ORIGINAL PAPER

Myocardial stunning. Morphological studies in acute experimental ischemia and intraoperatory myocardial biopsies

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Abstract

Myocardial stunning represent a consequence of brief ischemia with reversible regional contractile dysfunction dependent persist from minute to days after reperfusion, despite the absence of irreversible damage and restoration of coronary blood flow. The evolution of these new ischemic entity were described by experimental acute ischemia and repeated intraoperatory myocardial biopsies effectuated near 200 patients with heart disease, excluding those with cardiac failure and atrial fibrillation. Using histological histoenzymological and particularly ultrastructural methods, only reversible mitochondrial and sarcoplasmatic reticulum lesions, slight glycogen granules depletions and sporadical dissociation of myofilaments by edema were seen. Major mechanism for the state was suggested: generation of oxygen derived free radicals with consequent oxidative stress and impaired calcium homeostasis. Rare morphological appearance on stunned myocardium was signaled in the references, our collective first made these studies in Romania.

***Keywords*:** myocardial stunning, experiment, myocardial biopsy, pathogenesis.

Introduction

Myocardial stunning represent a consequence of a brief ischemia with reversible contractile dysfunction that persist after reperfusion despite the absence of irreversible damage and despite restoration of normal or near normal coronary blood flow [2, 3].

The concept of myocardial stunning becomes apparent after a period of experimental laboratory. Initially was described in 1975 in Vatner's group by Hendrickx GR *et al.* [1]. In the past decade, there has been an impressive increase in the number of publications, dealing with stunning at the experimental level [2–12].

During the 1980's however, the use of thrombolytic therapy and other forms of interventional recanalization under aortic cross clumping for treatment of acute and chronic syndromes with coronary artery bypass grafting, led to remarkable growth of interest in the phenomenon of myocardial stunning, recorded primarily as a laboratory curiosity. When this term was coined by Braunwald E and Kloner RA [3, 20] numerous clinical appeared after following years [12, 30].

New therapies designed to prevent stunning are being investigated in preclinical studies and soon will be tested in clinical trial. In short, the concept that myocardium can remain reversible depressed for extend periods of time after ischemia (1–15 minutes), is changing our understanding of the pathophysiology of coronary artery diseases and our appeared to the management of this disorders. The primarily purpose of this work is to describe the ultrastructural lesional evolution in course of stunned of experimental acute and transitory ischemia and in course of aortic cross clamping in various cardiac surgeries.

Materials and methods

The present paper concerns the morphologic aspects resulting from experimental myocardial ischemia (operated with N. M. Constantinescu in "Carol Davila" University, Bucharest) and repeated intraoperatory myocardial biopsies in various ischemic cardiomyopathies (surgical intervention effectuated by V. Cândea in Central Military Hospital and II[nd] Cardiovascular Clinic of "Prof. C. C. Iliescu" Cardiac and Cardiovascular Institute, Bucharest).

From experimental material were selected 60 dogs with complete and incomplete occlusions of left or circumflex coronary artery that presented in the first 15 minutes myocardial contractile dysfunction that remain few minutes or 2–3 hours intervals when observed appearance stunned myocardium states. From this zones on made biopsies after 2, 5, 10, 15, 20, 30, and 40 minutes and after the intraoperatory the disappearance of dyskinesis in ischemic zone. On made: histological staining (HE, VG, PAS–Alcian, Lie), histoenzymological testing (SDH, cytochromoxidase, LDH, MDH, ATP-ase) on samples and mitochondrial suspension with the help of Biochemistry Department,

from "Victor Babeş" Institute, Bucharest, head Sm. Constantinescu [20]. From over 200 patients with valvulopathies, congenital heart diseases and ischemic heart diseases; for this study were discarded the patients with cardiac failure and previous atrial fibrillation. All patients were several investigated especially EKG, coronarographic, echocardiographic, with dobutamide stress, regional fraction estimation on extracorporeal circulation (ECG). The surgical interventions under were effectuated by V. Cândea and his collective. To describe the evaluation of lesions on made, for control, before aortic cross-clumping myocardial biopsies and after 2, 5, 7, 10, 12, 20, 25, 30, and 40 minutes. The same histological, histochemical and especially transmission electron microscopic studies were made.

Results

In experimental studies within first 10 minutes, the histologic examination showed only circulatory changes and Lie staining rare fuchsinophil fibers. The ultrastructural alteration were slight: near normal aspects in first minutes (Figure 1) after 5–15 minutes reversible lesions were seen in the mitochondrial cristae at the sections (Figure 2) and suspensions (Figure 3), sarcoplasmatic reticulum (Figure 4), decrease of glycogen granules, perinuclear lysosomal hyperplasia (Figure 5). After 15 minutes of ischemia, these affected myocardial fibers alterned with others with accentuated reversible lesions (Figure 6): Z bands waves, glycogen depletions, unequal intermyofibrilary edema, dissociations of some sarcomeres, cause of excitation–contraction disturbance, more dilatation of the sarcoplasmatic reticulum and tendency to nuclear margination. Mitochondrial lesions consisted of changes in volume, swelling and rarefied cristae, good-looking in the suspension (Figure 3). After 30 minutes of ischemia numerous and progressive irreversible lesions was sees (Figures 7 and 8).

The patients with surgical interventions under extracorporeal circulation presented after aortic cross clumping globally ischemia and these reason for detecting the evolution of this lesions we are excluded in this study the patients with cardiac failure and previous atrial fibrillations. Because in these patients the heart is rendered globally ischemic during aortic cross clamping and then reperfused, and because in most cases no myocardial necrosis is demonstrable, the most likely explanation for this postoperative dysfunction is that it represent a form of global myocardial stunning, in analogy with observation made in experimental models of cardiopulmonary by-pass.

In first myocardium biopsy were made from right auriculum before of cardiac arrest for a control sample.

During surgical techniques on made from papillary muscle of the left ventricle successive biopsies after 3, 5, 6, 10, 12, 15, 20, 35, and 40 minutes and before aorta cross declamping when operation did not 40 minutes.

The histological method were non signification only myocyte fuchsinophilia accentuation.

Electronmicroscopical and also histoenzymological on remarked that lesions become slightly more accentuated within the first 5–6 minutes when somewhat more after 10–12 minutes, moderately after 15–20 minutes, following circulatory and metabolic disturbances. Within this context, progressive lesion of the cardiomyocytes and vascular membrane systems appeared.

The mitochondria manifested early changes clustered together with rarefaction of the cristae and tendency to ballooning. The tubular system especially take endoplasmic reticulum, presented unequal dilatation and here and there subsarcolemal ballooning. These capillaries showed turgescent endothelial cells and basement membranes with frequent pynocytosis vesicle and neighboring perivascular edema. The myofilaments appeared intact yet a tendency to breaking up in patches and progressive laceration by intrasarcoplasmatic edema, which however did not modify the periodicity of sarcomeres, interrupted at time by the Z bands. Lysosomal hyperplasia and activation, especially perinuclearly but without structural disintegration of the neighboring area. Glycogen granules were unequally agglomerated and dissociated in some zones by edema. Nuclear alterations were minor at this stage. When surgery had be prolonged, progress of these lesions to irreversibility were progressive observed.

The biochemical and histoenzymological investigations were centered on the determination of mitochondrial marker enzymes, cytochromoxidase, NADH, cytochrome C reductase that decrease in the intervals that were studied. Histoenzymologic progressive investigations brought supplementary data explaining some physiopathologic process in the ischemic myocardial stunning state.

Discussions

The stunned myocardium is a new pathological entity [2] within a framework of ischemic cardiac diseases as a prolonged reversible post-ischemic regional ventricular dysfunction that remains some minutes or hours after reperfusion. These new idea were regarded by experimental studies in 1975. In some times, during 1976–1979, in Romania at "Victor Babeş" Institute, a collective of Pathology Department (head D. Laky) published original papers in which described alone morphologic, electrocardiographic and biochemical appearances in experimental acute ischemia, with myocardial contractile dysfunctions.

During 1980s, however the wide spread of thrombolytic therapy and the interventional recanalization for treatment of acute ischemic syndromes, coupled with recognition patients with coronary artery experience spontaneous reperfusion after coronary spasm or thrombosis, lead to a remarkable growth of interest in the phenomenon of myocardial stunning, demonstrated by the number of full-length original manuscripts published on the topic of myocardial stunning only in the five years [13–25] sine 1982 when this term was coined by Braunwald E and Kloner RA [3].

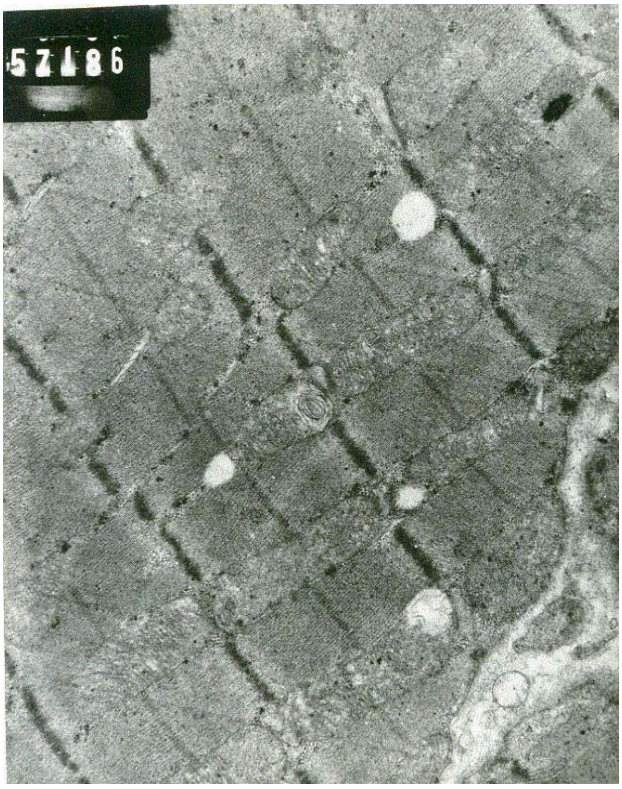

Figure 1 – ***Dog myocardium, biopsy from posterior papillary muscle after 2 minutes circumflex coronary occlusion: near normal aspects isolated dilated sarcoplasmatic reticulum (EM, ×9500)***

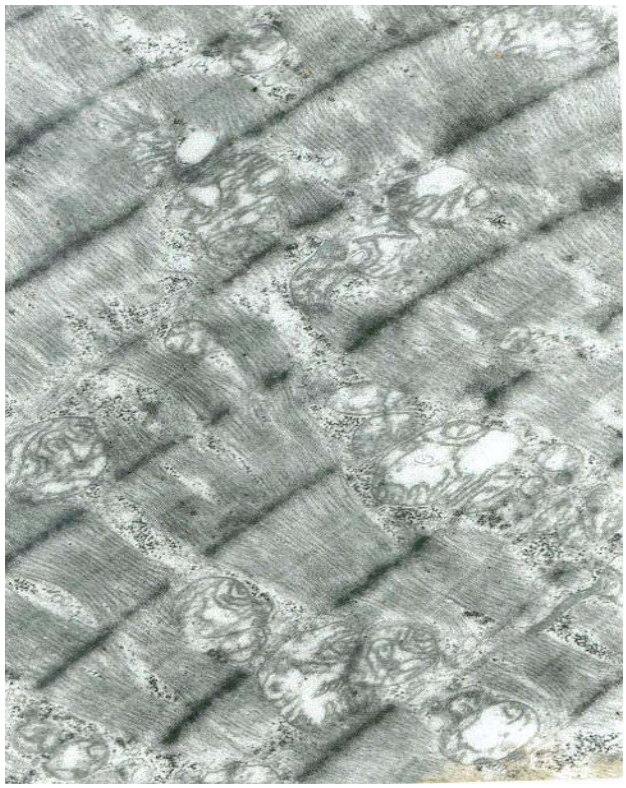

Figure 2 – ***Dog myocardium, transient myocardial ischemia section from left myocardium 10 minutes after aortic cross clumping : lesions of the mitochondrial cristae, some Z lines interruptions (EM, ×8000)***

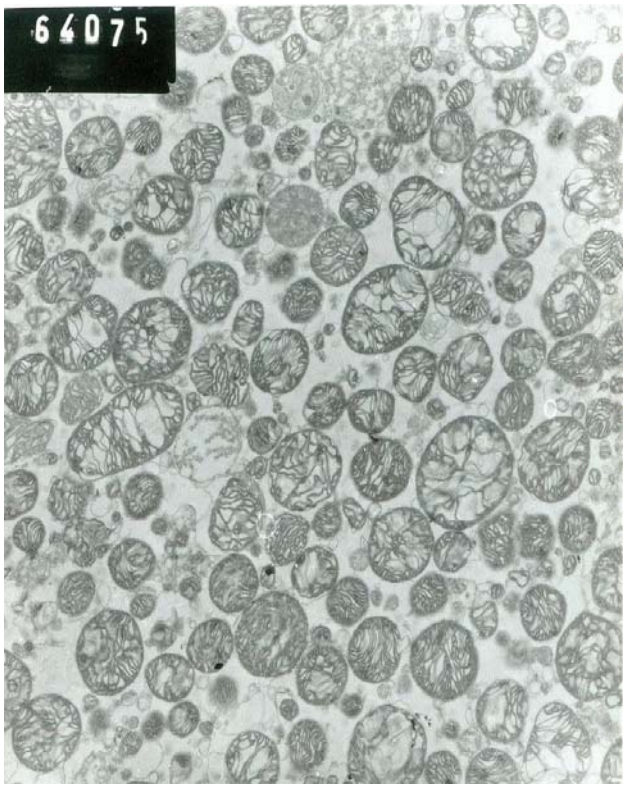

Figure 3 – ***Dog myocardium, idem, mitochondrial suspension (EM, ×4500) cristae lesions***

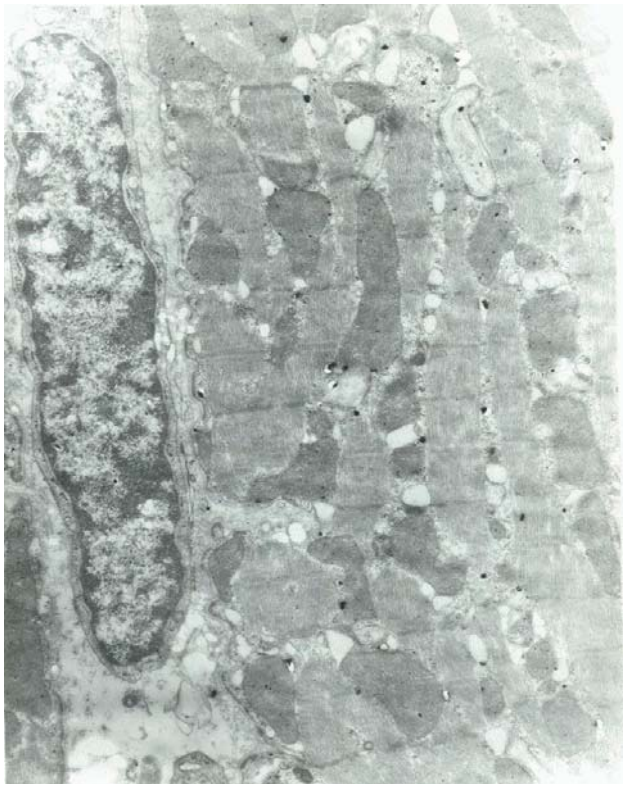

Figure 4 – ***Dog myocardium, transient myocardial ischemia 15 minutes: sarcoplasmatic tubular various dilatation, mitochondrial lesions (EM, ×4000)***

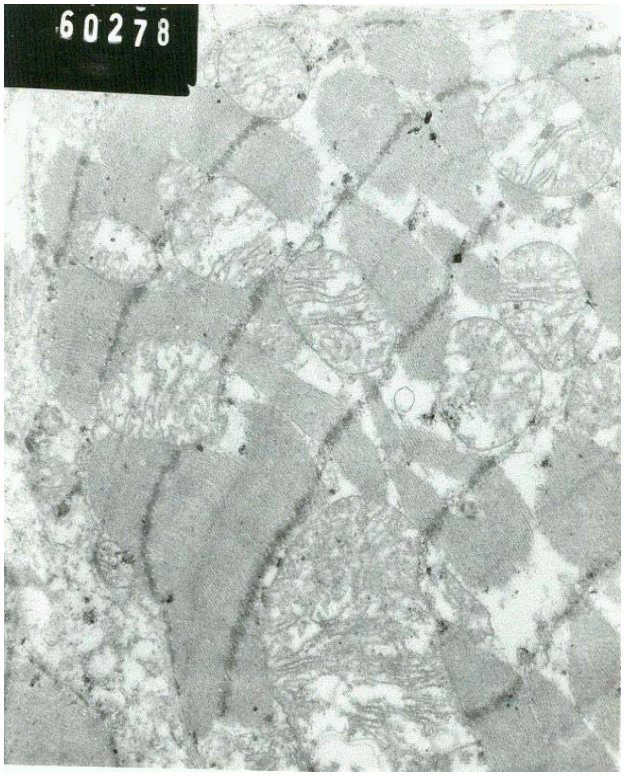

Figure 5 – ***Dog myocardium, idem: perinuclear lysosomas sarcoplasmatic reticulum dilatation (EM, ×9500)***

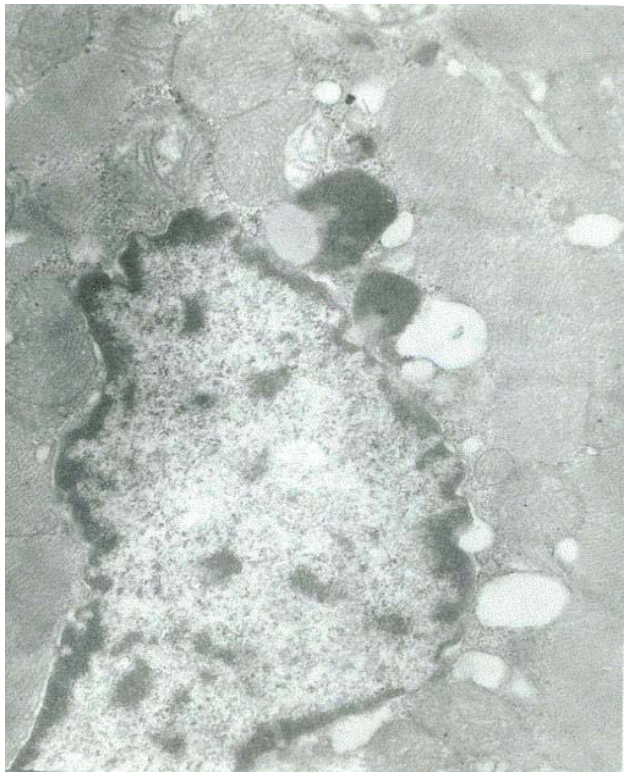

Figure 6 – ***Dog myocardium, transient myocardial ischemia 20 minutes: dissociation of sarcomeres by interstitial edema, Z line wave, increase of mitochondrial lesions (EM, ×9500)***

Figure 7 – ***Human myocardium, ischemic heart disease 57 years old male: irreversible sarcomeral lesions; intercalary disk and lysosome also on observed***

Figure 8 – ***Human myocardium ischemic heart disease 61-years-old male, myocardial biopsy after 40 minutes aortic cross clumping : necrosis of any sarcomeres, agglomeration of mitochondrial dilatation and breaking of sarcoplasmatic reticulum, edematous zones (EM, ×11000)***

Perhaps the major problem encountered in clinical studies [15–25] however is to discern whether a reversible defect of contractility is cause by stunning [17–20], silent ischemia [16] or hibernation.

Stunning may coexist in some cases with myocardial hibernation. This phenomenon were observed in the vicinity myocardial infarct zone and subendocardium Mechanism of myocardial stunning were information regarding the pathogenesis of post-ischemic dysfunction in lumen is quite scare.

Almost all data are derived from the setting of cardiac surgery because postoperative left ventricular dysfunction has been a major clinical concern for many years and because surgery provides easy to myocardial and coronary venous blood specimens for biochemical studies.

Experimental studies suggest two major mechanisms for myocardial stunning: (1) generation of oxygen-derived free radicals with consequent oxidative stress (oxyradical hypothesis [17, 28]), and (2) impaired calcium homeostasis resulting in transient calcium, overload, excitation–contraction uncoupling and/or decreased myofibrilar sensitivity to calcium ("calcium hypothesis"). A number of observations [29] are consistent with the concept that oxyradicals contribute to pathogenesis of postoperative LV dysfunction after coronary artery bypass grafting (CABG). For diagnosis of stunned myocardium scintigraphic and echocardiographic technique (in particular Tc–sestamibi imaging and low dose dobutamine echocardiography appear promising for the prospective diagnosis. Myocardial stunning occurs in several clinical situations and may be a cause of dramatic morbidity and mortality [25, 26].

As mentioned above, stunning may adversely affect the outcome of CABG in patients who are high risk because of conditions such as impaired baseline contractile reserve, long aortic clamping time, repeated CABG, unstable angina, left main coronary disease or concomitant valve replace; indeed, stunning is probably the most frequent cause of myocardial dysfunction and low cardiac output syndrome after CABG. Experimental studies have repeatedly demonstrated that stunning can be corrected with inotropic therapy [5]. In addition, there is considerable evidence that this from of contractile failure is preventable by calcium antagonists [30] or antioxidant therapy [5, 6, 28].

Conclusions

Stunned myocardium represent a prolonged contractile dysfunction that may persist for minutes to days after a brief episode of ischemia up to 15 minutes in condition of normal blood flow and the absence of necrosis. In experimental material and during cardiac surgery, the successfully myocardial biopsies revealed only the evolution of reversible mitochondrial and sarcoplasmatic reticulum lesions, slight glycogen granules depletions, sporadically dissociation of myofilaments by edema. The mechanism of stunning involves generation of oxygen free radicals as well as alterations in calcium homeostasis and possibly alterations in contractile protein structure. Stunning has been observed in several clinical scenarios, including after percutaneous transluminal coronary angioplasty, unstable angina, stress – induced ischemia, after thrombolysis, and after cardiopulmonary bypass. Oxygen radical scavengers and calcium channels blockers have been shown to enhance function of stunned myocardium in experimental studies and in few clinical studies calcium channel blockers have been show to ameliorate stunning. The future challenge is how to minimize the stunning phenomena in clinical practice.

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