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"Slow-Motion" Cinematographs of the Contraction of Single Cardiac Muscle Cells.

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The material for this study was supplied by tissue cultures of embryonic heart in which the cardiac muscle had grown out and become differentiated. The ventricles of 16 day rat embryos were cut into fragments 1 to 2 mm. in diameter and cultivated in a medium composed of rat plasma and embryo juice. In the course of 2 to 4 weeks a new growth of muscle appeared among the fibroblasts, taking the form of anastomosing bands similar to adult cardiac muscle. This new growth was closely applied to the under surface of the cover glass so that single cells could be observed with the highest powers of the microscope. Cross striations could be seen in many of the muscle cells of both explant and new growth after one month. The striated appearance in the living cell was produced by the arrangement of small brick-shaped bodies, apparently mitochondrial in nature, into more or less regular longitudinal and transverse rows. Since this picture was somewhat different from that presented by muscle from an adult animal it seems likely that we were dealing with a transition stage in which the differentiation was incomplete. The presence of myofibrillæ with alternating dark and light bands and "membranes of Krause" in fixed preparations, however, indicated that the process was well advanced. Although the muscle in these cultures contracted spontaneously, its activity was intermittent, especially when exposed to room temperature for the making of photographs. This irregularity was overcome by substituting Ringer solution for the usual medium.

Since the intracellular movements were too rapid to be followed by direct observation through the microscope, an effect of slowing was accomplished by making "slow-motion" cinematographs. Exposures were taken at the rate of 32, 48 and 64 per second. The camera, a Bell and Howell 16 mm model 70D, without lenses, was mounted over a compound microscope. The lens system of the microscope consisted of a long-range substage condenser and a Zeiss apo. 120, n.a. 1.3 objective without an ocular.

The following observations were made:

1. Myofibrillæ were not visible in the living cells. The clear hya-

line matrix lying between the longitudinal rows of brick-shaped bodies mentioned above gave an appearance suggestive of fibrillæ but they were not interpreted as such. The only other structures visible in the living cells were occasional spherical droplets of neutral fat imbedded in the matrix.

2. The hyaline matrix or sarcoplasm was the active substance during contraction. The mitochondrial bodies appeared to be forced together in a direction parallel to the long axis of the fiber. The strips of sarcoplasm at their ends, corresponding to the light band of cross striation, became markedly thinner, at times being almost obliterated, while the strips of sarcoplasm at their sides, whose presence suggested longitudinal striation or myofibrillæ, became somewhat thickened. The shape of the blocks themselves did not change. In other words, the blocks were comparatively rigid structures suspended in a relatively fluid matrix and as they were passively moved closer together by the shortening of the cell as a whole, the matrix was forced from its position at the ends of the blocks to the spaces at their sides.

3. No reversal of striation was observed, but it must be remembered that these fibers may present a primitive type of striation and that the pictures used for study were not made with polarized light.

4. The contraction traveled with a wave-like motion from one end of the cell to the other. Occasionally twitches were observed that involved only a part of the cell, in which case the wave-like appearance could not always be identified.

5. The period of time occupied by the contractile phase was approximately 3 times as long as that occupied by relaxation.

6. Although the hyaline matrix or sarcoplasm was of a fluid consistency it had much of the elasticity of a gel. Fluidity was indicated by the rapidity of movement of the intracellular blocks. A gel-like elasticity was suggested, on the other hand, by the fact that the interior of the cell did not become quiet immediately after contraction but quivered and frequently set up small reflected waves in a direction opposite that of the contractions.