Technical Papers

Physiological Damage under Emotional Stress as a Function of Early Experience[1]

Otto Weininger[2]

Department of Psychology, University of Toronto, Toronto, Canada

Studies at the Wistar Institute of Anatomy and Biology at Philadelphia (*1*) have suggested that the more that albino rats are handled and petted, the better they seem to thrive in the laboratory situation. Major hypotheses investigated in the present study were that albino rats, gentled for 3 wk for 10 min a day following weaning, would (a) show significantly greater mean weight, (b) show more activity and less fearful behavior in an open field situation, and (c) sustain less physiological damage to the endocrine, cardiovascular, and gastrointestinal systems under prolonged emotional stress as adults, than would a comparable group of controls.

Three litters of male albino rats, pure Wistar strain, were randomly sorted by litters into two groups immediately following weaning (23 days of age), and were housed in individual cages. A check on randomness of this procedure showed the mean weights of these two groups to be approximately the same. The 16 male animals in the experimental group were gentled for 10 min a day for the next 21 days. Gentling consisted of holding the animal in the experimenter's left hand with the hand placed against the experimenter's chest, so that the animal was nestled in the palm of the left hand, with the right thumb or forefingers stroking the back of the animal from the head to the base of the tail. During this time the control group of 16 male albinos was not handled at all, but in all other respects was treated exactly the same as the experimental group. Water and Fox Bar food pellets were available to all animals in both groups ad libitum.

At 44 days of age, that is, 3 wk after weaning, rats in both groups were weighed, and were weighed once a week thereafter. The animals were introduced to a brilliantly lighted open field situation at the ages of 58 and 65 days. At 60 days of age rectal temperature was taken, and finally, at 79 days of age each rat was immobilized and placed on its back for a period of 48 hr during which time it was deprived of food and water. At the end of the stress period, the animals were sacrificed and autopsies were performed by an independent laboratory technician who had no means of identifying the group to which the animals belonged.

Inspection of the data indicates support for all three major hypotheses. In respect to weight, for example, a check at 44 days showed the mean weight of the gentled group to be 161.06 g compared to 141.25 for the nongentled, a different significant at the 0.0002 level of confidence, although there had been no significant difference between the weights of these two groups at the start of the experiment, when the animals were 23 days of age. This trend continued throughout the experiment. At 79 days of age, just before introduction of the stress situation, mean weight of the gentled group was 319.00 g, compared to 264.75 for the nongentled. Critical ratio obtained here was 13.3. Further analysis indicated that this difference in weight between experimental and control groups was related to a significantly greater proportion of adipose tissue per 100 g of body weight for the former, and to a significantly greater skeletal length for the former. The gentled rats simply grew more than the nongentled.

Records of activity in the open-field test did not turn out in the direction predicted by the second hypothesis. In the first trial, the nongentled animals wandered over the grid to a greater extent than the gentled; in the second trial, no significant difference was obtained. But in both trials, the gentled rats ventured significantly closer to the brilliantly lighted center of the open-field setup, thus showing more of a tendency to ignore the natural habit of their species to cling to walls (thigmotaxis) and avoid light. Rectal temperatures were significantly greater for the gentled rats, which suggests that the basal metabolic rate of the experimental animals may have been altered by the gentling procedure.

Finally, autopsies of the 32 rats immediately following stress showed that relatively more damage had been sustained by the cardiovascular and gastrointestinal systems of the nongentled animals compared to the gentled. Results for the endocrine system were equivocal. In respect to the heart, examination showed that 12 out of 16 rats in the nongentled group, but only 5 out of 16 in the gentled group, had suffered severe heart damage, including distension of the surface blood vessels. This difference is acceptable at the 2% level of confidence. No statistically significant difference with respect to liver and kidney damage was observed. In regard to the gastrointestinal system, a count showed a total of 54 bleeding points in the stomach and duodenum of the nongentled rats and a total of 9 for the gentled. This difference is significant at the 0.01 level of confidence. In terms of weight per 100 g of body weight, the adrenals of the nongentled animals were significantly heavier, at the 0.001 level of confidence, than the adrenals of the gentled animals. No significant differences in weight were obtained in respect to kidneys, liver, or pancreas glands.

Cardiovascular damage under prolonged stress, as Selye has shown (*2*), may be considered an end product of the action of ACTH from the pituitary in re-

[1] This article is, in substance, the same as a paper delivered Dec. 28, 1953, at the annual meeting of the AAAS in Boston.

[2] This research was supported by a grant from the National Research Council of Canada, to Dr. E. W. Bovard, Jr.

leasing hormones from the adrenal cortex. The relative immunity to stress damage on the part of the gentled animals may, therefore, have resulted from a decreased ACTH output from the pituitary in response to the same alarming situation that also faced the nongentled animals. If this were the case, it could be expected that a comparison of adrenals from gentled and nongentled rats following stress would show the latter to be heavier, after being stimulated by more ACTH output. Such was indeed the case.

References

1. GREENMAN, M. J., and DUHRING, F. LOUISE. *Breeding and Care of the Albino Rat for Research Purposes.* Philadelphia: Wistar Institute of Anatomy and Biology, (2nd ed.), 1931.
2. SELYE, H. *Annual report on stress.* Montreal: Acta, 1951.

Manuscript received December 14, 1953.

The Energy Requirements for Bacterial Motility

Harold J. Morowitz[1]

National Bureau of Standards, Washington, D. C.

In light of recent advances on the structure of bacterial flagella (*1, 2*), it is of interest to calculate the energy expended by these "monomolecular muscles" in propelling the organism.

For the case of a small object moving through a viscous medium at low velocity, the force F necessary to balance frictional resistance is given by $F = fv$, where f is the frictional coefficient and v the velocity. If the bacteria under consideration are assumed to be prolate spheroids of equatorial semi-axis b and semi-axis of revolution a, then f, is given (*3*) by

$$f = \frac{6\pi\eta \sqrt[3]{ab^2}(1-\rho^2)^{1/2}}{\rho^{2/3} ln \left[\frac{1+(1-\rho^2)^{1/2}}{\rho}\right]}, \qquad (1)$$

where η is the coefficient of viscosity of the medium surrounding the bacterium and ρ is the ratio of axes, b/a. The energy expended per unit time, P, is given by

$$P = Fv = fv^2, \qquad (2)$$

where f is given by Eq. (1).

This formulation neglects the frictional resistance of the flagella which will be assumed to have the same value of that of the bacterium for order of magnitude calculations. An extension of the more exact hydrodynamical analysis of Taylor (*4*) should lead to a more precise value of P.

For cells of *Bacillus subtilis,* b is 0.5 micron, a is 1 micron (*5*) and v is 10 microns/sec (*6*). The coefficient of viscosity of water at 25° C is approximately 0.009 poise. Substitution of these values in Eq. (2) leads to a value of about 1.1×10^{-11} erg/sec. Doubling this figure to allow for the resistance of the flagella and converting to more convenient units yields a power output in motility of about 14 electron volts/sec. Further assuming that the conversion from chemical to mechanical energy is 25 percent efficient, one finds the total rates of energy expenditure for motility of one organism to be about 56 electron volts/sec.

The analogy between flagella and muscle fibers (1), and the observation that isolated flagella contract in the presence of adenosine triphosphate (2) makes it reasonable to assume that the energy of motility comes from energy rich phosphate bonds. In that case about 150 bonds reacting per sec would supply the necessary energy. Electron micrographs indicate that the organism has about 10 to 20 flagella, and analogous data from the flagella of larger organisms would indicate that each of the flagella flicks about 10 to 20 cps.

As the total number of flagellar flicks per sec, 100–400, is the same order of magnitude as the number of bonds reacting per sec, it is possible to consider each flagellar flick as the result of a small number of discrete chemical events (perhaps one), such as metabolic hydrolysis of energy rich phosphate bonds. In studying bacteria, one reaches a small order of size where a very few reacting molecules exert a large influence.

References

1. ASTBURY, W. T. *Scientific American* **184**, 20 (1951).
2. DE ROBERTIS, E., and FRANCHI, D. M. *Program IXth Meeting, Electron Microscope Soc. America,* 1951.
3. COHN, E. J., and EDSALL, J. T. *Proteins, Amino Acids and Peptides,* 404–406. New York: Reinhold, 1943.
4. TAYLOR, G. *Proc. Royal Soc. (London)* **211A**, 225 (1952).
5. KNAYSI, G. *Elements of Bacterial Cytology.* Ithaca: Comstock Pub. Co., 1951.
6. HEILBRUNN, L. V. *An Outline of General Physiology.* Philadelphia: Saunders, 1943.

Manuscript received November 6, 1953.

The Induction of Scab Lesions on Aseptic Potato Tubers Cultured *in vitro*[1]

W. G. Barker and O. T. Page

Department of Botany,
Ontario Agricultural College, Guelph, Canada

The production *in vitro* of tubers from etiolated potato shoots has been reported recently (*1*). It was considered that a significant contribution could be made concerning the inception of potato scab lesions by observing the action of a pure culture of a known pathogenic strain of *Streptomyces scabies* (Thaxter) Waks. and Henrici on sterile potato tissue. This would be of particular interest because it has not been possible to demonstrate that *S. scabies* alone could cause scab. In addition, there are reports citing the habitation of normal potato tissue by microorganisms (*2–4*, among others). It is with these considerations that this preliminary report is concerned.

A series of differential media was used in an attempt to isolate microorganisms that might be occurring as "normal" microflora of the cultured potato tissue. In addition, the medium used in culturing the potato tissue (*1*) will support the growth of many organisms. In no case was there a microorganism isolated from cultured potato tissue which appeared

[1] Present address: National Heart Institute, National Institutes of Health, Bethesda, Md.

[1] Supported in part by the Potato Scab Committee of Ontario.