enzymatic changes are enhanced by the drug-pretreatment. Also, the cardiac changes induced by endotoxin and histamine in rabbits were identical, suggesting that endotoxin shock might be mediated by histamine in vivo[8].

It was known that no significant blood pooling in the hepato-splanchinic system of rabbit after endotoxin infusion[7], and the observed myocardial SDH activity changes might have been brought about by primary action of endotoxin on the heart at the given period of time (15 min). Previously, streptolysin-O was the only known bacterial toxin to induce primary cardiac depression[10]. Even though no depression of function has been found after endotoxin infusion in the isolated rabbit heart[11], the present study is consistent with our recent findings made after coronary infusion of endotoxin in the canine heart, in situ. It was seen that cardiac mechanical and biochemical functional changes that occurred[12], strongly suggested that the endotoxin might react primarily on the heart as well as on the peripheral vessels. We also found that exogenous histamine infusion into the coronary artery resulted in myocardial functional changes similar to that of endotoxin shock[13,14].

Zusammenfassung. Escherichia coli-Endotoxin (1 mg per kg) wurde in die Ohrvene von Kaninchen injiziert: Die Succinat-dehydrogenase- und Cytochromoxydase-Aktivität in den Mitochondrien des Herzmuskels wurde nach 15 min verändert. Es ist wahrscheinlich, dass das Endotoxin sowohl auf das Herz, als auch auf die peripheren Gefässe direkt wirkt.

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Effect of Acute and Exhaustive Exercise Upon the Fine Structure of Heart Mitochondria

Mitochondria are among the most labile of myocardial structures, and the first to react to different stimuli. Most of the studies concerning the modifications of mitochondrial fine structure in different stages of cardiac functional overload have been made after long-lasting stimuli. Acute and exhaustive exercise represents perhaps one of the best ways to obtain cardiac functional overload in a short time. In the present report the modifications of mitochondrial fine structure after acute and exhaustive exercise are described.

The observations were performed on the hearts of 12 healthy adult dogs which were submitted to acute and exhaustive exercise forcing them to swim in a tank filled with warm water (24 °C) until they became exhausted and sank into the water. This occurred after variable periods of time, ranging between 40 and 90 min, according to the strength of the animals. Immediately after that the dogs were anaesthetized and a small piece of the tip of the heart was removed and fixed in cold 6.25% glutaraldehyde in cacodylate buffer[1]. The material was post-fixed in osmium tetroxide and embedded in Araldite[2].

The electron micrographs of the heart sections revealed the existence of mitochondria of unusual size. In some cases the mitochondrial mass represented more than one half of the whole myocardial area (Figure 1). The increase in mitochondrial size could be seen under three different aspects. In some cases there appeared to be a fusion of neighbouring mitochondria, appearing as long and slender

Fig. 1. Increase of the mitochondrial mass. In this micrograph most of the myocardium is represented by mitochondria with some swelling of the matrix. ×12,000.

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mitochondria with lengths comprising several sarcomeres (Figure 2). In other cases, bulky forms wider than normal were found, especially in those mitochondria with a subplasmalemmal or perinuclear localization, although occasionally the enlargement was noticed in those located among the myofibrils. The mitochondria appeared then as large bodies with a length comprising several sarcomeres and an almost equal width. They presented normal looking cristae of an unusual length, but in agreement with the increase in mitochondrial size (Figures 3 and 4). Lastly, the mitochondria increased in size as the result of an apparent swelling, with a decrease of the electron density of the matrix and of the number of cristae, which appeared peripherically located and occasionally disrupted. Sometimes a combination of the modifications was found. Giant mitochondria with partial vacuolization of the matrix and disruption of the cristae were frequently observed (Figure 5).

Increase of the mitochondrial size has been found in the dog myocardium under several experimental conditions. Functional overload after experimental aortic stenosis has been the procedure most usually employed[3,4]. However,

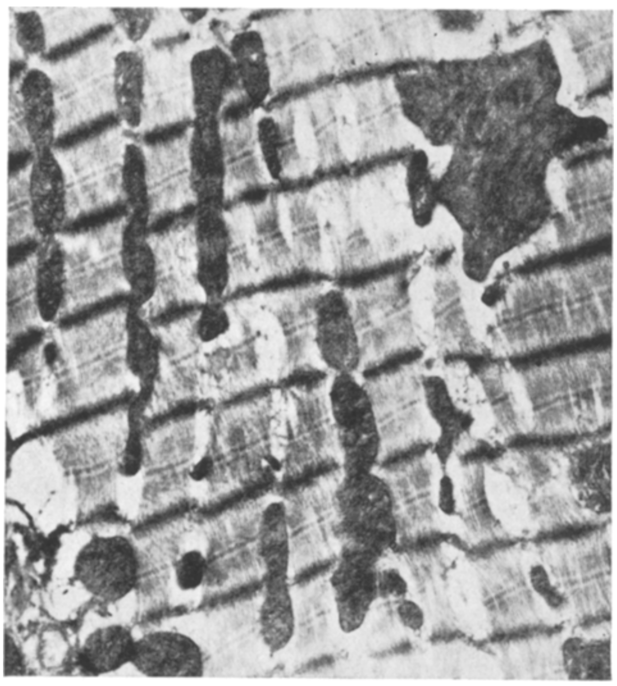

Fig. 2. Probable fusion of neighbouring mitochondria. In this area most of them exceed 3 sarcomeres in length. × 14,000.

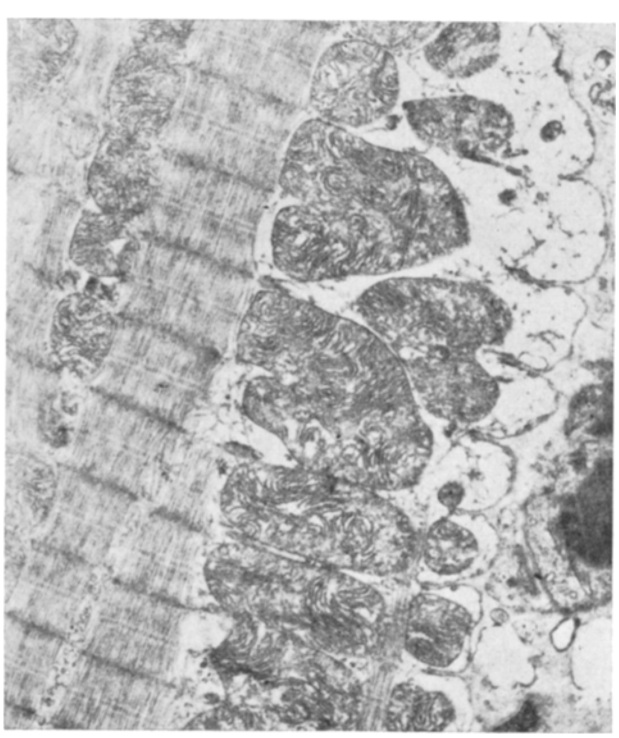

Fig. 3

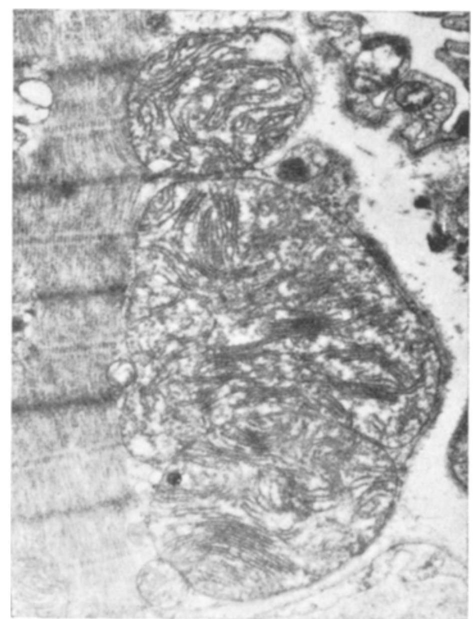

Fig. 4

Figs. 3 and 4. Increase in mitochondria individual size. The cristae are unusually long and appear oriented in an irregular fashion. Fig. 3 × 19,000, Fig. 4 × 21,000.

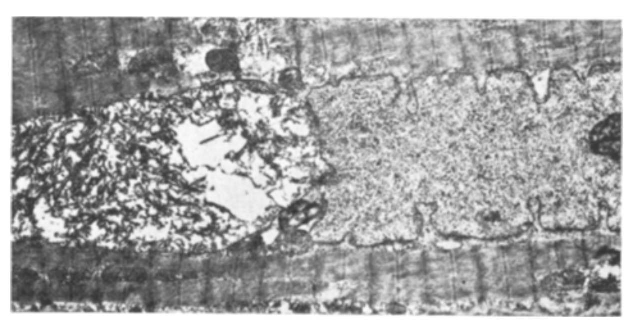

Fig. 5. Giant mitochondria with a juxtanuclear localization. Partial swelling of the matrix and disruption of the cristae may be seen. Neighbouring mitochondria are of normal size. × 5,000.

3 F. Z. Meerson, T. A. Zaletayeva, S. S. Lagutchev, and M. G. Pshennikova, Exp. Cell Res. *36*, 568 (1964).
4 A. Wollenberg and W. J. Schulze, J. Biophys. Biochem. Cytol. *10*, 285 (1961).

most of the studies have been made a long time after the stenosis was performed. To our knowledge, the only study made after a short time is that of MEERSON et al.[3], two days after the cardiac functional overload started. Swelling of mitochondria and vacuolization of the matrix have been observed after several degrees of ischaemia[5-7]. In this case, no increase of mitochondrial size was reported.

The modifications described in this paper are similar to those found after functional overload and ischaemia. It is probable that they are the result of both kinds of stimuli. However, what was striking was the rapidity of the alterations. Although some of them, such as the swelling, could reasonably occur in a few minutes, the fusion and appearance of giant mitochondria indicate in our opinion a true increase in mitochondrial mass, with active reproductive phenomena. To our knowledge, no biological system has yet been reported where such a rapid increase in mitochondrial mass occurs.

Apparently, the myocardial mitochondria possess the ability to reproduce in a short time, in response to stimuli that produce an acute functional overload.

Zusammenfassung. Eine deutliche Vermehrung der Mitochondrienzahl und -grösse in den Herzmuskelzellen wurde in Hunden durch erzwungenes Schwimmen (rasche und erschöpfende Leistung) festgestellt.

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Inhibition of Amphibian Egg Development by Histones

The histones are the basic nuclear proteins of animal and plant cells. They seem to be involved in the regulation of the gene activity[1,2]. However, some recent reports suggest a more important role in this respect for the non-basic nuclear proteins[3]. Biochemical evidence shows that histones inhibit DNA-primed RNA synthesis in pea seedlings, mammalia and bacteria[4-8]. Moreover, the activity of DNA-polymerase is diminished by the addition of histones at high concentrations[9-11]. This inhibitory effect is probably related to the formation of a DNA-histone complex, which is unable to act as 'primer' for the nucleic acids synthesis[4,12]. On the other hand, selective removal of the histones from the nucleus by mild trypsin treatment enhances *m*-RNA synthesis[5]. Very similar effect is obtained by acetylation and methylation of histones, and this reversible mechanism could be responsible for the regulation of genes in vivo[13].

MOORE[14] suggests that, during early embryonic development, before gastrulation, most of the structural genes are 'turned off' because they are closely associated with histones. After gastrulation, the genes are 'turned on' by changes in the DNA-histone association. In this respect the treatment of amphibian eggs by histones greatly inhibits gastrulation[15].

We treated the eggs of Anurans (*Discoglossus pictus*) with lysine-rich (HL) and arginine-rich (HA) fractions of calf thymus histones[16]. The eggs treated with 0.4–0.1 mg/ml of HL fraction at two blastomeres are blocked during cleavage; with 0.05–0.025 mg/ml the development stops at the gastrula stage.

In the first case the results are likely to be related to the inhibition of DNA-polymerase reaction. At lower concentrations, however, only the DNA-primed RNA-synthesis could be affected, and development stops at gastrulation, when new molecules of *m*-RNA are required for the differentiation[17]. After the gastrula stage, the eggs become relatively insensitive and the effects of the treatment are unimportant. The HA fraction has the same effect but at higher concentrations. The protamine from salmon milt is as effective as HL fraction.

With shorter treatment (2 h) between two blastomeres and late blastula stages, 0.05–0.025 mg/ml HL is able to inhibit the development: only a few eggs gastrulate and form abnormal tadpoles.

The developmental abnormalities are remarkable in every embryonic apparatus (microcephaly, cyclopia, adhesive organs fused, olfactory organs absent, vacuolar notochord, spina bifida and external intestine). At gastrulation the inhibitory effects begin at concentrations of 0.2 mg/ml, and the tadpoles have greatly reduced tails. After this stage only 10 h treatment can produce functional abnormalities in the tadpoles. The marked inhibitory effect on morphogenesis produced by brief treatment with histones from cleavage to the blastula stage, suggests that

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