TABLE II

Bond	Ba compound	compound	Mn compound	Marsh α glycine	Stosick Ni glycine dihydrate
	Å	Å	Å	Å	Å
C_1–O_1	1·293	1·248	1·254	1·252	1·25
C_1–O_2	1·172	1·247	1·278	1·255	1·29
C_1–C_2	1·526	1·519	1·512	1·523	1·50
C_2–N	1·507	1·481	1·508	1·474	1·42
Bond angles					
O_1–C_1–O_2	124°·2	124°·2	124°·8	125°·5	122°
O_1–C_1–C_2	119°·8	119°·4	119°·5	117°·4	115°
O_2–C_1–C_2	116°·0	116°·4	115°·7	117°·1	123°
C_1–C_2–N	111°·7	111°·5	110°·5	111°·8	103°

the bond lengths and bond angles in the glycine molecule as revealed from the three structures as compared with those got for α-glycine by Marsh[7] and for Nickel Glycine dihydrate by Stosick.[8] The structures seem to be stabilised by a set of hydrogen bonds.

Details of the structure will be published in due course after completing the refinement with diffractometer data.

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1. Albrecht, G. and Corey, R. B., *J. Am. Chem. Soc.*, 1939, **61**, 1087.
2. Kitaigorodsky, A., *Act. Physicochim. U.R.S.S.*, 1936, **5**, 749.
3. Ksanda, C. J. and Tunnell, G., *Am. J. Sci.*, 1936, **35 A**, 173.
4. Iitaka, Y., *Act. Cryst.*, 1958, **11**, 225.
5. Narayanan, P. and Shanta Venkata Raman, *Curr. Sci.*, 1967, **36**, 315.
6. — and —, *Z. Kristallogr.*, 1967, **124**, 460.
7. Marsh, R. E., *Act. Cryst.*, 1958, **11**, 654.
8. Stosick, A. J., *J. Am. Chem. Soc.*, 1945, **67**, 365.

EFFECTS OF PHOTOPERIODISM ON MOULTING IN *HEMIGRAPSUS NUDUS* DANA

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ABSTRACT

In the shore crab, *Hemigrapsus nudus*, light as an environmental regulator of moulting is considered. Absence of light inhibits proecdysis, though increased light conditions do not accelerate it.

INTRODUCTION

LIGHT intensity and periodicity are known to influence crustacean moulting. There is evidence that moulting will be inhibited for several months in *Gecarcinus* if it is subjected to constant illumination (Bliss, 1954), but that darkness would favour its proecdysial growth (Bliss, 1964). *Cambarus* responds to daily photoperiod by an increased tendency to moult in winter months (Stephens, 1955). In *Carcinus* constant illumination retards moulting without affecting the duration of proecdysis (Passano, 1963). Kurup (1963) observed that, in *Hemigrapsus*, continuous illumination is not favourable for moult preparation, though prolonged darkness blocks moulting altogether. Controlled experiments, reported here, have further shown that moult responses of this shore crab, exposed to photoperiod, are considerably varied.

MATERIAL AND METHOD

Specimens of *Hemigrapsus nudus*, collected from Cape Arago, Oregon, U.S.A, were kept in aquarium tanks (2½′ × 2′ × 1½′) containing sand-beds with sea-water. The tanks were continually aerated and were supported on black-painted metal jackets of constant-running freshwater to simulate tide-pool conditions. A thermostat was also suitably installed to maintain a temperature range of 13–15° C. in the tanks. The water jackets were also pro-

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vided with hinged light-proof lids having two 25-Watt fluorescent-lamp fixtures so as to produce 10–40 foot candles of illumination on the water surface in the tanks. The lights were controlled by automatic timers. The animals were fed with dog biscuits or fish scraps, and each day, the tanks were examined for food left overs and dead animals. The sand-beds were also cleaned and the sea-water changed on alternate days, the cleaning time never having exceeded 30 minutes for each tank. The stages of the crabs in the intermoult cycle were determined on the basis of criteria chosen from Kincaid and Scheer (1952), Scheer (1960) and Kurup (1964).

The experiment was done in the winter months, November to December. Crabs in diecdysis (*See* Bliss, 1964) or belonging to C_4 stage (*See* Drach, 1939), of about the same size (3·20–3·60 cm. carapace width) were selected for the experiment, irrespective of sex (all females were non-ovigerous). They were divided into 5 groups (Groups A, B, C, D and E)—each group contained in a separate tank and each tank accommodating 9 animals—and then subjected to different photoperiod responses. Group E functioned as a control as those animals were subjected to 9 hours photoperiod which approximated with the normal daylength of the place in relation to season and latitude. The daylength had a maximum variance of 22 minutes only during the experimental period (Data from Coast and Geodetic Survey, U.S. Department of Commerce). The crabs were numbered on the carapace with a wax pencil to follow up the changes in the intermoult cycle individually. The Drach stages of the test animals were estimated and recorded for the days of the experiment (any time from 8 a.m. to 10 a.m.) and the results on the 12th and 30th days analysed.

Results

Animals in Group A, kept in dark for full 24 hours a day did not show any proecdysial change even on the 30th day and 7 crabs survived the test period. Of the 9 crabs (Group B) maintained under 24-hour illumination, 3 changed themselves into D_0 by the 12th day. One more changed into D_0 by the 30th day, while proecdysis was not even initiated in the remaining 5. The maximum mortality was also observed in this group with only 3 animals (all in D_0) surviving the experimental period. In Group C subjected to 15-hour light and 9-hour dark each day, all the 9 animals were in D_0 on the 12th day; by the 30th day 3 crabs were in D_2 and 6 were in D_1, though 2 animals belonging to the D_1 stage died before completion of the experimental period. In Group D, to which light and dark alternated every 12 hours, 5 crabs showed no signs of proecdysis initiation till the 12th day, while 4 crabs moved on to the D_0 stage. By the 30th day, 3 animals (two in C_4 and one in D_0) were dead and the 6 survivors were all in D_0. Group E exposed to 9-hour light and 15-hour dark did not evince any response, and all the survivors (8 out of 9) remained in the C_4 stage even at the close of the experimental period. The one crab that died during the one-month observation was also in C_4.

For a quantitative assessment of moult induction, based on the results obtained for the 12th and the 30th days, a proecdysis index was calculated by the sum of the duration of stages of responsive animals (stage value) divided by the initial number of experimental animals of each group. The time run from one physiological state to another is approximately known (C_4 to D_0 = 15 days; D_0 to D_1 = 7 days; D_1 to D_2 = 6 days) for a closely related brachyuran genus belonging to the same *Grapsidæ* family, *Pachygrapsus* (Hiatt, 1948) and is applied here as the standard. As the intermoult specimens of *Hemigrapsus* and *Pachygrapsus* are about the same size, the duration of the intermoult cycle of these genera may not be varied (Scheer, 1960; Kurup, 1963). Accordingly, the total stage value of Group C, for example, can be worked out as under:

Days run by 9 C_4 crabs to reach $D_0 = 9 \times 15 = 135$

Days run by 6 D_0 crabs to reach $D_1 = 6 \times 7 = 42$

Days run by 3 D_1 crabs to reach $D_2 = 3 \times 6 = 18$

Total stage value = 135 + 42 + 18 = 195

The total stage value (*i.e.*, 195) divided by the initial number of experimental animals in the group (*i.e.*, 9), which is 21·67, will be the proecdysis index for Group C. Likewise, the index for Group B and D works out to 6·67 and 11·67 respectively and that for A and E to zero. The results of the experiment, including percentage mortality and proecdysis index of the various groups are tabulated in Table 1.

TABLE I

Group	Experimental state	Initial stage	Stage on 12th day	Stage on 30th day	Proecdysis index	Percentage mortality
A	24-hour dark	C_4	C_4	C_4	00·00	22·22
B	24-hour light	C_4	D_0	D_0	06·67	66·67
C	15-hour light	C_4	D_0	D_1-D_2	21·67	22·22
D	12-hour light	C_4	C_4-D_0	D_0	11·67	33·33
E	9-hour light	C_4	C_4	C_4	00·00	11·11

The data were analysed to test whether there is significant variation among the 5 treatments in so far as their effect on proecdysis index is concerned. The variation in the different stages is found significant at the 5% level. However, the treatments do not show significance and it may be due to non-availability of intermediate values. The analysis of variance is presented in Table II.

TABLE II

Source	S.s.	d.f.	m.s.	F calculated	F 5%
Treatments ..	110·10	4	27·5	2·34	3·84
Stages ..	120·80	2	60·4	5·16	4·46
Error ..	93·90	8	11·7	..	..
Total ..	324·80	14	..	..	..

Discussion and Conclusions

The mortality rates in the 5 groups of experimental animals are markedly different; the maximum mortality was observed at the maximum light conditions and the minimum at the shorter photoperiod (9-hour light) which corresponds to the normal daylength of the locality in winter months. Evidently, constant light is lethal to this crab, in spite of it initiating proecdysial responses. The animals are also more resistant to dark environs though it is not favourable to progress towards moult.

The experimental results also demonstrate that daily light ration does affect moulting in *Hemigrapsus*. In controls belonging to Group E (with 9-hour photoperiod) there is no indication of proecdysis and a stage change even in one animal has not been observed (all static at C_4) during the experiment. It might mean that at least in winter months (November to December) the intermoult cycle of this shore crab is longer, though in *Cambarus* the inclination to moult is only stronger in winter than in other seasons (Stephens, 1955). The maximal effect in *Hemigrapsus* is observed in the 15-hour light condition showing a proecdysial index of 21·67. However, the strength of the tendency to moult does not increase with increased light period in the crab; the 24-hour illumination did produce a proecdysis index of 6·7 only as against 11·67 by a 12-hour light period. This too is in contrast to the results on *Cambarus* (Stephens, 1955) where increased illumination was reported to have increased proecdysial changes. Again, continuous absence of light inhibits proecdysis in *Hemigrapsus*, though the condition obtained in *Gecarcinus* is just the reverse (Bliss, 1964). The results on *Hemigrapsus* indicate that the responses are quantitative in relation to photoperiod, a minimum threshold (somewhere between 9-hour and 12-hour daily light) needed to evoke a response notwithstanding. As moulting in crustaceans is controlled by the X-organ-Sinus gland-Y-organ complex (Kurup, 1963), it is also very likely that the pathway of photoperiodic moult induction is through the neuroendocrine system.

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1. Bliss, D. E., *Anat. Record.*, 1954, **120**, 742.
2. —, *Gen. Comp. Endocrinol.*, 1964, **4**, 15.
3. Drach, P., *Ann. Inst. Oceanog.* (Paris), N S. 1939, **19**, 103.
4. Hiatt, R. W., *Pacific Sci.*, 1948, **2**, 135.
5. Kincaid, F. D. and Scheer, B. T., *Physiol. Zool.*, 1952, **25**, 372.
6. Kurup, N. G., *J. Anim. Morphol. Physiol.*, 1963, **10**, 113.
7. —, *Biol. Bull. Woodshole.*, 1964, **127**, 97.
8. Passano, L. M. and Jyssum, S., *Comp. Biochem. Physiol.*, 1963, **9**, 195.
9. Scheer, B. T., *Ibid.*, 1960, **1**, 3.
10. Stephens, G. C., *Biol. Bull. Woodshole*, 1955 **108**, 235.