inappreciable. The response was the motion of the mercurial column of the capillary electrometer. This was observed by one of us (Mr. PAGE), who recorded the event on the cylinder by closing the current through the "signal" which the other had broken in the act of excitation. The interval between break and make of the signal current comprised: (1) the physiological delay to be measured; (2) the instrumental delay (that is the time lost, if any, in moving the mercurial column); and (3) the "personal time" or "reaction time" of the observer. The last we had no difficulty in eliminating, for Mr. PAGE's personal time was subject to very slight variation. The second we had not taken into account at all, assuming it to be non-existent. The consideration that excitatory difference in *Dionæa* probably begins gradually, and that if so the motion of the column is slow, and may therefore not be perceived until a longer period than the normal time after it has actually begun, led us later to conclude that our method was inadequate for the purpose, and that the only way by which the question could be settled was by the use of the rheotome and galvanometer.

*The after effect.*—The change which has been above described as occupying the greater part of the first 2" after excitation is followed by a period during which the surfaces are nearly, or absolutely, in the same condition electrically as they were before excitation, but sooner or later a secondary effect begins, which I propose to call the *after effect*. During the after effect the internal surface becomes negative to the external.

In order to bring the after effect into view, the rate of the rheotome must be reduced and the periods of closure much lengthened. In our first observation the rate of revolution was 1 in 78", and the closing time 1.4". The leaf was excited as before at intervals of three minutes, without being placed in the chamber, the temperature of the room being 18° C., the mode of excitation being the same as in the previous series. After the first excitation the galvanometer circuit was closed between 1.3" after excitation, and 2.6". In this case the deflection was negative, for, in consequence of the relatively low temperature at which the observation was taken, the first phase of the

measurement was made in the moist chamber at 32° C.—a result which exceeds the truth by about 0.05". Assuming that the error is to be set down, not of course to loss of time in transmission of the effect to the mercurial column, but to the lengthening of the "reaction time" when the motion to be perceived is gradual, it renders all the observations made by signalling with the electrometer useless for the purpose of measuring the duration of the delay, but does not interfere with the use of this method for other purposes, particularly for determining which of the two similar electrical responses occurs earliest.

variation was prolonged so as probably to cover a great part of the closing time. In the second experiment, the closing time being 1.3" later, namely, from 2.6" to 3.9" after excitation, there was a positive deflection which was not measured. In the third, 1.3" later, the positive deflection was 90, in the following ones, successively, +71, +70, +50, +45, +30, +30, and so on. On placing the leaf in the chamber and repeating the observation a perfectly similar set of results was obtained. Between 0.6" and 1.95" after excitation there was a negative deflection of -380. Between 1.3" and 2.6" a much smaller negative of -70, these being the expression of the first phase. Then followed between 2.6" and 3.9" a positive deflection of +6, after which, at successive consecutive periods, +100, +75, +70, +42, +20, and so on.

From these and other similar observations made by the same method, it appears that in the fundamental experiment the external surface which, 4" or 5" after excitation, attains its greatest positivity as compared with the internal, returns very slowly (if at all) to its previous state. Various other methods were used for the purpose of observing the progress of the after effect with more completeness, of which the best is exemplified in the following experiment.

The method employed was as follows :—It having been ascertained that the electromotive force of the after effect at 2" after excitation corresponded to a deflection of 160 galvanometer scale, of which the equivalent was 0.016 D., the galvanometer having been over compensated to that extent was closed at 1" after excitation. In this way the needle was brought so nearly to rest that it followed the changes of difference without oscillation, and could without difficulty be read at 5" intervals. The readings were :—

At 3",	at 8",	at 13",	at 18",	at 23",	at 28",
+9.	-50	-130	-142	-149	-160.

That is, at 3" after excitation the positivity of the external surface was a little more than 0.016, at 8" less, at 13" much less, and so on, so that the result in electromotive force might be expressed as follows: +0.017 D., +0.011 D., +0.003 D., +0.002 D., +0.001 D. Accordingly, in another leaf, after carefully compensating the existing difference and breaking the galvanometer circuit as before, we at once adjusted the compensator so that on closing there should be a negative difference of 0.01 D. We then excited, and 2" after closed the galvanometer circuit. Again the needle followed the slow electrical change, its positions at 5" intervals being as follows: +70, 0, -60, -120, -140, -150, -150, -160, &c. As the value of 0.01 D. with the preparation in circuit was 190 scale, the internal surface was still 30 scale (= about 0.0015 D.) negative. Repeating the same experiment with an over compensation of 0.015 D. the successive positions of the image were: +20, -40, -140, -200, -220, -240, -250, -260, &c. So that the observation indicated, as before, that in this instance the negativity of the internal surface exceeded the compensation used.

In a third leaf observed at the same time, the resistance being unusually low, it was

impossible to get the after effect on the screen, although the difference was not larger than in the other two. On interpolating as before a length of wire corresponding to 0·009 D., and closing the circuit 2" after excitation, the successive readings at 5" intervals were: +200, + off scale, +230, +60, -50, -130, -210, -260, -298, -320, -345, -350, -360, &c., the reading due to over compensation being -370; so that here, as in the other instances, the maximum difference was between 0·01 D. and 0·02 D.

*Influence of repeated excitations on the intensity and duration of the after effect.*—Whenever a freshly prepared leaf is observed with the capillary electrometer during the course of a succession of excitations, it is found that in the first two or three the negative movement of the mercurial column is followed by a positive one which equals or may even exceed itself in extent. As the excitations are repeated, the positive phase becomes smaller and smaller, until after eight or ten, it may very readily escape observation.

For the purpose of obtaining more exact information on this subject, the galvanometer and rheotome were used; for it was obvious that if in a succession of experiments the electrical condition of the led off surfaces could be compared, during a period after excitation corresponding to the maximum of the after effect, that is, the period during which the internal surface of the leaf is most negative, the successive deflections recorded would give a good indication of any such diminution as the observations with the electrometer indicated.

The period of closure chosen was between 3" and 4·5" after excitation. The deflections were, of course, all positive, but varied in extent from 175 to 21. In 21 successive excitations the mean of the first seven deflections was 157, of the second seven, 86, and of the third, 55·5. In this experiment the leaf was excited at intervals of not less than two minutes. If the excitations are repeated as often as every half minute it is found that the after effect completely disappears. This can be easily shown by the electrometer, but can be more exactly determined by the galvanometer. The best way to do this is, first to make with the galvanometer an observation of the kind described in the last paragraph (p. 30), then to subject the leaf to a succession of excitations at short intervals, and to follow these by a second galvanometer observation for comparison. The following results are obtained:—

(1.) The fourth excitation of a leaf showed an after effect of which the course was as follows, as indicated by the negative deflections observed at 5" intervals: +300, +262, +155, +100, +74, +66, +59, &c.

(2.) After ten effectual excitations at half minute intervals the after effect was: +160, +154, +70, +49, +40, +30, +12, &c.

(3.) After ten more effectual excitations it was: +25, +5, 0, 0, &c. The leaf was then left to itself for 15 minutes and the after effect again observed. It was: +85, +130, +62, +52, +49, +40, +36, &c.; and after ten minutes' further rest: +100, +200, +104, +70, +62, +52, +47, &c.

On repeating the excitations at half minute intervals, the after effect was again entirely abolished, reappearing again after a period of rest. It was further found, in another experiment, that if, after a succession of excitations at half minute intervals, a leaf was excited twice at longer ones, *e.g.*, two minutes and a half, the after effect had, by the time the second was made, already reappeared, or if previously diminished had much increased.

In Part III. it was shown that the influence of an excitation in increasing the previous difference (that is, the positivity of the external surface) or in other words, in rendering the external surface permanently more positive than it was before excitation, depended on the interval of time which had elapsed since any previous excitation. In a leaf which has been repeatedly excited at frequent intervals, excitation leaves behind no change in the electrical relation of the opposed surfaces, because there is no after effect. In other words, the persistent change of the potential, consequent on excitation, described in Part III., is a small remainder of the after effect, as it seldom amounts to more than 0.002 D, whereas the after effect itself often attains at its maximum an electromotive force of 0.02 D. It may be roughly stated that the latter is apt to leave behind a remainder of something like one-tenth of its amount. All that can be insisted on as resulting from observation is, that the subsidence of the after effect is extremely slow and incomplete, and that whenever it exists it leaves behind it a remainder—the excitatory increase of positivity of the external surface, already described.

*The excitatory process as observed when the leaf is led off by symmetrical (identical) surfaces of contact of opposite lobes.*—The excitatory variation which is observed in a leaf led off symmetrically, that is, by corresponding surfaces on opposite lobes, is the combined result of equal and opposite electromotive changes which have their seat in the tissues between the contacts. Consequently, if in an unsevered leaf so led off we suppose an excitation to take place at the same moment from each electrode, no variation would be observed, for the electrical changes due to the one excitation would exactly balance those due to the other. The appearance of a variation would indicate inequality of action between the two lobes. If the excitatory response of the right lobe were either stronger than that of the left, or were accomplished in a shorter time, the variation would express the difference. In the former case the right would have the advantage throughout; in the latter, the right would have the advantage at first, afterwards the left.

The case in which two lobes are led off and excited simultaneously and symmetrically has not been realised experimentally. The consideration of it, however, serves to facilitate the understanding of the case which actually presents itself in which the leaf is led off by symmetrically placed contacts on opposite sides, but excited on one side only. In this case experiment shows that there is always a galvanometric effect, but that the character of the variation observed differs in different leaves very considerably—so much so indeed that at first sight the differences appear irreconcilable. Before

giving an account of them it will be useful to consider what we should expect. This depends mainly on the question whether, in the transmission of the excitatory effect from the seat of excitation to the opposite lobe, time is lost or not. In the fundamental experiment we have evidence that the excitatory effect travels. For when we excite the left lobe and the right responds, some time is without doubt occupied in transmission. If, as Professor MUNK believes, the transmission is so extremely rapid that the excitatory change may be said to begin at the same moment at all parts, the case would resolve itself into the hypothetical one from which we started, viz.: that of excitation taking its rise at the same moment from both leading off contacts; and this being so, any galvanometric effect observed must be due to one of the two causes referred to above, namely, either to unequal intensity or to unequal duration of the electrical response of the two sides of the leaf. If, in addition to these inequalities, we have to consider the influence of propagation (*i.e.*, if the time lost in propagation is sufficiently considerable to tell), the interpretation of the galvanometric effect observed in an experiment in the form represented in fig. 13 becomes extremely complicated; for the modifications severally due to loss of time and to difference of intensity would interfere with each other in a way very difficult to estimate. It will be shown in a subsequent section that the rate of propagation in the excitatory disturbance is great, as compared with the duration of the excitatory effect; in other words, that its wave-length is considerable. This being so, the question whether or not the modification due to loss of time will show itself, will depend on the abruptness with which the disturbance commences, as will be seen from the following diagrams. If, for example, we suppose that the excitatory electromotive changes are so directed that the led off surface on either side becomes negative to the midrib, and that the rise of negativity on the right side is represented in two different leaves by the curve *a*, that of the left side by the curve *b*, the galvanometric effect during the period represented will be expressed in each figure by the shaded area, the depth of which is greater in the first case than in the second; so that if (as is represented in fig. 12, C) it were further diminished by a greater abruptness in the rise of negativity on the left side, it might escape observation. And it is easy to see that if, in addition to beginning more abruptly the excitatory disturbance on the left side were more intense, the influence of the loss of time in propagation would lose itself entirely in that of inequality. The two last suppositions (fig. 12, B and C) appear to afford the key to the understanding of what is actually observed when a leaf is led off by symmetrical points and excited near one of them. In this case there is always an excitatory effect, proving that the two lobes do not respond equally. Further, we have found that so far from its being true, as asserted by Professor MUNK, that it makes no difference what spot is chosen as the seat of excitation, it never happens that variations of the same character are obtained when, in two successive experiments, or in two series of experiments, the right and left lobe are alternately excited. At first sight it seemed as if this difference were referable to propagation, but more careful consideration has

led to the conclusion that inequality of response has a much larger share in its production than loss of time. Ground for this conclusion is to be sought, as regards each particular case in which a symmetrically led off leaf acts unsymmetrically, in the observation that each of its two lobes when separately and successively tested shows the same inequality. Unfortunately, the nature of the case did not present itself in its true relations until too late to do this in a sufficient number of cases, but I have abundant evidence that in general the electrical reactions of the two lobes of a leaf are unequal.

Fig. 12.

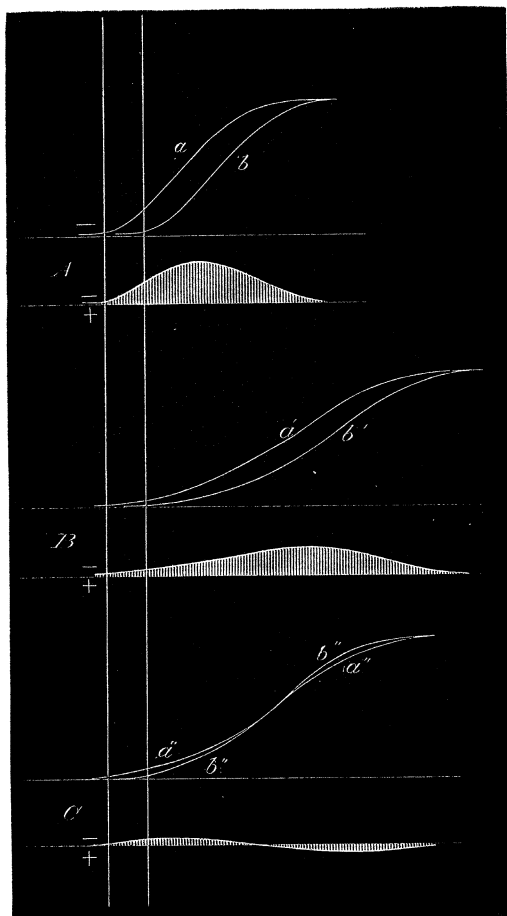


Diagram intended to illustrate the relations of the first phase of the excitatory variation when the leaf is led off by symmetrical contacts on the under surface.

In observations on symmetrically led off leaves the electrodes may be applied either to the internal or the external surface.

*Symmetrical contacts on the internal surface of the leaf between the two marginal sensitive hairs.*—In order to determine, if possible, the general character of the variation in this form of experiment, four observations were made in succession with the electrometer. The leaves used were led off by symmetrical contacts between marginal

hairs of opposite lobes, and subjected, first on one side and then on the other, in each experiment, to two series, each consisting of ten successive excitations. The table shows the results. The letter *f* or *m* indicates that, in the case to which it relates, the fixed or movable electrode was on the excited lobe. In all the experiments the excitation was effected by touching a sensitive hair with a camel-hair pencil. The hair touched is indicated. After each observation the difference was compensated. The mean differences are given in the last column. All of the leaves were tested by observing the excitatory effect in each lobe separately when led off as in the fundamental experiment.

No. of experiment.	Which lobe excited.	Which hair touched.	Which electrode on excited lobe.	Character of variation.	Difference.
I.	R.	Proximal .	M.	Monophasic, $-39.1$ . . . . .	+0.011 D.
	L.	Ditto .	F.	Diphasic, $+11.7, -21.2$ . . . . .	+0.007 D.
II.	L.	Proximal .	F.	Monophasic, $+6.2$ . . . . .	-0.01 D.
	R.	Ditto .	M.	Diphasic, $-2, +25.4$ . . . . .	-0.02 D.
III.	R.	Proximal .	M.	Diphasic, $-3.2, +18.8$ . . . . .	-0.007 D.
	L.	Ditto .	F.	Monophasic, $+22.2$ . . . . .	-0.005 D.
IV.	L.	Distal . .	F.	Diphasic, $+3.4$ pause . . . . .	+0.023 D.
	R.	Ditto . .	M.	$+7.4$ (from zero)* . . . . . Diphasic, $+9.8, -2.9$ . . . . .	+0.023 D.

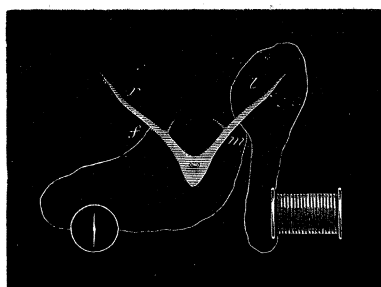
In leaves I. and II. the difference between the two lobes was so great that the leaves could scarcely be considered normal. In leaf III. the variation was normal on both sides, but the first phase was larger on the left than on the right ( $-8.5, +4.3$  on the left,  $-4.4, +7.8$  on the right). In leaf IV. the first phase was nearly equal on both sides ( $-30.0$  on the left,  $-31.6$  on the right), and was followed in both cases by a normal after effect. But these facts afford no clue to the understanding of the results recorded in the table, excepting in so far as they show that in a well-balanced leaf—*i.e.*, one which responds to excitation with some degree of equality—whichever lobe was excited directly had the advantage over the other in the compound variation. That this was so was confirmed by the observation that in leaf IV. the negative phase of the variation, as observed with opposite contacts (the fundamental experiment), was much smaller ( $-18.4$  on the left,  $-26.6$  on the right) when the opposite lobe was excited than when a hair adjacent to the leading off electrode was touched. The only general conclusion that I venture to draw from the experiments is a negative one, namely, first, that the differences which are observed between the excitatory effects in the two cases are as little due to mere inequality of response in the two lobes as to

\* To be read thus:—In the first phase, the meniscus moved from zero of the scale to  $-3.4$ , in the second, after a pause, from  $3.4$  to  $7.4$  in the same direction.

propagation ; for such inequality would not account for the effect produced by changing the seat of excitation. We must suppose that, independently of any difference in physiological activity which may exist between the lobes, the electrical disturbance is more intense in those parts of the leaf which are nearest to the seat of excitation than in those which are more distant from it. This being admitted, and account being taken of the complicated arrangement of the electromotive structures concerned, the incongruity of the results need no longer surprise us.

*Symmetrical contacts on the under surface, opposite the space between the sensitive hairs* (fig. 13).—The same considerations apply to this case as to that in which the leaf is led off from the upper surface ; but inasmuch as the under surface presents no appreciable difference of structure, whereas in the upper we have to do with the sensitive hairs, the prospect of obtaining congruous results is not so hopeless. It will be sufficient to refer to one or two experiments made during the autumn of 1880, in which the galvanometer and rheotome were used for comparison of the excitatory effects.

Fig. 13.



Leaf led off by symmetrical contacts on under surface of lobes.

The first leaf used for the purpose was one in which symmetrical results could hardly be expected. It was a large well-grown leaf which contained the remains of a digested fly ; and when, after having been excited an equal number of times on either side, it was allowed to close, the incurvature of the left lobe was much greater than that of the right. This leaf was excited electrically (the exciting contacts, *x*, being on the internal surface) and led off symmetrically, the electrodes touching the external surface exactly opposite the trigone on either side, the fixed electrode being on the right side. The rheotome gave the following result, the period of closure being 0·2'' :—

	<i>x</i> On left lobe.	<i>x</i> On right lobe.
	M.	F.
At 0·4'' . . . . .	− 37·5	+ 50
At 0·6'' . . . . .	− 9·0	+ 41
At 0·8'' . . . . .	+ 33·0	+ 40



The numbers given are the means of two accordant observations at each period, before each of which the difference was compensated.

In a second leaf, led off in precisely the same way, the general character of the excitatory effect was observed with the electrometer. The fixed electrode was on the left lobe. When that lobe was excited electrically (as in the last experiment) the variation was diphasic, the first phase being positive. When the right lobe was excited ( $x$  on right lobe), the variation was also diphasic, with opposite signs. The rheotome gave the following, the closing time being 0.075":—

	$x$ On left lobe.	$x$ On right lobe.
	F.	M.
At 0.10" . . . . .	+ 9	— 7
At 0.20" . . . . .	+24	—18
At 0.30" . . . . .	+27	—20
At 0.40" . . . . .	+20	— 9
At 0.50" . . . . .	+ 4	— 4

The numbers express in each instance the means of two observations, before each of which, as usual, the difference was compensated. The resistance exceeded 160,000 ohms, notwithstanding that the utmost pains were taken in making the leading off contacts.

In a third experiment we had at our disposal a large vigorous leaf. The fixed contact was on the right lobe. The leaf was subjected to 48 excitations during a period of 70 seconds, to all of which it responded. Its resistance was such that 100 scale of the galvanometer corresponded to 0.45 D. The excitations gave with the rheotome (the duration of closure of the galvanometric circuit being one-fifth of a second) the following results:—

	$x$ On right lobe.	$x$ On left lobe.
	F.	M.
At 0.20" . . . . .	+28	—33.5
At 0.4" . . . . .	+44	—24.3
At 0.6" . . . . .	+50	— 3.0
At 1.0" . . . . .	0	0

These observations show, even more strikingly than the previous ones, how completely Professor MUNK is mistaken in his statement that, provided the leading off is the same, the seat of excitation is a matter of indifference. They also serve to strengthen the conclusion that the intensity of the electrical disturbance is greater in

the neighbourhood of the seat of excitation than at a distance from it. No other explanation would account for the results.

*Rate of propagation of the excitatory disturbance.*—The only certain method of determining the rate of transmission of the excitatory change in *Dionæa* consists in measuring the interval of time after excitation at which the electrical effect first becomes appreciable, by a series of observations in which a leaf, led off as in the fundamental experiment, is excited alternately in the neighbourhood of the leading off electrodes, and at a corresponding part of the opposite lobe. In such an experiment the interval of time between excitation and effect in the one case will be greater than in the other by a fraction of a second. That fraction, multiplied by the distance in millims., will give the rate per second in millims.

In our former paper we attempted to arrive at a conclusion on the subject by a method which we now know to be inapplicable. I will first refer to certain experiments which were made on this principle in the Kew Laboratory in 1878, for the purpose of confirming the statements contained in our former paper (1876). A plant was placed in the chamber at 30° C., and led off as above stated. The excitatory variation on the electrometer scale was  $-12$  followed by  $+10$ . The leaf was excited by touching the sensitive hairs, alternately on opposite sides, the time of excitation and that of response being determined by the method referred to in Part II. During the period of observation the temperature rose to 37° C. The observations were made in four series, in which the means of the time-intervals between excitation and response were as follows:—

1.	Excitation of the hairs of the same side	. . . . .	0·08"
2.	" " " " opposite side	. . . . .	0·28"
3.	" " " " same side	. . . . .	0·116"
4.	" " " " opposite side	. . . . .	0·243"

In another leaf the experiment was made first at the temperature of the room, afterwards at that of the chamber 32° C. Six observations gave:—

1.	Excitation of the hairs of the same side	temp. 18° . .	0·10"
2.	" " " " opposite side	" 18° . .	0·27"
3.	" " " " same side	" 32° . .	0·05"
4.	" " " " opposite side	" 32° . .	0·12"
5.	" " " " same side	" 18° . .	0·43"
6.	" " " " opposite side	" 18° . .	0·78"

Here the excitation was by induction shocks, the external surface being punctured by fine electrodes for the purpose. This observation shows the combined effect of temperature and local diminution of excitability. The influence of the latter is still more strikingly shown in the following series, the temperature throughout being 18° C.

Leaf fresh :—

1. Excitation of the same side . . . . . 0·15"
2. „ „ opposite side . . . . . 0·26"

After repeated excitation :—

3. Excitation of the same side . . . . . 0·50'
4. „ „ opposite side . . . . . 0·84"

Later :—

5. Excitation of the same side . . . . . 0·53"
6. „ „ opposite side . . . . . 0·78"

Still later :—

7. Excitation of the same side . . . . . 1·54"
8. „ „ opposite side . . . . . 3·50"

These Kew experiments showed that at the ordinary temperature of summer the interval of time between excitation and response is considerable, and that its length depends on the distance of the seat of excitation from the led off surfaces, and is much longer in injured leaves than in fresh ones. But for reasons which I have already stated in discussing the fundamental experiment, it is certain that time measurements made by the method of signalling are incapable of giving an answer to the question of the rate at which the excitatory effect is propagated. During the hot weather of last summer (July, 1881) we had the opportunity of making experiments with the pendulum rheotome which proved in the clearest way that, under favourable conditions, the rate of propagation in the leaf of *Dionæa* is extremely rapid.

From the extreme difficulty which was met with last summer in obtaining plants in vigorous condition, I was prevented from making as many observations as I deemed desirable. Of those which were made I think it best to submit the results of one only, which was perfectly satisfactory :—

Period of closure.	Same lobe excited.	Opposite lobe excited.
0·02" to 0·04"*	0	0
0·02" „ 0·06"	—11	0
0·02" „ 0·08"	—22	0
0·02" „ 0·10"	—31	0
0·02" „ 0·125"	..	9
0·02" „ 0·15"	..	26
0·10" „ 0·20"	..	50

In this experiment the plant was placed in the chamber at 38° C. and remained at that temperature throughout. The leaf was led off as in the fundamental experiment. The excitation was by induction shocks. The exciting electrodes were applied on the

\* Trace at 0·05.

led off lobe close to the distal and proximal hairs respectively, so that a straight line connecting the contacts would pass through both of them. On the opposite lobe they were applied opposite to each other in the space between the hairs. The observations with excitation on the opposite side were made first, so that the effect of "exhaustion," if any, in increasing the delay, would tell against the result. Two observations were taken at each period, which coincided in most cases exactly. The weather was extremely warm and sunny. In other leaves the time interval between the two sets of results never exceeded that observed in the one recorded, so that I think I am justified in saying that the rate of transmission is at least 200 millims. per second. But it must be carefully borne in mind that this only applies to plants at hot-house temperature, as was strikingly shown in all of the experiments. A leaf, which in the chamber at  $32^{\circ}$  C. responded to an excitation of the opposite lobe when the galvanic circuit was opened at 0.08" after excitation, gave no trace of a variation with a closing time which lasted to 0.20", when the experiment was repeated as soon as possible afterwards in the room at  $21^{\circ}$ . On shifting K 2 to 0.20", so that the galvanometer circuit remained closed from one-fifth of a second after excitation onwards, the image shot off the screen.

#### PART V.

##### EXCITABILITY.

The investigation of excitability in *Dionæa* is a matter of great difficulty, partly on account of the rapid changes which it undergoes, partly on account of the difficulty of so applying any excitant that its exciting action can be assumed to be constant. The results of our observations as to the behaviour of the leaf when excited will be best stated in relation to the different methods of excitation.

1. *Mechanical excitation*.—It was observed by Mr. DARWIN ('Insectivorous Plants,' p. 289) that when a single human hair was fixed in a handle, and cut off so that one inch projected, and "the extremity of the hair was then brought by a slow movement laterally into contact with the tips of a filament, the leaf" excited "instantly closed," or, "after two or more touches of the same kind;" whence he concluded that they were extremely "sensitive to a momentary touch" which was best adapted to enable the trap to close on an insect. To this I have to add another observation, which is at first sight so improbable that I have become convinced of its truth with great reluctance, viz.: that the touch is much more efficacious when it is a living touch, that is, when communicated by an instrument (particularly a hair or a camel-hair pencil) held in the hand, of which the motion is muscular, than by any mechanical arrangement which I have been able to contrive. The two forms of mechanical excitation which have been used are (1) a lever, of which the axis works in a firm support of which the height can be varied at will, at the end of which a camel-hair pencil is fixed of which

the point is downwards, and so constructed that when it is depressed by the finger the signalling circuit is closed and the tip of the camel-hair pencil brought into contact at the same moment; and (2) a similar lever of which the motion is due to the action of an electro-magnet included in the signalling circuit. Whichever of these forms was employed, and whatever care was taken in setting the pencil so as to ensure its hitting the hair to be excited without fail, it was invariably found that after a few excitations no further response was obtained; whereas it is always easy to repeat an experiment which I made with Dr. FRANCIS DARWIN in 1873, in which we succeeded in exciting a leaf during an hour at two minutes' intervals, without a single failure of the electrical response. This may, I think, be due to the relative rudeness (if such a word may be applied) of the mechanical motion as compared with the other. I thought at first that it might be attributed to the sameness of the motion—*i.e.*, to the circumstance that when the blows are inflicted mechanically, however gently, they are all in the same direction and each acts on a part which has been acted on before. As, however, I have not found that altering the direction of the motion makes any perceptible difference in the result, this explanation fails. What is to be learned from the observation is this: the very slightest touch is an "adequate" stimulus; any beyond this is more than sufficient and injurious. Its exhausting influence is, however, evidently perfectly local; for the loss of sensibility of one hair fails to determine any similar loss in others which have not been excited. If in any leaf a single hair, excited repeatedly by mechanical means, has ceased to respond, you can at once reproduce the result by acting on another hair, which will respond just as readily as if the leaf were intact.

#### *Electrical excitation.*

1. *Single induction shocks.*—If dry metallic electrodes are applied to the internal surface of the leaf, induction shocks conducted through them are without effect, however near the electrodes may be—the reason being, no doubt, that the surface is so perfectly dry that but little current passes between the platinum points. This difficulty may be overcome, either by using the pointed wires so that by piercing they can be brought into contact with the cells beneath, or by covering the surface with a layer of conducting material such as kaolin, made into paste with half per cent. solution of common salt. I used the former of these methods in my first experiments in 1873, as well as in those of which the results were published in 1876. For obvious reasons it is preferable to use non-polarizable electrodes, to which the form described in Part II. is the best that can be given. I have always used the small induction apparatus of DU BOIS-REYMOND without removing the core, and one DANIELL cell in the primary circuit. In all experiments opening induction shocks have been exclusively used. As a rule, the electrodes are applied to opposite points of the upper and under surface of one lobe. With reference to the action of such currents, so applied, it has been ascertained: (1) That when in a succession of excitations the secondary coil is

gradually brought nearer to the primary, it is observed that no effect whatever follows the passage of the induction current until the secondary coil is approximated to a relatively short distance, usually about 10 centims. from the primary. (2) That the excitatory effect produced at this distance is unmodified by further approximation. (3) That the distance of the secondary coil at which the first response takes place, when in a succession of excitations it is gradually approximated, is much greater when the induction current is directed from the upper to the under surface than in the contrary case; in other words, that the leaf responds to a weaker current when it is directed downwards than when it is directed upwards; and, finally (4) That if one current of an intensity a little less than sufficient to evoke a response be followed at a short interval of time by a second current of similar intensity, the latter may be effectual. This last fact will be reverted to further on, under the head of "Summation."

*Excitatory influence of the voltaic current.*

For the purpose of studying the influence of a weak voltaic current, a rheochord of platinum wire was used, of which the length was 14 feet and the resistance 20 ohms; this was interpolated in the circuit of a DANIELL cell. Currents could be derived from it and conducted through the leaf by connecting one electrode with the end of the rheochord, the other with any point of its length by a slider. In this case the derived current might, in consequence of the relatively high resistance of the plant, be regarded as proportional to the length of wire between the end block and the slider. By comparing this rheochord with the compensator it was found that each foot of wire corresponded to a tension difference of about 0.06 D. and hence the whole wire to about 0.84 D. With this contrivance the following observations were made. In the first series the leaf was led off to the galvanometer as in the fundamental experiment, but a switch (a POHL'S Wippe, without cross wires) was introduced into the circuit, of such construction that for one or more seconds a derived current from the rheochord could be sent through the electrodes, the galvanometer circuit being for the same period broken. As care was taken to compensate immediately before the switch was thrown over, this produced no movement of the needle, but served to show in what way the passage of the current modified the electromotive properties of the leaf. The effects observed were as follows:—(1) When a very weak current was used (1 foot of wire, corresponding to a difference of potential of 0.06 D.) a slight effect, resembling polarization, was observed—*i.e.*, a transitory deflection indicating the existence in the leaf of a current opposed to the current which had been led through it, its direction being upwards when the anode was on the upper surface, and *vice versa*. It was ascertained that this was not due to polarization of the surfaces of contact of zinc with zinc sulphate solution, for when the experiment was repeated with the clay points in contact with each other, *i.e.*, without the interposition of the leaf, no

deflection was observed. (2) When the derived current was doubled (2 feet of wire) the after effect was always positive, whatever was the direction of the current. In other words, the effect showed itself in either case as an augmentation of the previous difference. Consequently the downwards current left behind it as its after effect, a current in the same direction as itself. By increasing the current, this effect, whether its direction is upwards or downwards, could be increased in about the same proportion. When the experiment was made in a leaf of which the opposite lobe was led off to the electrometer, no effect was observed during or after the passage of the current, showing that the effect was not propagated, that is, was not an excitatory effect and did not cause one. When similar currents were led in a similar way through the leaf stalk by electrodes on opposite surfaces, a polarization took place, but no effect resembling the one described was observable. (3) When (1) and (2) were alternated, that is when the current was passed through the leaf a number of times in succession in the same direction, but alternately with different strengths of current (1 foot and 2 feet of wire), the effect was, of course, alternately positive (with the stronger current) and negative (with the weaker). In both cases the effect was evanescent, and the previous difference was unaffected. (4) When the length of wire was increased to about 5 feet (a current five times as great as in (1)), different lengths, however, being required in different leaves, the effect was that of an excitation, that is, there was a negative deflection which was cut short by a larger one in the opposite direction. When this was compared with that of a mechanical excitation of one of the hairs of the opposite lobe the two variations, although resembling each other, were not identical. For if in a succession of observations a momentary (tenth of a second) closure of a downward current of sufficient strength to excite the leaf was alternated with mechanical excitation of a hair of the opposite lobe so as to compare the effects, it was found that in the variation which followed the electrical excitation, the first phase was smaller, the second phase larger than in those produced mechanically.

	Mechanical.		Electrical.
	Previous difference.	Variation.	Variation.
1st series . . . .	+0.0003	- 36 +192	- 3 + off scale ( <i>i.e.</i> , over 600)
2nd series . . . .	+0.0028	- 67 +132	-10 + ditto
3rd series . . . .	+0.0035	-102 +110	-11 +500

Each series consisted of six mechanical and as many electrical excitations. The mean results of each series are given in the columns headed "Variation."

(5) The excitatory effect produced in (4) was followed by an "after effect" which subsided very gradually in the ordinary way, leaving behind it a lasting increase of the previous cross difference, and when a leaf was subjected to successive excitations, the

variation gradually changed its character just as it does when mechanical excitations follow each other at short intervals. In a leaf of which the cross difference was, at the beginning of the experiment, abnormal, and which was excited 58 times in succession in half an hour, the variation being observed alternately with the electrometer and galvanometer, the cross difference changed sign from  $-0.009$  D. to  $+0.005$  D. The mean difference and variation were in three successive periods as follows :—

	Cross difference.	Galvanometer.	Electrometer.
1st period . . . .	$-0.0023$ D.	$-40.8$ +137	$-29.8$ +1.8
2nd period . . . .	0	$-51.0$ 0	$-23.2$ 0
3rd period . . . .	$+0.0036$ D.	$-61.4$ 0	$-21.0$ 0

In another similar experiment the cross difference changed, in a series of 32 excitations, from  $-0.011$  D. to  $+0.01$  D., the second phase (positive deflection) diminished from  $+210$  scale to  $+90$  scale, while the first phase (negative) increased from  $-15$  to  $-220$ . (6) When (3) was repeated with the difference that the lengths of wire were respectively 3 feet and 5 feet, instead of 1 foot and 2 feet in two series *a* and *b*, so that the currents conducted through the leaf were, in series *a*, only sufficiently strong to produce the effect (2), viz. : a single positive deflection, whereas in series *b* they were sufficient to give rise to an excitation, it was observed that in series *a* the electrical relation between the upper and under surface remained unaltered, but that in series *b* the diphasic variation was followed by an after effect which subsided with the usual increase of the cross difference already described and exemplified.

The preceding facts may be summed up as follows :—The closing of a voltaic current directed from the upper to the under surface of the leaf of *Dionæa* produces different effects according to its strength. Very weak currents give rise to feeble polarization. Currents of moderate strength produce a transitory increase of the cross difference, that is, make the under surface more positive to the upper surface than it was before. Strong currents produce this transitory increase of the cross difference, but, in addition, produce an excitation which is followed by a lasting increase of the cross difference. In the successive deflections observed, these two effects are summed ; so that the negative phase is diminished and the positive phase increased.

The explanation of these phenomena depends on the meaning to be assigned to the remarkable modification of the electrical state of the leaf produced by moderate currents. On this point we can only express ourselves negatively. It is not analogous to an excitatory effect, for it is not capable of propagation : it is not identical with the after effect for it is transitory : it is not a polarization effect, for its direction is always the same as that of the cross difference whatever the direction of the current which produces it. This fact also makes it impossible to identify it with KUNKEL's currents. The utmost that can be stated with reference to it, is, that it indicates the occurrence of a



local change in the path of the current, which is probably of the same nature as that which, when stronger currents are used, excites a propagable explosion.

By the experiments already related it has been shown that when a leaf is traversed by a voltaic current in a direction vertical to its surface for a short period (one-tenth of a second to a second) from the upper to the under surface, excitation follows as soon as the difference between the two electrodes applied to the two surfaces of a leaf at opposite points, amounts to something like half the electromotive force of a DANIELL cell. But they afford no information either as to the time or place at which the excitation occurs. It remains to be determined whether it occurs at the closure or opening of the circuit, and whether at the anode or cathode. The first of these questions was easily decided in experiments in which the leaf was led off as in the fundamental experiment and excited by currents for periods of sufficient length to render it possible to distinguish easily between the effects of opening and closing. The method consisted in making a series of observations in which the current was alternately directed upwards and downwards, and lasted from five to seven seconds, the opposite lobe being led off to the electrometer. A succession of experiments were made on different leaves in which various strengths were used, from one DANIELL, without rheochord, to two GROVES, always with the same result, namely, that the leaf responded at make, and at make only, whatever was the strength of the current or its direction. When strong currents were used, as will be noticed further on, the excitation at the closure of the circuit was succeeded by another from three to six seconds later, and might be easily mistaken for a break effect, if the moment at which it occurred happened to follow that at which the current was opened.

To the question whether excitation proceeds from the anode or cathode, we have failed entirely in our efforts to obtain an answer. The modes of experiment used were as follows :—The leaf having been led off to the electrometer as in the fundamental experiment, non-polarizable electrodes for excitation were applied on the surface of the opposite lobe and on the petiole. In some cases the lobe electrode was on the under surface, in others on the upper. A reverser was introduced into the exciting circuit, so that the current of two GROVES could be directed either towards the leaf or towards the leaf stalk. In either case the excitatory effect showed itself at make, and make only. But when the electrode was on the upper surface it was anodic, and when it was on the lower surface, cathodic. At first sight it seemed as if this indicated a difference of action between the poles, but on consideration it appeared to be another form of the result already recorded, viz. : that in the lamina the downwards current is more effectual than the upwards. A current *from* the leaf stalk which passes out at the under surface acts in the same way as a current *to* the leaf stalk which enters at the upper surface, because in the excitable part the direction of both is descending. The next attempt to settle the question was by a rheochord experiment, in which the leaf was excited by electrodes applied symmetrically to corresponding surfaces on the right and left lobes respectively, the one lobe (right or left as

the case might be) being led off to the galvanometer as in the fundamental experiment. The time of closure of the exciting circuit (two LECLANCHÉ'S) was one-twentieth of a second. The galvanometer circuit was also closed for the same period, an interval of 0.03" intervening between the opening of the one circuit and the closure of the other. In the first leaf experimented on, the results were remarkable. In a series of twelve observations in which the current was directed across the leaf, alternately from and to the led off lobe, the needle remained motionless when, as in the first case, the anode was next the leading off surface, but swung to the negative side from 12 to 14 scale when the cathode was in the same position. As repetition confirmed the result it was clear that, as regards the particular leaf under observation, the proximity of the cathode was favourable to the early occurrence of the excitatory effect. Doubt was, however, suggested by the consideration that here, when the cathode was on the left lobe, the direction of the current near the leading off electrodes was downwards, that is, favourable to the excitatory effect, whereas in the opposite lobe it was unfavourable, and this surmise we confirmed by the observation that in another leaf, in which an exactly similar experiment was made, with the exciting electrodes applied to the internal surface, the response was strongest when the anode was on the led off lobe. All that was learned from these attempts was, that by the method employed, which seemed to be the only one likely to yield an answer to the question, no answer could be obtained. The want of material made it impossible to proceed further, but even had it been forthcoming I should have been at a loss how to use it.

### *Influence of the Duration of the Current on the Effect.*

1. *Short periods.*—In order to determine the shortest period of closure of a voltaic current directed transversely from the upper to the under surface of a leaf, experiments were made in which the time of closure was gradually shortened by means of the pendulum rheotome. It was found that there was no diminution of effect until the duration of the closure was less than one-hundredth of a second. Beyond this, the response became uncertain when two DANIELL'S with a rheochord of 20 ohms were used, and disappeared when the closing time was reduced to 0.007. Want of material made it impossible to investigate the important question of the influence of strength of current on the minimum. It was, however, ascertained that it was necessary to lengthen it when weaker currents were used.

2. *Currents of long duration.*—When a voltaic current of five seconds' duration and of moderate strength is led through the leaf from the upper surface between the sensitive hairs to the under surface, there is a response at make, and no other. When stronger currents are used (from one DANIELL to two GROVES) the first variation is followed by a second. If the current is continued for a long period (30 seconds) a succession of excitatory variations takes place at irregular intervals, as shown in the following

examples. If the leaf is expanded and not restrained, it may close at the first excitatory variation, *i.e.*, one to one-and-a-half second after the closure of the current. But if the experiment is made at a low temperature ( $18^{\circ}$  to  $20^{\circ}$  C.) this event does not happen until after the third, fourth, or fifth variation. To observe these facts the current of a GROVE cell is led through one lobe, while the other is led off (in the usual way, to the electrometer, and the time after closure of the circuit noted by the watch, as in the following experiments:—

I. Current of one GROVE from above downwards for half a minute; excitatory variation at 3'', 9'', 13'', 19'', 23'', 30''.

Same current directed upwards—variation at closure only.

II. Current of one GROVE from above downwards for one minute; variation at 1'', 15'', 25'', 34'', 38'', 45''.

Same current directed upwards—variation at closure only.

III. Same leaf. Current directed through contacts previously used for leading off; leaf led off by lobe previously excited. Variation at 2'', 8'', 22'', 32''.

Same current in opposite direction—variation at closure only.

*Summation of stimuli.*—To prove summation it must be shown that two equal excitations, each of which alone is inadequate to evoke a response, do so by summation, that is, when they follow each other at a short interval. If this is so, there is evidence that the first, although it is apparently without effect, yet produces a change in the excitable structure which renders it more excitable than it was before. The best method consists in making two alternating series of excitations, in one of which single opening induction shocks, just inadequate to produce excitatory effect, are used, while in the other, two induction currents of equal strength follow each other at a variable interval.

1st Series. Distance of coil 7.6 centims. Temperature of chamber  $32^{\circ}$  C. Interval 0.02''. Ten excitations, of which Nos. 2, 4, 6, 8 were single, 1, 3, 5, 7, 9 were repeated. All of the latter were effectual; all of the former ineffectual.

2nd Series. Distance of coil 7.4 centims. Temperature  $32^{\circ}$  C. Interval 0.1''. Ten excitations; six single, five repeated ( $\frac{1}{10}$ ''). All of the latter were effectual; all but one of the former ineffectual.

3rd Series. Distance of coil 7.3 centims. Temperature  $32^{\circ}$  C. Interval 0.2''. Sixteen excitations, of which half single, the rest repeated ( $\frac{1}{5}$ ''). All of the latter were effectual; all except two of the former ineffectual.

4th Series. Another leaf. Distance of coil 8 centims. Temperature  $32^{\circ}$  C. Four excitations; two single, two repeated at intervals of  $\frac{1}{8}$ ''. Both of the latter effectual; the other ineffectual.

Four excitations; two single, two repeated at intervals of  $\frac{2}{8}$ ''. The latter effectual; the former ineffectual. In this series it could easily be seen that the excitatory effect followed the second excitation.

5th Series. Eighteen double excitations at intervals of half a second ; of these eight were fruitless. Of the ten responses, half occurred at the first excitation, the remainder at the second.

These experiments show that at all intervals between 0.02" and 0.4" summation occurs, and that at 0.5" the result becomes uncertain.

Another method may be used which consists in subjecting a leaf to repeated excitations by successive series of inadequate induction shocks at a fixed interval—*e.g.*, at a twentieth of a second, gradually increasing the number in each series until a response is obtained. In an experiment of this kind a leaf responded in a room at 20° C. when the number of opening shocks amounted to 23. In the chamber at 32° C. it responded after 7. It is to be noted that, in an experiment of this kind, the distance of the secondary coil from the primary must very slightly exceed that at which single shocks are responded to without fail. This method is useful as a means of testing excitability under different conditions.

## PART VI.

### RELATION BETWEEN THE EXCITATORY PROCESS AND THE MECHANICAL EFFECT.

The moment at which the mechanical effect begins can be easily measured graphically by the following method. A cork is prepared, with a hole across its axis for the reception of the round horizontal arm of a support resembling that shown in fig. 4 (electrodes). On this the cork can be rotated. The cork is cut so as to present a surface somewhat smaller than that of a lobe of the leaf. To this a leaf is cemented with plaster of Paris by the external surface of one lobe, leaving the midrib free. To the border of the opposite lobe a very light straw lever is fixed by the same means, in the position it would assume if it were a prolongation of the middle marginal hair. This having been done, the lever is made horizontal by rotating the cork which, with the leaf, is brought into such a position that its motions are inscribed on a blackened cylinder, of which the horizontal movement should be 1 centim. per second. The leaf is excited mechanically by a camel-hair pencil of which the motion breaks a circuit. The moment at which this happens is recorded on the same cylinder by a DEPRez' time marker. It is thus learnt that the mechanical response, at temperatures from 15° to 20° C., begins at an interval of from a minute-and-a-half to two minutes after excitation, so that in the fundamental experiment it coincides with the close of the first phase of the variation. If this observation is made at a low temperature, as *e.g.*, 15° C., and the hairs are touched with great care, the first excitation produces no perceptible motion. At the fifth or sixth the lever slowly rises and continues to do so for several seconds (see Proceedings 1876, p. 415). To record this effect, the best plan is to leave the lever free and record its position at the end of every half second on a smoked glass plate. The result is shown in the figure. It is seen that in each case the rate of ascent of the lever is greatest during the first half

second, after which it gradually declines, and that the total effect consequent on each excitation is very variable, being dependent on accidental differences in the way in which the sensitive hair is touched.

*The mirror experiment.*—The effect of series of frequently repeated mechanical excitations on leaves at relatively low temperatures may be observed with greater accuracy by another method. The leaf is attached to the rotating cork in exactly the same way as in the experiment last described, but instead of a lever a very light glass mirror, such as is used for reflecting galvanometers, is cemented to the external surface of the free lobe, near the margin. By means of this mirror the image of a horizontal slit is thrown on a vertical scale which is so graduated that the angular movement of the lobe can be measured with great delicacy. An experiment of this kind gives the following results. The leaf having been led off to the electrometer by electrodes applied to the upper surface of the attached lobe and to the midrib, was subjected to 22 excitations, each of which consisted in touching very gently one of the hairs of the attached lobe. The total angular movement (rotation) of the mirror was 167°. This was accomplished in 20 excitations, all of which were effectual in the sense that each produced a normal variation.

Number of excitation.	Effect.	Sum of effects.
1	°0	°0
2	°0	°0
3	0·5	0·5
4	0·4	0·9
5	0·4	1·3
6	0·8	2·1
7	1·0	3·1
8	4·1	4·5
9	3·5	8·0
10	4·0	12·0
11	5·5	17·5
12	7·5	25·0
13	13·0	38·0
14	15·0	53·0
15	42·0	95·0
16	34·0	129·0
17	10·0	139·0
18	11·0	150·0
19	13·0	163·0
20	4·0	167·0

The fact that in this and in other similar experiments it is possible to excite the leaf once, twice, or a greater number of times without any appreciable movement of the image, at first sight seems to indicate that the electrical effect is independent of the mechanical. But before accepting such an inference it must be remembered that we cannot be at all sure that the interstitial movement of liquid, which in the leaf as in all the moving organs of plants is the efficient cause of change of form, may not begin without making itself visible by any change in the curvature of the lobe, how-

ever delicate our means of observation may be. It appears to me probable that every effectual excitation—every excitation which is marked by electrical disturbance—is also followed by diminution of the water-charge of the excited protoplasm. At first, this may either expend itself entirely or produce only such small effects as, in the experiment last related, presented themselves in excitations 3 to 7.

In our previous paper the phenomena described above were referred to as evidences of summation. It will be better in future to limit the application of this word to the still more remarkable phenomenon which has been described under the heading “Summation” in Part V., p. 47; for those phenomena are strictly analogous to the effects to which the term “Summation of stimuli” has been applied by STIRLING.\* The term “Summation” is in itself as applicable to the process now under consideration as to the other, but it differs from it in this respect—that it is rather a summation of effects than a summation of excitations. The process with which it is most comparable in the physiology of animal excitable tissues is that which is known as the *Treppe* or staircase.† By this term is designated the fact that when the ventricle of the heart of the frog is subjected, after a period of rest, to a series of effectual excitations, the contractions become stronger and stronger, although the excitations are all of equal intensity. In both processes the mechanical effect of the first excitation is much smaller than that of the second, the second smaller than that of the third, and so on, but in the *Treppe* the difference which is large at first rapidly diminishes, whereas in the leaf it as rapidly increases. Further, the total mechanical effect of the first excitation is so small as to be barely perceptible, whereas in the heart the first contraction (*i.e.*, the weakest) is at least half as vigorous as the strongest. But in proceeding with our comparison we find that in both, the change of form is opposed by a resistance which has its seat in the structure of the organ, and is removed step by step when it is subjected to repeated excitations. In the heart this resistance diminishes rapidly at first, more slowly afterwards: in the lamina of the leaf it does not begin to yield until after several excitations, provided that they are only just strong enough to provoke an electrical response. This difference is unimportant, and becomes almost imperceptible if somewhat stronger excitations are substituted for the minimal ones. In this case the effect of the very first excitation may be large, that of the second larger, the third still larger, and so on, but with decreasing, not increasing increments, just as in the *Treppe*. The most fundamental difference is that which concerns the relation between the resistance and the mechanism of contraction in the two cases. In the ventricle only a fraction of the mechanical effect, even of the first contraction, is due to abolition of resistance. At each repetition of the excitation this fraction diminishes, until after a dozen or so of excitations the successive mechanical effects become sensibly equal. From the first, therefore, the change of form is due

\* STIRLING, “Ueber die Summation elektrischer Hautreize.” LUDWIG'S Arbeiten, 1874, p. 223.

† See BOWDITCH, “Ueber die Reizbarkeit der Muskelfasern des Herzens.” LUDWIG'S Arbeiten, 1871, p. 156.

to something more than mere diminution of resistance. In the leaf the removal of resistance is everything. Consequently, so soon as a sufficient number of excitations have occurred to accomplish this, it is not only incapable of resuming its original form, but of any further response to excitation until, after many hours, the resistance is restored.

If we go a step further and enquire what this resistance consists of, we are led, in accordance with the conclusion that has been arrived at by every physiologist who has investigated the mechanism of the changes of form of the excitable organs of plants, to identify it with turgescence. By turgescence we understand the power which living protoplasm possesses of retaining water. In the case of cells which are excitable, the immediate effect of excitation is suddenly to diminish this power, and thereby to produce a diminution of volume of the cells which is equal to that of the water (probably holding diffusible bodies in solution) which is discharged into the intercellular spaces.

#### ADDENDUM.

Received November 3, 1881.

*[The following is a Summary of the most important Experimental Results embodied in Parts III., IV., and V.]*

1. When different parts of the surface of the uninjured and unexcited leaf of *Dionæa* are compared by the method of compensation described in Part II., electrical differences present themselves even when the points selected are symmetrical. In this case the differences observed are accidental. They are due either to surface conditions which disappear when the leaf, with its leading off electrodes in position, is allowed to remain in saturated air (differences of turgidity of the surface layers of cells) or to accidental physiological differences between the two lobes, which do not disappear.

2. Similarly, when unsymmetrical points of either surface of the leaf are led off, the differences observed are for the most part accidental. In one case, however—that of the comparison of the middle of the under surface of either lobe with the under surface of the petiole—the negativity of the former to the latter is a result which occurs so constantly that it must be assumed to have a physiological meaning.

3. If any two points opposite to each other on the upper and under surface of either lobe are compared, it is usually found that the upper surface is negative to the lower. Whether this is so or not in the unexcited leaf, it becomes so after one or more excitations; in other words, the effect of mechanical or electrical excitation, whether applied in the neighbourhood of the surfaces compared or at a distance from them, is to produce a lasting change in their electrical relation of such a nature that on repeating the comparison of the two surfaces the *under surface* is always found to have become more positive (less negative) than it was before excitation.

4. If the same observation is made while the leaf is subjected to mechanical or electrical excitation, it is seen that at the moment that excitation takes place the under surface becomes suddenly negative to the upper. By the rheotome we learn that the change last mentioned does not begin until  $\frac{1}{20}$ th of a second after its cause, that it culminates about the middle of the first second, the maximum difference amounting to as much as 0.08 DANIELL, and then gradually subsides.

5. It is further learnt by the same mode of observation that in the course of the second second or towards its close, the electrical relation of the surfaces to each other is reversed, the under surface becoming positive to the upper. The difference of potential in this direction having rapidly augmented until it amounts to about 0.02 DANIELL, very slowly subsides into the lasting effect of excitation described in 1.

6. When two symmetrical points on the under surfaces of opposite lobes are similarly compared during excitation, the excitatory effect differs in character according to the relation between the seat of excitation and the leading off contacts, the surface of the lobe which is directly excited becoming negative to the other, consequently by changing the positive seat of excitation the direction of the first effect may be reversed.\*

7. When a voltaic current is led across the lobe from the upper to the under surface by electrodes applied opposite to each other near the sensitive hairs, but not touching them, at the same time that the electrical state of the opposite lobe is observed as in 4, a response occurs at the moment that the current is closed, provided that its strength is such that the available electromotive force amounts to about half that of a DANIELL cell and that the temperature is not below 30° C. No response occurs at opening the current. If a stronger current (two DANIELLS) is used and the direction is downwards, the response at closing the current is followed by several others. This effect does not happen when the current is directed upwards.

8. A voltaic current directed from the upper to the under surface, which is too weak to evoke an excitatory response, produces an increase of the positivity of the under surface, limited to the part of the lobe through which the current passes, which lasts several seconds after the current is broken. Its direction shows that it is not due to "polarization."

9. Voltaic currents of less than  $\frac{1}{200}$ th second duration, though of moderate strength, do not excite *Dionæa*. Weaker currents cease to act when their duration is less than  $\frac{1}{100}$ th second. But the relation between strength and duration has not been ascertained.

10. Opening induction currents such as are yielded by DU BOIS-REYMOND's smaller inductorium when the secondary coil is at a distance of 8 to 10 centims. from the

\* It cannot be concluded either from the result stated above or from the preceding ones that in *Dionæa*, as in muscle and nerve, the excited part becomes *negative* to other parts. For it is as consistent with the facts observed to say that the upper surface becomes positive as the immediate result of excitation, as to say that the under surface becomes negative.



primary, may for many purposes be used more advantageously than any other mode of excitation, for by means of them a leaf may be subjected to repeated excitations for several hours without failure or appreciable exhaustion.

The minimum effectual strength of an opening induction current depends on its direction, weaker currents being responded to when they are directed downwards than in the contrary case. Induction currents which are inadequate to evoke a response are yet followed by a local change in the electrical state of the surfaces through which they are led which resembles that produced by weak voltaic currents.

11. *Summation of stimuli*.—Two inadequate opening induction currents which follow one another at any interval greater than  $\frac{2}{10}$ "', or less than  $\frac{4}{10}$ ths of a second, may evoke a response. In this case the response follows the second excitation. When a leaf is subjected to series of inadequate induction currents at short intervals (*e.g.*,  $\frac{1}{20}$ th of a second) the response may occur after a greater or less number of excitations, according to the temperature at which the experiment is made and the strength of the current.

12. *Summation of effects*.—In a series of mechanical excitations each of which is just adequate to produce an electrical response, those which occur earliest are followed by no visible change of form. Of the later members of the series each produces a measurable movement, the extent of which becomes greater each time that the excitation is repeated until eventually the leaf closes.]

#### CONCLUSION.

In the preceding paragraphs all reference has been omitted to the physiological meaning of the electromotive phenomena which have been described. The moment has now arrived at which it appears necessary to offer such an explanation as the knowledge acquired justifies, as to their relation to the vital processes with which they are associated.

According to Professor MUNK the electromotive properties of the leaf of *Dionæa* may be accounted for on the supposition that in each cell the ends of the cell are, in the resting state, positive to the middle, and that in excitation the difference of potential between the ends (poles) of the cell and its middle (equator) undergoes a sudden diminution. This theory must be rejected at once, on the ground that it fails to explain the fundamental experiment. The electromotive forces it supposes to be in operation act in directions parallel to the surface of the leaf—at right angles, therefore, to the path of the currents which show themselves when the two opposite surfaces of the leaf are led off to the galvanometer. Under these conditions it is impossible that the latter can be the expression of the former.

On the other hand, I accept as fundamental the doctrine that whatever physiological properties the leaf possesses, it possesses by virtue of its being a system of living cells. The first question, therefore, to be determined is that of the electromotive endowments of the individual cell.

Each of the two lobes of the leaf of *Dionæa* is made up, for the most part, of cylindrical cells. The long axes of these cells (parenchyma cells) are all parallel to each other, and to the vascular bundles which run out from the midrib towards the margin. A transverse section of the leaf, made in such a way as to include a vascular bundle, shows that there are two or three layers of them, the most superficial of which are covered by the epidermis of the under and upper surfaces of the leaf. As they are alike in structure, it may be assumed that they have the same or similar functions.

If it were possible, without injury, to investigate the electromotive properties of one of these cells individually, by leading off its opposite surfaces, we should probably find them to be isoelectrical both at rest and when excited; for the electrical differences which exist in the cell are probably between the living inside and the non-living outside, and not between different parts of the surface. Consequently the electrical differences which show themselves between the opposite sides of the leaf blade which is made up of layers of such cells cannot be due to the summation of smaller differences between the two sides of the individual layer, for these are isoelectrical. They must, therefore, be referred to the contact of different layers of cells with each other. But if electrical differences exist at the surfaces of contact of the cells, they must be associated with physical differences of other kinds. With respect to the nature of this difference it appears to be very probable that the most important element is migration of water. For on the one hand we know that in consequence of the surface evaporation, migration of water certainly exists, while on the other we have proof in the experiments of Dr. KUNKEL, that such migration cannot occur without producing electrical differences.

As has been so conclusively proved by the experiments of PFEFFER, those motions of the organs of plants which follow excitation are attended by diminution of the turgor or water-charge of the protoplasm of the excitable cells. The displacement of water occasioned by this discharge must unquestionably be attended by electrical change. In *Dionæa* the mechanical effect of an excitation can be seen from one to two minutes after the moment at which the excitation takes place. It is at the same moment that the second phase, or after effect, begins. In their origin, culmination, progress, and duration the two changes are closely associated. It therefore appears extremely probable that the two are causally related—that the positivity of the external surface is an expression of the electrical differences which exist between contiguous strata in different degrees of turgor—the less charged being negative to the more.

The same theory affords us a complete and satisfactory explanation of the gradual change of potential which always results from repeated excitation, as well as for the fact so constantly observed, that the “after effect” which follows each of the first few excitations to which a leaf is subjected, is much greater in extent than those which occur after later ones, and that eventually the after effect fails entirely, reappearing after a period of repose.

I need scarcely say that the first phase of the variation—the effect which immediately follows excitation and has an opposite sign to the after effect and a much higher electromotive force—does not admit of a similar explanation; for it cannot be imagined that a change which spreads over the whole lamina in less than one-twentieth of a second can be dependent on migration of water. The excitatory disturbance which immediately follows excitation is an explosive molecular change, which by the mode of its origin, the suddenness of its incidence, and the rapidity of its propagation is distinguished from every other phenomenon except the one with which I have identified it, namely, the corresponding process in the excitable tissues of animals.

Of the nature of this preliminary disturbance (to which alone the term excitatory variation ought to be applied, it alone being the analogue of the “action current” of animal physiology) we know nothing. Just as in the case of muscle and nerve, the proof that it is attended by any chemical alteration or by the separation of any product of disintegration is wanting, so it is here. In the one case as in the other, we must regard the electrical change as a visible sign of an unknown molecular process. That it is not the primary change which occurs in protoplasm when it enters into the state of excitation we have evidence in two facts, first, that even when the exciting agent is an induction current which passes through one of the opposite surfaces by which the leaf is led off to the galvanometer, so that no time whatever is lost in transmission, there is still an interval between excitation and response of about 0.03”, during which interval molecular changes are obviously in progress; and, secondly, that excitation just insufficient to evoke a response gives effect to a second which follows it, provided that the interval between the first and the second does not exceed one-third of a second—a result which can only be explained on the supposition that the first excitation actually effectuates a molecular change, notwithstanding that the visible sign is wanting.

The direction of the excitatory effect in the fundamental experiment is such as to indicate that in excitation, excited cells become positive to unexcited, whereas in animal tissues excited parts always become negative to unexcited. The apparent discrepancy will probably find its explanation in the difference of the structural relations of the electromotive surfaces.