

ORIENTATION IN COMPOUND FIELDS OF EXCITATION; PHOTIC ADAPTATION IN PHOTOTROPISM.

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I.

One test of tropistic analysis is its usefulness as providing a description of oriented movement under controlled circumstances of deliberately contrived simplicity. A deeper test is found in its ability to predict measurable aspects of behavior in new situations of some complexity. It should be more obvious than it seems to have been, that an understanding of certain features of animal movements may be arrived at through the attempt to synthesize reasonably complex situations of which significant elements may be studied separately. It is in fact frequently easier to measure the effect resulting from controllable opposed sources of excitation than to quantitate single orienting tendencies. Experiments of this type should therefore give, in suitable instances, a convenient method for the precise characterization of orienting effects (Crozier, 1924-25, 1925-27; Crozier and Stier, 1927-28). They may also open a way toward the measurement of certain aspects of central nervous function (*cf.* Crozier, 1925-27; Crozier and Pincus, 1926, 1926-27, *a*) and of important features of quantitative variability in behavior (Crozier and Federighi, 1924-25, *a*; Crozier, 1925-27).

We have been interested to secure data upon additional instances to supplement the now scanty information bearing upon such questions. The treatment given by Szymanski (1911, 1911-12) does not meet our requirements; nor, for other reasons, does the interpretation of Weiss (1924), so far as we are able to understand it. With an organism which is both geotropically reactive and phototropic, a compound field of excitation may be devised such that in creeping the organism

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is forced to resolve a conflict between mutually opposed orienting tendencies. If, from previous knowledge of each tropism as a more or less isolated element, it be possible to predict the form taken by the resolution, then there is obtained good indication of the efficacy of the initial interpretations. For the phototropic orientation of young rats upon an inclined plane it was possible to predict that the intensity of the light just required to enforce horizontal creeping should be a power function of the sine of the inclination of the creeping plane (Crozier and Pincus, 1926-27, *c*); and it was possible, also, to account for the peculiar changes in the variability among the individual measurements at successively greater inclinations. Until it has been possible to carry analysis of this type into the treatment of cases still more complex, it would appear to be a waste of time to speculate extensively, as so many have done, regarding imponderable elements supposed to characterize behavior.

II.

The geotropic creeping of the slugs *Limax maximus* (Crozier and Pilz, 1923-24; Crozier and Federighi, 1924-25, *c*; *cf.* also Crozier and Pincus, 1926-27, *a*) and *Agriolimax campestris* has been carefully examined, particularly in the case of the latter species (Wolf, 1926-27). On a plane inclined at angle α to the horizontal the slug's orientation (upward or downward) is limited by an angle θ on the plane, such that, very nearly

$$\frac{\Delta \theta}{\Delta \log \sin \alpha} = \text{const.};$$

and, better,

$$(\Delta \sin \alpha) (\Delta \sin \theta) = \text{const.}$$

The phototropism of these slugs has also been studied extensively, especially in the case of *Limax* (Crozier and Federighi, 1924-25, *b*; Crozier and Libby, 1924-25; Crozier, 1925-27); data from experiments of Crozier and Cole relative to circus movements (*cf.* Crozier and Cole, 1922) as a function of light intensity are as yet unpublished, but a brief account has been given of orientation with opposed sources of light (Crozier, 1925-27). *Agriolimax* differs from *Limax* notably in the greater speed with which it becomes adapted to light

of ordinary intensities, as we shall describe. This in fact makes it possible to study here the kinetics of light adaptation. For *Limax* it was found that the amplitude of turning in circus movements, with constant speed of creeping, is very nearly proportional to the logarithm of the light intensity (Crozier and Cole). (This of course need not mean that the primary effect of the light follows this law, since the amplitude of turning is related to the speed of linear progression.)

Experiments were devised to examine the behavior of *Agriolimax* in a compound field of excitation such that the animal should at the same time be caused to orient upward upon a vertical glass plate, and laterally by a beam of light. The slug being initially dark-adapted, time enters as a variable under continuous exposure since photic adaptation is fast; the enforced deviation from vertical creeping therefore decreases. The operative problem thus consists in obtaining a measure of the orienting effect of the light, with the gravitational influence constant, at brief intervals during exposure to light until adaptation is effectively complete. While it would in some ways be preferable to measure at each chosen moment the intensity of light necessary to produce a certain constant deviation of the creeping path from the vertical, we have found it much easier in the present experiments to measure the deviation itself as a function of time. It is possible to account for the course of the observations quantitatively and very simply. The procedure may be reversed in a very obvious way for the study of dark adaptation.

In this manner it becomes possible to investigate the kinetics of photic adaptation in a phototropic animal.¹ The point is an important one, because few direct studies of light adaptation have been made. The present theory of the situation is based upon Hecht's general analysis of photic irritability, and, specifically with regard to light adaptation, upon his experiments with *Ciona* (Hecht, 1918-19). In this case it was shown that the decay of an initial mass of photosensitive substance, during repetitive stimulation by light, follows the law of a first order process. The further analysis of dark adaptation and of sensory equilibrium and intensity discrimination, in *Mya*

¹A slightly different method has been suggested in connection with circus movements (Crozier and Pincus, 1926-27, b).

and in the human retina, is beautifully consistent with this (Hecht, 1922-23, 1924-25, 1926-27). Hecht (1919-20, *b*) has pointed out that in such forms as *Ciona*, *Mya*, and others in which the time limit for the delivery of photic energy adequate for excitation is brief,—that is, exhibiting differential sensitivity,—the time limit is short because necessarily a function of the relative velocity of the “dark” reaction which regenerates photosensitive substance. This makes it possible to regard such cases as involving a mechanism which is fundamentally the same even when the excitation is continuous, as in phototropism. It is clearly important to determine if, in fact, similar mechanisms do underlie photic excitation in the two classes of reactions. When adaptation occurs very slowly it is almost impossible to devise the requisite experiments.

III.

A study of light adaptation here described was made with the slug *Agriolimax campestris*, already used for the investigation of geotropic conduct, so that the behavior in creeping on an inclined plane in darkness was well known. *Agriolimax* becomes adapted to light in a few minutes, so that the process of adaptation can easily be measured. The results obtained have to be carefully handled, however, because the animals are easily influenced in their response to gravity and light by food, for example, as shown by Crozier and Libby in *Limax* (Crozier and Libby, 1924-25). Furthermore, change of weather may influence the responses very strongly. The animals, even when kept for days and weeks under almost constant temperature and humidity, in a dark room, very often give no response whatever. The slugs used for the experiments were kept in darkness for at least 12 to 24 hours so that they were always well dark-adapted before the tests were made. They were then tested one after another. About 75 to 80 per cent showed a good negative orientation to gravity and to light. As pointed out before (Wolf, 1926-27), sometimes the slugs are positively geotropic. On the other hand, differences in the sensitivity to light can be observed. The animals which gave good responses were tested repeatedly. After being confined in the laboratory for 5 to 6 days, creeping and the tropistic responses may become too slow to permit proper determination of angles of orienta-

tion. During an experiment one animal was put upon a vertical plate of ground glass which was moistened every few minutes. In darkness the animals typically then creep straight upward. At one side of the creeping surface is a source of light which gives an equal distribution of parallel horizontal rays. Under these circumstances the animal moves upward, and at the same time away from the light with a certain angle of orientation (θ) (Fig. 1). After an interval of 1 minute the animal is taken away from the glass plate and put back, with vertical orientation, at the same place where it started its

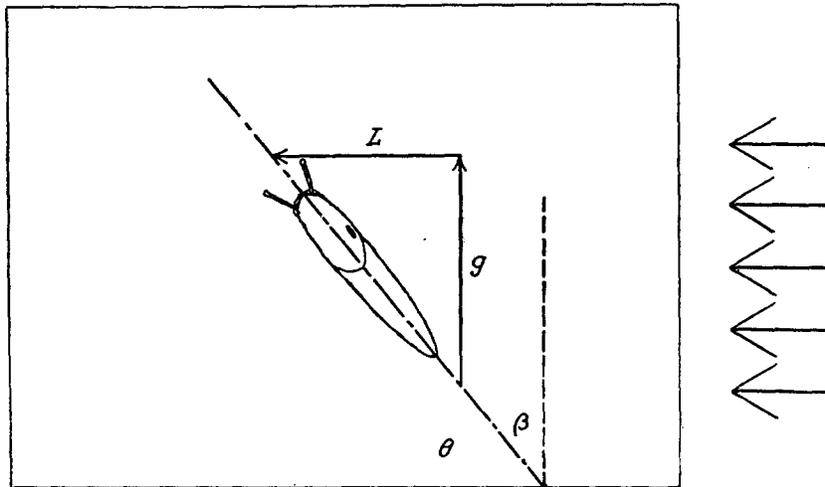


FIG. 1. Orientation of negatively geotropic and negatively phototropic *Agriolimax* on a vertical plate with light from the right,—to indicate terminology of the phototropic (L) and geotropic (g) vectors, the angle of orientation (θ), and its complement (β).

first run. The glass plate is divided in squares, so that the path of the slug can easily be copied on coordinate paper, where the angles of orientation are subsequently measured. The angle of deflection from the vertical decreases quickly. During tests with one animal the slug is repeatedly put back at the starting point, until finally the angle of deflection from the vertical becomes a minimum, sensibly zero. It is assumed that light adaptation is reached as soon as no deviation is apparent from the perpendicular path which ought to be

taken on a vertical plate in darkness; this means, of course, light adaptation to a degree which allows excitation to fall below the threshold required for effective opposition to the orienting force of gravity, as brought out subsequently. While it may appear that this method is crude, since it involves handling, it was always carried out with care and in a uniform way; the handling in fact assists in obtaining continuous creeping. The time lost during the manipulation in bringing the slug back to the starting point is about 5 to 7 seconds until it creeps again and shows a definite response. If handled carefully the eyestalks are not retracted during this operation, so that the eyes are continuously exposed to the light.

The progress of dark adaptation can also be observed in this way. For such studies the animals are first exposed to light until complete light adaptation is reached. The methods used during the experiments are the same as with light adaptation, but the slug is run at intervals after being placed in darkness. Dark adaptation is very much slower than light adaptation, and therefore does not interfere with the direct study of the light adaptation. In a later paper there will be given a fuller treatment of the results obtained during these observations. When tested with light of about 48 f. c. intensity, dark adaptation is not complete until approximately 4 hours.

It is to be noted that in these experiments the light acted always upon the *right* side of the slugs. Measurements of phototropism have shown that fluctuations do occur in the sensitivity of each eye; but the photic reactivity of the two sides, contrary to some earlier statements in the literature, is essentially the same (*cf.* Crozier and Cole, 1921-22).

The first experiments were made at three different times during winter and spring, with one constant light intensity of 29.48 f. c. Throughout these experiments the light intensities were measured by direct photometry, not computed. The initial series of experiments was made with individuals collected in the middle of October at various places. The time during which the animals crept away from the light, before they were brought back to the original starting point near the source of light, was 2 minutes. Each animal was tested at intervals of 1 or 2 days, between five and ten times in all. The results obtained from these more or less preliminary experiments showed

clearly the decrease of the angle of orientation. No great value can be given to these results, because at first the population used was not homogeneous enough; the temperature in the dark room was rather low and inconstant; and the interval of 2 minutes between the successive runs was too great, since the animals moved too far away from the light and decrease of its intensity was perhaps too great. The

TABLE I.

Mean angles of orientation (θ , β , as in Fig. 1) upon a vertical plate with light from one side, with their probable errors and measures of variability. Seven animals tested five to ten times each, at intervals of 2 minutes (see text). For convenience in further treatment the angle β , compliment of θ , is also tabulated.

Series	Light intensity	Time	θ	P.E. _m	C.V.	β
		<i>min.</i>			<i>per cent</i>	
I	29.48 f.c.	1.0	36.9°	±0.65	12.6	53.1°
		3.0	53.9°	±1.2	15.9	36.1°
		5.0	67.4°	±0.89	9.4	22.6°
		7.0	75.4°	±0.68	5.9	14.6°

TABLE II.

Mean angles of orientation (θ) of *Agriolimax* upon a vertical plate with light at one side, with P.E._m and measure of variability. Four individuals, tested six times each.

Series	Light intensity	Time	θ	P.E. _m	C.V.	β
		<i>min.</i>			<i>per cent</i>	
II	29.48 f.c.	0.5	17.9°	±0.26	9.6	72.1°
		1.5	38.7°	±0.49	8.3	51.3°
		2.5	60.6°	±0.98	10.7	29.4°
		3.5	74.5°	±0.39	3.4	15.5°

measurements are summarized in Table I. The data show (Fig. 2) the gradual approach to a condition of adaptation in which the light has no effect on the orientation. As already pointed out, if the inclination of the creeping plane be quickly decreased orientation away from the light is greater. And for some time after the slug is found to creep vertically as in darkness it will still be oriented by the light if the creeping surface is lowered to a horizontal position.

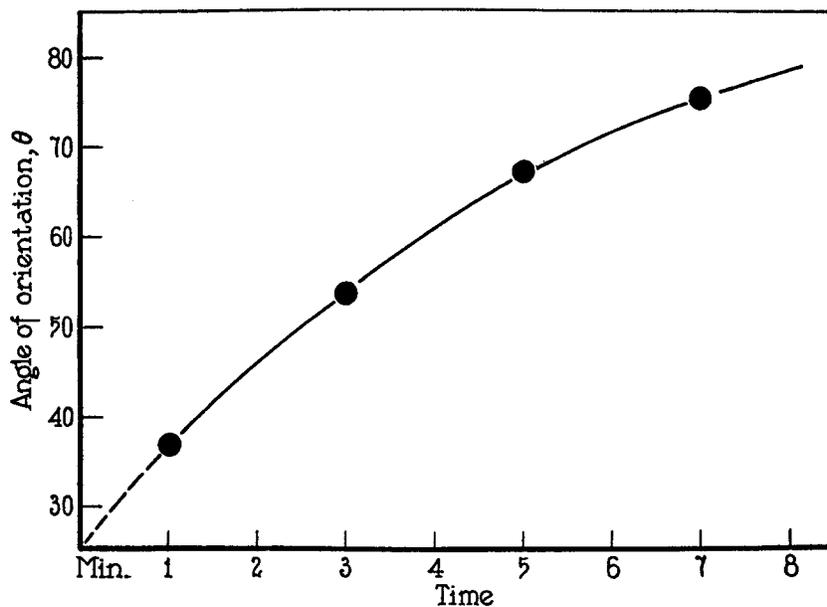


FIG. 2. The decrease of the angle of orientation (θ) with time during continuous exposure to light of 29.48 f.c. The time measurements are the mid-points of successive trails, in this case obtained at intervals of 2 minutes (Table I).

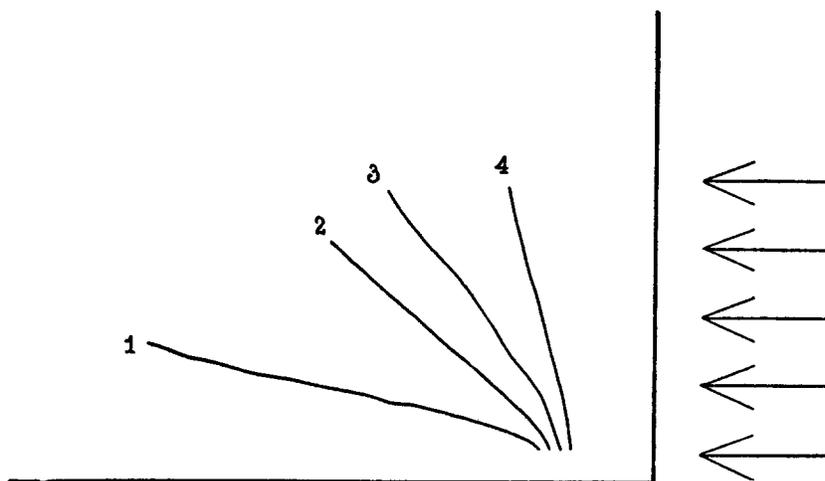


FIG. 3. Successive trials made by initially dark-adapted *Agriolimax* on a vertical plate, light from the right, at intervals of 1 minute (mid-points). (This gives the form of the records as available for analysis; actually the trails all began at the same point.)

For all later tests animals were collected at one place in the greenhouses of the Bussey Institution, which promised to give a more uniform population. The interval between the successive runs with each animal was only 1 minute. The path made during this time by the slug away from the light becomes therefore so short that the decrease of light intensity and the changing state of adaptation stay within limits which can be more easily neglected (Fig. 3).

The second series of experiments was made at the end of January and the beginning of February. For this series only six animals were used, which were tested six times each. Table II gives the measurements of the angles with the means and their probable errors. The light intensity was the same as used in the first series of tests.

TABLE III.

Mean angles of orientation (θ) of *Agriolimax* upon a vertical plate with light at one side, with P.E._m and measures of variability. Four animals tested six times each.

Series	Light intensity	Time	θ	P.E. _m	C.V.	β
		<i>min.</i>			<i>per cent</i>	
III	29.48 f.c.	0.5	27.9°	±0.69	17.4	62.1°
		1.5	46.7°	±0.48	7.2	43.3°
		2.5	60.6°	±0.34	3.7	29.4°
		3.5	72.6°	±0.41	3.7	17.4°
		4.5	79.0°	—	—	11.0°

The third series was carried on at the end of March; four animals were each tested six times. The results are summarized in Table III. They can properly be compared with those obtained 1 month before; even quantitatively the results are closely similar.

IV.

When a slug is creeping on a vertical plane illuminated from one side, two forces act upon it. The light tends to cause photonegative orientation, and the geotropic stimulus forces the animal to creep upward. If during such a test the light be removed, orientation is immediately upward. If, on the other hand, the plate be lowered, so as to be inclined to the horizontal, orientation is more completely

away from the light. Therefore the actual outcome depends upon concurrent continuous stimulations of the two kinds. The path actually described may then be regarded as the resultant of these two components. The deflection from vertical (β) is measured. Referring to Fig. 1, over a short interval

$$\tan \beta = \frac{L}{g}, \quad (1)$$

where L signifies the orienting effect of the light and g represents the geotropic response.

In describing geotropic orientation it has been pointed out that on a plane inclined at angle α to the horizontal upward movement is limited by an angle θ on the plane, such that

$$\frac{\Delta \theta}{\Delta \log \sin \alpha} = \text{const.}$$

(Crozier and Pincus, 1926, 1926-27, *a, d*; Wolf, 1926-27). But the attainment of such orientation does not mean that geotropic excitation ceases. The expressions relating to the angle θ merely define the extent of orientation. When orientation is achieved, the rate of progression (*cf.* Crozier and Pincus, 1926-27, *a*; Cole, 1925-27; Pincus, 1926-27) remains a function of the active gravitational component. This is true of *Agriolimax* and certain other forms, as well as of those cited, although extensive measurements have been made only with rats (Pincus, 1926-27). In this case, the rate of oriented creeping is also directly proportional to $\log \sin \alpha$. We may assume that this is likewise true for the slugs. It means that, θ and the rate of linear progression being each proportional to $\log \sin \alpha$, the rate at which the organism tends to lift its mass vertically upward is obviously proportional to $\log \sin \alpha$. From this it is fair to assume that the intensity of geotropic excitation is constant, for each value of α , regardless of the position of the animal's axis on the creeping plane—although the extent of upward orientation is limited by the distribution of tensions on its peripheral musculature.

If we then take g in equation (1) as constant, since the present experiments involve only $\alpha = 90^\circ$, we have

$$k \tan \beta = L. \quad (2)$$

But L changes with time, due to light adaptation. We assume that at any moment the photic irritability is proportional to the amount of photosensitive material S , only very slowly formed by the proc-

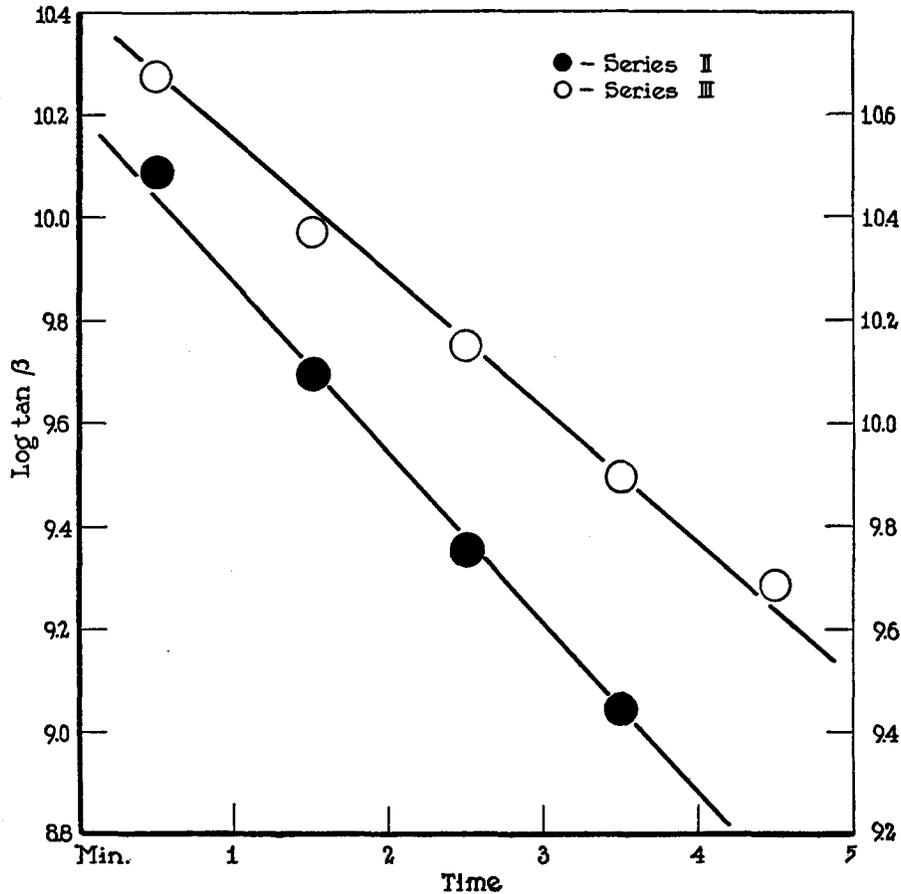


FIG. 4. The rate of light adaptation in two series of experiments made at different times (see text). Series II, scale at right. Series III, scale at left; intensity = 29.48 f.c. The results are plotted in terms of the equation deduced in the text, and if it be applicable they should give a linear decrease with time.

esses underlying dark adaptation, and that the photochemical destruction of S follows the kinetics of a first order reaction (*cf.* Hecht, 1922-23, 1923-24, 1924-25). This means, simply, that the freshly formed

décomposition products of S , requisite for excitation, will over any short interval be proportional to

$$S_t = S_0 e^{-K't}.$$

Since the light intensity is constant during an experiment we may write

$$K'' \tan \beta = e^{-K't}, \quad (3)$$

where K'' is a proportionality constant, K' a velocity constant. From this,

$$\log K'' + \log \tan \beta = -K'''t, \quad (4)$$

where K''' includes the modulus.

The constant K''' may be determined graphically; K'' includes so many proportionality factors that its discussion is postponed until experiments have been completed at various angles of inclination (α) of the creeping surface. The formula states that the logarithm of the tangent of the angle should decrease linearly with time during light adaptation. The results already given in Tables I to III, and subsequently, show excellent agreement with this expectation (Fig. 4).

V.

If the variation of the angle β with time during light adaptation may thus be used to follow the kinetics of the process, it is to be expected that K''' , the "velocity constant," should be a function of the light intensity—and presumably, over ordinary ranges of intensity, a logarithmic function (*cf.* Hecht, 1919–20, *b*, 1920–21). Experiments were therefore made with intensities, 73.69 and 16.32 f.c., respectively above and below the value initially used. The results are given in Table IV. It is to be noted that the data are closely comparable whether the averages are based upon a large number of individuals tested only once or twice, or upon a few animals tested repeatedly.

In Fig. 5 the data of Table IV are plotted in terms of equation (4). The agreement may be taken to be very good. The slopes of the lines in Figs. 4 and 5, given in Table V, change with the intensity

TABLE IV.

Mean angles of orientation (θ) of *Agriolimax* upon a vertical plate with light at one side, with P.E._m and measures of variability.

Series	Light intensity	Time	θ	P.E. _m	C.V.	β
17 animals tested once each.						
IV	16.32 f.c.	<i>min.</i>			<i>per cent</i>	
		0.5	58.0°	±1.18	12.3	32.0°
		1.5	71.4°	±0.75	6.3	18.6°
		2.5	79.5°	±0.61	4.5	10.5°
		3.5	84.0°	±0.75	1.8	6.0°
16 animals tested once each; same as used in Series IV.						
V	16.32	0.5	60.5°	±0.62	5.9	29.5°
		1.5	70.1°	±0.71	5.5	18.9°
		2.5	78.6°	±0.40	2.9	11.4°
6 animals tested six times each.						
VI	73.69	0.5	30.8°	±0.30	7.9	59.2°
		1.5	53.8°	±0.67	9.9	36.2°
		2.5	71.4°	±1.15	12.8	18.6°
		3.5	81.0°	±0.32	4.8	9.0°
25 animals tested once each.						
VII	73.69	0.5	22.5°	±1.35	44.4	67.5°
		1.5	48.3°	±1.49	21.9	41.7°
		2.5	67.8°	±1.10	11.8	22.2°
		3.5	78.7°	±0.65	4.1	11.3°
23 animals tested once each; same as used in Series VII.						
VIII	73.69	0.5	21.3°	±1.27	44.8	68.7°
		1.5	49.0°	±0.91	12.4	41.0°
		2.5	68.4°	±0.94	9.2	21.6°
		3.5	80.0°	±0.81	5.4	10.0°

TABLE V.

Values of the velocity constant for light adaptation (K''' of equation (4)), as related to intensity.

Intensity	K''' Mean
16.32 f.c.	2.6
29.48	2.92
73.69	3.53

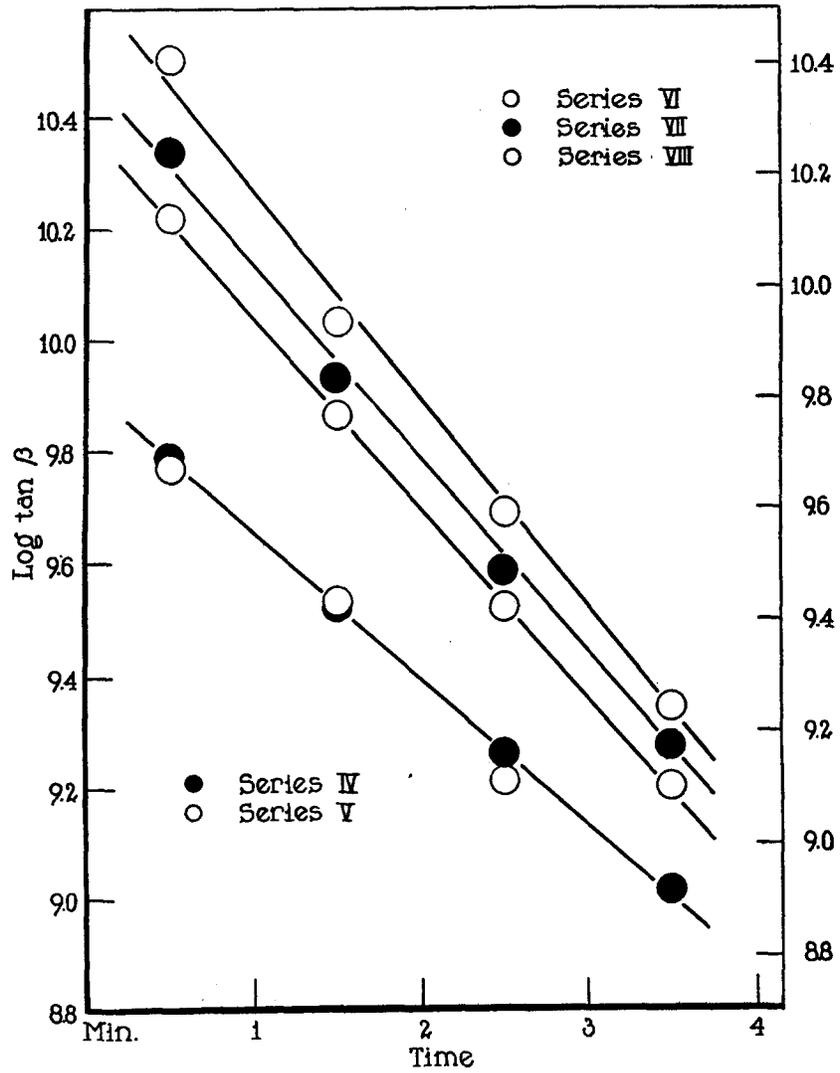


FIG. 5. The course of light adaptation in five series of experiments, at two different intensities: for Series IV, V, 16.32 f.c.; for Series VI to VIII, 73.69 f.c.

of the adapting light in such a way that K''' is directly proportional to the logarithm of the light intensity, as is shown in Fig. 6. The rate of light adaptation with intensities higher than 100 f.c. is so great as not to be measurable by this method; adaptation is complete in less than 2 minutes.

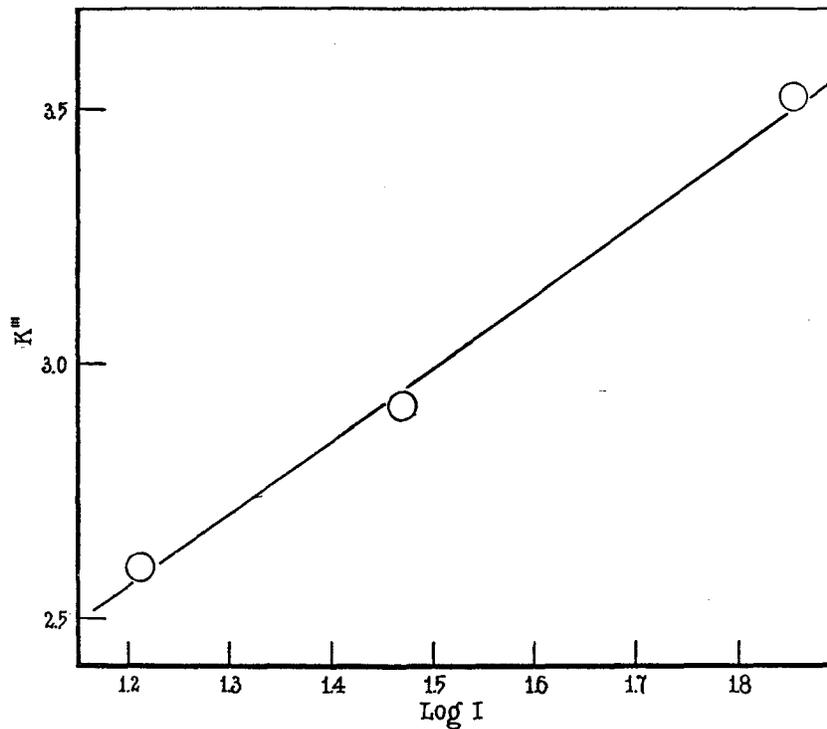


FIG. 6. The rate of light adaptation of *Agriolimax*, as measured through the opposition of photic and constant geotropic orienting forces, increases directly as the logarithm of the light intensity.

VI.

If a dark-adapted slug be placed at a point *A*, Fig. 7, on a vertical plane, with a source of parallel light rays at the right, its complete path, uninterrupted until light adaptation is attained, should be a curve more or less of the type shown. It is of interest to analyse such paths, briefly: because they give useful clues for the extension

of tropistic analysis to the consideration of non-rectilinear orientations. As such a slug creeps away from the light its own adaptation and the decreasing light intensity lead to creeping which is more and more nearly straight upward. With the experimental conditions so

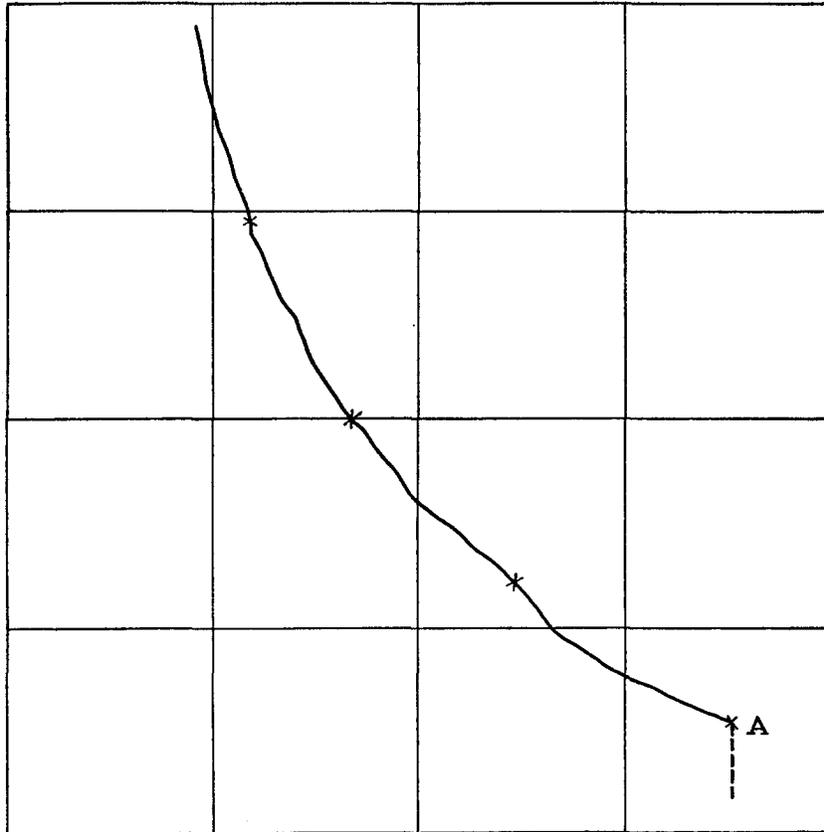


FIG. 7. Copy of the trail of a dark-adapted *Agriolimax* creeping on a vertical plate, at first in darkness (dashed line); light from the right is admitted at A. Successive minutes are marked on the trail. The coordinate units = 5 cm. (see text).

arranged that the light intensity does not decrease as the slug moves to the left, the slope of the curve at any point should be given by equation (3). On the other hand, if the light comes from a point

source, so that the intensity varies, the velocity "constant" for adaptation must include the logarithm of the intensity as a variable.

This could be tested by means of trails executed at different constant speeds of progression, but it is doubtful if conditions governing adaptation within the sense organ would be sufficiently uniform from time to time to permit more than a qualitative test.

SUMMARY.

During upward geotropic orientation upon a vertical plate the slug *Agriolimax* creeps vertically, in darkness. Horizontal light from one side produces orientation of dark-adapted slugs away from the vertical path, through an angle (β). The magnitude of this angle is a function of the light intensity and of time. The moderately rapid course of light adaptation is followed by measurements of β at fixed intervals. Simple assumptions as to the nature of the orienting forces lead to the conclusion that the logarithm of the tangent of β should decrease linearly with time, and that the rate of the decrease should vary directly with the logarithm of the light intensity. Both expectations are adequately realized. Certain implications of these results for behavior analysis are pointed out.

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