or water intake. Studies with other grasshopper eggs suggest that the froth may indeed shield the pod from excess water (16). During temporary flooding, a tanned pod might survive intact, whereas an untanned one would probably fall apart. In a submerged pod, the air trapped within the hard insoluble froth could possibly serve for respiratory purposes, as does the air carried in the "plastron" of certain aquatic insects (17). In the normally dried pod, this same trapped air could provide insulation against extreme temperatures. Severe climatic conditions are a reality in the life of R. microptera. In Florida, as elsewhere throughout the range of this insect in the southeastern United States, sudden torrential thunderstorms, alternating with periods of scorching heat, are the rule of the day during the summer months when the eggs are laid. Finally, accelerated tanning may also be of defensive value, since some predators might tend to ignore the pods once they have hardened.

The glands found in Romalea also occur in several related genera, all belonging to the subfamily Cyrtacanthacrinae (4). Species of other subfamilies (Acridinae, Oedipodinae, Pyrgomorphinae) lack the glands (4) for reasons unknown.

THOMAS EISNER* Division of Biology, Cornell University, Ithaca, New York 14850 **JULIAN SHEPHERD** Biological Laboratories, Harvard University, Cambridge, Massachusetts G. M. HAPP

Department of Biology, Catholic University of America, Washington, D.C. 20017

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- 7. By pulling on the ovipositor, ready access could be gained to the glands, and the crystals were inserted without contaminating the gonopore or other adjacent regions.
- The fact that the glands can be artificially everted by forcible distention of the abdomen with injected fluid (Fig. 2B) need not be taken to indicate that evagination is the normal mechanism whereby the glands deliver their secretion during oviposition. The glands of ovipositing females pulled from their burwere always found to be in the invaginated condition, despite the fact that the abdomen of such females was in its characteristic state (6) of sustained maximal dis-

tention. The muscle that attaches to the gland should not, therefore, be assumed to func-tion as a retractor. Instead, its action may consist of pulling taut the normally slack glandular pouch, thereby squeezing the pasty secretion from the lumen.

- 9. This was established by streaking isolated rom wounds, and comparing the rates at which the affected regions underwent tanning.
- 10. Blood or clots from wounds other than the operated gland sites was also found to promote egg tanning, thus ruling out the pos-sibility that the "wound factor" was simply residual secretion that had contaminated the gland sites. Unfortunately, the operated gland sites never healed completely with time, and it was therefore impossible to determine whether eggs laid by glandectomized females with *sealed* wounds might show retarded
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a phenol oxidase, and protein. As a result of the interaction of these substances, the phenol glucoside is hydrolized and the free phenol glucoside is hydrolized and the free phenol oxidized to quinone, and the quinone tans the protein of which the capsule is made. P. C. J. Brunet and P. W. Kent, *Proc. Roy. Soc. London Ser. B* 144, 259 (1955); P. W. Kent and P. C. J. Brunet, *Tetrahedron* 7, 252 (1959); M. G. M. Pryor, *Proc. Roy Soc. London Ser. B*, 128, 378 (1940); *B. Dursell A. B. Todd. Biochurg. L* 40, 677 Russell, A. R. Todd, Biochem. J. 40, 627 (1946).

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 - John Simon Guggenheim Memorial Founda-tion Fellow.
- 25 February 1966

Tetrodotoxin and Manganese Ions: Effects on Electrical

Activity and Tension in Taenia Coli of Guinea Pig

Abstract. Tetrodotoxin, at concentrations up to 5×10^{-6} gram per milliliter, has no effect on the spontaneous discharge in the smooth muscle of taenia coli. However, the spontaneous discharge is abolished by Mn^{++} at a concentration of 0.5 millimole per liter. The contraction induced by immersing the muscle in isotonic KCl solution is also suppressed in the presence of Mn^{++} . Because Mn^{++} is a specific suppressor of the spike induced by Ca^{++} and tetrodotoxin is an inhibitor of the spike induced by Na^+ , we suggest that Ca^{++} is a charge carrier in the production of spike potential in the smooth muscle and that the entry of Ca++ through the membrane acts as a trigger for the contraction of smooth muscle.

Holman (1) and Bülbring and Kurivama (1) found that spike height is apparently independent of the external sodium concentration but dependent on the external calcium concentration in the taenia coli. These findings suggest that increase in permeability to Ca++, rather than to Na+, is involved in the spike-generating mechanism in smooth muscle. In crustacean muscle, and particularly in the giant muscle fiber of the barnacle, it has been established that Ca++ is the charge carrier in production of the action potential (2). Further, Hagiwara and Nakajima (3)recently found that Mn++ at relatively low concentration specifically inhibits the Ca++-spike, while tetrodotoxin, a Na+-spike inhibitor (4), was ineffective in abolishing spike electrogenesis in the muscle fiber of the barnacle. Hence we examined the effects of tetrodotoxin and Mn++ on the taenia coli in the hope of throwing further light upon the role of Ca^{++} in the activity of smooth muscle.

Membrane potential and tension were recorded by means of a sucrosegap technique, similar to that employed by Bülbring and Burnstock (5). Taenia coli of the guinea pig was used throughout. The experiments were performed at room temperature (25° to 30°C).

Application of tetrodotoxin at very high concentrations, up to 5×10^{-6} g/ml, in normal Locke's solution or in hypertonic solution, did not change either the resting potential of the membrane or the amplitude of the spontaneous spikes. The drug had no effect on the spontaneously occurring spikes which persisted after the muscle was immersed in Na+-free solution (Bülbring and Kuriyama, 1). These results are summarized in Fig. 1. The effects of epinephrine, that is, the suppression of spontaneous discharge and the inducement of hyperpolarization, were



Fig. 1. Effects of tetrodotoxin on spike amplitude and resting potential. Spike amplitude is shown as the value relative to that of the control. Resting potential is shown as the deviation from the original value; positive values indicate depolarization. Tetrodotoxin was applied at time zero. (Closed circles) Tetrodotoxin $(10^{-6} \text{ to } 5 \times 10^{-6} \text{ g/ml})$ in normal Locke's solution; (crosses) tetrodotoxin $(3 \times 10^{-6} \text{ g/ml})$ in hypertonic solution with twice the concentration of NaCl; (open circles) tetrodotoxin $(3 \times 10^{-6} \text{ g/ml})$ in Na⁺-free solution with tris-hydroxy-methyl aminomethane in place of Na⁺.



Fig. 2. Effect of manganese ions (0.5 mmole/liter) on spontaneous spike discharge and tension. Upper trace is the membrane potential; lower trace, tension. Mn^{++} was washed out at the time indicated by W.



Fig. 3. Effect of manganese ions on contraction induced by potassium. Tension was produced by replacing sodium with isotonic potassium (153.8 mmole/liter) Locke's solution (K⁺-R). After 25 seconds, the potassium solution was washed out (W). Five minutes later, the potassium solution was again applied in the presence of 10 mM Mn⁺⁺ (10 mM Mn⁺⁺K⁺-R). Similar procedures were repeated at the various concentrations of Mn⁺⁺ indicated.

not modified by tetrodotoxin. Tetrodotoxin also lacked influence on the depolarizing action of acetylcholine. Some of these results have already been reported (6).

Though tetrodotoxin was ineffective, application of 0.5 mM MnCl₂ abolished the spontaneous discharge very rapidly, and a decrease of tension followed (Fig. 2). Hyperpolarization, similar to that produced by the application of epinephrine, was not observed. Hence the suppressing effect of Mn++ is quite different from that of epinephrine. If we consider these results together with the observations on the muscle fiber of the barnacle (3). we may suppose that the spike-generating mechanism in the taenia coli is the increase in Ca++-permeability of the membrane. From this view we can well explain the results obtained by Holman and by Bülbring and Kuriyama that the spike height is dependent on the external concentration of Ca++.

In Fig. 3, the inhibitory effect of Mn⁺⁺ on the contraction induced by isotonic KCl solution is illustrated. This result suggests that Mn++ has an inhibitory action on the coupling process between depolarization and tension, as is the case with crayfish muscle fibers (7). Furthermore, the contraction brought about by the application of acetylcholine to a completely depolarized muscle (Evans and Shild, 8) was also blocked by the presence of Mn^{++} . Therefore, in the presence of Mn^{++} , the smooth muscle behaves as though it were placed in Ca++-free solution (Yukisada and Ebashi, 8).

If one accepts the idea that the two different inhibitory actions of Mn^{++} in taenia coli—that is, the inhibition of the spike and the suppression of contraction—arise from one and the same action of Mn^{++} , namely, the inhibition of the Ca⁺⁺ entry through the membrane, it may be considered that the Ca⁺⁺-influx during the spike potential under normal conditions acts as a trigger for the smooth muscle contraction without another intervening process.

> Y. Nonomura Y. Hotta

H. Ohashi

Department of Pharmacology, Faculty of Medicine, University of Tokyo, Tokyo, Japan

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- 24 January 1966

Visual Disappearances Caused by Form Similarity

Abstract. Three forms were scaled for similarity by two groups of observers, who used different methods. A third group reported the duration of disappearances observed for each pair of forms. Duration of total disappearance increased with an increase in form-pair similarity. Neural overlap can explain the similarity judgments; cell fatigue, the disappearances.

Simultaneous disappearance of parallel line segments, and of identical forms, has been reported when a visual image is stabilized on the retina or when the visual stimulus is greatly simplified (1, 2). Identity or similarity is the obvious name for the cause, but the effect has not been demonstrated as a quantitative function of similarity. This experiment shows that a similarity scale predicts the disappearance duration of form pairs observed under reduced stimulation.

Forms were three polygons made by randomly connecting randomly chosen points, with the restriction that the result be a simple closed curve (Fig. 1). The task required subjects to observe either one form or a pair of forms monocularly for 10 minutes. They fixated on an "X" located between the pair of forms, or to one side of the single form.

Sixty-four paid high school and college students of both sexes were divided randomly into six groups. Each group observed one of the single forms or one of the pairs of forms. Subjects were instructed to report disappearances of the forms by saying "out" (single-form groups) or "left out," "right out," or "both out" (pairs), and

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reappearances by saying "in" (or "left in," "right in," or "both in"). The total duration of disappearance for each form was recorded. Subjects were told that they might see (i) no change, (ii) fading, or (iii) complete disappearance of one or both forms. They were instructed to report only complete disappearances. There were two 5-minute observation sessions, separated by a 1minute rest period. The position of the form (or forms) was changed between periods. Single forms were shifted from one side of the "X" to the other, and form pair members were reversed in position. Initial presentation position was counterbalanced within each group. The experimental room (1.8 \times 2.3 m) was illuminated by ceiling fluorescent lights; the walls were creamcolored cinder blocks. Subjects were seated in a chair 0.9 m from the fixation point, which was a 1.27-cm square "X" mounted 139 cm from the floor and centered on a piece of white cardboard (112 cm wide by 71 cm high) attached to the wall. The forms were centered 17.8 cm to either side of the "X," which gave them an angular separation of 15 deg at the viewing distance used. Subject's left eve was blindfolded; the experimenter sat directly behind the subject and recorded the subject's observations on a set of digital counters.

Thirty-two Cornell University undergraduates scaled the entire original set of ten forms (3) for similarity, using a multidimensional similarity ranking analyzed by Torgerson's scaling technique (4). Thirty-nine McGill graduate students and honors undergraduates scaled the three experimental forms for complexity and interstimulus distance, using a magnitude estimation technique. Pair B-C (Fig. 1) was chosen as a standard pair, with a fixed distance score of 10. A hypothetical identical pair was described as the zero anchor of no interstimulus distance. Observers were instructed to give a numerical distance score to pairs A-B and A-C, estimating the value by comparison to zero and pair B-C. Complexity was judged relative to shape C, which was given a standard score of 10. No zero anchor was suggested.

Mean total disappearance duration for each pair, summing disappearances of each form alone with both simultaneously, increased with an increase in form-pair similarity (Table 1, Fig. 1). All differences among mean duration were significant: for pairs AB-AC, t =9.79, df 18; for AB-BC, t = 7.00, df 19; and for AC-BC, t = 5.22, df 19; all P's < .001. Each form disappeared longer when it was paired with the more similar of the two remaining forms (Table 1). The difference between mean disappearance duration of A paired with $B(A_{\rm b})$ and A paired with $C(A_c)$ was significant (t = 2.27, df 18, $P < .05; B_{a}-B_{c}, t = 1.89, df 19, P < .1;$ C_a-C_b , t = 1.38, df 19, P > .1). The Cornell scaling results were transformed to the McGill scale by equating values for pair BC and were plotted with the McGill results against mean total disappearance duration in Fig. 1. The



Fig. 1. Mean disappearance duration as a function of form-pair similarity. 99