

seems to be due to the time relationship between EJP frequency and spontaneous cardiac rhythm, with regard to the membrane potential value, at the times when the evoked EJP's superimposed on the cardiac action potentials, and to refractoriness. Stimuli later than the second and third began to drive the cardiac action potential. There is a spurious impression of summation which is due to the slow rate of repolarization after each cardiac action potential. (The EJP evoked in the quiescent heart⁶ showed that facilitation and summation hardly occurred at a frequency of less than 0.5 cps.) The induced action potential has a plateau of shorter duration and of lesser potential level than the normal one. Furthermore it is noticed that the size of action potential changes alternately, larger and smaller. With a slightly higher frequency (not illustrated) the smaller action potential failed to occur, as the stimulus fell on a slightly earlier phase of the repolarization. Simultaneously, the 'one action potential to one stimulus' relation disappeared and 'one to one' and 'one to two' relationships occurred alternately.

When the interval between stimuli became 0.8 sec, each induced action potential corresponded to 5 or 6 stimuli. An example is shown in Figure C. Large amplitude deflections which occurred just before the spike initiation, are considered to be due to a local response superimposed on the EJP. The amplitude of EJP is variable during one cycle of the action potential. In quiescent preparations⁶ the amplitude of EJP's induced by such a stimulation series gradually increased and reached a steady level. Therefore, the variety of amplitude of EJP's may be attributed to the change of excitability and ionic flux in the muscle cell during one cycle of activity. The duration of the repolarizing phase of action potentials elongates during stimulation. It may be thought that the EJP's delay the repolarization of the membrane potential. In this case, intervals between action potentials before stimulation and during stimulation are about the same as in Figure A, measuring respectively 4.4 and 3.9 sec. Thus the effect of EJP's of such a high frequency on the rhythm of the action potential is weaker than at lower frequency (cf. Figure B), but its effect on the duration of the action potential is stronger.

When the stimulation frequency reached such a level that facilitation and summation occurred sufficiently to set up steady partial depolarization, the rhythm of action potentials was accelerated much as by overall depolarization of muscle membrane potential (unpublished work). This is shown in Figure D, in which the interval between

the stimuli is 0.1 sec. At this frequency apparently the EJP shows summation, and the maximum potential of the cardiac cycle during stimulation becomes less than that of the normal action potential. The individual EJP's could scarcely be distinguished from each other in Figure D. The interval between normal action potentials and those intervals during stimulation are about 4.4 sec and 4.0 sec respectively. Effect of the EJP on the repolarization of action potential is not clearly evident, but the tendency is for each action potential during stimulation to have a prepotential like that of the pacemaker potential. It may be thought that the acceleratory effect at high frequency was brought about chiefly by depolarization of overall membrane potential, due to a summation of EJP's. It was observed in many cases that, in the range of stimulation frequencies at which the EJP is long enough to overlap, increase in stimulation frequency caused increased depolarization and increase in the action potential frequency. Probably the depolarization caused by summation of EJP's, as in the case of depolarizing current applied to the muscle cell, diminishes the potential difference between the maximum hyperpolarizing potential and the threshold potential.

Thus, the acceleratory effect of EJP's on the rhythm of action potentials seems to be due to different modes of action: 1. Individual low frequency EJP's evoke a corresponding action potential, and 2. summation of high frequency EJP's, by depolarizing the overall membrane potential, increases the frequency of the spontaneous action potentials. The former is illustrated clearly in Figures A and B, and the latter in Figure D. Interaction between the two modes produces a complex effect at intermediate frequencies (Figure C)⁹.

Zusammenfassung. Nachweis, dass die Reizwirkung des Herz-Akzeleratornerven beim Molusken *Dolabella auricula* von der Reizfrequenz abhängt. Elektrische Ableitungen von einzelnen Herzkammerzellen zeigen, dass bei niedriger Reizfrequenz jedes nicht-summativ «excitatory junctional potential» ein Aktionspotential des Herzens hervorruft.

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⁹ Contribution from the Shimoda Marine Biological Station, No. 205.

Photoperiod Manipulation to Control Diapause in the Pink Bollworm, *Pectinophora gossypiella*

Light breaks introduced during the period of darkness in a 24-h photoperiodic regimen have a marked effect on continued growth and development or on the induction of diapause in some species of insects¹⁻¹². The present paper reports further information about light breaks that may prove useful to those who are attempting to develop this phenomenon for the control of lepidopterous pests. The pink bollworm, *Pectinophora gossypiella* (Saunders), an important pest of cotton, was chosen as the test insect because the diapause of this species is so highly sensitive to manipulated photoperiods^{9, 13-17}.

Eggs from adults reared in LL (continuous light) were shipped by air from Brownsville, Texas, to the laboratory at Beltsville, Maryland. There the freshly-hatched larvae

were placed in 1-dram glass vials (15×45 mm) three-quarters filled with artificial medium¹⁸, and the vials were then stoppered with cotton plugs. Supplementary medium was added on the tenth day. The vials were divided randomly into 8 lard can containers¹⁹ equipped with 5-watt fluorescent lights that gave 150 lux at insect level and placed in a dark room maintained at $26 \pm 1.5^\circ\text{C}$. The lights in the containers were programmed for the regimens listed in the Table. Insects that had not pupated in 51-55 days were considered to be in diapause. The results are given in the Table and may be summarized as follows:

1. When a short day (LD 12:12) was used, diapause was found to be reduced by the introduction of 15 l-min

Effects of manipulation of photoperiods on the growth and development or diapause of the pink bollworm. Newly hatched larvae were placed on an artificial diet at start of test

Test No.	Condition	Number of insects	Diapause (%)	Confidence limits
1	L 06.00–18.00; D 18.00–06.00 EST; with 15 1-min light pulses each 3 min (15 min total L) from 19.30 to 20.30	34	56	38–73
2	L 06.00–18.00; D 18.00–06.00; with 15 1-min light pulses as before from 03.30 to 04.30	36	33	19–51
3	L 06.00–18.00; D 18.00–06.00 with 15 1-min light pulses as before from 19.30 to 20.30 and from 03.30 to 04.30	71	23	13–34
4	L 06.00–18.00; D 18.00–06.00 with 15 1-min light pulses as before from 23.30 to 00.30	30	87	69–96
5	L 06.00–18.00; D 18.00–06.00	69	78	68–88
6	L 06.00–22.00; D 22.00–06.00	66	5	1–13
7	L 06.00–18.00; D 18.00–06.00 shifted to L 06.00–24.00; D 24.00–06.00 every 2 days	63	83	71–91
8	Multiple shifts			
	2 days L 06.00–18.00; D 18.00–06.00	68	91	82–97
	2 days L 06.00–24.00; D 24.00–06.00			
	2 days L 06.00–18.00; D 18.00–06.00			
	18 h L 06.00–18.00; D 18.00–24.00			
	2 days L 24.00–18.00; D 18.00–24.00			
	30 h L 24.00–18.00; D 18.00–06.00 (Repeat cycle)			

light pulses (separated by 3 min of dark) during the dark period as described in the Table, tests 2 and 3. The results were similar to those obtained by ADKISSON^{9,16} and MINIS¹³ though they used 1-h continuous light breaks rather than light pulses.

2. When a short day (LD 12:12) was used, similar light pulses introduced beginning at 5.5 h after the start of the dark period did not reduce or increase the percentage of diapause compared with an uninterrupted short day regimen (Test 4). These results were also similar to those obtained by ADKISSON^{9,16} who used continuous 1-h light breaks.

3. Alternating long day (LD 18:6) regimens with short day regimens (LD 12:12) every 2 days did not change the percentage of diapause (Test 7). This result is similar to the findings of BELL and ADKISSON¹⁷, who alternated 2 days of LD 12:12 with 1 day of either LD 14:10 or LD 16:8.

4. When an LD 12:12 was alternated with an LD 18:6 (Test 8) by shifting the start of the light period in every other 2-day regimen of LD 18:6, 91% of the insects went into diapause. The results resembled those obtained in Test 7.

5. Factors other than the total amount of light energy per day are involved in determining whether the pink bollworm continues growth and development or goes into diapause.

Short 1-min light pulses introduced over a 1-h period to prevent diapause have some practical advantages over continuous light breaks. No attempt was made to determine the minimum effective duration, number, or intensity of such pulses, but there is clearly an economic advantage to pulses of light instead of continuous light if the method is to be put into use in the field: the short pulses can reduce the total amount of light (energy and cost) without missing the time of peak sensitivity of the insect. Field tests are being conducted utilizing this information in an attempt to reduce the overwintering population of a number of agricultural insect pests by the use of light breaks in early and late portions of the night^{20,21}.

Zusammenfassung. Im 12-h Kurztag gehaltene *Pectinophora gossypiella* gehen zu 78% in Diapause. Zusätzlich

applizierte, einstündige Perioden mit 15 Lichtpulsen von je 1 min Dauer reduzieren die Diapause rate. Die besten Resultate ergeben 2 Pulsperioden, je 1,5 h vor und nach der Hellphase appliziert.

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- 21 We thank C. L. MANGUM, Plant Protection Division, Agricultural Research Service, US Department of Agriculture, Brownsville, Texas 78520, for supplying us with pink bollworm eggs.