Title: Direct protective effects of dexmedetomidine against myocardial ischemia-reperfusion injury in anesthetized pigs.

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Running head: dexmedetomidine and myocardial ischemia

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Abstract

Systemic administration of α_2-adrenergic agonists has been shown to protect ischemic myocardium, but the direct effects on ischemia-reperfused myocardium have not yet been clarified. This study was carried out to determine the effects of intracoronary dexmedetomidine (DEX) on the myocardial ischemia-reperfusion injury in anesthetized pigs. In open-chest pigs, the left anterior descending coronary artery (LAD) was perfused through an extracorporeal circuit from the carotid artery. They received intracoronary infusion of DEX at a rate of 1 ng/mL (group LD, n = 9), 10 ng/mL (group MD, n = 9), or 100 ng/mL (group HD, n = 9) of coronary blood flow or vehicle (group C, n = 12) for 30 min before ischemia. Myocardial stunning was produced by 12-min ischemia of the perfused area of LAD and 90-min reperfusion. The effect on reperfusion-induced arrhythmias was evaluated using the incidence of ventricular tachycardia or fibrillation after reperfusion. Regional myocardial contractility was evaluated with segment shortening (%SS). DEX significantly reduced the incidence of reperfusion-induced ventricular arrhythmias. DEX significantly improved the recovery of %SS at 90 min after reperfusion ($32.6 \pm 3.1\%$ in group C, $58.2 \pm 2.1\%$ in group LD, $61.1 \pm 1.8\%$ in group MD and $72.0 \pm 2.0\%$ in group HD. DEX suppressed the increase of plasma norepinephrine concentration after

reperfusion. The results indicate that DEX would exert the protective effect against ischemia-reperfusion injury by the direct action on the myocardium, which is not mediated through central nervous system.

Key words: dexmedetomidine, myocardial stunning, norepinephrine, α_2-adrenergic agonist, myocardial ischemia

Introduction

The progression of interventional recanalization for the treatment of acute coronary syndrome has been shown to reliably improve prognosis, on the other hand, it has been shown that the reperfusion itself causes a serious phenomenon termed reperfusion injury in clinical settings (1). Brief periods of coronary artery occlusion followed by reperfusion produces reversible contractile dysfunction, i.e., stunning, for several hours and are associated at the time of reflow with lethal arrhythmias, ventricular tachycardia (VT) or fibrillation (VF) (2, 3). Elimination of reperfusion injury may further improve the outcome of patients with coronary artery disease (1).

Experimental and clinical studies suggest that catecholamines promote the progression of myocardial injury, and arrhythmogenic effects of enhanced sympathetic activity and cardiac catecholamine concentrations are well documented (4). Previous studies reported that myocardial ischemia rapidly and massively increased the norepinephrine (NE) concentration in the myocardial tissue, but not in systemic blood sample (5, 6). Miura et al. (6) also reported that the elevation of NE concentration during ischemia was markedly attenuated by ischemic preconditioning.

α_2-Adrenergic agonists have been reported to have protective effects on the ischemic myocardium such as coronary artery stenosis or myocardial stunning, and

attenuate plasma NE levels, and preserve myocardial blood flow in the inner layers in the laboratory (7-9). It is well known that the central sympatholytic effect of α_2-adrenergic agonists may be beneficial during myocardial ischemia. α_2-Adrenoreceptors also exist at sympathetic nerve endings, and cardiac presynaptic α_2-adrenoreceptor stimulation decreased NE release from sympathetic nerve endings (10). Thus, cardiac presynaptic α_2-adrenoreceptor stimulation would attenuate the elevation of NE concentration in ischemic myocardium. However, the effects of cardiac presynaptic α_2-adrenoreceptor stimulation on ischemia-reperfused myocardium have not yet been clarified.

Dexmedetomidine (DEX) is a highly specific and selective α_2-adrenergic agonist. α_2-Adrenergic agonists are useful adjuncts to anesthesia because of their sedative, analgesic, sympatholytic, and specific hemodynamic effects (11). It has been shown that DEX was associated with a trend towards improved cardiac outcomes in a clinical setting (12). The aims of this study were to determine whether DEX could exert direct protective effects against myocardial stunning and reperfusion-induced ventricular arrhythmias, and to investigate the influence on the myocardial NE concentration in anesthetized pigs.

Materials and Methods

All experimental procedures used in this investigation were reviewed and approved by the Animal Care Committee of the Nagasaki University School of Medicine.

Instrumentation

Thirty-nine pigs (20–35 kg) of either sex were sedated with ketamine hydrochloride (20 mg/kg) intramuscularly. After intravenous (IV) access was established via an ear vein, the pigs were anesthetized with α–chloralose (100 mg/kg) and fentanyl (10 μg/kg) IV, followed by continuous infusion of α–chloralose (10 mg/kg/h) and fentanyl (5 μg/kg/h) throughout the study period. Through a midline cervical incision, the trachea was intubated for connection to a Harvard respiratory pump (Harvard Apparatus Co., South Natick, MA). Mechanical ventilation was facilitated by an intermittent IV infusion of vecuronium (0.2 mg/kg). Tidal volume, respiratory rate, and inspired oxygen concentration were adjusted to maintain the arterial carbon dioxide tension ($PaCO_2$) between 35 and 40 mmHg, and the arterial oxygen tension (PaO_2) between 100 and 300 mmHg. End-tidal CO_2 concentration was continuously monitored by using a gas analyzer (Capnomac Ultima, Datex, Helsinki, Finland). Lactated Ringer's solution was infused at a rate of 5 ml/kg/h. Sodium

bicarbonate was administered to maintain the base deficit within 5 mEq/L. Arterial blood glucose concentrations were measured before and during ischemia and maintained at baseline values with an IV infusion of 10% dextrose as needed throughout the study period. The esophageal temperature was maintained between 36°C and 37°C throughout the study period by using a warmer blanket and a heating lamp.

A heparin-filled catheter was inserted into the right carotid vein to administer fluid and drugs. A standard peripheral lead electrocardiogram was monitored continuously. A medial sternotomy was performed and the pericardium opened, exposing the heart. Systemic anticoagulation was achieved with sodium heparin (750 U/kg) IV, followed by continuous infusion of sodium heparin (250 U/kg/h). The left anterior descending coronary artery (LAD) distal to the first diagonal branch was cannulated with a stainless-steel cannula and perfused with blood from the left carotid artery through an extracorporeal circuit. Coronary perfusion pressure (CPP) was measured from the sidearm of the circuit, using pressure transducer-tipped catheter (PC500; Millar Instruments), and coronary blood flow (CBF) of the perfused area of LAD was measured with an ultrasonic flow probe (ADP17; Crystal Biotech, Hopkinton, MA) attached at the circuit. The circuit also contained a distal infusion port for drug administration. A 22-gauge catheter was inserted into epicardial vein at the same level

as was LAD to allow coronary venous blood sampling. The venous cannula was allowed to drain freely into a beaker to prevent venous stagnation. This venous blood was returned intermittently to the pig to maintain isovolemic conditions. A pressure transducer-tipped catheter (PC500; Millar Instruments) was inserted into left ventricular (LV) chamber through an incision in the apex for continuous recording of LV pressure (LVP). The peak rate of increase in LVP ($LVdP/dt_{max}$) was determined by electric differentiation of the LV pressure waveform. A pair of ultrasonic segment length transducers was implanted in the subendocardium of the perfused area of LAD to measure changes in regional contractile function (percentage segment shortening [%SS]). Segment length was monitored by ultrasonic amplifiers (VF-1; Crystal Biotech). End-systolic segment length (ESL) was determined 10 ms before maximum negative LVdP/dt, and end-diastolic segment length (EDL) was determined 10 ms before dP/dt first exceeded 140 mmHg•s^{-1} (immediately before the onset of LV isovolemic contraction). %SS was calculated using the formula; %SS = (EDL – ESL) × 100 × EDL^{-1}. All hemodynamic data were continuously monitored on a polygraph and digitized via a computer interfaced with an analog-to-digital converter (HEM; Physio-Tech, Tokyo, Japan).

Assay of plasma NE concentrations

Assay of plasma NE concentrations in coronary venous blood samples of the perfused area of LAD was undertaken at baseline and immediately after reperfusion, and determined by high-performance liquid chromatography with coulometric electrochemical detection.

Measurement of regional myocardial blood flow (RMBF)

Colored microspheres (Dye-trak; Triton Technology Inc., San Diego, CA) used to measure RMBF were injected into the extracorporeal circuit at baseline and before ischemia. Immediately before injection, the microspheres were suspended using Voltecs® (Scientific Industries, Bohemia, NY). Microspheres (15 μm in diameter) were injected into the extracorporeal circuit as a bolus during a 30-s period. Two milliliters of methylene blue was injected into the circuit, and the dyed area was resected as the perfused area after cardiac arrest with KCl injection. After the weight of the resected tissue was measured, the tissue was isolated and divided into four transmural sections, which were subsequently subdivided into inner, middle, and outer layers. After being weighed, each piece and the remaining myocardial tissue of the dyed region were separately dissolved by 4 mol•l^{-1} KOH, and colored microspheres

were collected by vacuum filtering. These microspheres were dissolved by dimethylformamide, and the photometric absorption of each dye solution was determined by an UV-visible recording spectrophotometer (UV-160A; Shimazu Co., Kyoto, Japan).

The composite spectrum of each dye solution was resolved into the spectra of the single constituent by a matrix-inversion technique incorporated into the spectrophotometer. By using calculated photometric absorption, RMBF was determined by using the equation; $Q_m = (A_m \times Q_t) / A_t$, where Q_m is blood flow of samples (mL/min/g), Q_t is total CBF (mL/min), A_m is photometric absorption in sample, 1 g, and A_t is total photometric absorption of the perfused area of LAD.

Experimental protocols

Figure 1 shows the experimental design. Thirty minutes after the instrumentation was completed, baseline systemic and coronary hemodynamics were recorded. Pigs were randomly assigned to one of four groups. If the pig was excluded before completion of the experiment, the next one was assigned to the same group. Each group received intracoronary infusion of DEX at a rate of 1 (group LD, n=9) or 10 (group MD, n=9) or 100 (group HD, n=9) ng/mL of CBF or drug vehicle (group C,

n=12) for 30 min before ischemia. Coronary blood concentration (ng/mL) of DEX was calculated by dividing the intracoronary infusion rate (ng/min) by the prevailing CBF rate (mL/min). The concentrations of low-dose DEX corresponded to clinical plasma concentrations for sedation in the intensive care (13). DEX was diluted with saline, and infused at a rate of 1.0-2.0 mL/min. All pigs were subjected to 12-min ischemic period followed by a 90-min reperfusion. The ischemia was produced by the complete occlusion of the extracorporeal circuit. The hemodynamics and the contractile function were monitored continuously throughout the experiment. In large mammals, dogs, and pigs, myocardial stunning can be induced by a single completely reversible episode of regional ischemia lasting less than 20 min. We carried out preliminary study to determine the ischemic period. We aimed at about 50% recovery from baseline after 90-min reperfusion.

If more than five premature ventricular contractions (PVC) per minute or multifocal PVC occurred, lidocaine (1mg/kg IV) was administered. This was repeated if necessary. VT and VF were treated with lidocaine (1 mg/kg IV) followed by direct defibrillation of the heart with 40 joules. If the animal did not recover from VT or VF after direct defibrillation once, the case was considered intractable and excluded from the study. Exsanguination was carried out by opening extracorporeal circuit under

anesthesia. The effect on reperfusion-induced arrhythmias was evaluated with the incidence of VT or VF and the total amount of lidocaine used within 10 min after reperfusion.

Statistical analysis

All data are expressed as mean ± SEM. One-way analysis of variance (ANOVA) for non-repeated measures followed by the Bonferroni's post hoc test was used to test for differences in baseline hemodynamics and %SS. Data within groups were analyzed with one-way ANOVA for repeated measures, and data between groups were analyzed with two-way repeated measures ANOVA followed by the Bonferroni's post hoc test. Incidences of VT or VF were compared using Fischer's exact test. P values <0.05 were considered statistically significant. Statistical analysis was performed using SPSS 15.0 software (SPSS Japan, Tokyo, Japan)

3. Results

There were no significant differences in demographic data among groups. Arterial blood gas values and blood glucose were maintained within physiologic range in all pigs.

Reperfusion-induced arrhythmias

Assessments of reperfusion-induced arrhythmias are summarized in Table 1. Eight pigs (67%) in group C, two pigs (22%) in group LD, and one pig (11%) in groups MD and HD exhibited VT or VF, and were treated with direct defibrillation. Four in group C, and one in groups LD and MD did not recover from VT or VF after direct defibrillation once; these cases were considered intractable and excluded from the study. The incidences of reperfusion-induced VT or VF in groups MD and HD were significantly lower than that in group C. Moreover, the DEX groups required less lidocaine than group C.

Hemodynamics

Changes of hemodynamics and %SS are summarized in Table 2. There were no significant differences in the systemic and coronary hemodynamics or %SS between

pre- and post-treatment of DEX (1 ng/mL or 10 ng/mL or 100 ng/mL). LVdP/dtmax were reduced significantly during ischemia and remained depressed throughout reperfusion in all groups. LVEDP increased significantly during ischemia and early reperfusion but returned to baseline values at 90 min after reperfusion in all groups. CBF significantly increased 5 min after reperfusion compared with the baseline values in all groups.

Dex improves myocardial stunning

Figure 2 shows the percentage changes of %SS from baseline (100 %) throughout the time course. In all groups, %SS markedly decreased and fell below 0% during ischemia, indicating bulging. The group C showed poor recovery of %SS during reperfusion, which was no more than 35% of baseline even after 90 min. All-dose DEX significantly enhanced the functional recovery from myocardial stunning compared with controls. Moreover, high-dose DEX significantly improved the functional recovery (72% of baseline at 90 min after reperfusion) compared with medium-dose (61% of baseline) and low-dose (58% of baseline) DEX.

Dex reduces plasma NE concentrations

Figure 3 presents the plasma NE concentrations in coronary venous blood sampling of the perfused area of LAD. In the group C, the NE concentrations significantly increased after reperfusion compared with the baseline. DEX significantly suppressed the increases of the NE concentrations after reperfusion.

Dex and RMBF

Figure 4 presents the subendocardial to subepicardial (endo/epi) blood flow ratio. In the group C, LD and MD, this ratio did not significantly change after the administration of the drug compared with the baseline. In contrast, high-dose DEX significantly increased this ratio compared with the baseline.

Discussion

The present results show that intracoronary infusion of DEX significantly improves regional myocardial contractility after 12-min ischemia and reperfusion of the perfused area of LAD in a dose-dependent manner, and suppresses reperfusion-induced ventricular arrhythmias in anesthetized pigs. These results suggest that DEX has direct protective effects against myocardial ischemia-reperfusion injury. In addition, DEX suppresses an increase in the plasma NE concentrations after reperfusion and increases the endo/epi blood flow ratio.

Myocardial reperfusion injury has four basic forms; lethal myocyte injury, vascular injury, stunned myocardium and reperfusion arrhythmias (1). Stunned myocardium and reperfusion arrhythmias are produced by brief periods (< 20 min) of coronary artery occlusion followed by reperfusion and are reversible injury in experimental studies (3, 14). Lombardi et al. (15) showed that a brief period of LAD occlusion followed by reperfusion caused sympathetic activation and a decrease in the VF threshold in dogs. Shindo et al. (16, 17) reported that both a short period (10 min) and a prolonged period (40 min) of coronary artery occlusion increased myocardial interstitial NE level in the ischemic region in cats. Chen et al. (18) showed that postischemic heart had a large amount of coronary NE overflow in an isolated working heart preparation. It is

considered that mechanism of stunned myocardium and reperfusion arrhythmias is associated with the increase of extracellular NE concentration within the ischemic myocardium irrespective of central sympathetic activity.

Several studies reported that intravenous administration of clonidine, an α_2-adrenergic agonist, and DEX could prevent a myocardial ischemia-induced NE release in anesthetized dogs (9, 19, 20). Mivazerol, α_2-adrenergic agonist, also suppressed the increase of plasma NE concentration and preserved myocardial blood flow in the inner layers during coronary artery stenosis in anesthetized dogs (10). Meissner et al. (9) showed that clonidine improved recovery from myocardial stunning and attenuated increases in catecholamine plasma levels in dogs. Thus it is considered that α_2-adrenergic agonists exert myocardial protection resulting from the attenuation of the catecholamine response to ischemic stress and redistributing myocardial blood flow. However, these protective effects might depend on central nervous actions by systemic administration of α_2-adrenergic agonists. In the present study, intracoronary DEX administration suppressed an increase in NE release in the coronary venous blood sampling of the ischemic region, and exerted protective effects against reperfusion injury. Cai et al. (21) reported that α_2-adrenergic receptors were present in the heart in vitro, and α_2-adrenergic stimulation prevented reperfusion-induced VT/VF in vivo and

that this effect is not centrally mediated. The release of NE from the isolated human papillary muscle was inhibited by xylazine, an α_2-adrenergic agonist (10). Thus, the cardioprotective effects of intracoronary DEX administration should be mediated through cardiac presynaptic α_2-adrenoreceptor stimulation.

The direct effects of α_2-adrenergic agonists on coronary vasculature are controversial. Heusch et al. (22) showed that activation of coronary vascular α_2-adrenergic receptors induced poststenotic coronary vasoconstriction and myocardial ischemia. However, previous studies indicated that α_2-adrenergic coronary vasoconstriction exerts a favorable effect on ischemic myocardium preventing a transmural redistribution of blood flow away from the endocardium and improving endo/epi blood flow ratio (23, 24). Moreover, Kitakaze et al. (25) demonstrated that intracoronary α_2-adrenergic stimulation increased CBF during ischemia by enhancing the vasodilative effects of adenosine released from the ischemic myocardium. In non-ischemic heart, systemic administration of DEX induced a pronounced decrease in CBF in dogs (26). However, Meissner et al. (9) reported that clonidine increased endo/epi blood flow ratio in normal condition. In the present study, CBF was not changed after intracoronary DEX administration, but in the group HD endo/epi blood flow ratio was significantly increased in normal condition. Lawrence et al. (27) also

reported that DEX at 10 μg/kg but not 0.1 or 1 μg/kg increased endo/epi blood flow ratio without changing CBF in halothane- and fentanyl-anesthetized dogs. This effect of high-dose DEX may cause more improvement of the functional recovery from myocardial stunning.

The application of cardioprotective therapy after reperfusion is clinically feasible because the onset of reperfusion is predictable and is under the clinician's control. The present results show that intracoronary infusion of DEX before ischemia enhanced the functional recovery of stunned myocardium and suppressed reperfusion-induced ventricular arrhythmias. Okada et al. (28) reported that administration of DEX during the pre-ischemic period reduces myocardial infarct size in isolated rat heart. On the other hand, Guo et al. (29) reported that administration of DEX pre-hypoxia, but not post-hypoxia, exerts a direct protective effect on the left ventricular dysfunction caused by hypoxia-reoxygenation in isolated rat heart. Mimuro et al. (30) also demonstrated that administration of DEX after reperfusion did not influence hemodynamics or CBF, but increased myocardial infarct size in isolated rat heart. The mechanisms of this discrepancy are not known; however, administration of DEX after reperfusion might not be beneficial or deleterious.

The doses of DEX using in this study were set on the basis of a previous study

using 0.7–14.7 ng/ml DEX in volunteers (31), and the concentrations of DEX in groups

LD and MD corresponded to 10 and 100 times clinical plasma concentrations for

sedation in intensive care (13). It is possible that a factor other than α_2-adrenergic

agonist activity affected the present results in group HD. The present results show that

high-dose DEX significantly increased endo/epi blood flow ratio and caused greater

improvement in terms of functional recovery. In addition, we suggest that the greater

improvement in terms of functional recovery is mediated by an increase in endo/epi

blood flow ratio. These results are consistent with previous studies using other

α_2-adrenergic agonists, such as mivazerol or clonidine (8, 9). It is likely that the

effects of a factor other than α_2-adrenergic agonist activity are only small, even with the

use of high-dose DEX in this study. The esophageal temperature (36-37°C)

maintained in this study was slightly lower than the normal temperature. Hypothermia

would affect myocardial injury (32). However, mild hypothermia is thought to occur

below 35°C, and there were no significant differences in terms of esophageal

temperature among the groups in this study. The body temperature should not have

affected our results.

In the present study, while the LAD was perfused with blood containing DEX,

venous effluent containing DEX (that portion not collected via the implanted coronary

venous catheter) returned directly to the systemic circulation. However, this venous return is the relatively small size compared to the total systemic venous return. In addition, systemic hemodynamic parameters did not vary during the intracoronary infusion of DEX. Therefore, although measurements of aortic blood concentrations were not available to confirm, we assumed that its concentrations in the systemic arterial circulation remained low and did not exert central nervous actions.

In conclusion, intracoronary DEX enhanced the functional recovery of stunned myocardium and suppressed reperfusion-induced ventricular arrhythmias. These effects are possible due to preventing an increase of myocardial NE level in ischemic region through cardiac presynaptic α_2-adrenoreceptor stimulation, but this is not centrally mediated.

List of abbreviations

VT: ventricular tachycardia; VF: ventricular fibrillation; NE: norepinephrine; DEX: dexmedetomidine; IV: intravenous; $PaCO_2$:arterial carbon dioxide tension; PaO_2: arterial oxygen tension; LAD: left anterior descending coronary artery; CPP: coronary perfusion pressure; CBF: coronary blood flow; LV: left ventricular; LVP: left ventricular pressure; $LVdP/dt_{max}$: peak rate of increase in left ventricular pressure; %SS: percentage segment shortening; ESL: end-systolic segment length; EDL: end-diastolic segment length; RMBF: regional myocardial blood flow; PVC: premature ventricular contractions; endo/epi: subendocardial to subepicardial

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Figure legends

Figure 1: Time course of the experimental protocol.

All pigs were subjected to 12-min ischemia of the perfused area of left anterior descending coronary artery (LAD) and subsequent 90-min reperfusion. Drug vehicle and dexmedetomidine (DEX) were administered by an intracoronary infusion for 30 min before ischemia. During experiment, hemodynamics, percentage of segment shortening (%SS), plasma norepinephrine concentration in coronary venous blood sampling, and regional myocardial blood flow (RMBF) were measured at times indicated by the closed circles. The effect on reperfusion-induced arrhythmias was evaluated at 10-min after reperfusion.

Figure 2: Percentage segment shortening (%SS) of the ischemic-reperfused area (% of baseline).

Values are mean ± SEM. and represent all surviving pigs. Group C (n=8): drug vehicle, group LD (n=8): low-dose dexmedetomidine (1 ng/mL), group MD (n=8): medium-dose dexmedetomidine (10 ng/mL), group HD (n=9): high-dose dexmedetomidine (100 ng/mL). †: P < 0.05 vs. group C. #: P < 0.05 vs. group LD. ƒ: P < 0.05 vs. group MD.

Figure 3: Plasma norepinephrine concentrations in coronary venous blood sampling of the ischemic-reperfused area.

Values are mean ± SEM. and represent all surviving pigs.   Group C (n=8): drug vehicle, group LD (n=8): low-dose dexmedetomidine (1 ng/mL), group MD (n=8): medium-dose dexmedetomidine (10 ng/mL), group HD (n=9): high-dose dexmedetomidine (100 ng/mL).   *: P < 0.05 vs. baseline.   †: P < 0.05 vs. group C.

Figure 4: Subendocardial to subepicardial (endo/epi) blood flow ratio in the ischemic-reperfused area.

Values are mean ± SEM. and represent all surviving pigs.   Group C (n=8): drug vehicle, group LD (n=8): low-dose dexmedetomidine (1 ng/mL), group MD (n=8): medium-dose dexmedetomidine (10 ng/mL), group HD (n=9): high-dose dexmedetomidine (100 ng/mL).   *: P < 0.05 vs. baseline.
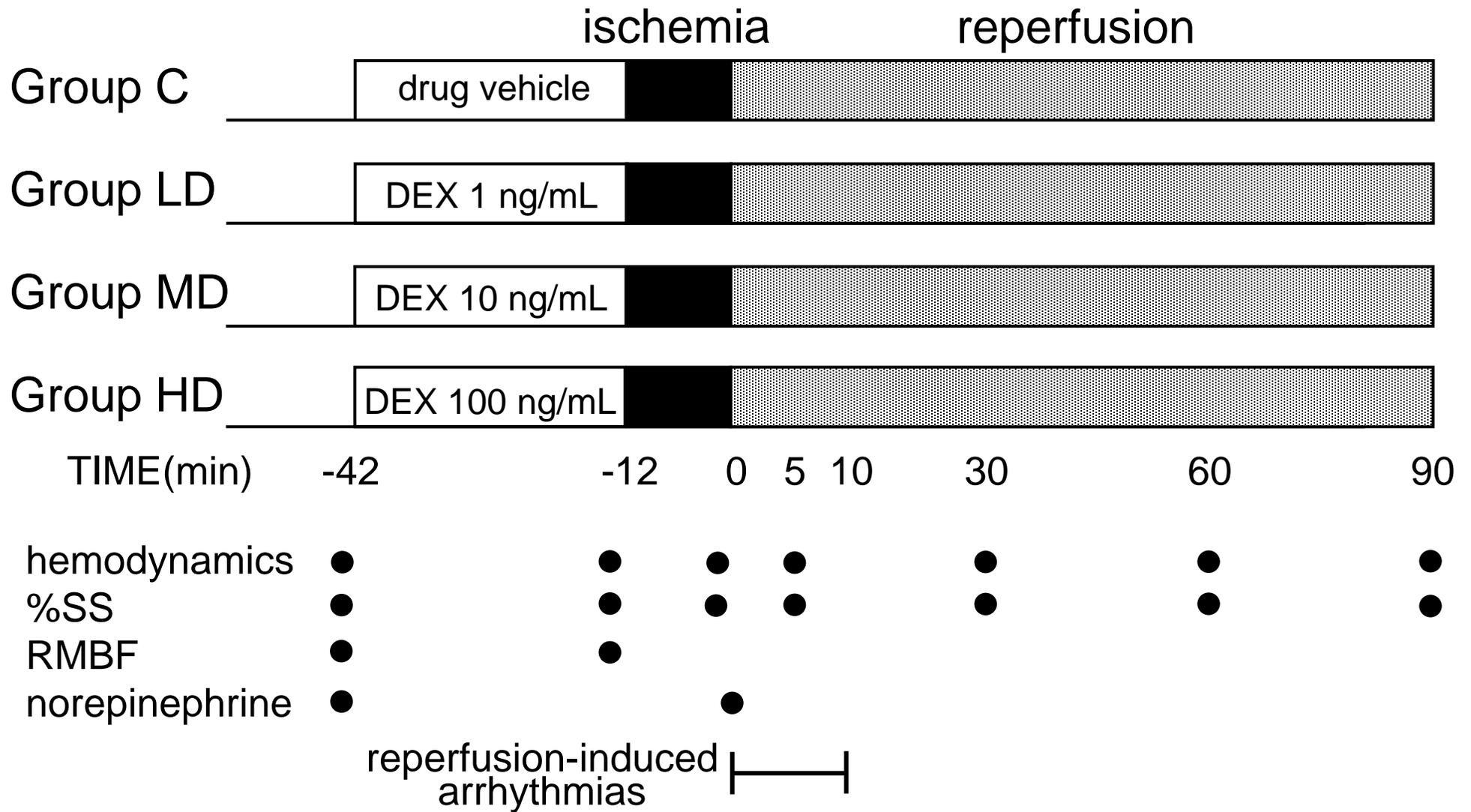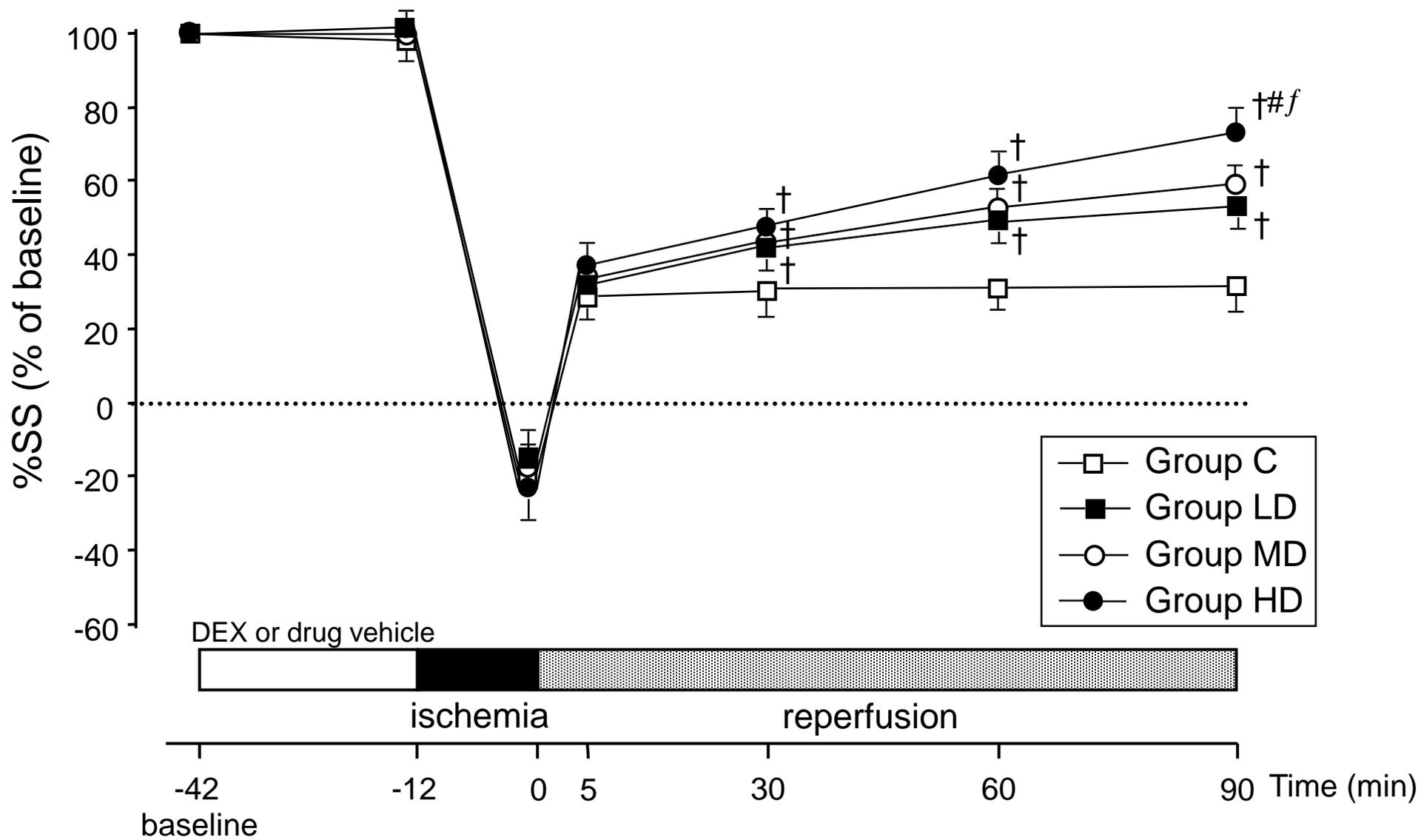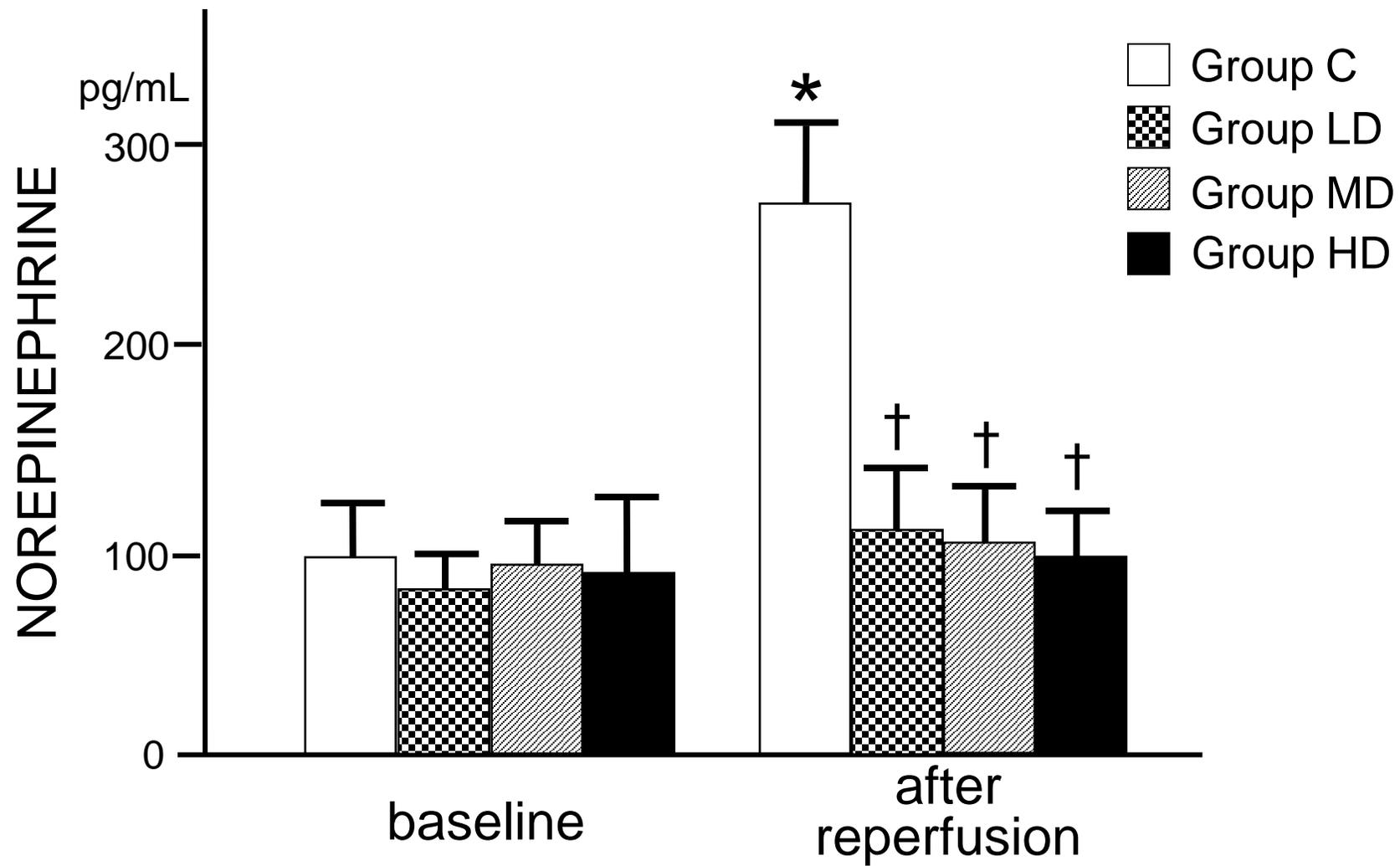
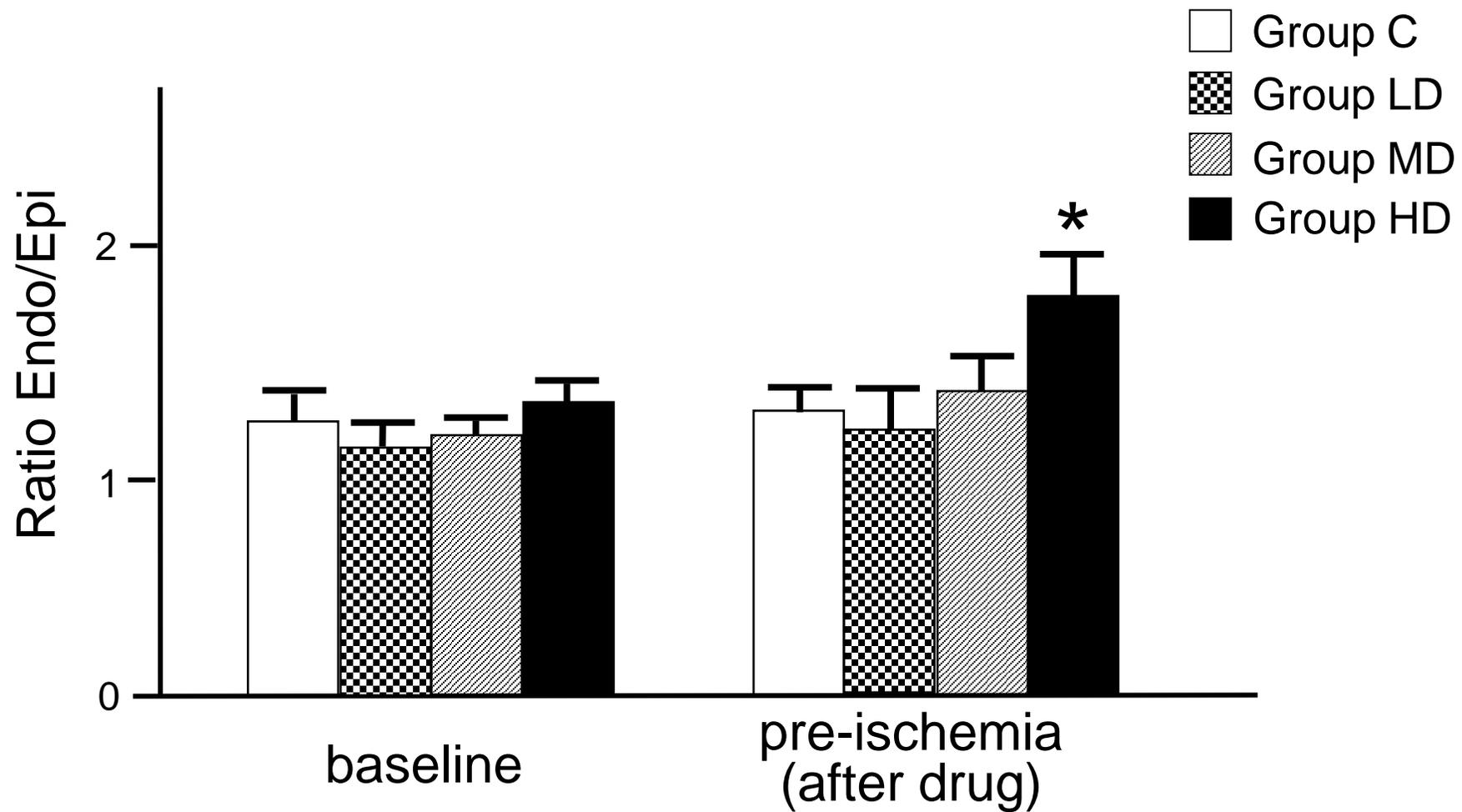Figure 1

Figure 2

Figure 3

Figure 4

Table 1: Assessments of reperfusion induced arrhythmias

	Incidence of VT/VF (%)	Exclusions (Intractable VT/VF)	Final number	Lidocaine (mg/kg)
Group C (n=12)	67	4	8	3.75 ± 0.41
Group LD (n=9)	22	1	8	2.63 ± 0.63 †
Group MD (n=9)	11†	1	8	2.13 ± 0.13 †
Group HD (n=9)	11†	0	9	1.56 ± 0.41 †

Values of lidocaine are mean ± SEM. and represent all surviving pigs. VT = ventricular tachycardia; VF = ventricular fibrillation; Group C = drug vehicle; Group LD = low-dose dexmedetomidine (1 ng/mL); Group MD = medium-dose dexmedetomidine (10 ng/mL); Group HD = high-dose dexmedetomidine (100 ng/mL).

†: Significantly different from the corresponding value in Group C ($p < 0.05$)

Table 2: Hemodynamic Data

	Baseline (Pre-treatment)	Pre-ischemia (Post-treatment)	Ischemia	Reperfusion 5 min	30 min	60 min	90 min
HR (beats/min)							
Group C (n=8)	89±4	89±4	88±5	94±5	95±6	92±7	91±5
Group LD (n=8)	82±6	81±6	83±5	83±7	84±6	81±7	80±7
Group MD (n=8)	87±5	85±4	81±4	84±4	86±4	87±4	88±3
Group HD (n=9)	89±6	86±7	83±6	84±7	86±7	88±7	89±7
LVPP (mmHg)							
Group C (n=8)	132±7	130±7	127±8	123±9	125±10	127±10	129±10
Group LD (n=8)	129±8	129±7	124±7	125±9	126±8	129±8	134±8
Group MD (n=8)	136±4	135±2	134±4	135±4	134±5	134±6	133±5
Group HD (n=9)	140±5	140±4	134±5	133±4	138±4	138±4	140±4
LVEDP$_{max}$ (mmHg)							
Group C (n=8)	8.5±1.0	8.7±0.9	10.4±0.8*	9.8±1.1*	9.0±0.9	8.1±0.7	7.8±0.7
Group LD (n=8)	9.1±1.0	9.3±1.1	10.6±1.1*	11.6±1.2*	9.3±1.0	8.9±0.9	8.8±0.8
Group MD (n=8)	7.9±0.8	7.9±1.0	9.5±1.0*	9.2±1.1*	8.5±1.0	7.9±0.9	7.9±0.8
Group HD (n=9)	8.6±0.8	8.7±0.7	10.2±1.0*	9.9±0.8*	8.9±0.5	8.5±0.8	8.2±0.7
LVdP/dt$_{max}$ (mmHg/s)							
Group C (n=8)	2908±161	2938±154	2360±151*	2288±184*	2414±182*	2431±174*	2520±186*
Group LD (n=8)	2664±161	2572±168	2266±141*	2014±127*	2157±167*	2284±148*	2429±145*
Group MD (n=8)	2915±145	2902±162	2416±148*	2410±137*	2612±173*	2781±194	2800±141
Group HD (n=9)	3080±172	3054±169	2458±132*	2460±134*	2565±131*	2807±153	2965±165
mCPP (mmHg)							
Group C (n=8)	121±7	121±6	117±8	114±9	116±9	116±9	119±9
Group LD (n=8)	112±7	109±6	109±6	108±5	112±6	112±6	112±6
Group MD (n=8)	124±4	122±4	122±4	121±3	122±4	122±5	121±5
Group HD (n=9)	126±5	127±4	123±4	119±4	123±3	124±4	125±4
CBF (ml/min)							
Group C (n=8)	22.2±3.2	22.4±3.4		35.4±4.0*	21.5±2.7	21.6±3.0	21.0±2.6
Group LD (n=8)	24.8±1.8	24.6±2.1		39.1±3.6*	24.7±2.2	25.2±2.6	24.5±2.4
Group MD (n=8)	21.4±2.8	21.7±3.2		38.7±4.5*	22.3±3.5	21.4±3.0	21.2±2.9
Group HD (n=9)	20.3±2.0	20.1±2.3		32.9±2.8*	18.9±2.1	19.4±1.9	19.9±2.0
%SS							
Group C (n=8)	22.0±2.2	21.9±2.2	-3.7±1.2*	6.5±1.1*	6.4±0.9*	6.9±0.8*	7.2±1.1*
Group LD (n=8)	26.1±2.4	25.9±2.1	-2.7±1.0*	9.2±0.8*	11.7±0.9*†	13.4±1.1*†	15.1±1.2*†
Group MD (n=8)	24.1±2.4	24.2±2.4	-3.8±0.8*	8.0±1.0*	11.1±1.0*†	13.0±1.0*†	14.6±1.2*†
Group HD (n=9)	25.7±2.4	26.4±2.1	-4.3±0.7*	8.4±0.9*	12.5±1.5*†	15.1±1.7*†	18.4±1.6*†

Values are mean ± SEM. and represent all surviving pigs. HR = heart rate; LVPP = left ventricular peak pressure; LVEDP = left ventricular end-diastolic pressure; LVdp/dt = rate of increase of left ventricular pressure; mCPP = mean coronary perfusion pressure; CBF = coronary blood flow; %SS = percentage of segment shortening; Group C = drug vehicle; Group LD = low-dose dexmedetomidine (1 ng/mL); Group MD = medium-dose dexmedetomidine (10 ng/mL); Group HD = high-dose dexmedetomidine (100 ng/mL).

*: Significantly different from baseline ($p < 0.05$). †: significantly different from the corresponding value in group C ($p < 0.05$).