A Comparison of the Methods of Molt Staging According to Drach and to Adelung in the Common Shore Crab, Carcinus maenas

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(Z. Naturforsch. 29 c, 754-756 [1974]; received August 5, 1974)

Molt Staging, Molting Hormone, Crabs

Molt stages of juvenile *Carcinus maenas* were determined independently according to the methods of Drach and of Adelung. With the exception of stage DO the two systems agree rather well over the whole intermolt cycle. The molting hormone contents of animals regenerating several limbs and whose molt stages were determined according to Drach are in the same range as the values published by Adelung.

A prior condition for studying the molting physiology of crustaceans was the subdivision of the molting cycle into a series of accurate molt stages. Criteria for this subdivision were first given by Drach ¹ and then applied to several malacostraca ²⁻⁵ and even to the so-called entomostraca ⁶⁻⁸. For different crustacean species the initial concept of Drach was modified and more defined ^{9, 10}.

Another method for subdividing the molting cycle in crustaceans was introduced by Adelung ¹¹ for his studies on juvenile Carcinus maenas. Adelung used the time, the rigidity of parts of the body and especially the regeneration of amputated legs as criteria for his staging method. Amputation of the legs was done in order to shorten the molting cycle and to synchronize the crabs. Animals treated in this way show a very constant time interval from one exuviation to another, the length of which is size-dependent. This allows a subdivision of the intermolt cycle into 21 stages.

So far no direct comparison of the two methods of molt staging had been made. The purpose of this investigation was therefore to compare both methods for one species in order to determine whether they correspond or not and to show advantages and disadvantages of each method.

Material and Methods

Carcinus meanas specimens with a carapax width of 18 to 28 mm were used. The animals had completed at least two molting cycles under controlled laboratory conditions. 7 of the 8 walking legs were amputated immediatly after the exuviation. The intermolt stages were determined independently by

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the methods of Drach and of Adelung. The animals were weighed and stored at $-30\,^{\circ}\text{C}$ until extracted for hormone determination. The extraction procedure was the same as used by Adelung ¹². The hormone content of the extracts was determined using the house fly bioassay ¹³.

Results

Fig. 1 shows the results of a comparison of the two methods. With the exception of DO, the other stages determined by the method of Drach are directly comparable to the stages determined by

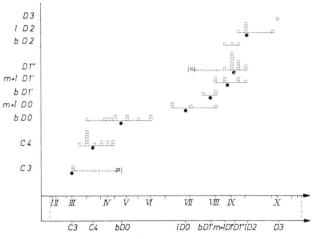


Fig. 1. Molt staging in Carcinus maenas according to Drach (C3 to D3; b = beginning, m = middle, l = late) and to Adelung (I to X). One square corresponds to one determination.

■ Means of the stages determined according to Drach. Two determinations are figured in brackets, they are not included in the corresponding means.

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Adelung. The molting hormone content was determined from 26 out of the animals of Fig. 1. The results of these determinations were compared with the titer curve previously published by Adelung ¹¹. Only stage C4 shows higher values as compared to the surrounding stages III b and III c. In all other stages the new determined values are in the same total range as has been found by Adelung.

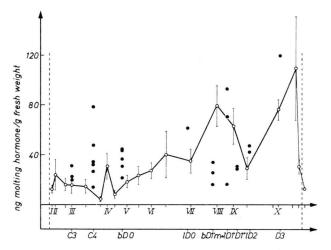


Fig. 2. Molting hormone content of Carcinus maenas during the molting cycle (○-○). Means ± S.E.M. Titer curve published by Adelung 11. ■ Determinations of the molting hormone content of Carcinus maenas. The molt stages were determined according to Drach, using the same extraction and bioassay as Adelung. One point represents one determination. The same x-axis as in Fig. 1.

Discussion

So far no direct comparison of the molt staging methods according to Drach and to Adelung has been made. It was therefore difficult to compare titers of metabolic and endocrine parameters of crabs during a molting cycle obtained by using two different systems of molt staging. From our results it is now possible to compare such investigations since we were able to show that these two systems are in good agreement, at least in the crab Carcinus maenas. With the exception of the range from C4 to late DO both systems show very close time intervals from one molting stage to the next one and only between C4 and late DO does the method of Adelung give a better separation.

From our few determinations of molting hormone content in *Carcinus maenas* at different molt stages it cannot be concluded whether the subdivision of the molting cycle according to Drach would give

the same curve for hormone titer during the whole intermolt cycle as the one published by Adelung. There may be a different trend at C4 and beginning DO as compared to the corresponding range III b, c to V since this is the only period in which the new determinations are not in the same range as the values published by Adelung. It should be noticed that within this period (stages III b to V) stage IV is at the middle point between C4 and beginning DO. From the animals staged "IV" according to Adelung, some can be staged "C4" and the others "beginning DO" according to Drach. Since the higher values in hormone content, that are cumulated at stage IV, spread over C4 and beginning DO, the rise in hormone content seems to be in a closer relationship with the onset of the regeneration process (which is a criterium for stage IV) than with the epidermal freeing from the old cuticle in the branchial epipodites (which is the criterium for beginning DO).

Since both systems of subdividing the molting cycle are directly comparable the question is whether one of the two systems shows advantages over the other one and in what case they should be used. Drach's staging method appears now as a basic reference and it is useful for comparisons between the results from different workers and different species. It can be immediately applied on freshly captured animals but in brachyourans it cannot provide a continuous control of the molt cycle of individual specimens because a small part of a maxilliped has to be cut off, and this can be done only twice on the same animal. Molt staging according to Adelung gives a closer subdivision at the end of the intermolt and the beginning premolt. The determination is very rapid, easy to learn and does not demand any microscopical investigation of parts of the body. There is nearly no disturbance at the moment of molt staging. This method is advantageous in investigating rapidly changing endocrine and metabolic events in late intermolt and early premolt. On the other hand the method of Adelung cannot be used generally. It is only applicable when constant laboratory conditions are given including constant temperature, pH, salinity and nourishment. Furthermore 7 of the 8 walking legs must be amputated when using the method of Adelung. This has the advantage of shortening the molting cycle and of synchronizing the animals but simultaneous regeneration of numerous walking legs is known to promote molt preparation in some species, almost as effectively as eyestalk removal 14 and even, as reported recently 15, in Y-organless specimens of Pachygrapsus. Intensive regeneration, which is at least in part a molting hormone requiring process, may cause a rise of the molting hormone content as compared to normal animals. Whether this is true

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or not cannot be answered since no experimental study on this subject has been made so far.

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