Artificially Induced Pearl Production*

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THE studies of the last dozen years or so directed to the elucidation of the mechanism of pearl-production, and of the pathological conditions which induce or favor it, have made it increasingly clear that the production of a pearl is not, as was formerly largely believed, the direct result of the effort of the mollusc to cover over with nacre an irritating foreign body (e. g., a grain of sand or a parasite), but rather of the presence in the subcutaneous tissues of a portion of the shell-secreting epidermis, in the form of a closed sac, which need not necessarily be associated with any foreign body. The factors that determine the presence or otherwise of such an epidermal "island" seem to differ in different species of molluscs, and in the same species in different localities. The case in which the chain of causation is most fully known is that of the pearls formed through the agency of the trematode *Gymnophallus* in the common mussel, *Mytilus edulis*, worked out by the present writer in 1902 (*P.Z.S.*, 1902, vol. I, pages 140-166), in which case the mollusc normally surrounds the parasite, while still alive, with a closed sac composed of the outer shell-secreting epidermis; and this sac, on the death of the parasite, or on its migration to another part of the host, lays down concentric layers of the shell substance, so forming a pearl. The fact that *Gymnophallus* invariably becomes surrounded by such a sac, after settling down in the mussel, and that other parasites and foreign bodies do not normally share the same fate, pointed to the conclusion that the formation of the pearl-sac, and consequently of the pearl, is due to some peculiar or specific stimulation on the part of the parasite, and not to mere mechanical irritation, such as would arise from the presence of any foreign body. But the nature of this stimulation, whether mechanical (as e. g., by the parasite habitually carrying in with it a portion of the epidermis) or toxic (i. e., resulting from an abnormal growth of epidermal cells induced by some specific secretion of the parasite and comparable to the production of plant-galls), is a question which still awaits further investigation.

The artificial production of pearly excrecences, or blisters, on the interior of the shell, by introducing between the mantle and the shell beads or metal images, which become coated over with nacre, has long been practised by the Chinese, and has been developed into a flourishing industry by the Japanese, whose "culture pearls," formed over a core of mother-of-pearl, are now familiar objects in cheap European jewelry. The secret process devised by Linnaeus for producing pearls in

fresh-water mussels was analogous to these methods, though never commercially developed; and similar enterprises have been attempted in Burma and elsewhere. All these processes have, however, so far only resulted in the production of pearly excrecences, or blisters, on the shell; and the successes in the produc-

Fig. 1.—Section through an artificially produced pearl-sac in *Anodonta*. $\times 160$. The pearl-sac is not yet completely formed, the epidermal epithelium (*Ps*) being absent from one side of the cavity (at the bottom of the figure). Pearly substance, however, is already in process of formation. *aB*, autochthonous connective tissue; *FB*, transplanted connective tissue; *jB*, young transplanted connective tissue. Five days after the operation.

tion of free pearls in this manner, which have been claimed from time to time, have not been scientifically demonstrated. Recently, in Japan, Mr. Mikimoto is stated to have produced a few small free pearls by artificial means, but the method adopted has not been disclosed, and success on anything like a commercial scale has not been achieved.

The production of free pearls by mechanical treatment—not, of course, on a commercial scale as yet—has, however, recently been successfully accomplished by Dr. F. Alverdes, working under Prof. Korschelt at Marburg, and is described in a paper in the *Zoologischer Anzeiger* for September 12th of last year.¹ The method adopted by Dr. Alverdes was the introduction, by injection into the mantle-parenchyma, of fragments of the shell-secreting epidermis. The molluscs upon which he operated were the fresh-water pearl-mussel (*Margaritana margaritifera* L.), which is still fished for pearls in some of the rivers of Scotland and Ireland, the common pond-mussel (*Anodonta*) and the river-mussel (*Unio pictorum* L.).

The method of treatment was as follows: The shell was held open with a wedge, a portion of the mantle-margin was detached from the shell (to which it is normally bound by the reflected margin of the periostracum), and fragments of the outer shell-secreting epidermis of the mantle were scraped off with a knife; in other cases a small disk of tissue, containing both the shell-secreting epidermis and the ciliated lining of the mantle-cavity, was cut out. These fragments of epidermis, with the connective tissue which remained adherent to them, were then injected into the loose connective tissue of the mantle of the mussel.

After treatment the molluscs were returned to their native waters, where they were kept in suitable enclosures. They were killed, and examined at intervals ranging from 2 days to 27 weeks. The wound caused by the injection was quickly healed by the migration of wandering cells, to form a scar, leaving intact the introduced "island" of epidermis (see Figs. 1 to 3), with such connective tissue as happened to have remained attached to it. These "islands" did not, as might have been expected, fuse with and become indistinguishable from the autochthonous tissues, but remained sharply marked off from them throughout, and no actual confluence between the transplanted and the autochthonous connective tissue seems to have taken place. Dr. Alverdes found that, when the injected epidermis came to lie with the free surfaces of the cells against the autochthonous connective tissue, it died in a short time. If, however, a fragment of epidermis found its way into one of the cavities which occur in the parenchyma

it survived, and spread out around the cavity as though endeavoring to cover over the exposed connective-tissue surface with epidermis, in a manner recalling that figured by the present writer in the developing trematode-sac in *Mytilus* (*loc. cit.*, Plate XIV, Figs. 2 and 3). Alverdes considers that the process here involved is not due to a multiplication of the epidermal cells by mitosis, but to a flattening out of the cells, mitotic figures first appearing after the cavity is completely lined with epidermis. By the growth of the epidermis around the cavity in the parenchyma a closed pearl-sac is formed; and then the growth therein of a pearl follows naturally from the normal secretion of this epidermis. If the space lined with epidermis is more or less spherical, so is the pearl; if it is irregular, a pearl of irregular shape results.

The growth of the pearl-sac takes place with remarkable rapidity, being completed in from 2 to 14 days. This point is of interest as helping to account for the great difficulty that has been experienced by all workers on the subject in searching for the earliest stages of pearl-sac development in nature.

Cysts were also formed by the ciliated epidermis which lines the mantle-cavity when successfully injected. It would be interesting to pursue experiments with this tissue, to ascertain whether these ciliated cells would retain their original characters or whether under any circumstances they are capable of giving rise, either by direct transformation or by cell-division, to shell-secreting epidermis.

The size of the pearls which Dr. Alverdes produced depended, in the first instance, on the sizes of the cavities into which the injected epidermal fragments found their way. They ranged from 30 μ to 1 millimeter in diameter. In some cases the secretion of pearly substance in the sac began a few days after the operation; in others it had not started after seven and a half weeks (see Fig. 3).

Quite apart from the interest which attaches to so important a step in the direction of the solution of the economic problem of artificial pearl-production and to the achievement of an end which has long been foreseen by zoölogists, the theoretical value of these experiments is considerable, as adding one more link to the chain

Fig. 3.—Section through an artificially produced pearl-sac in *Unio*, in which a pearl has not yet begun to form. $\times 300$. Remains of dead cells are shown in the center of the epidermal sac. *tB*, transplanted connective tissue. Seven and a half weeks after the operation.

Fig. 2.—Section of an artificially produced pearl (decalcified) about 1 millimeter in diameter *in situ* in the mantle of *Margaritana*. $\times 54$. A dark foreign "nucleus" accidentally introduced is seen in the center of the pearl. *FB*, transplanted connective tissues. Twenty-seven weeks after the operation.

¹ Versuche über die künstliche Erzeugung von Mantelperlen bei Süßwassermuscheln. *Zool. Anzeiger*, Bd. xlii, Nr. 10, SS. 441-458.