Levosimendan as a Rescue Drug in Experimental Propranolol-Induced Myocardial Depression: A Randomized Study

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Study objective: Severe β-blocker intoxication remains a clinical challenge despite a variety of treatment options. Because of its unique mechanism of action, the new calcium sensitizer levosimendan may provide more prominent cardiac support compared with current medications used to reverse negative inotropy. We hypothesize that levosimendan could reverse propranolol-induced severe negative inotropy in a porcine model of β-blocker intoxication.

Methods: Twenty-four pigs were anesthetized and monitored. After severe propranolol intoxication was completed, animals were randomized into 3 groups. With a double-blind procedure, 9 animals received a 1.25-mg levosimendan bolus, followed by saline solution infusion, 9 animals received mean arterial pressure–targeted dobutamine infusion after saline solution bolus, and 6 animals received a saline solution bolus followed by saline solution infusion. Hemodynamic and laboratory data were collected during a follow-up period of 120 minutes.

Results: All 9 pigs in the levosimendan group survived. In contrast, 4 of 6 (67%) and 7 of 9 (78%) pigs died during the experiment in the placebo and the dobutamine groups, respectively. The levosimendan group showed improved change in the maximum positive slope of the left ventricular pressure, cardiac output, stroke volume, and mean arterial pressure compared with the dobutamine and the placebo groups.

Conclusion: Levosimendan improved hemodynamic function and survival in this animal model of severe propranolol intoxication. The potential clinical application of levosimendan in propranolol intoxication warrants further investigation. [Ann Emerg Med. 2009;54:811-817.]

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0196-0644/$-see front matter

doi:10.1016/j.annemergmed.2009.06.512

INTRODUCTION

Background

More than 9,000 toxic exposures to β-blockers were reported to US poison centers in 2007. Of those exposures, 692 (7.4%) were associated with severe bradycardia, hypotension, and acute negative inotropy; 3 deaths were associated with β-blocker overdose. The majority of the exposures were unintentional (86.2%), but a significant (10.8%) proportion of cases were suicide attempts.[1]

The flow of calcium across myocardial cell membranes is necessary for cardiac automaticity, pulse conduction, and contraction. β-blockers indirectly decrease cyclic adenosine monophosphate production, with a subsequent decrease of calcium influx through L-type calcium channels. Interruption of calcium flux leads to decreased intracellular calcium ion concentration; this produces cardiovascular dysfunction because of negative inotropy, which can result in cardiovascular collapse.[2]

Importance

The therapeutic goal in the treatment of β-blocker intoxication is to support myocardial function and thereby restore critical organ perfusion. Various treatments have been attempted, including volume expansion, atropine, cardiac pacing, insulin, dopamine, dobutamine, glucagon, enoximone, hemodialysis, continuous venovenous hemodiafiltration, and gastrointestinal decontamination with activated charcoal.[3-6]

The calcium sensitizer levosimendan is a targeted therapy for the treatment of acute decompensated heart failure. It is available in more than 40 countries but not yet in the United

Editor's Capsule Summary

What is already known on this topic

Severe β-blocker poisoning is treated with multiple pharmacologic agents, with limited success. Levosimendan increases the sensitivity of cardiac myocyte contractile proteins to calcium.

What question this study addressed

The hemodynamic effects of levosimendan were tested in a porcine animal model of severe β-blocker intoxication.

What this study adds to our knowledge

Compared with dobutamine and saline solution treatments, the levosimendan group showed improved survival, cardiac contractility, cardiac output, and mean arterial pressure. All 9 pigs in the levosimendan group survived, whereas 4 of 6 and 7 of 9 pigs died in the placebo and the dobutamine groups, respectively.

How this might change clinical practice

It will not yet, but levosimendan offers a promising new approach for the treatment of severe β-blocker poisoning.

States. By increasing calcium sensitivity of contractile proteins, levosimendan induces a positive inotropic effect mediated through calcium-dependent binding to troponin C.[7,8] Levosimendan has a direct contractility-enhancing effect [9] and also improves function of the stunned myocardium.[10] Levosimendan causes coronary dilation and systemic vasodilatation through opening of adenosine triphosphatase–sensitive potassium channels.[11,12] It seems that levosimendan works under extreme systemic conditions, such as acidosis and sepsis.[13,14]

Goals of This Investigation

Our study assessed the effect of levosimendan on hemodynamic performance in a porcine model of propranolol-induced negative inotropy.

MATERIALS AND METHODS

Study Design

The National Animal Ethics Committee of Finland approved this methodology. Animal care, welfare, and procedures were carried out in accordance with the regulations of the Council of Europe.

Twenty-four landrace pigs (median 29 kg; interquartile range (28, 30)) were fasted, but allowed access to water, for 12 hours before the experiments. Premedication with medetomidine 50 μg/kg, midazolam 0.10 mg/kg, ketamine 10 mg/kg, and

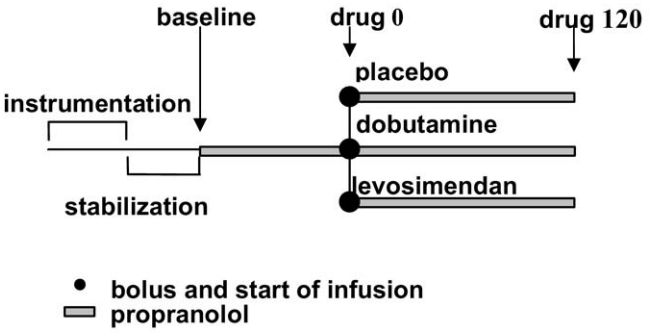

Figure 1. Experimental protocol.

fentanyl 5 μg/kg intramuscularly was followed by cannulation of an ear vein and intravenous administration of propofol 2 mg/kg for intubation. Anesthesia was maintained with propofol (10 mg/kg per hour) and fentanyl (3 μg/kg per hour). The animals were ventilated in volume-controlled mode (Servo 900; Siemens, Elema AB, Solna, Sweden) with 5 cm H_2O of positive end-expiratory pressure. The fraction of inspired oxygen was maintained between 0.3 and 0.6 to keep PaO_2 levels between 13.3 kPa (100 mm Hg) and 20 kPa (150 mm Hg). Tidal volume was kept at 10 mL/kg, and the minute ventilation was adjusted to maintain $PaCO_2$ levels between 4.5 and 5.5 kPa (34 to 41 mm Hg).

Interventions

A right femoral artery was cannulated and a pulmonary artery catheter (7.5F flow-directed, Arrow; Arrow International Inc., Reading, PA) introduced through the right internal jugular vein. The angiography catheter was inserted into the left ventricle through the left femoral artery to measure left ventricular pressures. A femoral vein was cannulated for fluid administration. During instrumentation, the animals received 3 mL/kg per hour infusions of 0.9% saline solution and 1.5 mL/kg per hour gelatin (Gelofusine; B. Braun Medical, Melsungen, Germany). Additional saline solution was administered if necessary to keep pulmonary artery occlusion pressure between 5 and 8 mm Hg. Body temperature of the animals was kept above 38°C (100°F) using an operating table heater and warmed fluids.

After instrumentation, a stabilization period of at least 30 minutes was followed by baseline measurements (Figure 1). Propranolol intoxication was then induced. The target was 40% of baseline cardiac output for 15 minutes. The intoxication model was initiated with 1 mg/kg of propranolol intravenously, followed by an intravenous infusion containing 5 mg/mL of propranolol at 180 mL/hour until target cardiac output was reached. Thereafter, the rate of propranolol infusion was reduced to 90 mL/hour and maintained until the end of the experiment. This method was chosen to mimic oral intake and was based on a pilot trial of 3 pigs.

After target cardiac output was reached, the pigs were randomized into 3 groups: 6 pigs in the placebo group, 9 pigs in

the dobutamine group, and 9 pigs in the levosimendan group. Randomization (sealed opaque envelopes) was performed by a research nurse at the Department of Intensive Care at Kuopio University Hospital. The research nurse also prepared and diluted the study drugs, thereby ensuring that the investigators and research assistants were blinded to the group assignment. All 3 groups received a 10 mL fluid bolus within 2 minutes, followed by an infusion of 0 to 15 mL/hour, which was adjusted to maintain mean arterial pressure above 65 mm Hg.

The placebo group received a 10 mL fluid bolus consisting of saline solution colored with thiamine, followed by the same saline solution infusion. The dobutamine group received a 10 mL bolus of saline solution colored with thiamine and a 2.5 mg/mL dobutamine infusion. The levosimendan group received a bolus with 1.25 mg of levosimendan in 10 mL of 5% glucose (Simdax; Orion Pharma, Espoo, Finland) and a saline solution infusion. All the fluids were similar in appearance.

Methods of Measurement

Left ventricular pressure, mean arterial pressure, central venous pressure, and pulmonary artery occlusion pressure were recorded with quartz pressure transducers and displayed continuously on a multimodular monitor (S/5 Compact Critical Care Monitor; Datex-Ohmeda, Helsinki, Finland). All pressure transducers were calibrated simultaneously and zeroed to the level of the heart. The inotropic effect was measured as a change in the maximum of the positive slope of the left ventricular pressure. It was recorded once a minute, and the mean value during 5 minutes was calculated to be used in the analysis. Cardiac output was measured by bolus injectates in triplicate with 10 mL of room-temperature 0.9% sodium chloride. The mean value of 3 measurements was used. Systemic vascular resistance was calculated from maintained mean arterial pressure, central venous pressure, and cardiac output. Pulse rate was measured from the ECG, which was also continuously monitored (Datex-Ohmeda). A median value of 2 minutes is presented in the "Results."

Blood samples were drawn at baseline, when the target cardiac output was reached (drug 0), and immediately before clinically estimated collapse of hemodynamics or at the end of the experiment (drug 120).

Primary Data Analysis

Sample size was estimated as 6 animals per group, with a 2-sided *P* level of .05 and a power of 0.80 to detect a 1 L/minute difference in cardiac output, with SD of 0.7 between levosimendan and placebo groups at 30 minutes after bolus of levosimendan or placebo. We chose a larger sample size for the treatment groups to detect possible differences between levosimendan and dobutamine groups. The difference in cardiac output and the SD were based on a previous study of calcium channel blockade intoxication in a pig model (unpublished data).

The outcome measures were normally distributed, as tested by the Kolmogorov-Smirnov test at points when there were more than 3 animals alive. When there were fewer than 3 animals alive, the distribution was not tested. The mixed models were used to examine the differences between the study groups from preintoxication (baseline) to postintoxication (from drug 0 to drug 120) phases. Time point and group were treated as fixed effects (factors) and subject (pig) as a random effect. Models were constructed separately for each parameter (response variable). Estimated marginal means were calculated from the model parameters, and post hoc comparisons were based on these estimated marginal means and were Bonferroni corrected to account for multiple testing. Analyses were not adjusted for baseline differences. Values are presented as medians and interquartile ranges. The statistics and *P* values are for the living animals. The effects of propranolol intoxication are presented as percentage change from the baseline to the drug 0 level. Statistical analyses were performed with SPSS for Windows, version 16.0 (SPSS Inc., Chicago, IL). *P* values less than .05 were considered statistically significant.

Table 1. Measures of hemodynamic performance.

	Baseline	Drug 0
MAP, mm Hg		
Levosimendan	110 (105; 113)	60 (49; 68)
Dobutamine	105 (105; 111)	63 (42; 66)
Placebo	107 (104; 111)	53 (49; 62)
CO, L		
Levosimendan	4.6 (4.3; 4.8)	2.3 (2.1; 2.5)
Dobutamine	4.6 (3.8; 4.6)	2.1 (1.8; 2.4)
Placebo	4.7 (4.6; 5.0)	2.3 (1.5; 2.4)
LV dP/dt, mm Hg/s		
Levosimendan	2,230 (2,008; 2,418)	621 (600; 834)
Dobutamine	2,058 (1,886; 2,140)	831 (493; 885)
Placebo	2,090 (1,868; 2,266)	693 (490; 760)
Pulse rate, beats/min		
Levosimendan	94 (86; 101)	93 (91; 96)
Dobutamine	97 (95; 103)	98 (89; 101)
Placebo	134.5 (128.5; 140.5)	100 (85.25; 108)
CVP, mm Hg		
Levosimendan	8 (7; 9)	10 (9; 10)
Dobutamine	7 (7; 9)	10 (10; 14)
Placebo	8 (8; 8)	11 (10; 11)
SV, mL		
Levosimendan	47 (42; 50)	24 (23; 28)
Dobutamine	40 (37; 46)	21 (18; 24)
Placebo	37 (34; 40)	22 (17; 23)
SVR, dyn·s/cm^5		
Levosimendan	1,849 (1,744; 1,888)	2,131 (1,967; 2,394)
Dobutamine	1,855 (1,710; 2,091)	2,091 (1,681; 2,674)
Placebo	1,668 (1,608; 1,776)	2,090 (1,962; 2,242)

MAP, Mean arterial pressure; *CO,* cardiac output; *LV dP/dt,* maximum of the positive slope of the left ventricular pressure; *CVP,* central venous pressure; *SV,* stroke volume; *SVR,* systemic vascular resistance at baseline and drug 0 (median and interquartile range).

RESULTS

There was no difference between the groups in baseline data, except in hemoglobin and pulse rate (Tables 1 and 2). Hemoglobin level was higher in the dobutamine group, at 74

Table 2. SvO_2, calcium, and lactate values at baseline, drug 0, and drug 120 or just before hemodynamic collapse.

	Median (Interquartile Range)		
	Baseline	**Drug 0**	**Drug 120**
SvO_2, %			
Levosimendan	69 (68; 72)	40 (32; 45)	56 (44; 63)*
Dobutamine	70 (67; 70)	39 (38; 41)	22 (19; 40)
Placebo	65 (61; 71)	30 (26; 39)	22 (19; 34)
Calcium, mmol/L			
Levosimendan	1.33 (1.29; 1.35)	1.25 (1.24; 1.34)	1.33 (1.26; 1.34)
Dobutamine	1.34 (1.26; 1.37)	1.30 (1.26; 1.30)	1.27 (1.24; 1.35)
Placebo	1.32 (1.32; 1.34)	1.29 (1.29; 1.32)	1.26 (1.17; 1.28)
Lactate, mmol/L			
Levosimendan	0.6 (0.5; 0.7)	1.20 (0.60; 1.20)	0.5 (0.5; 0.6)*
Dobutamine	0.6 (0.6; 0.8)	1.20 (1.10; 1.70)	2.0 (1.2; 2.5)
Placebo	0.6 (0.5; 0.8)	1.45 (0.8; 2.0)	1.9 (1.7; 2.1)

*$P<.05$ levosimendan versus dobutamine and levosimendan versus placebo. There were 9 animals in the levosimendan and the dobutamine groups and 6 in the placebo group.

g/L (71; 78), versus the placebo group, at 58 g/L (54; 61). Pulse rate was higher in the placebo group, at 135 beats/min (129; 141), versus the levosimendan group, at 94 beats/min (86; 101), and dobutamine group, at 97 beats/min (95; 103).

The hemodynamic parameters and laboratory data of ionized calcium, lactate, and mixed venous saturation (SvO_2) at the time point when intoxication was complete (drug 0) are presented in Tables 1 and 2. When intoxication with propranolol was complete, there were no differences between the groups except in hemoglobin between the dobutamine and placebo groups: 73 g/L (67; 76) versus 53 g/L (48; 55), respectively (Tables 1, 2). In each animal, cardiac output decreased at least by 40%: in the levosimendan group, the reduction was 50%; in the dobutamine group, 54%; and in the placebo group, 51%.

The group receiving levosimendan had higher cardiac output, maximum of the positive slope of the left ventricular pressure, stroke volume, mean arterial pressure, and pulse rate compared with the dobutamine and placebo groups at each time point (Figure 2; $P<.05$ for each time point). There was no difference in systemic vascular resistance between the groups throughout the study (P=not significant). The median dose of dobutamine was 6.9 μg/kg per minute (6.8; 19.5) during the experiment (Appendix E1, available online at http://www.annemergmed.com).

The final blood samples were taken immediately before the clinically estimated collapse of hemodynamics in each pig or at drug 120 minutes. In the blood samples, there was a decrease in SvO_2 in both dobutamine and placebo groups compared with the levosimendan group, in which SvO_2 increased from drug 0 to drug 120 (Table 2; $P=.003$ for levosimendan versus dobutamine; $P=.004$ for levosimendan versus placebo).The laboratory values of sodium, potassium, and glucose were within the normal range in all groups throughout the experiment (Appendix E1, available online at http://www.annemergmed.com).

All pigs in the levosimendan group survived the experiment. In contrast, 4 of 6 and 7 of 9 pigs died during the experiment in the placebo and the dobutamine groups, respectively (Figure 3).

LIMITATIONS

We used an animal model, which may not simulate oral propranolol intoxication in humans. This was a model of severe or near-fatal intoxication, and the hemodynamic changes in our animal model are similar to those seen with human propranolol intoxication.

Various rescue drugs and interventions are currently used to treat propranolol intoxication; in this model, we chose dobutamine as the control treatment, which may not be the method of choice by some physicians. Dobutamine is widely used in low cardiac output, and we chose to compare levosimendan to a clinically used inotropic drug. Comparing levosimendan in combination with other drugs or alone versus drugs or interventions other than dobutamine might be relevant.

DISCUSSION

The main finding in our study was that levosimendan provided a survival benefit compared with that of control treatments. All animals receiving levosimendan survived, whereas most animals died in the placebo and the dobutamine groups. Survival was associated with preserved myocardial function, as measured by maximum of the positive slope of the left ventricular pressure, cardiac output, and stroke volume. In contrast, we did not find evidence of excessive vasodilation in response to levosimendan. Therefore, the major beneficial effect of levosimendan may be due to its positive inotropic effect, mediated through calcium sensitization of contractile proteins.

Levosimendan enhances cardiac contraction by improving the use of available calcium, rather than by inundating the cell with excessive calcium.[15] This effect can maintain or reverse cardiac contractile function, even under severe β-receptor blockade. Similar findings were reported in guinea pigs.[16]

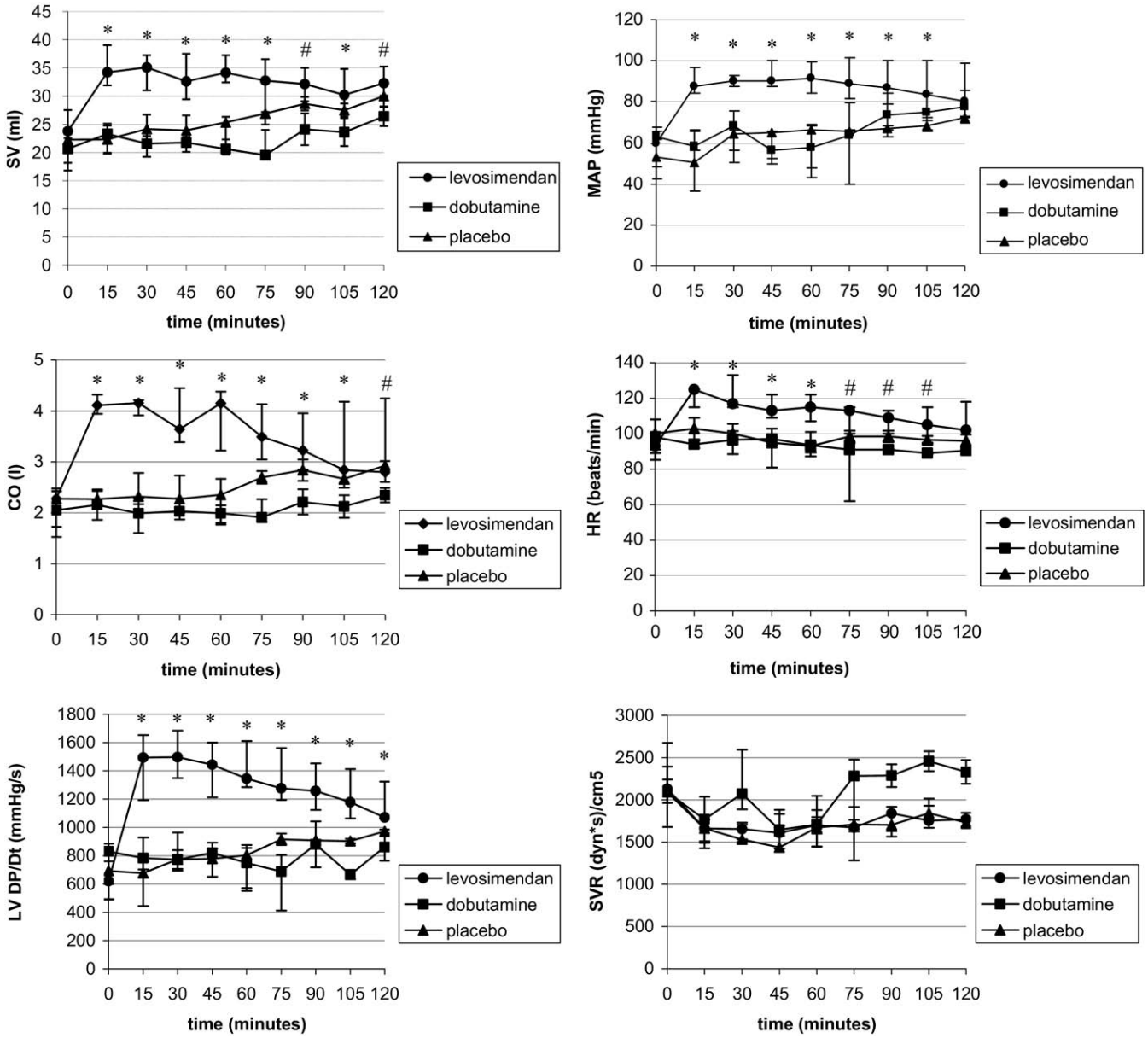

Figure 2. Hemodynamic changes in a porcine model of propranolol intoxication. Stroke volume (SV), cardiac output (CO), maximum of the positive slope of the left ventricular pressure (LV dP/dt), mean arterial pressure (MAP), pulse rate (HR), and systemic vascular resistance (SVR) between time 0 and time 120 in different groups (median and interquartile range; * $P<.05$ between levosimendan vs. placebo and dobutamine groups; # $P<.05$ between levosimendan and dobutamine groups). The statistics and *P* values were calculated for the animals that were alive at each time point. See "Materials and Methods" for details.

Levosimendan causes vasodilatation through opening of adenosine triphosphatase–sensitive K^+ channels,[19] which may contribute to coronary and systemic vasodilation with intravenous administration of levosimendan.[17,18] Even though the vasodilating effect of levosimendan has been well documented,[19-21] it is noteworthy that in this experiment it had a positive effect on mean arterial pressure. It is conceivable that the inotropic effect of levosimendan was more prominent than the vasodilatory effect.

Dobutamine is a direct-acting sympathomimetic amine that has its primary effect on β_1-receptors and relatively minor effects on β_2- and α_1-receptors. Our results indicate that nonselective blockade of β-receptors can cause dobutamine to be ineffective in reversing cardiac contractile function. This finding is supported also by previous study in humans.[22] Therefore, the effect of dobutamine on inotropy in the presence of total β-blockade (intoxication) is limited. Furthermore, levosimendan was significantly more effective than dobutamine in patients on a β-blocking agent for heart failure

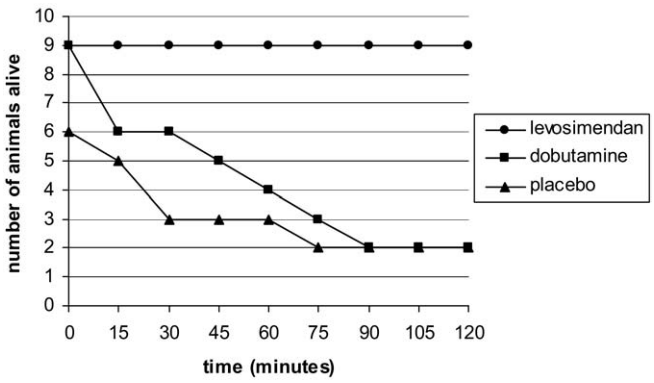

Figure 3. Animal survival between drug 0 and drug 120.

in the LIDO study.[23] Levosimendan may outcompete propranolol for β-receptor binding sites, but dobutamine cannot outcompete the propranolol.

In summary, treatment with levosimendan in this experiment improved hemodynamic measures and survival in a porcine model of propranolol intoxication. It may provide an alternative to presently recommended pharmacologic therapy in cases of severe propranolol intoxication. Confirmation of the effectiveness of levosimendan in humans requires further preclinical and clinical study. Appendix E1, available online at http://www.annemergmed.com.

Supervising editor: Richard C. Dart, MD, PhD

Funding and support: By *Annals* policy, all authors are required to disclose any and all commercial, financial, and other relationships in any way related to the subject of this article that might create any potential conflict of interest. See the Manuscript Submission Agreement in this issue for examples of specific conflicts covered by this statement. Financial support provided by Orion Pharma Inc.

Author contributions: HL, ER, LL, and JK designed the study and obtained research funding. HL, JR, and JK performed the study. HL, VK, and JK performed statistical analysis. HL, ER, LL, and JK prepared the article. JK takes responsibility for the paper as a whole.

Publication dates: Received for publication December 23, 2008. Revisions received March 6, 2009; April 9, 2009; and April 30, 2009. Accepted for publication June 24, 2009. Available online September 19, 2009.

Reprints not available from the authors.

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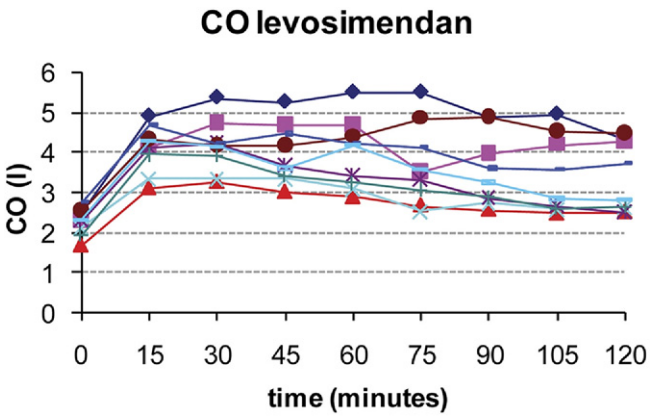

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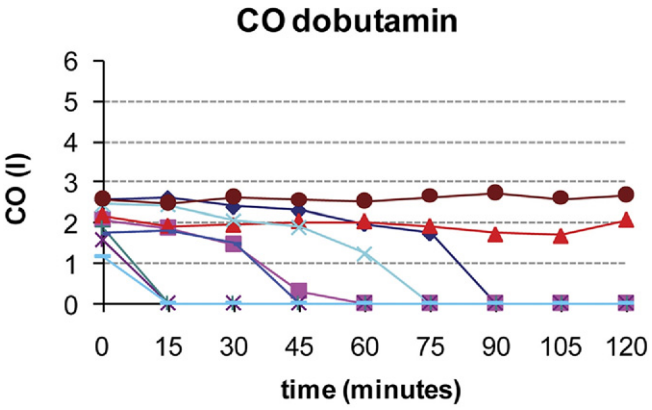
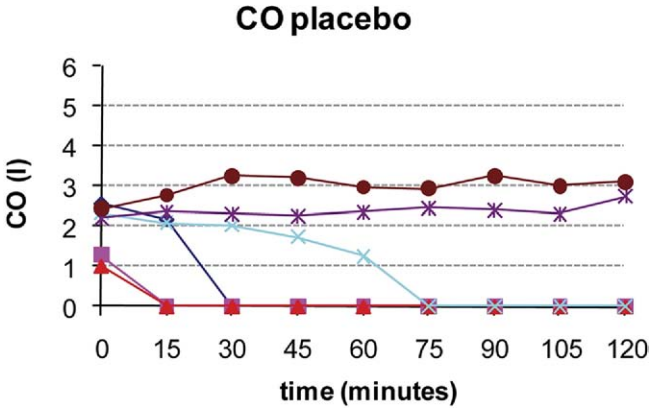


Figure E2.

APPENDIX E1. Hemodynamic changes of individual animals in a porcine model of propranol intoxication.

The changes in CO, LVdP/dt and MAP of each animal in levosimendan, dobutamine and placebo groups from Drug 0 to Drug 120. Figure E1-9



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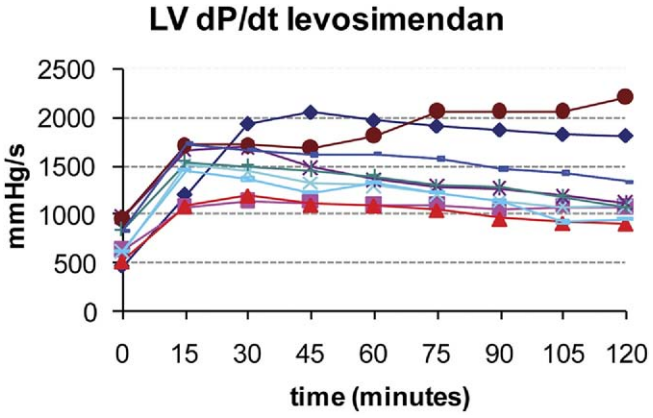

Figure E4.

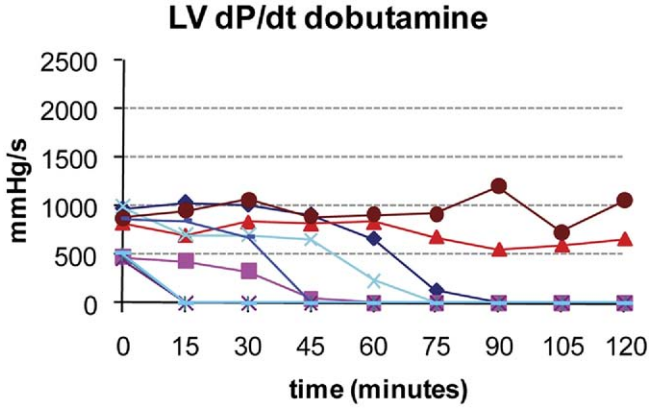

Figure E5.

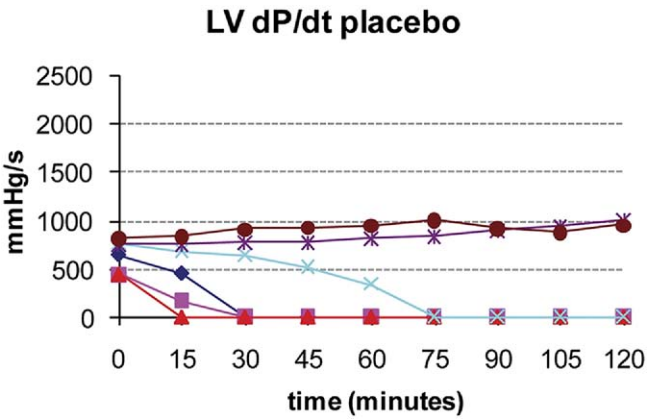

Figure E6.

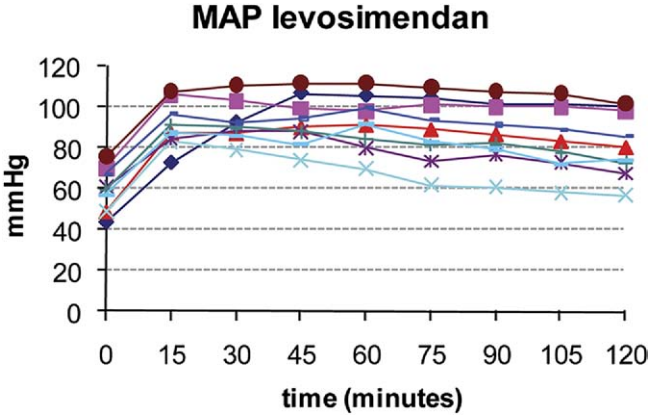

Figure E7.

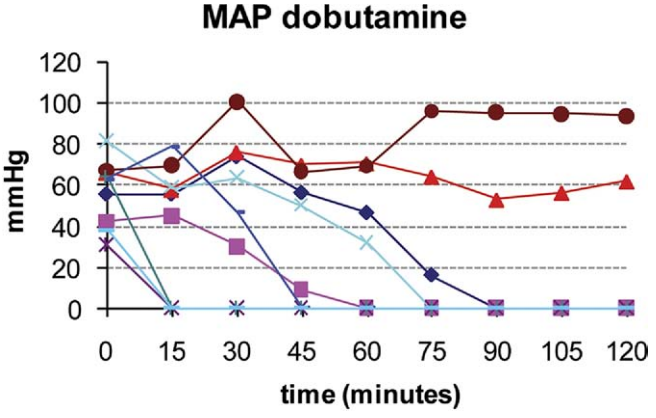

Figure E8.

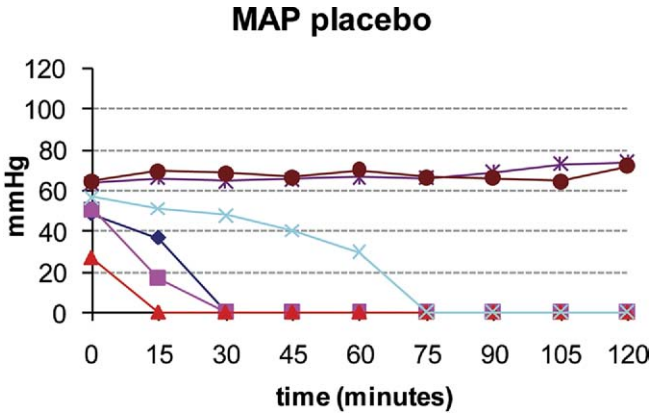
MAP placebo
120
100
80
60
40
20
0
mmHg
0
15
30
45
60
75
90
105
120
time (minutes)

Figure E9.