ANALYSIS OF THE GEOTROPIC ORIENTATION OF YOUNG RATS. VII

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I

Measurement of the geotropic orientation of young rats creeping upon an inclined surface has lead to a theory of the orientation. For a number of well-inbred lines of rats tested, the relationship between angle of orientation $\theta$ on the inclined surface and the slope of the surface ($\alpha$) is a specific, characteristic, reproducible property. The curve connecting $\theta$ with $\alpha$, for a standard age of individual (13–14 days), defines the tropistic orientation as a type of performance, in terms of an essential condition of the performance.\(^1\) The curve for each race of rats is compound, comprising three segments; each of these segments behaves as an independent heritable unit in crosses between different races (Crozier and Pincus, 1929–30a, b, 1931–32e). The complete curve, extending from a threshold slope of surface to an upper slope beyond which no further significant increase in extent of upward orientation is apparent, is interpreted as resulting from the fact that two elements are concerned in the sensory excitation which results in the oriented creeping. One of these is the magnitude of the gravitational pull experienced by the legs which support and propel the body mass; the second is the frequency with which this gravitational pull acts upon the legs. Each of these elements is directly controlled by the slope of the surface upon which creeping takes place; the speed of

\(^1\) In a discussion of this matter which is not at all clear, von Buddenbrock (1931, pp. 716 et seq.) claims that this performance is not tropistic in character, but is a manifestation of a “higher activity;” but he completely ignores the development of the quantitative treatment which derives from the elementary assumption as to the nature of a tropistic performance (Crozier, 1929), namely, that orientation is maintained when excitation is sensorially equivalent upon the two sides of a bilaterally symmetrical organism.
creeping is almost a rectilinear function of $\log \sin \alpha$; this permits differentiation of the curve connecting $\theta$ with $\log \sin \alpha$, in such a manner as to reveal the distribution of thresholds for the excitation of diverse sensory elements (excitation units) as depending upon the slope of surface (Crozier, 1929; Crozier and Pincus, 1929–30b, 1931–32b, 1931–32d). The role of the magnitude of the gravitationally induced pull supported by the legs in creeping is tested directly by the imposition of additional loads, and the predictable distortions of the curves connecting $\theta$ with $\log \sin \alpha$ are obtained (Crozier and Pincus, 1929–30b, 1931–32d).

It is a matter of considerable importance to provide an independent test of the propriety of the assumption that the speed of creeping is a separate element in the determination of the magnitude of the excitation. The speed of creeping is taken as the only practicable measure of the frequency with which the legs are called upon to support the gravitationally induced pull. The appropriate test is provided by treatments of the young rats which, without the imposition of additional loads which must be carried, or other rearrangements of leg-supported tensions, will automatically force the rats to creep at higher speeds. The most convenient way of doing this is by the injection of suitably adjusted doses of adrenalin. Qualitatively, the expected effect is produced. In terms of the antecedent analysis it is to be expected that in a broad way the effect of forcing the legs to move more frequently should be entirely similar to that produced by the imposition of an added load. It is to be expected, furthermore, that the threshold slope of surface for the elicitation of definite orientation should be found lower than in the absence of higher speeds of progression, but that at the new threshold slope the orientation angle obtained should be statistically identical with that characteristic of the threshold slope of surface which obtains for the untreated rats.

Not only is it true that the orientation angle $\theta$ as a function of the slope of surface is a characteristic property of each pure line tested, but it has been demonstrated in addition that the variability of orientation is also a constitutional property. The scatter of the individual determinations of $\theta$ at each of a number of slopes of surface, measured
by the respective $\sigma$'s, is a rectilinear function of $\sin \alpha$ for the un-weighted, untreated rats; the relative variation of performance ($\sigma/\theta$) is also a rectilinear function of $\theta$; and the quantity which has been termed "the proportionate modifiable variation" (Crozier and Pincus, 1931-32a, etc.) is likewise a characteristic racial property. The several indices of variability behave in a characteristic manner in individuals produced by cross breeding (Crozier and Pincus, 1931-32e, and Paper X).

Since these methods of measuring and expressing variation of performance permit a very precise indication that the observations concerned which satisfy these criteria are free from individual bias or idiosyncrasy due to the observer, and are competent to express an organic invariant of a particular race under the conditions thus far explored, it becomes possible to raise as a fundamental issue the question as to what is really meant by a statement to the effect that a given method of experimental treatment, or an alteration due to a spontaneous or other organically determined constitutional change of an organism, has "increased" or "decreased" this organism's variation of performance. The whole question of the nature and meaning of "variation of response" is indeed at issue (cf. Crozier, 1929). The problem is somewhat analogous to that which we have already discussed (Crozier and Pincus, 1927-28, 1929-30a, b) in the case of measures of total performance. The fact that for certain of our lines the curves connecting $\theta$ with $\alpha$ cross one another shows that it is impossible to characterize the respective geotropic performances except by the complete representation of the creeping curves—no mere measure of maximum performance, for example, is anything but misleading; the behavior of the segments of the respective curves in cross-breeding tests amplifies the ground for this position. So also in the consideration of variability, variation of performance must be considered as a function of the performance before a significant index of variability can be obtained. That significant measures are obtainable by this procedure is amply demonstrated by the fact that the indices of variation secured on this basis are constant within narrow limits for each race, but differ pronouncedly for the several races. In practice, then, it will not be adequate to state that under a given set of constant conditions the root-mean-square deviation of the measures of performance has been
increased or decreased with reference to a chosen standard. The behavior of the total variation of the performance, and the measurement of that fraction of the total which is modified according to the intensity of excitation, is the necessary basis for any discussion of changes which may be induced in variation of performance. It is easy to show that divergent results of completely contradictory superficial significance may be obtained unless this procedure is followed.

This paper is concerned primarily with these two points: (1) the distortion of the fundamental \( \theta - \alpha \) curve as the result of injection of young rats of standard age with a sufficient dosage of adrenalin chloride to produce an appreciable increase in the speed of progression during geotropically oriented creeping; (2) the modifications thereby induced in the measures of variation of performance. An important additional point which the latter consideration enables us to deal with, is connected with the suspicion (Crozier and Pincus, 1931–32c) that the total variation of performance, or the fraction thereof which is a function of the intensity of excitation, may be determined by the total number of effective sensory elements affected through tension in the musculature which supports and moves the animal’s mass.

The discussion thus far has been restricted to the consideration of the performance of young rats of standard age (13–14 days after birth). It becomes of very considerable interest to extend these considerations to the treatment of the orientation of adult rats. In this case, two possibilities are apparent, and there is no means of deciding beforehand, nor any necessity to decide, which of the two may be expected: either the total number of effective “sensory units” or units of variation may change with age, or these numbers may appear as racial invariants. To this aspect of the problem we shall turn in the succeeding paper.

III

The present experiments are concerned with young rats of race A, 3 individuals (2 males, 1 female) from the 23rd generation of this inbred line with which many previous observations have been made (cf. Crozier and Pincus, 1931–32a; and subsequent observations). The method of observation has been discussed previously (Crozier and Pincus, 1931–32a). These rats received intraperitoneal injections of 2/5
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cc. of adrenalin chloride 1:50,000 in sterile Ringer solution before each set of measurements. Control observations, employing injection of Ringer solution only, showed no disturbances whatever of the fundamental $\theta - \alpha$ relationship to be traceable to the fact of handling and injection. Antiseptic precautions were employed. Beginning 1/2 hour subsequent to the injection and continuing for several hours thereafter, creeping on the inclined surface is "steadier," swifter; creeping is relatively uninterrupted; there are practically no long waits between periods of spontaneous creeping such as are involved in observations on the uninjected, "normal" young rats. Repeating the in-

| Order of measurement | $\alpha$ | $\theta$ | P.E.$\theta$
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<tr>
<td></td>
<td>degrees</td>
<td>degrees</td>
<td>degrees</td>
</tr>
<tr>
<td>1</td>
<td>12.5</td>
<td>54.05</td>
<td>±1.99</td>
</tr>
<tr>
<td>2</td>
<td>15</td>
<td>59.85</td>
<td>1.87</td>
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<tr>
<td>3</td>
<td>20</td>
<td>61.70</td>
<td>1.87</td>
</tr>
<tr>
<td>4</td>
<td>25</td>
<td>65.46</td>
<td>1.60</td>
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<tr>
<td>5</td>
<td>30</td>
<td>70.11</td>
<td>1.40</td>
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<tr>
<td>6</td>
<td>40</td>
<td>73.89</td>
<td>1.22</td>
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<tr>
<td>7</td>
<td>50</td>
<td>73.43</td>
<td>1.38</td>
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<tr>
<td>8</td>
<td>60</td>
<td>78.14</td>
<td>0.86</td>
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<tr>
<td>9</td>
<td>70</td>
<td>78.21</td>
<td>0.79</td>
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jections after 24 hours leads to an identical succession of events. The sequence of observations was so arranged (Table I) that no unfair influence upon the readings at any one slope was to be looked for.

In Fig. 1 the data of Table I are plotted, as well as the standard curve for uninjected A rats; the latter is taken from the material in previous papers, with which sundry subsequent determinations show quantitative agreement. The curve for the injected rats is possibly modifiable at its lower end, but as drawn it passes through the standard deviations of the two lowest points. The effect of the injection with this particular dosage of adrenalin is clearly such that (1) it lowers the threshold slope of surface from approximately $\alpha = 20^\circ$ to $\alpha = 12.5^\circ$;
(2) nevertheless, at this new, lower threshold slope the threshold orientation angle, the threshold response, is statistically identical with that obtained at the threshold slope of surface effective for the uninjected rats; (3) the curve connecting $\theta$ with $\alpha$ has been distorted in precisely the same manner as it is (Crozier and Pincus, 1929–30$b$, 1931–32$b$) by the addition of a mass of approximately 2.5 gm. on the saddle region; the lower portion of the curve has been pulled out to the left, the slope has been decreased; this is the effect which we have attributed (Crozier and Pincus, 1929–30$b$, 1931–32$b$) to the bringing into action of additional receptor units at slopes of surface much below those characteristically required for their normal activation; (4) this implies a flattened, intermediate region in the curve, as Fig. 1 shows to be in fact demonstrated; and it requires further, in terms of the antecedent analysis, that the uppermost portion of the $\theta - \alpha$ curve should under these conditions lie below the standard curve for un-

![Graph](image_url)

**Fig. 1.** Modification of the relation between mean orientation angle $\theta$ and slope of surface ($\alpha$) with young rats of race $A$, by intraperitoneal injection with 2/5 cc. adrenalin chloride 1:50,000. The standard curve is derived from many previous measurements with litters of race $A$. The effect is exactly analogous to that obtained by the carrying of an additional load of approximately 2.5 gm. on the saddle of such young rats.
treated young rats. Qualitatively, therefore, the effect of adrenalin, involving the increased frequency in the use of the legs during the execution of oriented trails, is precisely that which the assumption underlying our differentiation of the $\theta - \log \sin \alpha$ graphs (Crozier and Pincus, 1929–30b) has implied.

IV

The theory of the measurement of variability for cases of this type requires that the relative variation of performance should be a rectilinear function of the intensity of excitation. The latter is measured by the orientation angle $\theta$ (Crozier and Pincus, 1929–30b, 1931–32a). Fig. 2 shows that for the series of measurements in which adrenalin has been injected, the ratio $100 \frac{P.E.}{\theta}$ is a declining rectilinear function of $\theta$. From this graph, extending from threshold $\theta$ to maximal $\theta$, the following indices are directly computed: (1) the variation number, $V.N.;$ (2) the total relative variation, namely, the area under the graph; (3) and the proportionate modifiable variation, namely the fraction of the total area under the graph which is included in the upper, triangular portion. From the earlier experiments with untreated A rats we already know that for this race each of these quantities is a statistical constant, at the standard age. The respective values are given in Table II, together with the values obtained from the series of measurements with the adrenalized rats.

The variation number, $V. N.,$ corrected for size of sample in the usual way (Crozier and Pincus, 1931–32a, 1931–32e), is 3.64 for the adrenalized rats. For the untreated A rats (Crozier and Pincus, 1931–32d) this index has the value 2.56 in the same units. We have already demonstrated that the magnitude of this index is in no manner disturbed when the young rats are forced to carry added loads (Crozier and Pincus, 1931–32e). Fig. 3 shows that with samples of equal statistical weights the estimation of variation of performance, as by a P. E., can give misleading results when the comparison is restricted to a single slope of surface. With the adrenalized individuals the relative variation of performance is more sharply restricted by increasing the intensity of excitation through elevation of the surface on which creeping is allowed to occur. Consideration of the total variation of performance, that is, of the area under the variation plot,
Fig. 2. The relative variation of performance as related to the magnitude of the response, for young rats of race A injected with adrenalin. The variation number (V. N. φ) is 3.64, the total relative variation 127 units.

### TABLE II

Indices of variation of orientation angles for young rats of race A, with and without injection of adrenalin.

<table>
<thead>
<tr>
<th>Index of variability</th>
<th>Without adrenalin</th>
<th>With adrenalin</th>
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<tr>
<td>V. N. φ</td>
<td>2.56</td>
<td>3.64</td>
</tr>
<tr>
<td>Total relative variation</td>
<td>84 units</td>
<td>127 units</td>
</tr>
<tr>
<td>Proportionate modifiable variation</td>
<td>56 per cent</td>
<td>56 per cent</td>
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demonstrates that the total variation of performance, between identical sets of limits of intensity of excitation (θ), is markedly enhanced in the case of the adrenalinized young rats, namely from 84 units to 127 units. In this respect the action of adrenalin is to be compared with that apparent when genetic influences modify the variation of performance (Crozier and Pincus, 1929-30a, b, 1931-32d, e). The chance of affecting the total variation of performance, in the present experiments, of course involves variations connected with the process of injection, the absorption of the hormone, the reactivities of the individuals, and the nature of the time course of the action of the adrenalin. In fact, however, the measurements show unexpectedly excellent agreements among the several individuals. Corrected for size of sample and for number of individuals, the total variation (100 P. E. θ/θ) vs. θ, for normal young A rats and for young A rats carrying additional loads is identical, and amounts to an average of 84.4 units; for the adrenalinized young rats in the present series of observations the total variation is 127 units. It is to be concluded, therefore, that the effect of injection with adrenalin, under the conditions stated, gives uniform, reproducible distortion of the θ - α curve, and at the same time significantly increases the total variation of θ manifest over the entire range of slopes of surface within which geotropic orientation occurs.

The percentage of the total variation which is modifiable as a function of θ is completely unaffected, however, by comparison with the findings for the normal young A rats. For the latter, with and without additional loads carried, the proportionate modifiable variation ranges from 56 to 59 per cent. For the adrenalized young rats the percentage modifiable variation is still 56 per cent. We have already indicated that there appears to be a connection of some sort between the proportionate modifiable variation of θ and the number of sensory units, when diverse races are compared (Crozier and Pincus, 1931-32 c). It has been shown, also, and the demonstration is amplified in the succeeding paper, that the proportionate modifiable variation appears to be a constitutional property in the sense that it is independent of

2 It is obvious that the procedures here outlined should permit examination of the existence and nature of individual differences in the effects due to experimental treatments.
age of individual. This would be taken to indicate that while the effect of injection with adrenalin has been to increase the relative variation of the measured orientations, this has affected equally those generalized aspects of variation which concern the relative variation of $\theta$ at all intensities of excitation (the unmodifiable variation) and that

![Graph showing relative variation of performance](image)

**Fig. 3.** The relative variation of performance, corrected for the number of observations on each individual ($n$) and for the number of individuals in the set ($N$); data upon a standard litter ($A_{13}$, Crozier and Pincus, 1931–32) are taken from an earlier account.

fraction of the total variation which is limited and controlled (modifiable) according to the intensity of excitation.

It has been pointed out that the $\theta - \log \sin \alpha$ curve is distorted, under adrenalin, in a fashion exactly analogous to that evidenced when a weight of approximately 2.5 gm. is attached in the saddle posi-
tion (Crozier and Pincus, 1929–30 b, 1931–32 b); the differentiation of
$\theta - \log \sin \alpha$ curve shows that the area under the curve obtained by
plotting $(\Delta \theta / \Delta \log \sin \alpha)$ against $\sin \alpha$ is a little lower than with that
for the curve given by the untreated rats; the curve so obtained, giving
a picture of the distribution of the tension thresholds for the receptor
units involved, is of course entirely similar to that gotten with posterior-
ly located additional weights of 2.1 gm. (Crozier and Pincus, 1929–
30 b, 1931–32 b), although a little more extreme. The area under this
curve cannot be directly compared with other analogous determina-
tions for the A race, since the proportionality factor involved in the
differentiation is different, and consequently the magnitude of the
units on the ordinate scale. The fact that the proportionate modi-
ifiable variation of performance remains fixed under these diverse
conditions indicates once more (Crozier and Pincus, 1931–32 b) that
it reflects a fundamental property of the race of individuals considered.

Since the curve connecting $\theta$ with $\log \sin \alpha$ for the adrenalin-in-
jected rats is almost rectilinear, one may consider $P. E. \theta / \theta$ as a func-
tion of $\log \sin \alpha$; V. N. $\log \sin \alpha = 2.5$, slightly lower than with the
untreated A's (2.82); the percentage modifiable variation computed
on this basis is about 52 per cent. We also expect that $P. E. \theta$ should
be nearly a rectilinear function of $\sin \alpha$ (Crozier and Pincus, 1931–32 b)
and this is practically the case. At given slope of surface, however, the
variation of $\theta$ is higher with adrenalin (Fig. 3).

It is pertinent to inquire as to the manner in which the effect of the
injection of adrenalin could be expected to influence the geotropic
orientation. We have been more particularly concerned to obtain
an instance illustrating the fashion in which the analysis of variation
of performance must be approached, than in deciding upon the nature
of the effect of any particular substance when injected. It is clear,
however, that the mere fact of increased frequency of stepping could
be regarded as bringing into action more frequently, and consequently
with increased "mass" effect, an influx of self-generated (propriocep-
tive) impulses which could be conceived to modify the state of excit-
ability of the central nervous system in a very general way. This
could not be traced to a mere increase in the frequency of stepping,
as the experiments with added loads clearly demonstrate the absence of any such effect (Crozier and Pincus, 1931-32 b). An influence of the adrenalin upon muscular tonus, such as is actually observable in the slightly different, more flexed, posture of the legs, might be appealed to in such a case. This could be expected to contribute about equally to the possibility of several kinds of deviations from the exactitude of the oriented responses. In a following paper we shall have occasion to point out that with suitably prepared adult rats of race  A this effect of adrenalin is not at all apparent; in this case, the total variation of performance, for a given magnitude of \( \theta \), is in no respect different from that with untreated adult  A  rats, although the effect of adrenalin upon the \( \theta - \alpha \) curve is in character entirely similar to that which the present experiments with the young rats reveals.

It is difficult to see why the effect of injecting adrenalin should be analogous in all respects (save that involving variation) to the effect of an added mass located posteriorly at the saddle position on the back. One must suspect that the adrenalin acts differentially, under these conditions, upon the excitation of posteriorly located tension receptors, that is, receptors in the posterior pair of legs. Why this should be the case we cannot at the moment decide, and it is immaterial for the present argument.

**SUMMARY**

The intraperitoneal injection of standard young rats of race  A  with 2/5 cc. of adrenalin chloride 1:50,000 results in increased speed of geotropically oriented creeping upon an inclined surface. It was expected that the effect of such increased frequency of stepping must be analogous to that due to imposition of added loads carried by the rats during geotropic progression. This is verified. The curve connecting \( \theta \) with log sin \( \alpha \) is distorted, under adrenalin, so as to be comparable to that obtained with an added mass of approximately 2.5 gm. upon the young rat's saddle; the threshold slope of surface for orientation is accordingly lowered, from \( \alpha = 20^\circ \) to \( \alpha = 12.5^\circ \); at the new threshold slope of surface the mean orientation angle \( \theta \) is the same as in the absence of adrenalin at the corresponding threshold slope of surface.
The total variation of performance is significantly increased in the injected rats, and at given slope of surface the variation is slightly increased. The proportionate modifiable variation of response is quite unaffected by the distortion of the \( \theta - \alpha \) curve, and is the same as in standard young \( \lambda \) rats untreated or carrying additional loads.

It is pointed out that for the consideration of the problem as to whether a given experimental treatment, or a given natural situation, affects in any way the variation of performance of a living system, it is necessary to obtain indices of variability which involve the expression of variation of performance as a function of measured conditions governing the performance.

REFERENCES