PLATE 6.

E. Xanthophylls obtained from an extract of *Ficus Repens* in the month of May:—

(1) Chrysophyll obtained from the crude solution of the xanthophylls.
(2) Crude solution of the xanthophylls.
(3) A mixture of chrysophyll and the alcoholic portion.
(4 and 5) The alcoholic portion of different strengths, showing a slight obscuration.

F. Xanthophylls obtained from an extract of *Ficus Repens* in the month of December:—

(1) Crude solution of the xanthophylls; a case in which the fourth band is almost, if not, absent.
(2 and 3) The alcoholic portion of different strengths. This is the usual appearance of this spectrum, showing the bands more or less obscured.
(4) The alcoholic portion after standing a little time, the spectrum being the same as that produced immediately by the action of HCl.

G. The action of HCl on the xanthophylls:—

(1) The first CS_2 fraction.
(2) The first CS_2 fraction + HCl.
(3) Crude solution of the xanthophylls.
(4) Crude solution of the xanthophylls + HCl.
(5) Alcoholic portion (F − 2) + HCl.

"On Skin Currents.—Part I. The Frog's Skin." By AUGUSTUS D. WALLER, M.D., F.R.S. Received May 29,—Read June 6, 1901.

The principal object of the following observations was to investigate in the case of skin an electrical reaction by which it is in general possible to determine whether an animal or vegetable tissue is alive or dead.*

A side issue raised in connection with the general inquiry was whether or no the test is applicable to the human body; this obviously led to a detailed study of skin effects upon man and upon animals.

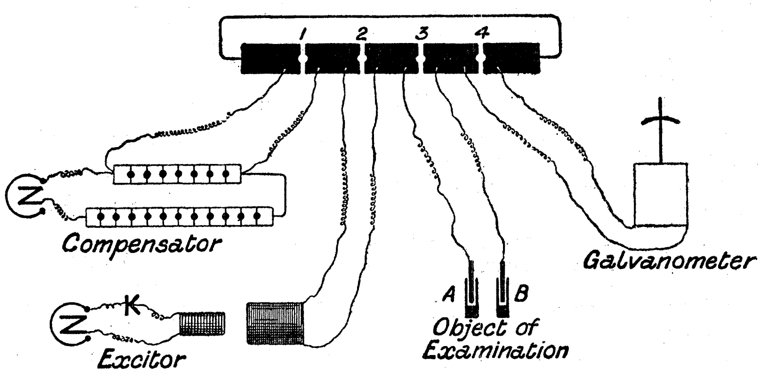
In the case of the frog, previous observations on skin currents are numerous and conflicting; but in so far as my present theme is concerned, the results have come out with the utmost regularity and quite clear of any suspicion of physical fallacy. In the case of man, the question has proved to be less simple, and although it is easy to distinguish between an assuredly living and an assuredly dead piece of skin, it is far from easy in doubtful cases to make sure that the skin is completely dead. The difficulty is caused by polarisation currents

* 'Roy. Soc. Proc.,' vol. 68, p. 79. References to previous papers are given there—p. 92.

with or against a reaction of low E.M.F., and is not eluded as easily as might have been anticipated by the use of alternating currents. Thus, *e.g.*, while it is easy to assure oneself that a healthy skin may survive for at least a week, one may not feel assured that it is absolutely dead at that time; and in the case of skin obtained from the post-mortem room 24 hours after death, while one may be quite sure that a given skin is still alive, one may not be so sure that another skin is completely dead.

For these reasons I have preferred in the present communication to describe only the very clear and easily demonstrated results of direct excitation of the frog's skin. And in connection with those clear and regular results, I take the opportunity of describing the more variable and debateable results of the indirect excitation of the same skin through nervous channels.

METHOD.—The method by direct excitation is as has been previously described and figured in the case of a vegetable tissue,* a piece of frog's skin laid on a perforated glass or ebonite plate in place of the seed between the unpolarisable electrodes, which serve for the exciting



current and subsequently for the excited current. For the purposes of the description to follow, the skin is to be pictured as if with its superior or external surface A directed upwards, in which case a current from the internal surface B to the external surface A, or an "outgoing" current is ascending or positive, and an "ingoing" current from A to B descending or negative. Excitation was made by single induction shocks, by series of alternating induction shocks, and by condenser discharges. The direction of exciting currents was always determined, the effects of polarisation were tested for, the electrodes in particular being always examined for polarisation, "anomalous" or positive, as well as ordinary or negative.

* A. D. W., *loc. cit.*, p. 82, fig. 1.

To obtain the effects of indirect excitation two kinds of nerve-skin preparation were used—(1) That of Roeber* and of Engelmann,† consisting of the sciatic nerve, knee, and skin of leg; (2) that of Hermann,‡ consisting of spinal column and skin of back.

In the case of indirect excitation, the response was observed during and after excitation. In the case of direct excitation, the accidental skin-current was exactly compensated, and the skin was excited while the galvanometer was short-circuited; the galvanometer was put into circuit between 1 and 2 seconds after excitation.

RESULTS.—1. The normal current is negative (ingoing). It regularly increases during the first 15 to 30 minutes after the skin is put upon the electrodes. The ordinary value of its E.M.F. is from 0·01 to 0·10 volt, *e.g.*—

Time.	Voltage of current.
0 min.	– 0·0010
10 ,,	– 0·0080
20 ,,	– 0·0265
30 ,,	– 0·0330

A lively skin gives greater current than a poor skin. Nevertheless, the former may, at the outset, exhibit a small current by reason of a positive (outgoing) effect due to manipulation. The latter gradually subsides, and negative current therefore gradually augments.

2. The normal response to direct excitation is positive (outgoing). The excitation may be mechanical or electrical, by a condenser discharge or by an induction shock, in a positive or in a negative direction.

The response is greater and smaller with stronger and weaker excitation. The initial positive frequently gives place to a subsequent negative phase, or a positive interrupted by a negative phase may be witnessed. In such cases comparatively weak excitations were used. With strong excitation the positive response is very persistent, and there is a marked "deflection-remainder."

The positive response to negative excitation generally exceeds the positive response to positive excitation.

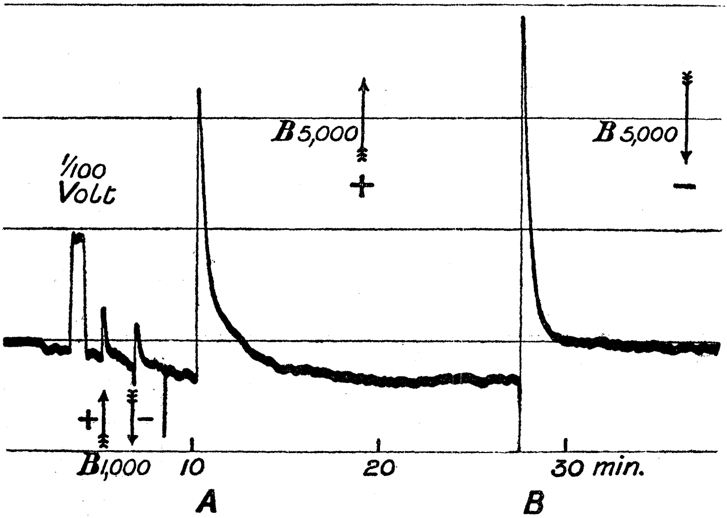
Tetanising currents of alternated direction give positive response.

The response to a single break shock exceeds that to the corresponding make shock with the ordinary arrangement of an induction coil.

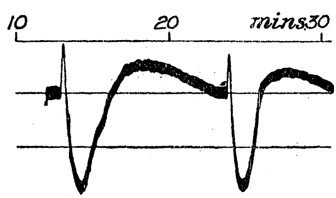
The response exhibits the phenomena of summation and of fatigue.

It is abolished at temperatures above +45° or below –6° and by mercuric chloride.

* Roeber, 'Du Bois-Reymond's Archiv,' p. 635. 1869.
† Engelmann, 'Pflüger's Archiv,' vol. 6, p. 127. 1872.
‡ Hermann, 'Pflüger's Archiv,' vol. 17, p. 292. 1878.



A, outgoing response to outgoing excitation (outer surface kathodic); B, outgoing response to ingoing excitation (outer surface anodic). The outgoing response, B, is preceded by a brief ingoing effect, homodrome with the exciting current. The excitation is by single break induction currents, 1000 +, 1000 −, 5000 +, 5000 −.



Polyphasic effect of direct excitation. Out—in—out. Response of this type is infrequent. The usual effect (or after-effect) is a strong or predominant outgoing effect, as shown in preceding figure.

Influence of Raised Temperature upon Direct Response of Frog's Skin.

Time.	Temp.	Tetan. 1000 +.	Tetan. 1000 −.
0 mins.	20°	+ 0·0200	+ 0·0230
40 ,,	30°	+ 0·0260	+ 0·0230
50 ,,	40°	+ 0·0020	+ 0·0005
55 ,,	45°	+ trace	− trace

Influence of Lowered Temperature upon Direct Response of Frog's Skin.

Time.	Temp.	Normal current.	Tetan. 1000 +.	Tetan. 1000 −.
0 mins.	18°	−0·03	+0·0142	+0·0083
	10°		+0·0125	+0·0085
	0°	−0·01	+0·0075	+0·0085
30 „	−2°		+0·0042	+0·0060
	−4°		+0·0028	+0·0035
	−6°		[spontaneous	+0·0035]
60 „	−6°	−0·00	+0·0015	−0·0010*
	−6°		−0·0005	+0·0005

* To single break shocks 10,000 + and 10,000 −. There was no response to 1000 + and −.

Note.—At −6° there was a sudden positive deflection, of electromotive source and not due to any sudden alteration of resistance, presumably indicative of excitation at the instant of congelation.

The signs + and − as regards tetanisation by alternating induction currents, refer to the direction of the break shock. Thus 1000 + signifies 1000 units of Berne scale, break shock outgoing (and make shock ingoing) through the skin.

With a dead skin, the deflections due to polarisation are in the direction of the break current, presumably by reason of superior polarisation by makes over breaks. (*Cf.* 'Proc. Physiol. Soc.,' November 12, 1898.)

With skin in this state, strong single shocks give rise to the ordinary polarisation counter currents.

Some Data regarding Magnitudes of Effects of Direct Electrical Excitation of Frog's Skin.

(Interval between Excitation and Galvanometer Closure = 2 Secs.)

Excitation.		Response.
1. Break induction current....	100 +	+ 0·0050 volt
	100 −	+ 0·0010 ,,
	1000 +	+ 0·0550 ,,
	1000 −	+ 0·0850 ,,
	10,000 +	+ 0·0700 ,,
	10,000 −	+ 0·0900 ,,
2. Break ,, ,, ...	1000 +	+ 0·0330 ,,
	1000 −	+ 0·0420 ,,
	5000 +	+ 0·0260 ,,
	5000 −	+ 0·0320 ,,
3. Make ,, ,, ...	1000 +	+ 0·0045 ,,
	1000 −	+ 0·0015 ,,
Break ,, ,, ...	1000 +	+ 0·0140 ,,
	1000 −	+ 0·0160 ,,
4. Make ,, ,, ...	500 +	nil
	500 −	nil
Break ,, ,, ...	500 +	+ 0·0085 ,,
	500 −	+ 0·0135 ,,
Make ,, ,, ...	1000 +	+ 0·0150 ,,
	1000 −	+ 0·0065 ,,
Break ,, ,, ...	1000 +	+ 0·0370 ,,
	1000 −	+ 0·0500 ,,
5. Condenser discharge	8 volts 1 mF. + (= 640 ergs)	+ 0·0100 ,,
	,, ,, −	+ 0·0100 ,,
	8 volts 0·1 mF. + (= 64 ergs)	+ 0·0015 ,,
	,, ,, − ,,	+ 0·0008 ,,

N.B.—The + sign signifies outgoing direction, the − sign ingoing direction.

3. The electrical response to indirect excitation of the nerve of a nerve-skin preparation is of three types—

I. Positive or outgoing.

II. Mixed { (*a*.) Positive interrupted by negative.
(*b*.) Negative followed by positive. }

III. Negative or ingoing.

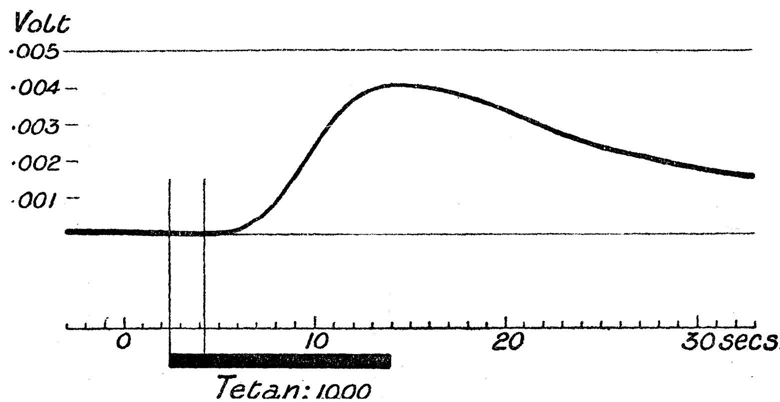
I have in no one instance witnessed the three types upon the same preparation, and may not therefore definitely say that they form three progressive stages. Nevertheless, I regard a positive response of type I as being the most normal, it having presented itself with the best preparations; and type III as the most enduring, it having exhibited least decline in consequence of repeated stimulation. I

have seen a response, positive at first, give place to a negative effect; and in the case of a mixed response of type II, I have seen a decreasing positive phase with an increasing negative phase. The entire series of responses is strongly suggestive of the theory that each effect is an algebraic sum of two opposite effects.

The positive effect by indirect excitation through nerve is less enduring than the negative effect. A second is always much smaller than a first positive effect.

Skin giving a mixed or a negative effect by indirect excitation has nearly always given a pure positive effect in response to direct excitation of whatever direction.

4. The interval of time between excitation of nerve and electrical response of skin is about 2 seconds.



Electrical response of skin of frog's leg to tetanic excitation of the sciatic nerve. (N.B.—The response is ingoing, *i.e.*, "Hermann's variation.")

5. The electrical conductivity of the skin is greatly augmented by direct excitation. This point is not in itself very remarkable since the alteration might be simply due to electrolysis. But the physiological origin of the change is indicated by the fact that dead skin similarly excited exhibits little or no change and by the fact that

6. The electrical conductivity of skin is greatly augmented by indirect excitation through nerve.

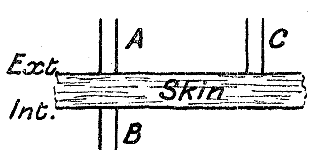
Influence of Excitation of Nerve upon Electrical Resistance of the Skin.

Exp.	Resistance before excitation.	Resistance after excitation.
1	2500 ohms	1000 ohms
2	2800 „	1400 „
3	2500 „	1500 „
4	4300 „	2400 „
5	3000 „	3000 „
6	4000 „	1300 „
7	3900 „	1200 „

Note.—In all except the 5th experiment, excitation of the nerve gave a large positive response. In the 5th experiment, there was no response and no diminution of resistance.

7. Atropine injected into the dorsal lymph sac has not in my hands abolished the electrical response of the skin produced by excitation of nerve. But by direct application to the skin the effect of such excitation has been promptly abolished. There has at such time been no perceptible alteration of the positive response to direct excitation in either direction. Such direct positive response has been promptly abolished by pencilling the external surface of the skin with a solution of mercuric chloride. In several instances the skin, before ceasing to respond altogether, has manifested a small negative response to both directions of excitation. The reaction is rapidly abolished by $HgCl_2$ solution of decimolecular strength, more gradually but completely abolished by $HgCl_2 \frac{M}{100} = (2{\cdot}7 \text{ per } 1000)$. Prolonged ($\frac{1}{2}$ hour) soakage of the skin in a freshly made 1 per cent. solution of atropine sulphate has produced diminution of the direct response—not much more marked, however, than may sometimes be observed after soakage in normal saline.

8. The electrical response of the skin to direct electrical excitation is at or near its external surface. This fact is indicated by the result of pencilling with mercuric chloride solution, and conclusively demonstrated by the following experiments:—

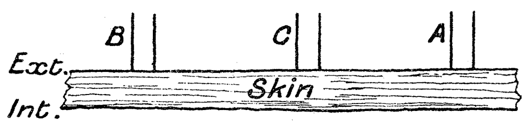


Excitation of the skin through A and B, subsequent lead-off to galvanometer A and C. A large after-effect is witnessed, from A to C

through the galvanometer, whatever had been the direction of the exciting current—*i.e.*, with A previously anodic or previously kathodic. On repeating the experiment, with lead-off through B and C there is little or no effect. The results are independent of the position of C, which may be transferred to the lower surface without altering them. The inefficacious combination B C is at once rendered efficacious by transferring B to the upper surface. (It is of course understood that any accidental current between A and C and B and C is compensated before each excitation.)

The experiment may be further varied in several ways, of which the most obvious is that in which all three electrodes are external or internal.

With external exciting electrodes A and B and subsequent effects led off from A C and B C, the direction of deflections indicates current in the skin from C to A and from C to B, *i.e.*, outgoing in A and B respectively, for both directions of excitation A to B or B to A. With internal exciting electrodes and the same (moderately strong) excitation there is little or no effect between C and A, or C and B, or even A and B.



Conclusion.—The two facts that I consider to be of principal importance as regards the further study of skin-phenomena are—

1. That the normal current of the unexcited skin is ingoing.
2. That the normal response of the excited skin is outgoing.

The hypothesis or figment in accordance with which these facts may be understood, or at least remembered, can be expressed as follows:—In a passive mass of living (animal) matter acted upon by its environment, there must be greater chemical change at any external point of its surface than at any internal point of its mass, and therefore an ingoing current. In an active mass of living (animal) matter giving out energy to the environment, chemical change must be greater within the mass than at the surface, and therefore an outgoing current. In the passive state any point of the surface is electro-positive to any point of the interior; in the active state internal points become less electronegative or actually electropositive in relation to the external surface.

Bibliographical Note.

Normal Current, or Current of Rest.—Du Bois-Reymond,* in connection with his investigation of muscle currents, was the first to definitely

* 'Thierische Elektricität,' 1854–57, *passim.*

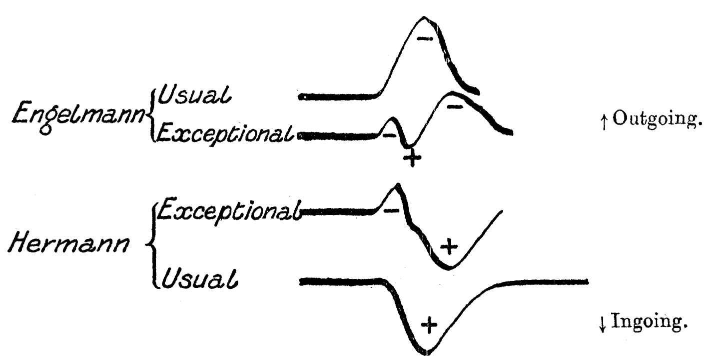
state that the normal current of the frog's skin is directed from without inwards. All subsequent observers have confirmed this point.

Indirect Excitation.—Roeber,* acting upon a suggestion of Rosenthal, was the first to make nerve-skin preparations of the sciatic nerve and skin of the leg, and to show that excitation of the sciatic nerve gave rise to an electromotive variation of the skin. He observed in the great majority of instances "a negative variation of the gland currents in consequence of non-electrical as well as of electrical excitation of the sciatic nerve." He mentions as an exceptional phenomenon, p. 644, a positive variation of the normal current.

Engelmann,† using the same method, comes to a similar conclusion, viz., that the usual effect of indirect excitation is negative variation of an ingoing current. He gives measurements of the effect (p. 130), from which may be gathered that a good response in his hands had the value 0·025 Daniell. The latent period is given as being from $\frac{1}{2}$ to 4 seconds. He describes the course of the variation as being very usually triphasic (– + –), which in the terminology used in the present communication reads + – +.

He considers that skin currents are "myogenic," the effects of the muscular investment of skin glands. He studies with particular care the influence upon the currents of variations in moisture of the skin, (imbibition and concentration currents).

Hermann‡ contradicts Engelmann's theory, and, to a certain extent his statement of fact as regards the action current. He gives the usual and principal effect as being a positive variation of the normal current.



He states, however, that such positive variation is sometimes preceded by a negative effect, and that, in rare cases, a pure positive effect is

* Du Bois-Reymond, 'Archiv,' 1869, p. 633.
† 'Pflüger's Archiv,' vol. 6, p. 97, 1872.
‡ 'Pflüger's Archiv,' vol. 17, p. 291, 1878.

observable. The opposition between Engelmann's and Hermann's statements is therefore not absolute enough to justify the statement that Engelmann's variation is negative and Hermann's positive. The difference of statement is one of degree only, Engelmann having been more prominently impressed by the outgoing phase, Hermann by the ingoing phase. Hermann considers that the chief (ingoing) phase is due to glandular activity, while the preliminary outgoing phase is due to a short circuiting, *viâ* gland ducts, of an epithelial current of action attributable to keratinisation.

Bach and Oehler,* under Hermann's guidance, observed that superficial cauterisation of the skin with saturated solution of $HgCl_2$ abolishes the normal current, and leaves the action current intact. Hermann's view is that normal current depends upon epithelial investment as well as upon glandular epithelium, whereas action currents through nerve stimulation depend upon glands.

Bayliss and Bradford,† employing Hermann's nerve-skin preparation, found Hermann's variation (ingoing) during January, Engelmann's variation (outgoing) during March. Their attention was particularly attracted during the last three months of the year to a triphasic character of variation − + − (or, according to the terminology of the present communication, + − +).

Direct Excitation of the Skin.—The first mention of definite direct excitation of the skin is to be found in Engelmann's paper of 1872.‡

Strong induction shocks were passed through the electrodes applied to opposite surfaces of the skin.

Compensation of its current was previously established, the galvanometer was cut out of circuit during excitation, and the effect upon the skin was observed immediately afterwards. The direction of excitation was not distinguished.

Biedermann§ approaches the question from the general standpoint of Hering's theory of opposite movements, dissimilation and assimilation, employs more particularly the frog's tongue, finds that during direct tetanisation (tongue and galvanometer in series) the response of the living tongue may be either positive or negative according to circumstances, the principal of these being temperature and moisture.

Bohlen,‖ under Biedermann's guidance, studied the gastric mucosa, *i.e.*, one epithelial layer in place of two, as in the case of the tongue, and obtained results confirmatory of Biedermann's.

Reid¶ and Reid and Tolput,** using the skin of the eel, found that

* 'Pflüger's Archiv, vol. 22, p. 30, 1880.

† 'Jour. of Physiology,' vol. 7, p, 217, 1886.

‡ 'Pflüger's Archiv,' vol. 6, p. 136.

§ 'Pflüger's Archiv,' vol. 54, p. 209, 1893.

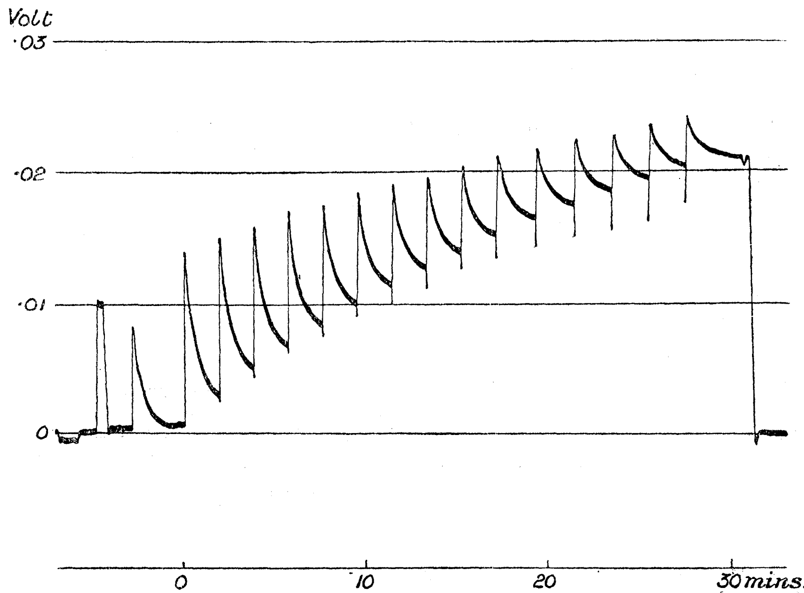
‖ 'Pfluger's Archiv,' vol. 57, p. 97, 1894.

¶ 'Phil. Trans.,' B, 1893, p. 359.

** 'Jour. of Physiology,' vol. 16, p. 217, 1894.

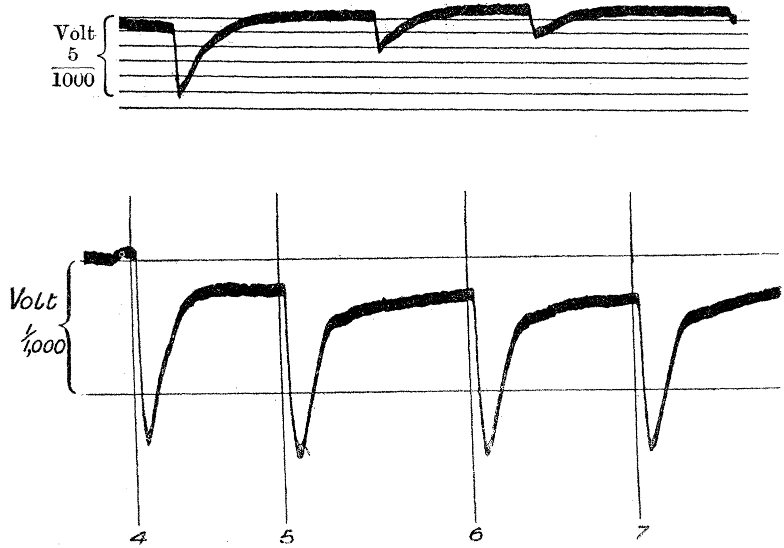
mechanical excitation and electrical excitation by induction shocks in either direction caused ingoing effects, occasionally preceded by outgoing effects.

Waller* finds that the normal and regular response of the frog's skin to any sort of disturbance—mechanical, chemical, or electrical—consists in a positive (outgoing) current.



Frog's skin. Summation of effects of direct excitation. Compensation is established at the outset of experiment, and left unaltered during its progress. The first deflection is that of 1/100th volt. The next is a trial deflection in response to a single break shock, 1000 +. The subsequent effects are by single break shocks, 2000 −, at 2 minute intervals. At each excitation the galvanometer is short-circuited for about 2 seconds, and the deflection therefore drops. The summating series of positive (outgoing) effects approximate towards a maximum of about 0·03 volt.

* 'Proc. Physiol. Soc.,' 1900.



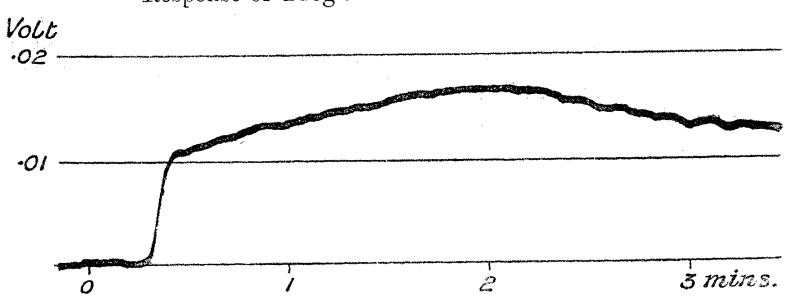
Frog atropinised by repeated injections into the dorsal lymph-sac of a 1 per cent. fresh solution of atropine sulphate.

1st nerve-skin preparation put up 2 hours later. Initial skin-current = – 0·0030 volt. Tetanisation of sciatic nerve by Berne coil at 1000 units for 15 seconds at intervals of 10 minutes. Series of ingoing effects.

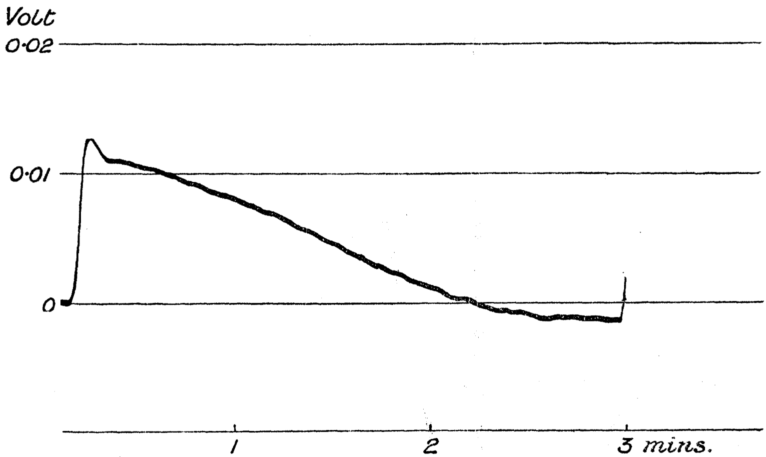
Time............	0	10	20	30	40	50	60 min.
Effects.	– 0·0040,	0·0020,	0·0015,	0·0013,	0·0011,	0·0011,	0·0010 volt.

In the first three responses of the upper line the galvanometer was shunted; in the next four responses of the lower line the galvanometer was unshunted.

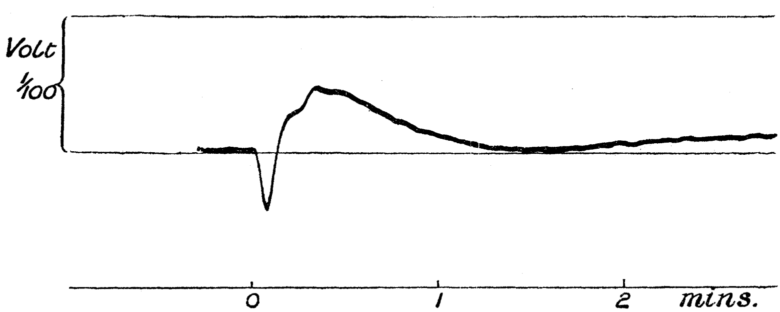
Response of Frog's Skin to Indirect Excitation.



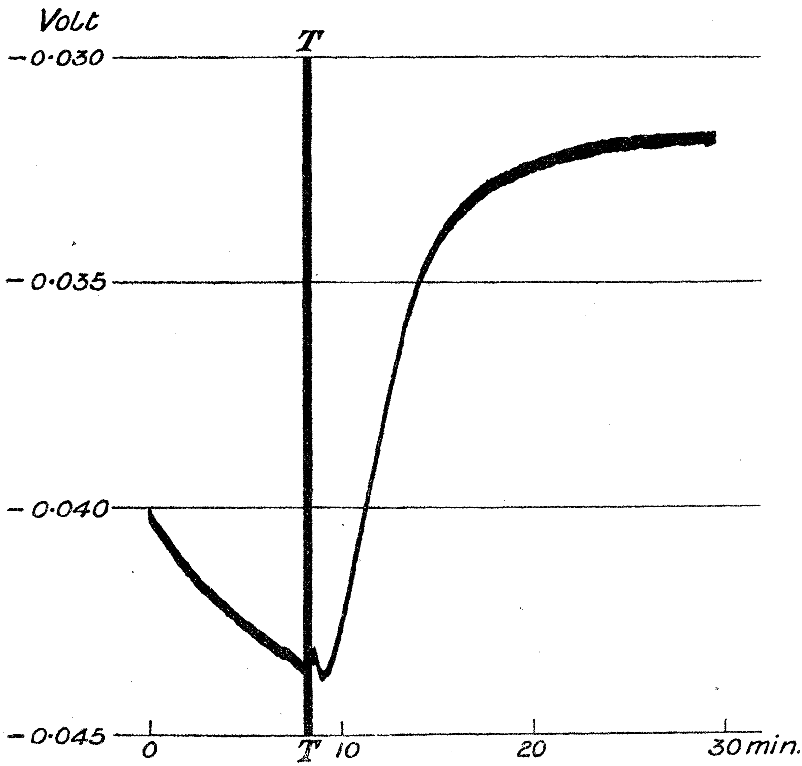
Type I.—Outgoing or positive response.



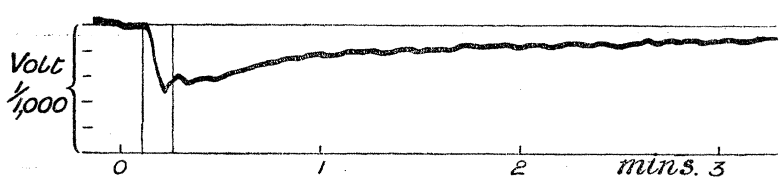
Type I.—Outgoing or positive response.



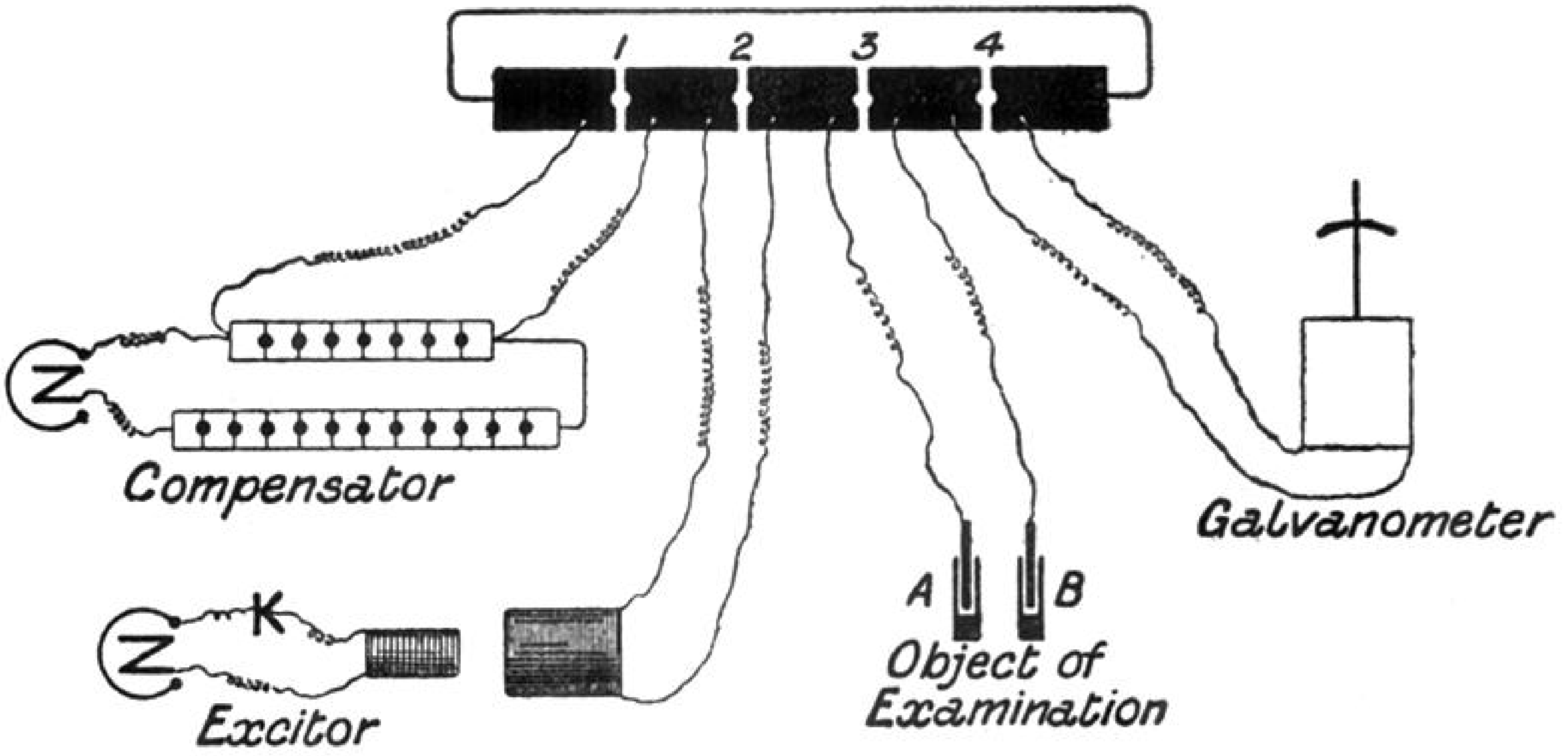
Type II.—Mixed response.



Type II.



Type III.—Ingoing or negative response.



1
2
3
4
Compensator
Galvanometer
A
B
K
Object of Examination
Excitor