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# MOULTING HORMONES IN *PALAEMON* (=*LEANDER*) (CRUSTACEA DECAPODA)

## II. DIFFERENCES BETWEEN POPULATIONS

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(Text-figs. 1–3)

Eye-stalk removal in certain species of Brachyura and Astacura has almost invariably led to the initiation of proecdysis. In other decapod crustaceans, however, there have been significant variations in the results of this operation. Thus Travis (1951) found that eye-stalk ablation had no effect on the duration of the moult cycle in *Panulirus argus*. And in the Natantia, even in a single species, quite different results have been reported. Drach (1944) found that eye-stalk removal in *Palaemon* (= *Leander*) *serratus* (Pennant) led, as in crabs, to the initiation of proecdysis and to accelerated moulting. In ignorance of this work I repeated some of the same experiments and found that eye-stalk removal in *P. serratus* led to slower moulting (Carlisle, 1953*a*)—a result quite opposite to that of Drach. Scheer & Scheer (1954) confirm my results in the same species.

Three main types of hypothesis may be advanced to explain these contradictions: (1) that the conditions were different in the laboratories of the three groups of workers, that is to say in the laboratories of Roscoff, Plymouth and Naples; (2) that the experimental techniques adopted were different—for instance, animals might be fed or starved; (3) that the population of prawns in the three laboratories might be different and the differing results be inherent in the material.

In order to decide between these hypotheses some hundreds of *Palaemon serratus* collected at Roscoff—the laboratory where Drach performed his experiments—were brought to Plymouth and duplicate experiments were performed on these and on indigenous Plymouth specimens.

At the outset it is worthy of remark that after a little practice it was possible to distinguish between the prawns from Plymouth and from Roscoff on the basis of the colour patterns (Carlisle, 1955). Certainly about 30% of each population could be confused, but the remaining 70% were clearly different. Moreover, in colour photographs of Naples prawns, taken by Sir Francis Knowles, it was easily seen that these prawns were different again. The

differences were slight and not such as would satisfy a systematist for specific, or even subspecific, characterization, but they were nevertheless real, despite the overlap between the populations.

## EXPERIMENTAL DATA AND CONCLUSIONS

The prawns were kept in running sea water in the aquarium tanks at Plymouth, either as a group of prawns in a large tank or singly in cages. The cages were made of $\frac{1}{4}$ in. mesh stainless steel, and each was 1 m square by 10 cm deep and divided into 100 cubical compartments of 10 cm side. A single prawn was placed in each compartment and the whole cage lowered into an aquarium tank. Prawns were fed daily on the flesh of *Mytilus*. For experimental purposes prawns were selected between 55 and 70 mm length measured from the tip of the rostrum to the tip of the telson. Eye-stalk ablation was performed by electrocautery; a single eye-stalk was removed on one day and the other on the succeeding day. Under these conditions the survival rate throughout the experiment was better than in earlier experiments (see Carlisle, 1953*a*).

Four groups of prawns were used: group P1—Plymouth prawns with eye-stalks removed; group P2—intact Plymouth prawns; group R1—Roscoff prawns with eye-stalks removed; group R2—intact Roscoff prawns.

In Fig. 1 is illustrated the variation of intermoult period in these four groups with alteration in temperature (small symbols). The points for the Plymouth prawns represent my own earlier experiments and those for the Roscoff prawns are plotted from the data of Drach (1944). The lines were drawn through the points by eye.

In the first experiment with these four groups of prawns a number of prawns were kept through two successive moults and the interval between these moults recorded. The temperature throughout this experiment was $13{\cdot}7 \pm 1{\cdot}1^{\circ}$ C. The results are summarized in Table 1 and diagrammatically in Fig. 2. The mean intermoult periods for the four groups are:

$$\bar{t}_{P1} = 51{\cdot}140 \pm 0.933,\ \bar{t}_{P2} = 35{\cdot}513 \pm 0{\cdot}715,$$
$$\bar{t}_{R1} = 17{\cdot}356 \pm 0{\cdot}494,\ \bar{t}_{R2} = 22{\cdot}206 \pm 0{\cdot}492.$$

where $t$ is time in days. From this it may be calculated that the probability, $P$, that these four groups are drawn from the same population is less than 0·001. For all the individual comparisons, except $\bar{t}_{R1} \sim \bar{t}_{R2}$, $P < 0{\cdot}001$; for $\bar{t}_{R1} \sim \bar{t}_{R2}$, $P = 0{\cdot}001$. It is thus quite evident that all four groups are very significantly different. These four means are entered into Fig. 1 as the four large symbols in line with the temperature 13·7° C. It will be seen that they fit well with the curves drawn from the earlier data.

In the second experiment a larger number of prawns was used for a shorter period and the moulting rate followed over 15 days. The temperature during

this experiment was 12·1 ± 0·4° C. The numbers of deaths and of moults in each group on each day of the experiment are listed in Table 2 and the moult rate illustrated graphically in Fig. 3. From these data mean intermoult periods can be calculated for the four groups:

$$\bar{t}_{P1} = 103{\cdot}077, \bar{t}_{P2} = 42{\cdot}118,$$
$$\bar{t}_{R1} = 17{\cdot}092, \quad \bar{t}_{R2} = 23{\cdot}567$$

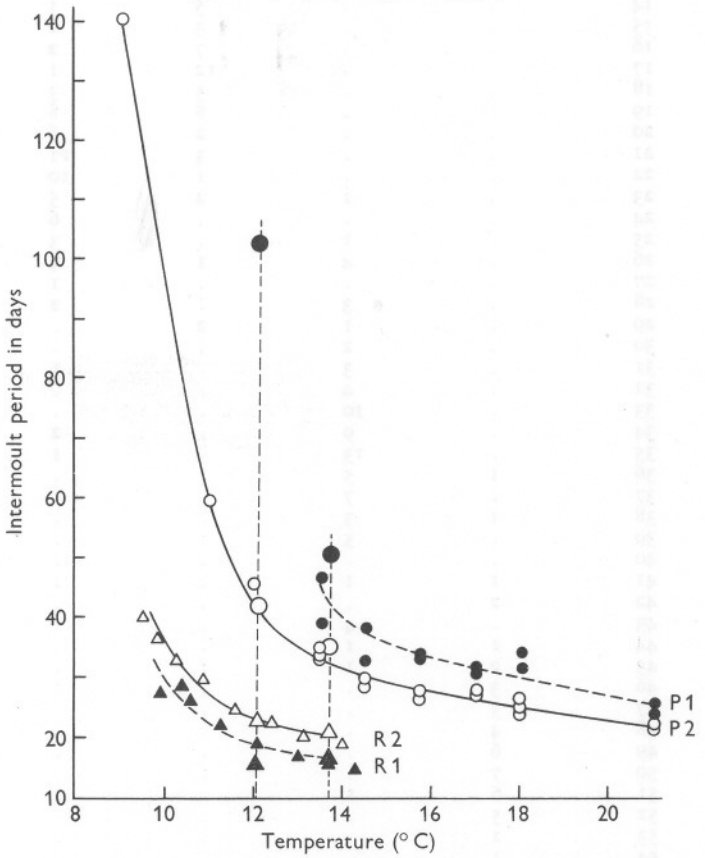


Fig. 1. Graph of the intermoult period of *Palaemon serratus* plotted against temperature. ●, P1: prawns from Plymouth whose eye-stalks had been removed; ○, P2: intact prawns from Plymouth; ▲, R1: prawns from Roscoff whose eye-stalks had been removed; △, R2: intact prawns from Roscoff. The lines are drawn through the small symbols by eye. Lines P1 and P2 represent my data; lines R1 and R2 are replotted from the data of Drach (1944). For the large symbols see the text.

These means are entered into Fig. 1 as the four large points in line with temperature 12·1° C. Once more the fit of the curves from the earlier data, extrapolated where necessary, is good. To calculate the significance of the differences in the moult rates in this experiment it is necessary to compute

TABLE 1

| Days between successive moults | Number of prawns in | | | |
|---|---|---|---|---|
| | Group P1 | Group P2 | Group R1 | Group R2 |
| 10 | . | . | 1 | . |
| 11 | . | . | 2 | . |
| 12 | . | . | 1 | . |
| 13 | . | . | 4 | . |
| 14 | . | . | 5 | 1 |
| 15 | . | . | 3 | 1 |
| 16 | . | . | 7 | 2 |
| 17 | . | . | 12 | 1 |
| 18 | . | . | 5 | 3 |
| 19 | . | . | 8 | 8 |
| 20 | . | . | 2 | 6 |
| 21 | . | . | 3 | 11 |
| 22 | . | . | 1 | 10 |
| 23 | . | 1 | 2 | 5 |
| 24 | . | . | . | 6 |
| 25 | . | 1 | . | 3 |
| 26 | . | 4 | 1 | 4 |
| 27 | . | . | . | 1 |
| 28 | . | 3 | . | 2 |
| 29 | . | 1 | 2 | . |
| 30 | . | 2 | . | . |
| 31 | . | 3 | . | 1 |
| 32 | . | 4 | . | . |
| 33 | . | 10 | . | . |
| 34 | . | 9 | . | 2 |
| 35 | . | 13 | . | 1 |
| 36 | 1 | 5 | . | . |
| 37 | . | 7 | . | . |
| 38 | 1 | 3 | . | . |
| 39 | . | 5 | . | . |
| 40 | . | . | . | . |
| 41 | 1 | 1 | . | . |
| 42 | 2 | . | . | . |
| 43 | . | 4 | . | . |
| 44 | 1 | 1 | . | . |
| 45 | 2 | 1 | . | . |
| 46 | 3 | . | . | . |
| 47 | 3 | . | . | . |
| 48 | 4 | . | . | . |
| 49 | 6 | . | . | . |
| 50 | 7 | . | . | |
| 51 | 6 | 1 | . | . |
| 52 | 5 | . | . | |
| 53 | 2 | . | . | . |
| 54 | 1 | . | . | . |
| 55 | 1 | . | . | . |
| 56 | 3 | 2 | . | . |
| 57 | . | . | . | . |
| 58 | . | . | . | . |
| 59 | 1 | . | . | . |
| 60 | . | . | . | . |
| 61 | 2 | . | . | . |
| 62 | 1 | 1 | . | . |
| 63 | . | . | . | . |
| 64 | 1 | . | . | . |
| 65 | . | . | . | . |
| 66 | . | . | . | . |
| 67 | . | . | . | . |
| 68 | 1 | . | . | . |
| 69 | 1 | . | . | . |
| 70 | . | . | . | . |
| 71 | . | . | . | . |
| 72 | . | . | . | . |
| 73 | . | . | . | . |
| 74 | 1 | . | . | . |

parallel probit lines and obtain probabilities by the modified mean probit difference method outlined by Carlisle & Dohrn (1953). The various possible comparisons and the corresponding probabilities are:

$$\Delta_{(a_{P1} \sim a_{P2})} = 0{\cdot}47 \pm 0{\cdot}14;\ P < 0{\cdot}001,$$
$$\Delta_{(a_{P1} \sim a_{R1})} = 0{\cdot}97 \pm 0{\cdot}15;\ P < 0{\cdot}001,$$
$$\Delta_{(a_{P1} \sim a_{R2})} = 0{\cdot}79 \pm 0{\cdot}14;\ P < 0{\cdot}001,$$
$$\Delta_{(a_{P2} \sim a_{R1})} = 0{\cdot}50 \pm 0{\cdot}12;\ P < 0{\cdot}001,$$
$$\Delta_{(a_{P2} \sim a_{R2})} = 0{\cdot}32 \pm 0{\cdot}11;\ P < 0{\cdot}01,$$
$$\Delta_{(a_{R1} \sim a_{R2})} = 0{\cdot}18 \pm 0{\cdot}11;\ P = 0{\cdot}1,$$

where $a$ is the mean probit and $\Delta$ the mean probit difference. All these differences except the last were highly significant.

We may conclude from these two experiments that the differences found between the four groups of prawns by independent workers in different laboratories are genuine differences inherent in the prawns, and not merely an expression of differing conditions or differing experimental techniques.

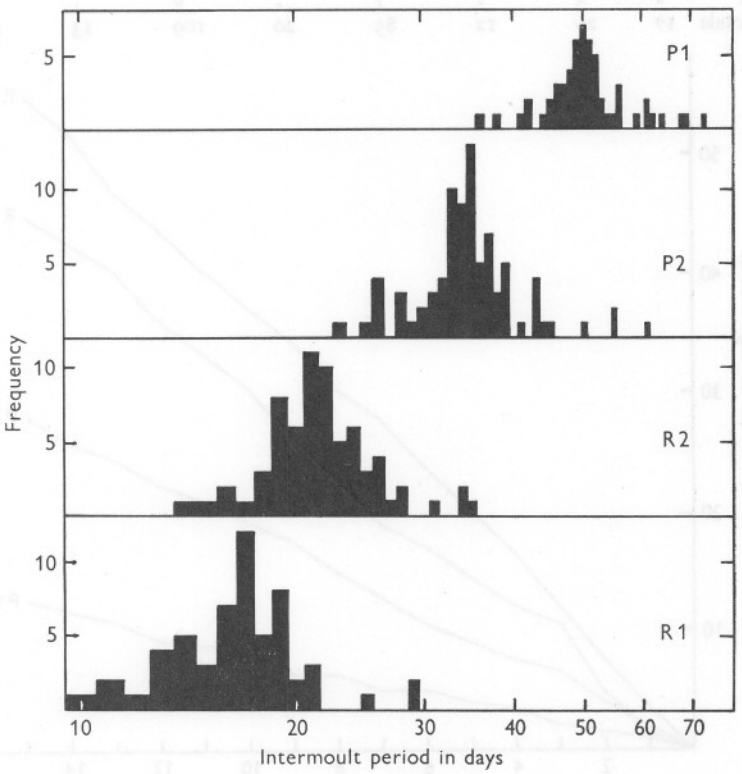


Fig. 2. Histograms drawn from the data of Expt. 1. The four groups of prawns are numbered as in Fig. 1. The time scale is logarithmic. Temperature 13·7° C.

TABLE 2. DEATHS AND MOULTS IN EXPERIMENT II

The number of animals which died without first moulting and of those which moulted on each day in each group.

| Group... | P1 | | P2 | | R1 | | R2 | |
|---|---|---|---|---|---|---|---|---|
| No. of animals... | 205 | | 301 | | 198 | | 297 | |
| Day | Dead | Moults | Dead | Moults | Dead | Moults | Dead | Moults |
| 1 | 5 | 2 | 2 | 6 | 3 | 7 | 3 | 4 |
| 2 | 3 | 1 | 5 | 6 | 4 | 8 | 5 | 10 |
| 3 | . | 1 | . | 8 | 3 | 9 | . | 17 |
| 4 | 3 | 3 | . | 3 | 2 | 9 | . | 5 |
| 5 | . | 1 | . | 4 | . | 7 | 2 | 7 |
| 6 | . | 2 | 1 | 5 | . | 7 | . | 8 |
| 7 | 2 | . | . | 4 | 2 | 8 | . | 6 |
| 8 | . | 3 | . | 9 | . | 5 | . | 10 |
| 9 | . | . | . | 8 | 3 | 8 | 1 | 11 |
| 10 | 2 | 5 | 3 | 5 | . | 6 | . | 13 |
| 11 | 1 | . | . | 5 | 1 | 6 | . | 8 |
| 12 | . | 1 | . | 4 | . | 7 | 1 | 9 |
| 13 | . | 3 | 1 | 7 | . | 6 | . | 12 |
| 14 | . | 2 | . | 4 | 2 | 10 | 1 | 7 |
| 15 | 1 | 2 | . | 7 | . | 6 | . | 7 |
| Totals | 17 | 26 | 12 | 85 | 20 | 109 | 13 | 134 |

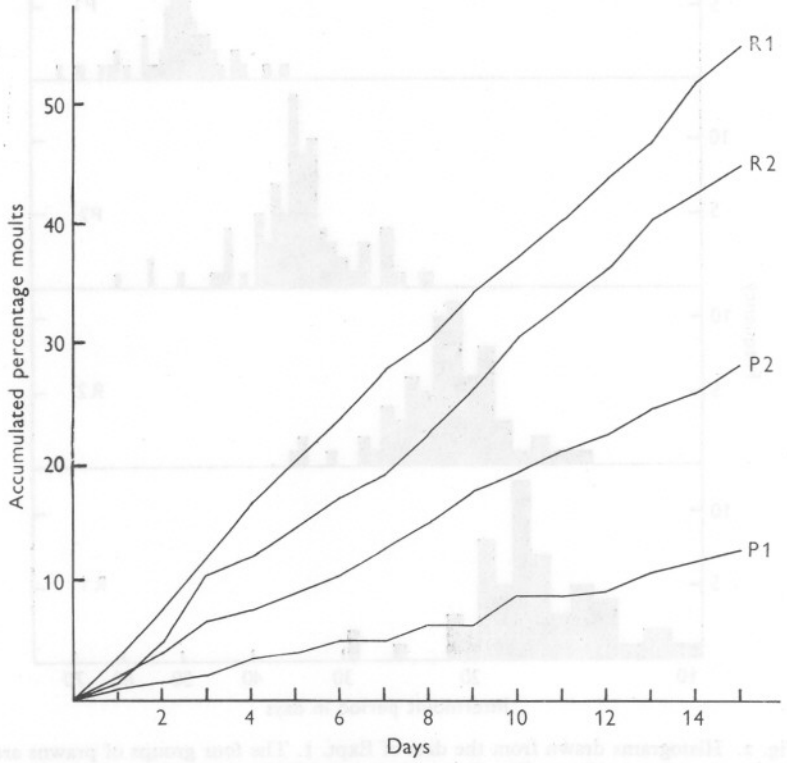


Fig. 3. Graph of the cumulative percentage of moults in Expt. 2. The four groups of prawns are numbered as in Fig. 1. Temperature 12·1° C.

There is a real difference in moult rate between intact *Palaemon serratus* from Plymouth and from Roscoff and the operation of eye-stalk ablation performed on these two populations has quite opposite effects leading to a longer intermoult period in Plymouth animals and to a shorter intermoult period in Roscoff prawns.

## DISCUSSION

There can be little doubt that the results of the experiments of Drach (1944) indicate the existence of a moult-inhibiting hormone in the eye-stalks of *Palaemon serratus*. On the other hand, the results of Carlisle & Dohrn (1953) and Scheer & Scheer (1954) indicate the presence of a moult-accelerating hormone in the eye-stalks of *Palaemon*, a hormone which accelerates the processes of proecdysis. It must not be thought that these two hormones are mutually antagonistic, for the moult inhibiting hormone inhibits the onset of proecdysis, while the moult accelerating hormone accelerates the progress of proecdysis once this has begun. Both hormones, however, will affect the duration of the intermoult period; the moult inhibiting hormone by lengthening diecdysis will increase the intermoult period, while the moult-accelerating hormone by shortening proecdysis will shorten the intermoult period. Thus, in a certain measure, the length of the intermoult period will depend on the relative rates of secretion of these two hormones, and on the rates of secretion of the moult-inhibiting and moult-accelerating hormones which are known to be produced in other parts of the body (Carlisle, 1953*b*, 1954; Scudamore, 1947; Stephens, 1951). The effects of eye-stalk ablation on the duration of the intermoult period will also depend on the relative rates of secretion of these various hormones. Here then we have a possible mechanism for the different moult rates in two populations of prawns and for the opposite effects that eye-stalk ablation has on these two populations. Presumably the differences between the populations are genotypic, for besides the differences in moult rate there are also differences in colour pattern and even in the precise topography of the endocrine organs of the eye-stalk—those organs which are responsible for the secretion of the moulting hormones. This is true also for Naples prawns which are different in both these respects from Plymouth and from Roscoff prawns.

I wish to thank Prof. P. Drach for his helpful criticisms and comments and the staff and fishermen of the Station Biologique de Roscoff for their friendly collaboration.

## SUMMARY

The populations of *Palaemon* (= *Leander*) *serratus* from Roscoff, Plymouth and Naples are noticeably different in the range of colour patterns, though there is some overlap between the populations from Plymouth and Roscoff at least. The mean intermoult period at any one temperature is different for these

two populations, even if kept in adjacent tanks in the Plymouth aquarium; the intermoult period is shorter for prawns from Roscoff. Eyestalk ablation leads to significant shortening of the intermoult period in prawns from Roscoff but to significant lengthening of this period in prawns from Plymouth, even when duplicate experiments are carried out in the one laboratory on collections of prawns from the two localities. An endocrinological explanation of these experiments is proposed.

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## APPENDIX

Some of the data from which curves P1 and P2 in Fig. 1 are drawn have been published (Carlisle, 1953*a*). Most of them, however, have not. As explained in the paper mentioned above it is possible to measure the intermoult period either directly or by calculation from the moult rate measured in a group of prawns over a period of days. By the latter method it is not possible to assign a standard error to the mean intermoult period, although it is possible to assign one to the difference between moult rates. Points on the graph (Fig. 1) have been obtained by both these methods. In Table 3 are given the mean intermoult periods corresponding to each of the points on curves P1 and P2, together with a standard error where this is appropriate.

### TABLE 3. INTERMOULT PERIODS

Table of the data which generated the curves P1 and P2 in Fig. 1. Where a standard error of the mean is given the mean intermoult period was derived from a number of direct observations of individual intermoult periods. Where no standard error is given the intermoult period is calculated from the moult rate measured in a number of animals over the stated number of days.

| | P1 | | P2 | |
|---|---|---|---|---|
| Temperature (° C) | Intermoult period (days) | Number | Intermoult period (days) | Number |
| 9·0 | . | . | 141·12 ± 11·27 | 93 |
| 10·0 | . | . | 94·63 ± 11·01 | 112 |
| 11·0 | . | . | 60·10 ± 7·33 | 108 |
| 12·0 | . | . | 45·95 ± 4·95 | 101 |
| 13·5 | 39·40 ± 2·71 | 5 | 33·78 ± 1·58 | 9 |
| | 35·46 | (100 × 26 days) | 34·62 ± 3·19 | 127 |
| | | | 47·30 | (100 × 26 days) |
| 14·5 | 33·10 | (150 × 15 days) | 29·13 ± 2·01 | 18 |
| | 38·74 ± 3·01 | 16 | 30·10 | (150 × 15 days) |
| 15·8 | 33·87 | (100 × 15 days) | 26·28 ± 1·99 | 12 |
| | 34·30 ± 2·76 | 9 | 27·00 | (100 × 15 days) |
| 17·1 | 31·12 ± 2·81 | 15 | 27·22 | (100 × 15 days) |
| | 32·13 | (100 × 15 days) | 27·84 ± 2·02 | 20 |
| 18·0 | 31·94 ± 2·11 | 21 | 24·23 ± 0·76 | 132 |
| | 34·50 | (100 × 15 days) | 25·66 ± 1·94 | 24 |
| | | | 26·47 | (100 × 15 days) |
| 21 | 24·11 ± 0·83 | 33 | 21·81 | (100 × 15 days) |
| | 25·97 | (100 × 15 days) | 22·79 ± 0·68 | 35 |