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Pulmonary morphofunctional effects of acute myocardial infarction

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ABSTRACT: Acute myocardial infarction (AMI) may yield several respiratory changes. Nevertheless, no comprehensive pulmonary morphological/physiological correlation has been performed under this condition. The aims of the present investigation were: 1) to determine the respiratory parameters in an experimental model of coronary artery occlusion, 2) to relate these results to findings from lung histopathology, and 3) to evaluate the effects of propranolol used prior to AMI.

Twenty-eight rats were anaesthetized and mechanically ventilated. In the control group (C), a suture line was passed around the left anterior descending coronary artery (LADCA). The infarct group (I) was similarly prepared but the LADCA was ligated and infarct resulted. In the control/propranolol (CP) and infarct/propranolol (IP) groups, propranolol was intravenously injected 5 min before surgery as performed in groups C and I, respectively. Lung static ($E_{L,st}$) and dynamic ($E_{L,dyn}$) elastances, airway resistance ($R_{L,int}$), and viscoelastic/inhomogeneous pressure ($\Delta P2_L$) were determined before and 30, 60 and 120 min after surgery.

In group I, $E_{L,st}$, $E_{L,dyn}$, $R_{L,int}$ and $\Delta P2_L$ increased progressively throughout the experiment, and were higher than those found in groups C, CP and IP. All respiratory parameters but $E_{L,st}$ remained unaltered in group IP. Lung histopathological examination demonstrated alveolar, interstitial and intrabronchial oedema in group I. Group IP showed only interstitial oedema.

Acute myocardial infarction yields lung resistive, elastic and viscoelastic changes. The last two results from alveolar and interstitial oedema, respectively. The previous use of propranolol diminishes respiratory changes.

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Acute myocardial infarction (AMI) may yield several respiratory changes, *e.g.* reduced pulmonary compliance, increased resistance [1], and decreased functional residual and vital capacities [2]. These changes are generally associated with ventricular dysfunction and subsequent pulmonary flooding. Several investigators have reported that early interstitial oedema does not appear to influence airway resistance, which augments only with alveolar flooding [3]. Other authors, however, have suggested that airway resistance may rise early, occurring at the level of either small [4] or large airways [5]. The mechanisms invoked to explain the mechanical changes include direct airway luminal narrowing by interstitial fluid [6], vagally mediated reflexes [7] and compression of the airways by adjacent congested arteries [4]. However, recent reports have proven the latter mechanism to play a relatively minor role in causing airway obstruction [8]. A comprehensive study evaluating the temporal evolution of lung resistance, elasticity, viscoelasticity and other mechanical unevennesses after AMI has hitherto not been performed. Thus, the first two aims of this investigation were: 1) to determine these respiratory parameters in an experimental model of coronary artery occlusion [9, 10], and 2) to relate these results to findings from lung histopathological analysis.

Pretreatment with a β-adrenergic blocker appears to yield beneficial effects on the ischaemic myocardium, both in experimental animals and patients. β-Blocker therapy is associated with lower mortality and better prognosis [11], less frequent arrhythmias [12] and reduction of infarct size [13]. Although there are many reports concerning cardiovascular effects resulting from β-blocker use prior to AMI, no data have been previously reported regarding pulmonary mechanics. Thus, as a third goal, this work intended to determine to what extent pretreatment with propranolol modifies respiratory mechanical parameters and lung histopathological findings after AMI.

Material and methods

Twenty-eight male Wistar rats were randomly divided into four groups. In the control group (C) (n=7, body weight (BW) 348±12 (mean±SD) g), sham surgery was performed: after anterior chest wall resection, a suture was passed around the left anterior descending coronary artery (LADCA), vascular occlusion being avoided. In the infarct group (I) (n=8, BW 347±17 g), a protocol similar to that used in group C was performed followed by LADCA ligature and infarct induction. In the control/propranolol group

(CP) (n=61, BW 277±14 g), the animals received an *i.v.*, bolus injection of propranolol (2 mg·kg^{-1}) before sham surgery. In the infarct/propranolol group (IP) (n=7, BW 315± 35 g), propranolol (2 mg·kg^{-1} *i.v.*) was injected 5 min prior to coronary occlusion. β-Adrenergic blockade was confirmed by cardiac frequency reduction, as registered on an electrocardiogram (ECG) 5 min after propranolol injection.

Experimental protocol

The rats were sedated (diazepam 5 mg *i.p.*), anaesthetized with pentobarbital sodium (20 mg·kg^{-1} *i.p.*) and maintained with an hourly dose of 10–15% of the initial dose. Then, the animals were tracheotomized, and a snugly fitting cannula (1.4 mm internal diameter (ID)) was introduced into the trachea. The animals rested in the supine position on a surgical table. A polyethylene catheter (0.2 mm ID) filled with heparinized (10 IU·mL^{-1}) saline solution (0.9% NaCl) was introduced into the right femoral artery. The catheter was connected to a transducer (Statham P23Db; Statham, Hato Rey, Puerto Rico) for blood pressure measurements. The right femoral vein was dissected and cannulated for propranolol administration.

The anterior chest wall was surgically removed. Mechanical ventilation (Model 683; Harvard Apparatus, Southnatick, MA, USA) at a frequency of 80 breaths·min^{-1} a tidal volume (VT) of 7 mL·kg^{-1} and an adequate amount of positive end-expiratory pressure (PEEP) was applied immediately before the pleural cavity was entered. The PEEP level was determined as follows. Before the pleural space was opened, the ventilator was disconnected at end expiration and the airways were occluded. After pleural incision there was an increase in tracheal pressure (Ptr) which corresponded to the elastic recoil pressure of the lung at its relaxation volume. Thereafter, the same pressure was applied to the lung (PEEP 2.0–2.5 cmH_2O).

An adequate pneumotachograph (1.5 mm ID, length 4.2 cm, distance between side ports 2.1 cm) constructed according to MORTOLA and NOWORAJ [14] was connected to the tracheal cannula for measurement of airflow (V'), and change in lung volume (VT). The pressure gradient across the pneumotachograph was determined by means of a Statham PM15E differential pressure transducer (Stratham, Hato Rey, Puerto Rico). The flow resistance of the equipment (Req), tracheal cannula included, was constant up to V' of 26 mL·s^{-1} and amounted to 0.13 cmH_2O·mL^{-1}·s. Equipment resistive pressure (Req × V') was subtracted from pulmonary resistive pressure so that the results represented intrinsic values. Because abrupt changes in diameter were not present in the circuit, errors in measurement of flow resistance were avoided [15]. The equipment dead space was 0.4 mL. Ptr was measured using a Hewlett-Packard 270 differential pressure transducer (Waltham, MA, USA). All signals were conditioned and amplified in a Beckman type R Dynograph (Schiller Park, IL, USA). V' and pressure signals were also passed through eight-pole Bessel filters (902LPF; Frequency Devices, Haverhill, MA, USA) with the corner frequency set at 100 Hz, sampled at 200 Hz using a 12-bit analogue-to-digital converter (DT-2801A; Data Translation, Marlboro, MA, USA) and stored on a personal computer-compatible microcomputer. All data were collected using LABDAT software (RHT-InfoData, Montreal, Quebec, Canada).

Muscle relaxation was achieved with gallamine triethyliodide (2 mg·kg^{-1} *i.v.*), and artificial ventilation was provided by a Salziner constant flow ventilator (Instituto do Coração, São Paulo University, São Paulo, Brazil). During the test breaths, a 5-s end-inspiratory pause could be generated by adjusting the ventilator settings, whereas during baseline ventilation no pause was used. In order to avoid the effects of different V' and VT [16], and thence inspiratory duration [17], on the measured variables, special care was taken to keep VT (2 mL) and V' (10 mL·s^{-1}) constant in all animals.

Pulmonary mechanics

Pulmonary mechanics were measured using end-inspiratory occlusions after constant flow inflations [16, 18, 19]. In an open chest preparation, Ptr reflects transpulmonary pressure (PL). After end-inspiratory occlusion there is an initial fast drop in transpulmonary pressure (ΔP1L) from the preocclusion value down to an inflection point (Pi,L), followed by a slow pressure decay (ΔP2L) until a plateau is reached. This plateau corresponds to the elastic recoil pressure of the lung (PL,el) (fig. 1). ΔP1L selectively reflects pressure dissipated against airway resistance in normal animals and humans [18, 19], and ΔP2L reflects

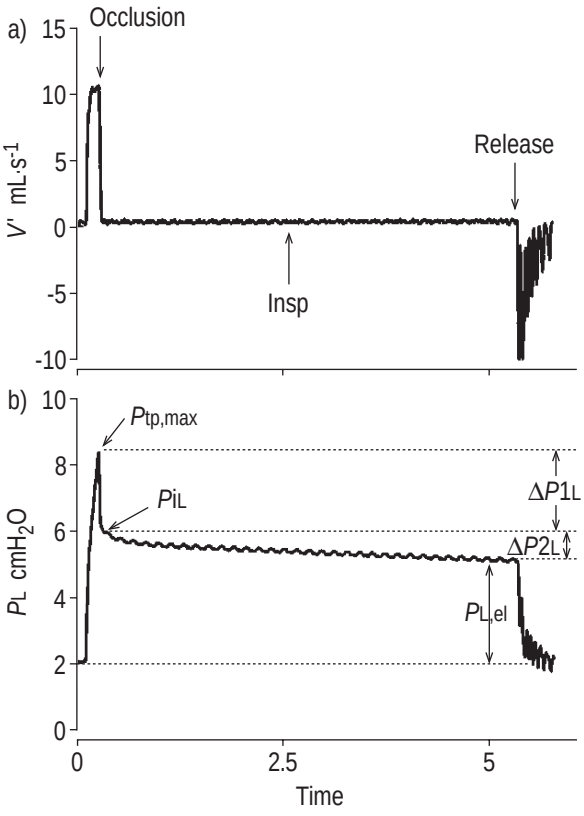

Fig. 1. – Trace of flow (V') and transpulmonary pressure (PL) from a rat with acute myocardial infarction. End-inspiratory occlusion is indicated with an arrow and was maintained for 5 s and then the airways were released. A positive end-expiratory pressure of 2 cmH_2O was used. ΔP1L: pressure dissipated against airway resistance; ΔP2L: pressure dissipated against viscoelastic and/or inhomogeneous lung component; PL,el: lung elastic recoil pressure; PiL: inflection point; Ptp,max: maximal transpulmonary pressure; Insp: inspiration.

the viscoelastic properties (stress relaxation) and/or inhomogeneities of lung tissues [19], together with a tiny contribution of pendelluft in normal situations [18, 20]. The total pressure drop ($\Delta P_{L,tot}$) is equal to the sum of $\Delta P1_L$ and $\Delta P2_L$. Airway resistance ($R_{L,int}$) was computed by dividing $\Delta P1_L$ by V'. Static lung elastance ($E_{L,st}$) was calculated by dividing $P_{L,el}$ by V_T. Dynamic lung elastance ($E_{L,dyn}$) was obtained by dividing Pi,L by V_T. ΔE was calculated as the difference $E_{L,dyn}$ - $E_{L,st}$. The data concerning lung elastance were presented in terms of $E_{L,st}$ and E_L instead of $E_{L,dyn}$ since they represent, respectively, the elastic and viscoelastic properties of the lung. Pulmonary mechanics were measured before and at three different occasions (30, 60 and 120 min) after the end of infarct induction or sham surgery. Before each measurement period, the lungs were hyperinflated three times to a P_L of 30 cmH_2O in order to keep volume history constant. Values are presented as mean±SD of eight–10 determinations on each occasion in all groups, and were corrected for BW. The experiments did not last >3 h.

The delay between the beginning and end of valve closure (10 ms) was allowed for by back extrapolation of the pressure records to the time of occlusion, and the corrections in pressure, although very minute, were performed as previously described [16].

Data analysis was performed using ANADAT software (RHT-InfoData, Montreal, Canada).

Continuous records of transcutaneous carbon dioxide tension and arterial blood oxygen saturation (S_{a,O_2}) were made using a SensorMedics FasTrac (Yorba-Linda, CA, USA), and ranged 4.9–5.6 kPa and 95–98%. The ECG was continuously monitored and registered on a chart recorder (Beckman type R Dynograph) at a paper speed of 25 mm·s-1.

After resting control data had been collected, the surgical procedure was performed. S_{a,O_2}, transcutaneous carbon dioxide tension, blood pressure and ECG were registered before and at 15, 30, 60 and 120 min after surgery.

Infarct induction

Infarct induction was achieved as previously described [9, 10]. Briefly, the anterior chest wall was resected in order to better visualize and manipulate the coronary artery, so avoiding accidental bleeding that could lead to mechanical respiratory changes [21]. Then, the heart was partially immobilized, with its apex held gently between the thumb and index finger of the left hand, and pointed craniad and slightly to the right side of the rat. In this position, a small curved needle (propylene 6.0 G, cardiovascular VT-22961) could readily carry a thread between the point of entry of the left margin of the pulmonary cone and the middle of an imaginary line connecting this point with the closest point on the insertion line of the left auricular appendage. After ligation, virtually the whole lateral wall of the left ventricle becomes ischaemic [10]. In addition, an immediate electrocardiographic change was observed.

Histology and morphometry

Morphometric analysis was performed in excised lungs at functional residual capacity. Immediately after the removal of the lungs *en bloc*, which took ≤90 s, they were frozen by rapid immersion in liquid nitrogen. Frozen lungs were fixed in Carnoy′s solution (ethanol/chloroform/acetic acid, 70:20:10) for 24 h at -70°C. Solutions with progressively increasing concentrations of ethanol at -20°C were then substituted for Carnoy′s solution until 100% ethanol was reached. The tissue was maintained at -20°C for 4 h, warmed to 4°C for 12 h, and then allowed to reach and remain at room temperature (25°C) for 2 h [22]. After fixation, the tissue block obtained from midsagittal slices at the level of the axial bronchus of the left lung was embedded in paraffin. Four-millimeter-thick slices were obtained by means of a microtome, and were stained with haematoxylin/eosin. Morphometric analysis was performed using an integrating eyepiece with a coherent system made of a 100-point grid consisting of 50 lines of known length coupled to a light microscope (Axioplan; Zeiss, Jena, Germany). Interstitial and alveolar oedemas were determined in each rat by the point-counting technique [23] across five random noncoincident microscopic fields at a 40× magnification. The eyepiece comprised 100 points and 50 lines. Points falling on oedematous alveolar lumina were counted and divided by the total number of points in each microscopic field; thus, data were reported as the fractional area of alveolar oedema. In order to quantify interstitial oedema, five transversely sectioned arteries were analysed in each rat: the cuff area was determined by the number of points falling on the interstitial space, and then divided by the vessel diameter; thus, data were reported as cuff-to-vessel area ratio. Bronchial oedema and constriction were evaluated by counting bronchi with oedema and/or constriction in relation to the total number of bronchi present in the slide (10× magnification); data were reported as fractions of the total number of bronchi.

The excised heart was immediately cut open and the left ventricle examined for an area of gross swelling and paleness. Three slices of 2 × 2 mm were cut from these pale areas, and then fixed with glutaraldehyde 2.5% and phosphate buffer 0.1 M (pH 7.4) for 60 min at -4°C. The slices were then rinsed in phosphate buffer, post-fixed with 1% osmic tetroxide in phosphate buffer for 30 min and rewashed three times in phosphate buffer. Finally, the slices were dehydrated in an acetone series and then placed in a mixture of 1:1 acetone:Epon overnight before embedding in Epon for 6 h. After fixation the material was kept for 48 h at 60°C before being submitted to ultramicrotomy for transmission electron microscopy in order to confirm infarction.

All animals received humane care in compliance with the "Principles of Laboratory Animal Care" formulated by the National Society for Medical Research and the "Guiding Principles in the Care and Use of Animals" approved by the Council of the American Physiological Society, USA [24].

Statistical methods

Statistical analysis was used to compare the results gathered from each group of rats as a function of time. Firstly, the normality of the data (Kolmogorov-Smirnov test with Lilliefor′s correction) and the homogeneity of variances (Levene median test) were tested. If both conditions were satisfied, repeated measures one-way analysis of variance

Table 1. – Mean blood pressure and cardiac frequency (f_C) in rats with and without propanolol pretreatment

	Time min				
	0	0 (Prop.)	30	60	120
BP mmHg					
C	92±10		93±20	97±33	89±29
I	97±03		82±11	80±10	78±16
IP	90±14	90±14	88±25	83±18	90±10
f_C beats·min^{-1}					
C	352±34		337±38	322±33	315±00
I	335±31		312±44	293±14	293±14
IP	336±03	242±09*	215±02*	206±02*	206±02*

Data are presented as mean±SD of six–eight animals per group, in control (C), infarct (I), and infarct/propranolol (IP) groups, observed prior to (0 min), and at 30, 60 and 120 min after, the surgical procedure. BP: blood pressure; (Prop.): propranolol); *: $p<0.05$ *versus* presurgery (0 min) value.

(ANOVA) was used. Otherwise, Kruskal-Wallis ANOVA was selected instead. If multiple comparisons were required, the Student-Newman-Keuls test was applied.

All mechanical parameter data gathered from groups C, I, CP and IP were compared using multivariate analysis (SuperANOVA). If multiple comparisons were then required, Scheffe's test was applied.

In order to compare the morphological variables among all groups, firstly, data angular transformation ($X' = \text{sen}^{-1}X$) was performed for percentage variables, and then one-way ANOVA was used. Multiple linear regression was performed in order to identify the relationships between functional and morphological parameters (dependent variables were represented by each functional parameter: $E_{L,st}$, E_L, ΔP_{2L}, and $R_{L,int}$; predictive variables were represented by morphological data, *i.e.* alveolar, interstitial, and bronchial oedema, and bronchoconstriction). The significance level was always set at 5%.

Results

Mean arterial blood pressure (BP) and cardiac frequency remained unaltered in groups C and I as a function of time (table 1). In group IP propranolol administration did not decrease BP, but cardiac frequency fell by 30.5% 5 min

Table 2. – Mechanical variables before and after infarct with and without propranolol pretreatment

	Time min			
	0	30	60	120
V' mL·s^{-1}				
C	9.74±0.28	9.99±0.27	9.97±0.23	9.856±0.26
CP	9.93±0.21	9.88±0.22	10.24±0.55	10.01±0.49
I	10.06±0.20	9.98±0.22	9.83±0.24	10.04±0.28
IP	10.13±0.17	10.07±0.15	10.10±0.27	10.07±0.25
V_T mL				
C	2.01±0.01	2.03±0.05	1.99±0.03	2.01±0.04
CP	1.96±0.07	1.98±0.03	2.03±0.04	1.98±0.03
I	2.01±0.04	2.00±0.02	2.00±0.02	2.01±0.03
IP	1.98±0.03	1.98±0.02	1.99±0.03	1.98±0.03
$E_{L,st}$ cmH_2O·mL^{-1}·kg				
C	0.560±0.07	0.573±0.13	0.621±0.15	0.649±0.15
CP	0.549±0.08	0.674±0.12	0.710±0.23	0.744±0.26
I	0.591±0.14	0.627±0.14	0.749±0.15	0.829±0.15*
IP	0.644±0.11	0.701±0.03	0.756±0.11	0.856±0.10*
E_L cmH_2O·mL^{-1}				
C	0.279±0.06	0.298±0.03	0.330±0.09	0.289±0.03
CP	0.356±0.10	0.354±0.06	0.371±0.10	0.354±0.05
I	0.297±0.05	0.326±0.12	0.399±0.10	0.503±0.18*,+
IP	0.321±0.10	0.289±0.05	0.328±0.09	0.336±0.10
$\Delta P_{L,tot}$ cmH_2O				
C	1.782±0.32	1.669±0.19	1.688±0.30	1.526±0.20
CP	2.031±0.49	2.065±0.23	2.159±0.30	1.901±0.24
I	1.717±0.35	1.772±0.44	1.922±0.34	2.413±0.72*,+
IP	1.706±0.28	1.528±0.232	1.724±0.32	1.695±0.24
ΔP_{2L} cmH_2O				
C	0.560±0.12	0.603±0.06	0.658±0.19	0.579±0.06
CP	0.615±0.09	0.699±0.12	0.755±0.21	0.699±0.11
I	0.596±0.10	0.650±0.23	0.797±0.20+	1.015±0.38*,+
IP	0.634±0.20	0.573±0.10	0.655±0.18	0.666±0.19
$R_{L,int}$ cmH_2O·mL^{-1}·s·kg				
C	0.043±0.01	0.037±0.01	0.036±0.01	0.034±0.01
CP	0.037±0.01	0.039±0.01	0.039±0.01	0.034±0.01
I	0.040±0.01	0.040±0.02	0.041±0.01	0.050±0.02*,+
IP	0.034±0.01	0.030±0.01	0.034±0.01	0.032±0.01

Data are presented as mean±SD of six–eight animals per group (eight–10 determinations per animal), in the control (C), control/pro-pranolol (CP), infarct (I) and infarct/propranolol (IP) groups, observed before (0 min), and at 30, 60 and 120 min after, surgery. V': flow; V_T: tidal volume; $E_{L,st}$: pulmonary static elastance; E_L: difference between pulmonary dynamic and static elastances; $\Delta P_{L,tot}$: total pulmonary pressure variation; ΔP_{2L}: pulmonary viscoelastic/inhomogeneous pressure change; $R_{L,int}$: pulmonary resistance. *: $p<0.05$ *versus* presurgery (0 min) value; +: $p<0.05$ *versus* IP group.

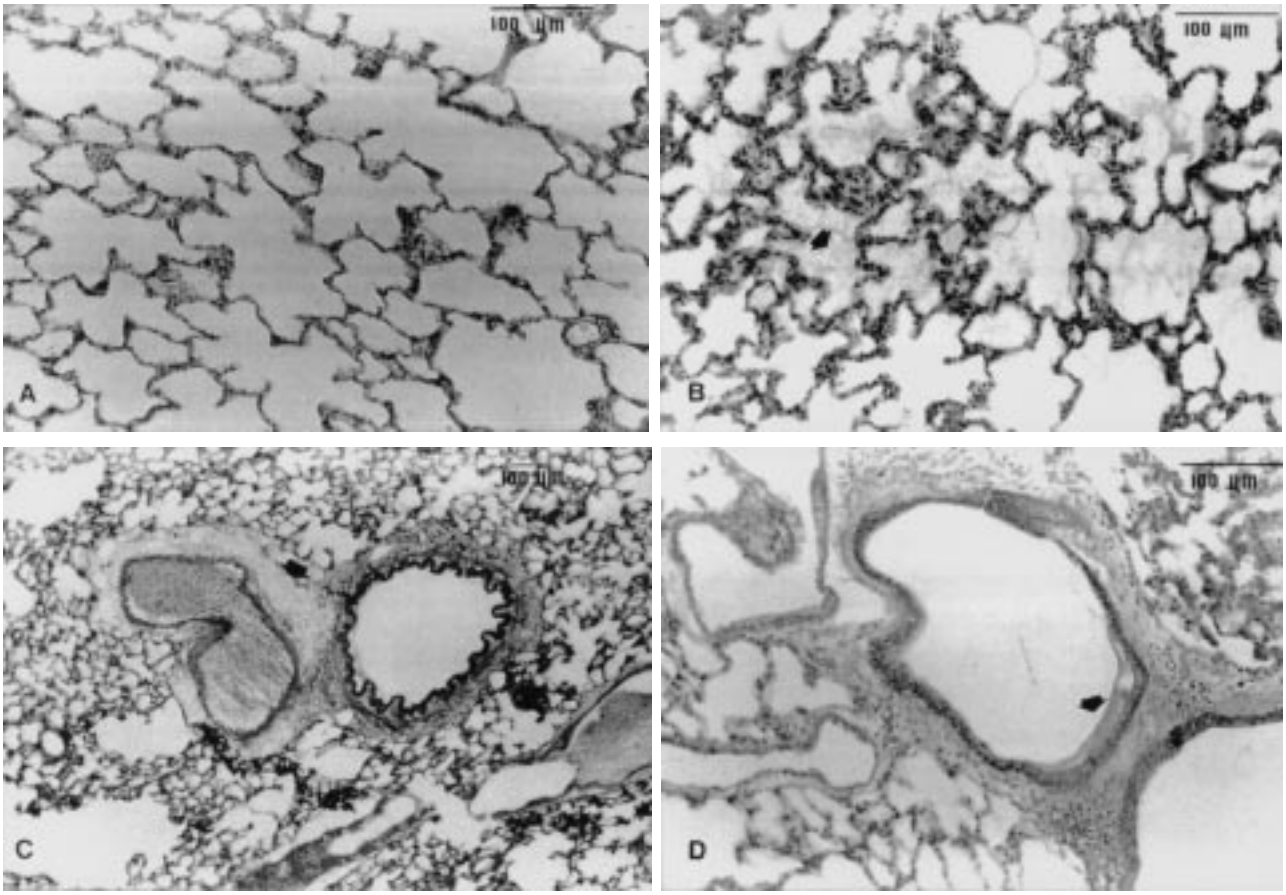

Fig. 2. – Photomicrographs of: A) normal pulmonary parenchyma (control group); and B–D) pulmonary parenchyma 120 min after acute myocardial infarction with: B) alveolar oedema (arrow); C) interstitial oedema (arrow); and D) intrabronchial oedema (arrow).

after its injection; thereafter cardiac frequency did not change (table 1).

Myocardial infarction was confirmed in all cases by the occurrence of an S-T segment elevation on the ECG just after coronary occlusion. Six out of eight animals in group I developed ventricular arrhythmia (ventricular tachycardia was present in four animals). These arrhythmias were observed until 20 min after infarct induction. and the sinusal rhythm spontaneously resumed after 30 min. No arrhythmia was observed in groups C and CP. The group pretreated with propranolol (IP) presented fewer arrhythmic episodes than group I (one animal with ventricular tachycardia and one rat with atrioventricular block).

Table 3. – Morphometric data in the presence and absence of infarction in rats with and without propranolol pretreatment

	Alveolar oedema fractional area	Interstitial oedema cuff-to-vessel area ratio	Broncho-constriction proportion of bronchi	Intrabronchial oedema proportion of bronchi
C	0.18±0.08	0.44±0.13	0.18±0.13	0.13±0.08
CP	0.26±0.09	0.55±0.25	0.10±0.09	0.23±0.08
I	0.50±0.13*	1.65±0.61*	0.23±0.15	0.44±0.17*
IP	0.23±0.10	1.46±0.56*	0.27±0.08	0.17±0.13

Data are presented as mean±SD of six–eight animals per group (five fields per animal) in the control (C), control/propranolol (CP), infarct (I) and infarct/propranolol (IP) groups at 120 min after surgery. *: p<0.05 *versus* presurgery (0 min) values.

Inspiratory V' and volume remained unaltered in groups C, I, CP and IP (table 2). All mechanical variables in groups C and CP presented no statistically significant changes.

$E_{L,st}$, ΔE_L, $\Delta P_{L,tot}$, $\Delta P2_L$ and $R_{L,int}$ increased during the course of the experiment in group I, reaching statistically significantly higher values at 120 min after induction of infarction (table 2). In group IP $E_{L,st}$ augmented progressively with time (significant at 120 min), but the other mechanical parameters remained unaltered.

At 120 min, ΔE_L, $\Delta P_{L,tot}$, $\Delta P2_L$ and $R_{L,int}$ were significantly higher in group I than in groups C, CP and IP. $E_{L,st}$ was higher in groups I and IP than in groups C and CP, respectively (table 2).

Histopathological examination of the lungs demonstrated alveolar, interstitial and intrabronchial oedema in group I (fig. 2). Group IP showed only interstitial oedema (table 3). No bronchoconstriction was found in any group.

Regression analyses between ΔE_L and $\Delta P2_L$ and interstitial oedema in groups I and C showed significant correlations (p=0.006 and 0.007; r=0.69 and 0.68, respectively). $E_{L,st}$ correlated with alveolar oedema (p=0.020; r=0.62), as depicted in fig. 3). Propranolol-pretreated groups (CP and IP) did not show any correlation between morphological and mechanical data.

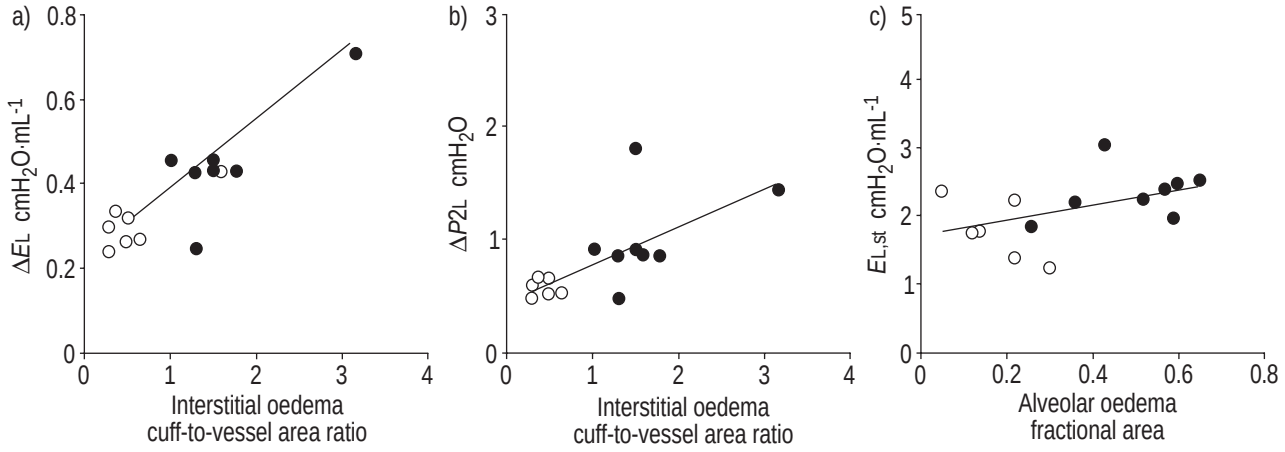

Fig. 3. – Correlation between pulmonary mechanical parameters and histopathological findings obtained in control (○) rats and in the presence of infarction (●): a) difference between pulmonary dynamic and static elastances (EL); p=0.006, r=0.69; b) pulmonary viscoelastic/inhomogeneous pressure changes (ΔP2L); p=0.07, r=0.68; and c) pulmonary static elastance (Est,L); p=0.020, r=0.62. The slope represents linear regression.

The transmission electron microscopy performed on the hearts of group I rats showed larger mitochondria with a decreased density of matrix and rarefaction of cristae, characterizing a very important tumefaction; glycogen particles, in this group, were almost absent. In group IP the mitochondria generally presented a normal configuration, scattered glycogen particles and preserved filament organization (fig. 4).

Discussion

The method used for the determination of pulmonary mechanics allows the identification of its elastic, resistive and viscoelastic and/or inhomogeneous components [18, 25]. In group I, EL,st, ΔEL, ΔPL,tot, ΔP2L and RL,int tended to increase progressively as a function of time, but were significantly higher than control values only at 120 min postinfarction (table 2). It should be emphasized that positive pressure ventilation could have postponed the beginning of the pulmonary mechanical changes.

Airway resistance and viscoelastic and/or inhomogeneous changes were abolished by pretreatment with propranolol. In group IP only EL,st was found to be significantly higher than in the control group at 120 min. Although infarction in animals injected with propranolol also resulted in some degree of ventricular dysfunction, it was less important, less liquid was accumulated in the lungs and pulmonary mechanical modifications were thus minimized (table 2). The findings observed in group IP could be explained by the cardiovascular actions of β-blocker agents. As β-blockade lessens oxygen consumption, ischaemic damage is reduced, and probably also the necrotic area [13], thus allowing better cardiac performance.

Pulmonary fluid accumulation has been implicated as a major cause of pulmonary changes after AMI [1, 26, 27]. The intensity of lung liquid accumulation is determined by left ventricular function [28], and its sequence of accumulation was well described by STAUB *et al.* [29]. In the present study, histopathological analysis showed significant alveolar and bronchial liquid accumulation only in the presence of infarction in animals not being administered propranolol (group I). In addition, interstitial oedema was detected in groups I and IP (table 3). These findings suggest that infarct-dependent ventricular dysfunction was reduced by β-blocker action.

RL,int represents airway resistance [19]. The increase in RL,int was probably influenced by bronchial intraluminal oedema (fig. 2), suggesting increased pulmonary liquid accumulation, in accordance with STAUB *et al.* [29]. In fact, a weak correlation between RL,int and intrabronchial oedema (p=0.027, r=0.51) was detected in the present study, which clearly must be cautiously considered. On the other hand, WAGNER and MITZNER [8] found that vascular engorgement of the airway wall *per se* has a negligible effect on airway obstruction, and suggested a reflex origin for the bronchoconstriction of the small airways. The present results evidenced no bronchoconstriction (fig. 2, table 3), and pulmonary liquid accumulation was the probable cause of increased RL,int in AMI.

Variations in ΔP2L can be closely related to the stress relaxation properties of the lung and pendelluft [18–20]. The increase in ΔP2L (66.7%) suggests a greater pressure dissipation in order to overcome pulmonary viscoelastic and/or inhomogeneous components in group I than in group C. This result is in agreement with previous studies [1, 4, 8, 26] reporting peripheral airway obstruction as of paramount importance to the mechanical changes found after AMI. The ΔP2L increment was well correlated with interstitial oedema. Interestingly enough, ISHII *et al.* [30] described a progressive increase in peripheral RL,int in the presence of pulmonary oedema. Along this line HOGG *et al.* [4] demonstrated greater peripheral than central RL,int, which could be explained by small airways compression due to vascular engorgement. However, recent work [8] has shown that vascular engorgement *per se* yields only a minute amount of small airways obstruction. Finally, HALES and KAZEMI [1] have shown fluid retention in the small airways after myocardial infarction. Clearly, further studies are required to verify the reason for the higher pressure losses in the lung periphery after AMI.

The effects of lung congestion on pulmonary compliance have been studied in models of pulmonary oedema [3], resulting in a direct correlation between the degree of pulmonary vascular congestion and progressive compliance reduction. In the present work EL,st and ΔEL also

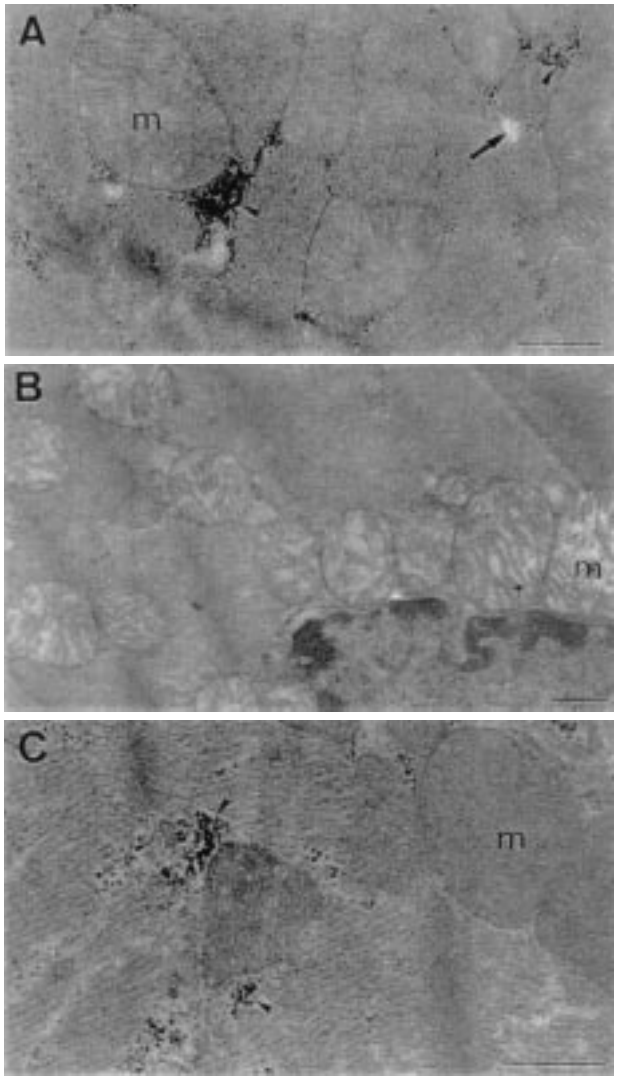

Fig. 4. – Photomicrograph of myocardial cell of: control group, with parallel myofilaments separated by elongated mitochondria (m), tubules of sarcoplasmic reticulum (arrow) and scattered glycogen (arrowheads); and B,C) in the presence of infarction: B) without propranolol pretreatment, showing swollen mitochondria and the virtual absence of glycogen; and C) with propranolol pretreatment, showing a myocardial cell similar to control with normal mitochondria and scattered glycogen particles. (Internal scale bars=1 μm.)

had their histopathological counterparts: the former was well correlated with alveolar oedema, whereas the latter presented a positive correlation with interstitial oedema. Alveolar flooding could have diminished the gaseous volume in the lung, but not total lung volume, thus stiffening the lung. In addition, surfactant action may have been thwarted by intra-alveolar fluid accumulation, resulting in elevated $E_{L,st}$. The reasons for the increase in $\Delta P2_L$ also pertain to ΔE_L, since both reflect pulmonary viscoelastic/inhomogeneous behaviour [19, 20]. In group IP $E_{L,st}$ was also increased, although no physiological/morphological correlation could be found. Nevertheless, these animals showed interstitial oedema (table 3), suggesting that pulmonary fluid accumulation could be the primary cause of $E_{L,st}$ changes.

After coronary occlusion there was neither significant blood pressure reduction nor a decrease in cardiac frequency (table 1), in agreement with VATNER [31]. In the present investigation, the area which had undergone infarction bulged after coronary artery occlusion, assuming a paradoxical motion, as previously described [31].

Upward S-T segment displacement on ECG was observed immediately after coronary occlusion in all animals which had undergone infarction (groups I and IP). Arrhythmias were observed mainly in group I. Nevertheless, these arrhythmias reverted spontaneously 30 min after infarction, the rhythm remaining unaltered until the end of the experiment. This biphasic behaviour of cardiac rhythm has been previously reported in dogs [32], but scarcely seen after coronary occlusion in rats [9]. The animals pretreated with propranolol showed a lower incidence of arrhythmias after infarction. This advantageous effect of propranolol could be secondary to its anti-arrhythmic action [12], or, mainly, to a reduction in the area undergoing infarction (its anti-ischaemic action) [33].

Infarct confirmation by histopathological analysis using light microscopy is only possible after 6–8 h of ischaemia [34], but ultramicroscopy can identify infarct lesions as soon as 20 min after coronary flow interruption [35]. Some early irreversible damages, *i.e.* glycogen loss, cellular oedema and mitochondrial engorgement (with increased matrix volume and eventual rupture) were detected in group I, as previously reported [36].

The beneficial effects of β-blockers administered just after infarction are well established [11]. In the present study, the animals were treated with propranolol before infarction induction, supporting the relevance of the use of β-blockers in populations at high risk of AMI [13]. To the authors′ knowledge, no previous experimental studies focusing on β-blocker effects on myocardial infarction have dealt with respiratory mechanics. Although various doses and conditions of administration (before or after infarction) have been used, in this study, venous administration of propranolol (2 $mg \cdot kg^{-1}$) was chosen, since it leads to a more uniform blood concentration, and had been previously and successfully employed in rats [37]. In the present study, there were neither haemodynamic nor respiratory complications related to β-blocker injection, as depicted in tables 1 and 2.

In conclusion, pulmonary resistive, elastic, and viscoelastic and/or inhomogeneous components increased 2 h after acute myocardial infarction. The last two findings were significantly correlated with alveolar and interstitial oedema, respectively. The higher pulmonary resistance might be dependent on the degree of intrabronchial oedema. Intravenous propranolol (2 $mg \cdot kg^{-1}$) *per se* did not alter pulmonary mechanics in normal rats. However, the previous use of propranolol in rats with acute myocardial infarction minimized respiratory changes, allowing elevations in only pulmonary elastance and interstitial oedema. Finally, pulmonary mechanical changes after myocardial infarction resulted mainly from pulmonary fluid retention.

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References

1. Hales CA, Kazemi H. Small airways function in myocardial infarction. *N Engl J Med* 1974; 290: 761–765.
2. Kidner PH. Respiratory function following myocardial infarction. *Acta Cardiol* 1981; 1: 1–6.
3. Cook CD, Mead J, Schreiner GL, Frank NR, Craig JM. Pulmonary mechanics during induced pulmonary edema in anesthetized dogs. *J Appl Physiol* 1959; 14: 177–186.
4. Hogg JC, Agarawal JB, Gardiner AJS, Palmer WH, Macklem PT. Distribution of airway resistance with developing pulmonary edema in dogs. *J Appl Physiol* 1972; 32: 20–24.
5. Lai-Fook SJ, Beck KC, Sutcliffe AM, Donaldson JT. Effect of edema and height on bronchial diameter and shape in excised dog lung. *Respir Physiol* 1984; 55: 223–237.
6. Milic-Emili J, Ruff F. Effects of pulmonary congestion and edema on the small airways. *Bull Eur Physiopathol Respir* 1971; 7: 1181–1196.
7. Lloyd TC. Reflex effects of left heart and pulmonary vascular distention on airways of dogs. *J Appl Physiol* 1980; 49: 620–626.
8. Wagner EM, Mitzner W. Effects of bronchial vascular engorgement on airway dimensions. *J Appl Physiol* 1996; 81: 293–301.
9. Johns TNP, Olson BJ. Experimental myocardial infarction. I. A method of coronary occlusion in small animals. *Ann Surg* 1954; 140: 675–682.
10. Selvey H, Bajusz E, Grasso S, Mendell P. Simple technique for the surgical occlusion of coronary vessels in rat. *Angiology* 1960; 11: 398–407.
11. ISIS-1 (First International Study of Infarct Survival) Collaborative Group. Randomised trial of intravenous atenolol among 16.027 cases of suspected acute myocardial infarction: ISIS-1. *Lancet* 1986; 2: 57–66.
12. Ahumada GG, Karlsberg RP, Jaffe AS, Ambos HD, Sobel BE, Roberts R. Reduction of early ventricular arrhythmia by acebutolol in patients with acute myocardial infarction. *Br Heart J* 1979; 41: 654–659.
13. Rasmussen MM, Reimer KA, Kloner RA, Jennings RB. Infarct size reduction by propranolol before and after coronary ligation in dogs. *Circulation* 1977; 56: 794–798.
14. Mortola JG, Noworaj A. Two-sidearm tracheal cannula for respiratory airflow measurements in small animals. *J Appl Physiol* 1983; 55: 250–253.
15. Chang HK, Mortola JP. Fluid dynamic factors in tracheal pressure measurement. *J Appl Physiol* 1981; 51: 218–225.
16. Kochi T, Okubo S, Zin WA, Milic-Emili J. Flow and volume dependence of pulmonary mechanics in anesthetized cats. *J Appl Physiol* 1988; 64: 441–450.
17. Similowski T, Levy P, Corbeil C, *et al*. Viscoelastic behavior of lung and chest wall in dogs determined by flow interruption. *J Appl Physiol* 1989; 67: 2219–2229.
18. Bates JHT, Rossi A, Milic-Emili J. Analysis of the behavior of the respiratory system with constant inspiratory flow. *J Appl Physiol* 1985; 58: 1840–1848.
19. Bates JHT, Ludwig MS, Sly PD, Brown K, Martin JG, Fredberg JJ. Interrupter resistance elucidated by alveolar measurement in open-chest normal dogs. *J Appl Physiol* 1988; 65: 408–414.
20. Otis AB, McKerrow CB, Bartlett RA, *et al*. Mechanical factors in distribution of pulmonary ventilation. *J Appl Physiol* 1956; 8: 427–443.
21. Martins MA, Zin WA, Younes RN, *et al*. Respiratory system mechanics in guinea pigs after hemorrhage: role of adrenergic stimulation. *Crit Care Med* 1990; 18: 515–519.
22. Nagase T, Lei M, Robatto FM, Eidelman DH, Ludwig MS. Tissue viscance during induced constriction in rabbit lungs: morphological-physiological correlations. *J Appl Physiol* 1992; 73: 1900–1907.
23. Weibel ER. Sterological methods. *In*: Practical Methods for Biological Morphometry. London, Academic Press, 1979; Vol. 1.
24. Bozzonalla JJ, Russel LD. Electron Microscopy: Principles for Biologists. Sudbury, Jones and Bartlett publishers| 1992.
25. Bates JHT, Baconnier P, Milic-Emili J. A theoretical analysis of interrupter technique for measuring respiratory mechanics. *J Appl Physiol* 1988; 64: 2204–2214.
26. Interiano B, Hyde RW, Hodges M, Yu PN. Interrelation between alterations in pulmonary mechanics and hemodynamics in acute myocardial infarction. *J Clin Invest* 1973; 52: 1994–2006.
27. Biddle TL, Yu PN, Hodges M, Chnace JR, Kronenberg MW, Roberts D. Hypoxemia and lung water in acute myocardial infarction. *Am H J* 1976; 92: 692–699.
28. Biddle TL, Khanna PK, Yu PN, Hodges M, Shah PM. Lung water in patients with acute myocardial infarction. *Circulation* 1974; XLIX: 115–123.
29. Staub NC, Nagano H, Pearce ML. Pulmonary edema in dogs, especially the sequence of fluid accumulation in lungs. *J Appl Physiol* 1967; 22: 227–240.
30. Ishii M, Matsumoto N, Fuyuki T, *et al*. Effects of hemodynamic edema formation on peripheral *versus* central airway mechanics. *J Appl Physiol* 1985; 59: 1578–1584.
31. Vatner SF. Correlation between acute reductions in myocardial blood flow and function in conscious dogs. *Circ Res* 1980; 47: 201–207.
32. Lazzara R, El-Sherif N, Hope RR, Scherlag BJ. Ventricular arrhythmias and electrophysiological consequences of myocardial ischemia and infarction. *Circ Res* 1978; 42: 740–749.
33. Aronow WS, Ahn C, Mercando AD, Epstein S, Kronzon I. Decrease in mortality by propranolol in patients with heart disease and complex ventricular arrhythmias is more an anti-ischemic than an antiarrhythmic effect. *Am J Cardiol* 1994; 74: 613–617.
34. Jennings RB. Early phase of the myocardial ischemic injury and infarction. *Am J Cardiol* 1969; 24: 753–765.
35. Fishbein MC, Meerbaum S, Rit J, *et al*. Early phase acute myocardial infarct size quantification: validation of triphenyl tetrazolium chloride tissue enzyme staining technique. *Am Heart J* 1991; 101: 593–600.
36. De Micheli A, Aranda A, Chavez E. Sustratos celulares del sindrome de infarto miocardico agudo. *Arch Inst Cardiol Mex* 1995; 65: 79–88.
37. Mill JG, Vassalo DV, Leite CM. Time course of changes in heart rate and sympathetic tone after coronary artery ligation in rats. *Braz J Med Biol Res* 1991; 24: 855–858.