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THE NEMATOCYST OF HYDRA (Part V).

THE EXCHANGE OF CNIDOBLAST CELLS BETWEEN TWO DIFFERENT SPECIES OF HYDRA

by

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INTRODUCTION.

There appears to be no single case on record whereby it has been possible to produce a permanent graft between hydra of two different species. BRIEN (1955) has conclusively demonstrated that although two species of hydra may show a great tolerance for one another after they have been grafted, there is no free tissue exchange between the species in question. This author demonstrated, for instance, that a hydra of one species in not able to furnish interstitial cells for the maintenance and repair of the

tissues of another animal. Although the grafted portions may remain in contact for several weeks, there is never cellular compatibility; eventually one member of the graft will grow and thrive while the other member becomes progressively smaller and eventually atrophies. In other instances the grafted portions may separate after a few weeks time. Moreover, BRIEN has shown that some species of hydra show virtually no tolerance for one another and separate within a day or two after the graft has been accomplished. The author has found this to be quite true in cases where an attempt has been made to graft the green hydra, *Chlorohydra viridissima,* to hydra of other species.

Although it has been clearly demonstrated that unspecialized cells such as interstitial cells are not able to function in another species of hydra, it appeared that there may, in fact, be a possibility that a specialized cell from one species of hydra is capable of carrying on its normal function in the tissues of another species.

The cell chosen for the present study was the cnidoblast cell. Unlike other specialized cells such as gland cells and nerve cells, the cnidoblast cell of one species may be readily identified from the cnidoblast of another species because of structural differences in their enclosed nematocysts. In certain cases these differences may be striking and can easily be recognized even under low magnifications; in other instances a critical examination is required before a positive identification can be made.

In our laboratory we have cultured two different species of hydra whose stenoteles differ radically. This paper will report a series of experiments designed to demonstrate that cnidoblast cells from one species of hydra are capable of functioning normally in the tissues of another species.

MATERIALS AND METHODS.

Two species of hydra served as experimental animals for the present study. One species was *Pelmatohydra oligactis* (Pallas). The other was a new species which has not been classified at the present time; in this paper it will be referred to as Species X. For a general description of Species X the reader may refer to the previous paper in the present series (*The Nematocyst of Hydra,* Part IV).

Numerous grafts were performed throughout the course of these experiments. These grafts were similar to the grafts described in the previous paper of this series (*The Nematocyst of Hydra,* Part IV). This paper also gives a description of the histological techniques which were employed in the present investigation.

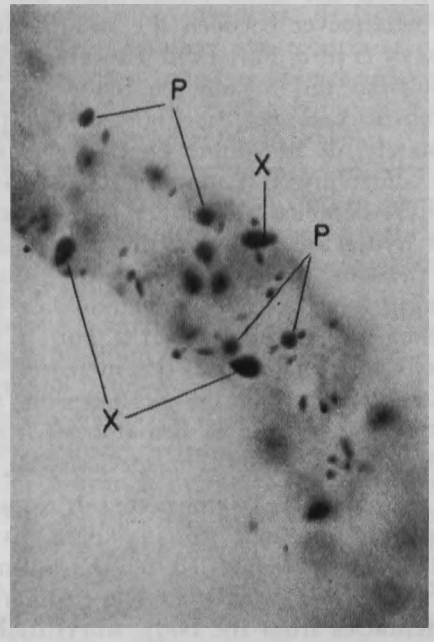

FIG. 1.
This photograph shows several nematocysts (stenoteles) characteristic of *P. oligactis* in the tentacle of Species X six days after the distal portion of Species X had been grafted to the proximal portion of *oligactis*.
P : stenotele of *oligactis*. - X : stenotele of Species X.

RESULTS AND CONCLUSIONS.

A method has been described (See *The Nematocyst of Hydra,* Part IV) whereby it is possible to inhibit the formation of nematocysts in Species X by grafting the distal portions of this species to the proximal portions of *Pelmatohydra oligactis*. Six days following the graft the tentacles of Species X are devoid of nematocysts except at their extremities. During the course of these investigations, however, it was noticed that in many animals, although there was no further production of nematocysts

in the growth region of Species X, that numerous stenoteles were present in the lower regions of the tentacles after a period of five days. Close examination of whole mounts, squash preparations of living animals, and serial sections revealed that the stenoteles in the lower regions of the tentacles of Species X were nematocysts characteristic of *P. oligactis.*

In many cases, on the other hand, there was no migration of nematocysts whatsoever between the two species (See Fig. 2, *The Nematocyst of Hydra,* Part IV). However, when the nematocysts of *P. oligactis* did invade the tissues of *Species X* the migration was an intense one (See Fig. 1). The present experiments, unfortunately, do not afford enough evidence to explain the mechanism controlling this more or less « sporadic » migration. Jones (1941) demonstrated quite conclusively that when the tentacles of hydra are removed, the interstitial cells in the body region of the animal respond by dividing many times and eventually forming large numbers of cnidoblast cells. On the basis of somewhat meager evidence, the author would like to offer a hypothesis to explain first, the increased production of nematocysts after removal of the tentacles of a hydra, and secondly, the intense migration of nematocysts from one species of hydra into the tissues of another species.

To begin, it is generally assumed that it is not possible to determine by present methods the fate of a given interstitial cell. When gland cells are needed by the animal, interstitial cells transform into gland cells. If nematocysts are removed by an excision of tentacles from the body, interstitial cells respond by forming new cnidoblast cells. It is predicted that every cnidoblast cell or nematocyte in the hydra is constantly secreting a substance which inhibits to a small extent the further production of cnidoblast cells by interstitial cells. In other words it is a substance which inhibits the differentiation of interstitial cells in a particular direction. When cnidoblast cells are removed, the inhibitory substance disappears and interstitial cells begin to differentiate in large numbers into cnidoblasts. Apparently, a single interstitial cell contains all of the enzymes and substances necessary to transform into any specialized cell type found in hydra. It is hypothesized that under normal conditions these enzyme systems are blocked by inhibitory substances which are produced by specialized cell types. If the hypostome of a hydra is removed, interstitial cells in the growth region immediately transform into mucus cells which normally line the gastrodermis of the hypostome.

The present experiments strongly suggest that the inhibitory substances produced by specialized cell types are quite similar between different species of hydra. When the distal portion of Species X is grafted to the proximal portion of *P. oligactis* there is a suppression of nematocyst formation in Species X. It is quite possible that these same substances which suppress the formation of nematocysts in Species X also perform in the same function in the tissues of *P. oligactis*. It is possible, moreover, that these inhibitory substances are more active or exert more influence in the tissues of another species than they do in the species in which they were formed originally. Conversely, once the nematocyst formation process in Species X has been inhibited by diffusion of inhibitory substances from the tissues of *oligactis*, there is an increased production of nematocysts in the tissues of *oligactis*. Examination of serial sections has revealed that whenever there is a migration of nematocysts from the tissues to *oligactis* to the tissues of Species X, there is, at the same time, a tremendous nematocyst production in the epidermis of *oligactis*. This production is often so great that the epidermis of *oligactis* actually bulges and huge « nests » of cnidoblasts resembling a small spermary develop. It appears that the loss of the inhibitory substance from the tissues of *oligactis* to the tissues of Species X results in the inhibition of nematocyst formation in Species X and the increase of cnidoblast formation in *oligactis*.

It was previously stated that on several occasions there is no migration of nematocysts from *oligactis* to the tissues of Species X after the graft of the two species was performed. The reason for this is not known at the present time, although it may be stated again that whenever there is no migration from *oligactis* to Species X, there is also no increased cnidoblast production in the epidermis of *oligactis*.

It has been assumed that the migration of nematocysts from the tissues of *oligactis* to the tissues of Species X is via the gastrovascular cavity. However, the fact that large numbers of *oligactis* stenoteles have been observed in the growth region of Species X as well as in the tentacles suggest strongly that the migration might be an epidermal one. If a cnidoblast cell is capable of passing between epidermal cells, traversing the mesogloea, and then passing between gastrodermal cells, it is not unreasonable to assume that cnidoblasts are also capable of migrating along the entire epidermis of the gastric and growth regions.

During the course of these studies it remained to be determined whether the nematocysts migrating from *oligactis* to Species X were functional once they had reached these « foreign » tissues. Two animals in which such a migration had occurred were squashed lightly and examined under an oil immersion objective while a few drops of dilute NaOH were added to the edges of the coverslip. The stenoteles characteristic of *oligactis* were discharged immediately in great numbers. This is taken as strong evidence for the fact that migrating nematocysts are normal, functional nematocysts.

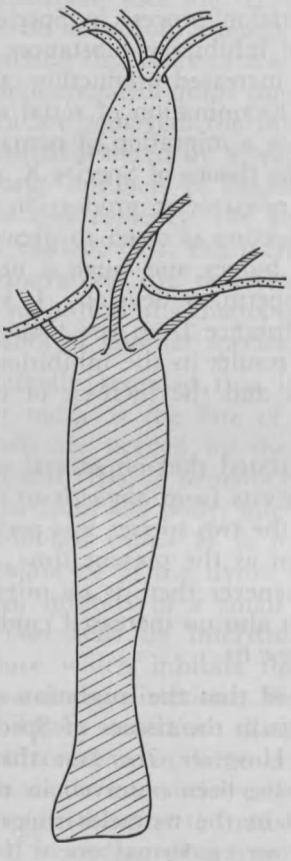

FIG. 2.

This drawing illustrates a reversal of polarity occurring in the proximal region of *P. oligactis* a few days after a graft consisting of a proximal portion of Species X (cross hatching) and a distal portion of *oligactis* (stippled area). Note the tentacles growing from the basal portion of the oligactis fragment.

Several grafts which were the reverse of the type previously described were performed. In this case the proximal portion of Species X was grafted to the distal portion of *oligactis*. As was shown in the preceding paper (*The Nematocyst of Hydra*, Part IV) there is no inhibition of the nematocyst formation process in *oligactis* after such a graft, but in other respects this graft combination was most interesting. If a proximal portion of *oligactis* is grafted to a distal portion of Species X the pieces

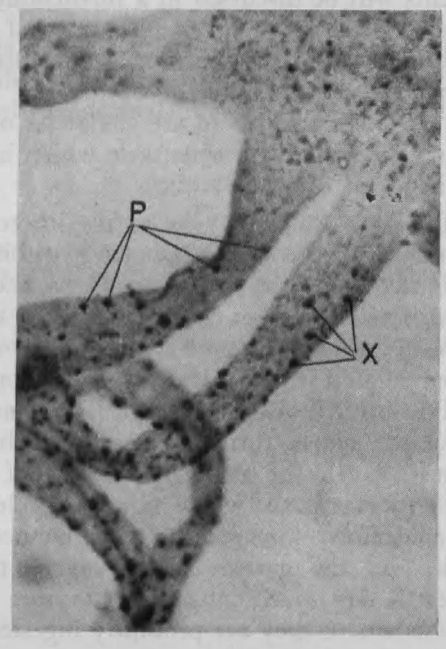

FIG. 3.

This photograph reveals the presence of tentacles at the basal portion of *P. oligactis* after the hypostome, growth region, and tentacles of this animal had been grafted for a few days to the proximal regions of Species X. Note that there are stenoteles from Species X in the tentacles of oligactis.

P : stenotele of oligactis. - X : stenotele of Species X.

become firmly united after two hours. A new head usually does not forms from the distal portion of the *oligactis* fragment, and after approximately one week and a half the grafted portions separate at the graft site. The *oligactis* portion then grows a new head, and the Species X portion produces a new base. However, if Species X represents the proximal portion of the graft, a new distal region, including fully developed tentacles and

hypostome, always forms at the junction of the two grafts from the tissues of Species X. These newly formed tentacles always contain a normal number of nematocysts. What is more interesting is that tentacles always grow at the same time from the proximal portion of oligactis (See Fig. 2). It would appear that *oligactis* would normally form a base in this region but the result is invariably the opposite. A true reversal in polarity of *oligactis* has been effected by this graft, but the implications of these experiments will be discussed in a forthcoming paper. For the present, however, it may be mentioned that the new tentacles produced in the proximal portion of oligactis contain several dozen stenoteles characteristic of Species X. An examination of Figure 3 reveals that there are stenoteles of both species in roughly equal proportions in a tentacle which has been formed from the tissues of *oligactis*.

It is concluded that there may be a free interchange of cnidoblast cells containing stenoteles between two different species of hydra. It is not known at the present why the stenoteles of Species X migrate to the tissues of *oligactis* when the reversal of polarity occurs. It is obvious that a new region of high metabolism has been created in *oligactis* at the junction of the grafted portions. Possibly the tissues of oligactis are not able to sufficiently supply the newly formed tentacles with nematocysts. Thus, the digestive cells of the gastrodermis of the tentacles will ingest any nematocysts which are available and deposit them in the epidermis. Since there is a connection between the two species via the gastrovascular cavity, the cnidoblast cells of Species X are easily able to pass into the digestive cavity of *oligactis* where they are promptly ingested by digestive cells of the gastrodermis.

It has been established that nematocysts from one species of hydra are capable of integrating themselves into the tissues of another species. This integration necessarily involves the cnidoblast cell or nematocyte which houses the nematocysts. It is possible to speculate a great deal on the evolutionary implications of these observations. Are all cnidoblasts found in one species of hydra capable of existing in all other species of hydra, or does this phenomenon occur only in closely related species? The author shooses not to speculate on this point at the present time. It is hoped that future experimentation will shed more light on this problem.

Summary and Conclusion.

It has been clearly demonstrated that cnidoblast cells plus their enclosed nematocysts of one species of hydra are capable of being integrated into the tissues of another species. These « exchanged » nematocysts appear to be normal in every respect. A theory which attempts to explain the general mechanism and direction of interstitial cell differentiation is offered.

The present experiments suggest that when two different species of hydra are grafted the exchange of nematocysts between their tissues is via the gastrovascular cavity. However, these experiments also lend support to the idea that the migration may be a purely epidermal migration on the part of the cnidoblast cell.

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