DIFFERENCES BETWEEN METHODS OF REGENERATION

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UDC 612.6.03

Regeneration of the anal filaments in larvae of the Ephemeroptera, taking place by endomorphosis, stimulates molting. During regeneration of the extremities (by morphallaxis) this stimulation is less marked.

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At the present time three principal types of methods of reparative regeneration are known: morphallaxis, epimorphosis, and endomorphosis.

In morphallaxis, regeneration takes place by redevelopment of the remnant of the organ into a complete structure of correspondingly smaller size. In this process cell proliferation plays a minimal role, and the process is accomplished mainly by displacement of cell material. This method is found more frequently in lower animals and is completely unknown for vertebrates. It is probably one of the primitive, oldest types of regeneration.

In epimorphosis, the organ is restored by the formation of a regeneration bud on the wound surface, sharply demarcated from the stump, and becoming differentiated only gradually. This process takes place by migration and intensive proliferation of cell material. Epimorphosis is the most widespread method and is found in nearly all groups of animals capable of regeneration.

Endomorphosis is the type of regeneration which takes place by proliferation of cell material in the remnant of an organ, but not of a wound surface. For this reason, the line of amputation usually remains clearly visible for a long time, and the regeneration affects the mass of the organ rather than its shape. One variant of this method of regeneration was first described by M. A. Vorontsova [2] during restoration of the internal organs of vertebrate animals (regeneration hypertrophy). Endomorphosis is widespread as a means of regeneration of the internal organs of vertebrate animals, but in invertebrates it has been described only among larvae of the Ephemeroptera [3].

It must be emphasized that these various methods of regeneration are frequently combined. However, as a rule, one type remains predominant. An example of this is regeneration in the turbellarian Dugesia lugubris [7]. Sometimes even homonymous organs in different animals regenerate by different methods [3, 5, 8, 9]. Finally, in animals of the same species, the same organ may regenerate by different methods under different conditions [4, 6].

It will be clear from the definitions given above that the various methods of regeneration differ significantly in their morphology, and it may therefore naturally be assumed that they may differ equally in other manifestations. To discover whether this is so, suitable indicators must be found to allow the appropriate differences to be readily discovered.

In the present investigation the effect of regeneration of different types on the time of onset of molting was investigated in insect larvae.

The earlier onset of molting under the influence of reparative regeneration has often been demonstrated in various insects [3, 5, 8]. However, it is not yet clear whether this applies to all types of regeneration or simply to one of them.

Course in General Biology, Faculty of Medical Biology, N. I. Pirogov Second Moscow Medical Institute (Presented by Active Member of the Academy of Medical Sciences of the USSR N. A. Kraevskii).
Translated from Byulleten' Éksperimental'noi Biologii i Meditsiny, Vol. 65, No. 4, pp. 98-100, April, 1968. Original article submitted September 15, 1966.

EXPERIMENTAL METHOD

Larvae of the ephemeropteran Cloeon dipterum were used, in which the limbs regenerate by morphallaxis and the anal filaments by endomorphosis [3]. Larger larvae, in which the rudimentary wings were still small, were chosen for the experiment. In addition, the most careful examination was made to ensure that no specimens with even the slightest defects of the anal filaments, limbs, tracheal gills, and antennae were included in the experiment.

Two series of experiments were performed. In series I, two anal filaments were amputated at the base by an ophthalmic scalpel. In series II, two limbs were removed by inducing autotomy at the junction between the femur and trochanter. The two anal filaments correspond approximately in mass to the two limbs. In series III, the control, the larvae were intact. All the animals were kept individually in small jars with 30 ml water. A few filamentous algae were placed into each jar. Twice a day, in the morning and evening, the jars were examined and larvae which had molted were immediately operated upon or transferred to the control series. In this way the experiment began immediately after molting. Regeneration of the removed organs took place normally, as described in a previous paper [3].

Subsequent examinations were carried out with an accuracy of up to 12 h in order to determine the length of the interval between molting. The numerical results obtained in all three series were subjected to statistical analysis by Student's method.

EXPERIMENTAL RESULTS

In the control series (19 larvae) molting took place on the average after 5.84 days, in series I (53 larvae) after 4.98 days, and in series II (49 larvae) after 5.43 days.

The difference between the molting times in series I and II was 0.45 days, which is statistically significant ($P = 0.0007$). The difference between these times for series I and the control is also significant, amounting to 0.86 days ($P < 0.0001$). The difference between this index for series II and the control was 0.41 days ($P = 0.045$). In this case the difference is not significant. In other words, no evidence of stimulation of molting by removal of the limbs was obtained.

The results thus confirm the view that differences exist between regeneration processes taking place by endomorphosis and morphallaxis. Endomorphosis stimulates molting considerably, while morphallaxis, if it stimulates molting at all, does so to a much lesser degree.

It is difficult to state the causes of these differences. Since molting in insects is dependent on the hormones of the corpora allata, it may be suggested that regeneration by endomorphosis has a stimulant effect on them, whereas in morphallaxis this effect is hardly present. Possibly during regeneration by morphallaxis, when migration and redifferentiation of cells play a leading role, the regenerative processes are more local in character. On the other hand, with the intensive proliferation and hypertrophy typical of endomorphosis, the process is more general in character. This hypothesis is confirmed, in particular, by the fact that after removal of a large proportion of the liver, which regenerates by endomorphosis, from mice, changes take place in the mitotic activity in the cornea and epidermis [1], i.e., regeneration by endomorphosis has a remote action on various processes in the body.

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