lishing Company. 1913. Pp. viii + 210; 1 plate; 101 figures in text. Price, ninety cents.

This little volume presents in an interesting manner those facts concerning rocks which are of interest to the student of general geology. The author has in mind a pocket manual which may be of service in the field. The treatment is from the standpoint of the macroscopic properties of rocks and is thoroughly modern. The book is well printed. The illustrations are excellent.

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## SPECIAL ARTICLES

## THE CULTIVATION OF TISSUES FROM THE FROG

In a series of experiments on the culture in vitro of tissues of the frog it was found that several kinds of tissues show a marked outgrowth after being kept for a few days in lymph or plasma. Small pieces of the tissues were mounted according to the usual method in hanging drops of the culture medium and sealed with vaseline in hollow slides. Cells may remain alive under these conditions for several weeks.

Spleen, bone-marrow and pseudothyroid give rise to a fringe of outwandering cells resembling leucocytes which extend farther and farther into the surrounding medium. Larger connective tissue cells wander out later, and both types of cells exhibit amœboid changes. Small pieces of tissue may almost entirely disintegrate into wandering cells.

The epithelial cells of the skin extend generally as a broad thin sheet of tissue. The cells move out in contact with the cover slip or the lower surface of the drop. Individual cells of the epidermis may become isolated and creep out alone, but there is a marked tendency for the cells to keep together in a continuous membrane. In a previous paper on the movements of the ectodermic epithelium of amphibian larvæ<sup>1</sup> it was shown that the ectoderm cells actively creep out by an amœboid movement of

<sup>1</sup> Univ. of Calif. Pubs. Zool., 1913.

the very thin and transparent protoplasm of their free borders. The method by which sheets of epithelium extend in the adult frog is essentially the same as in the embryo or larva.

In several cases black pigment cells were seen to isolate themselves and wander out along the cover slip or lower surface film of the drop. In some cases, especially in the smaller pigment cells, the changes in form were fairly rapid. Pseudopods were thrust out and retracted very much as in the common amœba, and in some instances the cells were seen to migrate nearly across the field of the microscope. The processes of the pigment cells of the adult, unlike those of the larvæ, may be nearly transparent, and they usually are so when first formed; frequently, however, they are very soon invaded by pigment granules. Outwandering cells may show branching processes characteristic of the expanded melanophores of the frog's skin. The change in form of the pigmented mass within the cell is due in part to changes in the outline of the whole cell and in part to the flowing back and forth of pigment granules within the cell processes. There is a measure of truth, therefore, in both the rival theories of the changes of the chromatophores in the skin of the frog.

In some preparations the peritoneal epithelium wandered out in the form of a sheet of tissue considerably greater in area than the original preparation. For the most part the extension consisted of flattened cells arranged in a single layer and showing a hexagonal contour like the cells of the shed cuticle. Many of these cells were furnished with cilia which beat actively for two weeks. The ciliated cells frequently became ameboid and wandered free from the rest, sending out fine processes several times the original diameter of the cell. Sometimes the processes branched repeatedly. One would not suspect these cells to be derived from ciliated epithelium were it not for their tuft of beating cilia, and the fact that one can actually observe their transformations. Follicle cells of the testis may creep out and give the appearance of giant amœbæ.

Fuller details of the behavior of various

types of tissue cells will appear in later papers. molt Similar experiments were tried with the tissues is th of crayfishes and crabs with little result beyond the keeping these cells alive for several weeks. great

alive and active for three months. S. J. HOLMES

## NOTE ON THE ABSORPTION OF CALCIUM DURING THE MOLTING OF THE BLUE CRAB, CALLINECTES SAPIDUS

The blood corpuscles of the crayfish were kept

THE problem of molting in crabs has thus far been investigated, with one exception,<sup>1</sup> only from the morphological point of view.<sup>2</sup> The following observations bear on certain chemical phases of the process of hardening following normal molting in the common blue crab.

The crab hardens by the deposition of CaCO. within the tissues of the soft shell. Has this Ca been absorbed and held in reserve during the period of preparation for molting,<sup>3</sup> or is it absorbed from the sea-water during the actual period of hardening? To test this matter, the following procedure was employed. Three pairs of crabs were chosen, each pair consisting of a recently shed individual and of a hard-shell individual of nearly the same size.<sup>4</sup> A comparison of the Ca content of the individuals of the same pair should throw light on the alternatives suggested. If the Ca content of the two members of each pair is equal, then the Ca must be absorbed before molting and held in reserve. If the Ca content of the hard specimen is very much larger than that of the soft, then the Ca must be absorbed after

<sup>1</sup> Irvine and Woodhead, Proc. Roy. Soc. Edinb., Vol. 16, pp. 324-354, 1888-89.

<sup>2</sup> For a review of the literature on the natural history of molting in Crustacea, see Herrick, Bull. U. S. Bureau of Fisheries, Vol. XV., pp. 1-252, 1895. For this species of crab, see Hay, App. Rep. U. S. Comm. Fish., pp. 395-413, 1904.

<sup>8</sup> Cf. Smith, Quart. Journ. Microsc. Sci., Vol. 59, p. 272, 1913.

<sup>4</sup> These were collected at the Beaufort, N. C., station of the U. S. Bureau of Fisheries. The writer is indebted to Dr. H. M. Smith, the commissioner, for the privilege of staying at the station. molting. Furthermore, if the first alternative is the true one, the Ca content of a crab in the act of casting its shell should be much greater than that of a normal hard crab. If, however, they have the same Ca content, then the second alternative is indicated.

Each crab was ashed separately, and the Ca in the ash determined by precipitating it as the oxalate, igniting and weighing as the oxide. The results of the analyses are indicated in the table. In each pair the hardshell specimen contains about twenty times the amount of Ca contained in the soft one. Also, Crab No. 9, which was in the act of casting its shell, has a Ca content comparable to that of a normal hard individual.

This shows clearly that the Ca used by the soft-shell crab for the purpose of hardening its new shell is not present at the time of the molt, but is absorbed from the sea-water during the hardening.

The mechanism by means of which a molting crab is enabled to absorb such abnormally large quantities of Ca is at present obscure, and in view of the meager data at hand, a discussion of this problem is best postponed until more work shall have been done.

TABLE

Crab No.	Condition	Width, Cm.	Weight, Gm.	Weight, Ca, Gm.	Ca Content, Per Cent.
6 11	soft hard	8.3 8.3	37.20 34.54	$0.0720 \\ 1.845$	0.19 5.34
$\frac{3}{7}$	soft hard	9.7 9.5	$\begin{array}{c} 56.53\\ 61.62 \end{array}$	$\begin{array}{c} \textbf{0.1468} \\ \textbf{2.963} \end{array}$	0.26 4.81
$\begin{array}{c} 13\\12\end{array}$	soft hard	$10.5 \\ 11.0$	70.00 70.90	0.2197 3.617	$0.31 \\ 5.17$
8 9	hard molting	$8.7 \\ 8.5^5$	$\begin{array}{c} 54.75\\ 67.93 \end{array}$	$2.861 \\ 2.520$	5.22 $3.72$

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<sup>5</sup> The width of the new shell was 9.8 cm. The per cent. of Ca in this specimen is low because the molting crab weighed more than an ordinary 8.5 cm. crab, and also because the old shell had two legs missing, which were being regenerated. The actual weight of Ca, however, is very close to that of a normal hard crab.