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Insulin is a Superior Antidote for Cardiovascular Toxicity Induced by Verapamil in the Anesthetized Canine[1]

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ABSTRACT

Because of its positive inotropic effects that are independent of cyclic AMP, insulin was compared to epinephrine and glucagon as a novel treatment for cardiac toxicity from verapamil. Twenty-four α-chloralose-anesthetized mongrel canines of either sex were instrumented to monitor standard hemodynamic and cardiodynamic parameters and maximum elastance at end systole, *via* the transit-time technique, as our index of contractility. Toxicity was induced by 0.1 mg/kg/min of verapamil (i.v.), until 50% reduction in mean arterial blood pressure or complete AV dissociation for 30 min. This was followed by continuous infusion of 1.0 mg/kg/hr of verapamil during one of four treatment protocols: 1) control (0.9% NaCl, 2.0 ml/min); 2) epinephrine (1.0 μg/kg/min); 3) hyperinsulinemic-euglycemic (HIE) clamp (recombinant insulin at 4.0 U/min with 20% dextrose, arterial glucose clamped); or 4) glucagon (0.2–0.25-mg/kg bolus infusion followed by 150-μg/kg/min infusion). Treatments were continued until death or 240 min after which time surviving animals received a 3.0-mg/kg additional bolus of verapamil. Verapamil decreased all hemodynamic parameters during titration. All controls died within 85 min. All treatments tended to improve hemodynamics; however, HIE significantly improved maximum elastance at end systole, left ventricular end diastolic pressure and coronary artery blood flow *vs.* other treatments ($P < .05$, repeated measures). Glucagon transiently restored sinus rhythm (four animals), but in all cases reverted to A-V dissociation, coincident with sharp decreases in circumflex artery blood flow and contractility. Hyperinsulinemic-euglycemic significantly improved survival post-bolus: C, 0%; epinephrine, 33%; HIE, 100%; and glucagon, 0% ($P < .05$, log rank statistic). HIE is a superior treatment for severe verapamil toxicity in the anesthetized canine and its ability to sustain survival appears to result from its positive inotropic effects.

CCB toxicity causes significant morbidity and mortality in the U.S. In 1991 there were 5705 toxic exposures involving CCBs reported to the American Association of Poison Control Centers (Litovitz *et al.*, 1991). Over 60% (3425) of these patients required treatment in a health care facility and 31 cases resulted in death, the majority from verapamil ingestion (18 of 31). With the introduction of new CCBs, coupled with expanded indications for these drugs, and the aging of the U.S. population, clinical use of these drugs will increase. As a result, a greater number of toxic reactions from these drugs is likely. Unfortunately, the treatment of such toxicity has been less than satisfactory to date, with a poor response to standard Advanced Cardiac Life Support drugs and no clear antidotes identified (Horowitz and Rhee, 1989; Ramoska *et al.*, 1993).

Verapamil, the most potent cardiovascular toxin of the CCBs, produces toxicity by blocking calcium entry at the slow, or L-type calcium channel, which are found in both smooth and cardiac muscle membranes (Glossman *et al.*, 1988). The net effect of verapamil on the blockage of the L-type channel includes cardiac conduction delays, negative chronotropic and inotropic effects and reduced systemic vascular resistance.

No antidote consistently reverses both the bradycardia and hypotension seen with verapamil toxicity. Increasing extracellular calcium would appear to be a logical treatment for CCB toxicity. In the canine model, calcium infusions reverse some of the negative inotropy, but not the bradycardia and peripheral vasodilation, associated with verapamil toxicity (Hariman *et al.*, 1979; Gay *et al.*, 1986). Use of calcium in clinical cases of severe verapamil overdose often has little effect on clinically measured parameters such as blood pressure and HR (McMillan, 1988; Orr *et al.*, 1982; van der Meer and van der Wall, 1983; Goenen *et al.*, 1986; Ramoska *et al.*, 1993).

Other treatments for verapamil overdose include catecholamines and GLC, which increase transmembrane calcium flow at the L-type channel by increasing intracellular cAMP concentrations (Colucci *et al.*, 1986; Rasmussen, 1986a,b; Lewis *et al.*, 1990; Murad and Vaughan, 1969; Chernow *et al.*, 1986, 1987;

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ABBREVIATIONS: CCB, calcium channel blocker; HR, heart rate; GLC, glucagon; EPI, epinephrine; cAMP, cyclic AMP; MABP, mean arterial blood pressure; RBF, renal artery blood flow; CO, cardiac output; LVP, left ventricular systolic pressure; LVEDP, left ventricular end diastolic pressure; CABF, circumflex artery blood flow; E_{max}, maximum elastance at end systole; BC, basal control; TITR, titration; HIE, hyperinsulinemia-euglycemia; TX, treatment; ANOVA, analysis of variance.

Lucchesi, 1968). Neither catecholamines nor GLC has consistently reversed toxicity of verapamil in animal studies or in humans (Lipman *et al.*, 1982; Horowitz and Rhee, 1989; Jolly *et al.*, 1987; Ramoska *et al.*, 1993).

Insulin's positive inotropic characteristics and its metabolic effects may have beneficial actions in reversing verapamil toxicity. Verapamil is known to inhibit insulin release from pancreatic islet cells (Devis *et al.*, 1975; DeMartinis and Barbarino, 1980), and several human cases of verapamil toxicity have required insulin to treat hyperglycemia (Enyeart *et al.*, 1983; Spurlock *et al.*, 1991). Insulin increases cardiac contractility during drug and ischemic-induced cardiac failure (Reikeras *et al.*, 1985a; Weissler *et al.*, 1973; Farah and Alousi, 1981). Although the mechanism of this positive inotropic effect is unclear, indirect evidence suggests that increased calcium entry may be responsible (Kones and Phillips, 1993).

The purpose of our study was to compare the efficacy of insulin in reversing verapamil toxicity compared to standard treatments (EPI and GLC), in a reproducible, anesthetized canine model of acute sustained verapamil toxicity.

Methods

Surgical procedure. Twenty-four fasted mongrel dogs of either sex weighing between 19.8 and 27.5 kg were anesthetized with sodium thiamylal (15 mg/kg i.v.) followed by α-chloralose (30 mg/kg/hr). Animals were intubated and ventilated with a Harvard respirator, with 5 to 7 cm H_2O positive end expiratory pressure. Catheters were placed in a femoral artery for measuring MABP and collecting blood samples for chemical analyses. Femoral veins were cannulated and used for infusion of α-chloralose and experimental drugs. *Via* a left subcostal incision, a pulse doppler-ultrasound flow probe (Triton Technology, San Diego, CA) was positioned around the left renal artery near the aorta to measure RBF. The subcostal incision was closed with towel clamps.

A left thoracotomy was made in the fifth intercostal space, and the pericardium was incised longitudinally to expose the heart. CO was measured with an electromagnetic blood flow probe (In Vivo Metric, Healdsburg, CA) positioned around the ascending aorta. A pneumatic vascular occluder (In Vivo Metric) was placed around the aorta distal to the flow probe and used to partially occlude the aorta for separating left ventricular pressure-dimension loops. To collect cardiac venous effluent as part of a separate experiment, a silastic catheter was inserted *via* the right atrial appendage, into the coronary sinus. Instantaneous LVP, LVEDP and HR were measured with a Konigsberg micromanometer transducer inserted *via* an apical stab incision and secured with a purse string suture. LVP was differentiated with a Gould differentiator amplifier and used to measure *+dP/dt*. A doppler flow probe (Triton Technology) was positioned around the left circumflex coronary artery at its origin from the left main artery to measure CABF.

The LV anterior-posterior internal minor axis diameter was measured by using the ultrasonic transit time method. Piezoelectric crystals (20 MHz, Triton Technology) were placed through stab incisions in the left ventricular anterior and posterior walls. Internal left ventricular dimensions were measured by using a Triton Technology sonomicrometer.

Measurements. All cardio- and hemodynamic measurements were recorded on an Astro-Med direct writing polygraph recorder equipped with Gould amplifiers. Data were acquired on-line through a Po-Ne-Mah digitalizing data acquisition system. The slope of the LV pressure-dimension relationship, E_{max}, was used as our index of contractility as described previously (Altimari *et al.*, 1986; Kober *et al.*, 1985). Briefly, the respirator was shut off at end of expiration and the aorta was occluded incrementally in order to generate 7 to 10 cardiac cycles. End systole was defined as the point of maximum elastance and was determined by computer analysis (Calcview) of each loop for the point of maximum pressure/dimension ratio. During this analysis, extrasystolic beats were deleted from analysis. Subsequently, a linear least-squares regression was used to calculate E_{max}.

External analysis and calculations. During each measurement period (outlined in experimental protocol), 2 ml of arterial blood were collected and analyzed as follows: 1) glucose and lactate with a Yellow Springs Instruments, Yellow Springs, OH, 2300 Stat analyzer and 2) electrolytes (Na^+, K^+ and ionized Ca^{++}) with a NOVA 6 electrolyte analyzer.

Experimental protocol. After surgical instrumentation, all animals received approximately 20 ml/kg of 0.9% NaCl as a bolus. This was followed by approximately a 60-min equilibration period. Two basal control measurements were then performed within 20 min to generate average BC data for cardio- and hemodynamic, electrolyte, glucose and lactate measurements. After the second basal measurement, (±)-verapamil hydrochloride (Sigma Chemical Co., St. Louis, MO), 1.0 mg/ml, was infused (0.1 mg/kg/min) *via* a femoral venous catheter until at least one of two hemodynamic set points were achieved: 1) a 50% reduction in average basal MABP or 2) complete atrioventricular dissociation. Once one of these parameters was achieved, the verapamil infusion was titrated to maintain the product of the MABP and the HR (rate pressure product) constant for 30 min before treatment. At 15 and 30 min during verapamil titration ($TITR_{15}$ and $TITR_{30}$), measurements were obtained. After this 30-min titration period, the verapamil infusion was changed to a constant infusion of 1.0 mg/kg/hr for the duration of the experiment. The animals were then assigned randomly to one of four experimental treatment groups: 1) Control ($n = 6$) −0.9% NaCl at 2.0 ml/min. 2) EPI ($n = 6$; Abbott Laboratories, Chicago, IL) was begun at 1.0 μg/kg/min i.v. and was titrated to maintain LVP and *+dP/dt* equal to basal levels. The concentration of EPI was adjusted for each animal's weight such that 1.0 μg/kg/min required a 2.0-ml/min infusion rate. 3) GLC ($n = 6$; Eli Lilly, Indianapolis, IN), a loading dose of GLC (0.2–0.25 mg/kg i.v.) was injected, followed by a maintenance infusion of 150 μg/kg/hr in an adjusted concentration to maintain a 2.0-ml/min infusion rate. 4) HIE ($n = 6$), calcium-free recombinant insulin (NOVOLIN, Bagsvaerd, Denmark) was infused at 4.0 U/min. *Via* a separate catheter, 20% dextrose was simultaneously infused at 0.7 to 1.0 ml/min to maintain arterial glucose concentration within 10.0 mg/dl of the basal level. Potassium chloride (0.2 mEq/ml) was infused at approximately 10 to 20 ml/hr to maintain serum potassium within 1.0 mEq/l of the average basal level. Arterial glucose and $[K^+]$ were analyzed every 10 min during the 1st hr of treatment and infusion rates were adjusted appropriately. To maintain a total infusion rate at 2.0 ml/min, 0.9% NaCl was infused simultaneously.

All animals were monitored until either death or 4 hr of treatment. During treatment, measurements were taken at 15, 30, 60, 120, 180 and 240 min (TX_{15}, TX_{30}, TX_{240}). All animals that survived to TX_{240} min received a rapid 3.0 mg/kg i.v. verapamil bolus followed by hemodynamic and cardiodynamic measurements 15 min later (TX_{bolus}), if the animal survived the bolus. The purpose of this additional bolus was to evaluate the ability of each treatment to sustain its antidotal effects.

Statistical analysis. All hemodynamic data were recorded initially by the Po-Ne-Mah data acquisition system and were subsequently transferred to a MacIntosh Quadra 950 computer for statistical analysis by using the SuperAnova statistical program (v.1.1). All data were tested for homogeneity of variance using Bartlett's test. Nonhomogeneous data were transformed by appropriate techniques. Basal and verapamil titration data were tested for differences among the four groups (control, EPI, HIE and GLC) by using one-way ANOVA. Overall significance between the four groups for continuous data (*e.g.*, E_{max}) was determined using repeated measures ANOVA (Sibley *et al.*, 1990; Dunnett, 1955), followed by Duncans Multiple Range test with $P \le .05$ considered significant. When repeated measures ANOVA found no overall difference for a variable, individual times were compared between groups by using contrast analysis (Sibley *et al.*, 1990). Bonferroni adjustments of "P" were made for multiple comparisons. The type III sums of squares was used in the general linear model used to calculate *F* values which allows for reduction in the number of animals over the experiment (Hendren *et al.*, 1989). Data are presented as means ±

S.E.M. Differences in survival rates were compared by using the log-rank statistic with $P \leq .05$ considered significant.

Results

Base line and titration. All cardio- and hemodynamic data (table 1) as well as arterial electrolytes, glucose and lactate (table 2) at BC were not significantly different between groups. Similarly, data at $TITR_{15}$ and $TITR_{30}$ were not significantly different between groups. Data from only $TITR_{30}$ are presented, representing time of maximal toxicity before treatment (tables 1 and 2). All hemodynamic data were significantly different at $TITR_{30}$ compared to BC groups except for CABF. Changes in electrolytes and glucose were not significant at $TITR_{30}$ compared to BC. Arterial lactate increased significantly for all groups except for those animals assigned to GLC (tables 1 and 2). The amount of verapamil required during titration was also not significantly different between groups (1.40 ± 0.26 mg/kg), and was consistent with previous studies which induced similar degree of verapamil toxicity in canines (Hariman *et al.*, 1979; Gay *et al.*, 1986). All animals eventually demonstrated complete AV dissociation, in most cases with bradycardic junctional rhythms in absence of any sinus electrical activity, before starting treatment protocol. No animals died during titration in this study.

Treatment

Survival. All control animals (*i.e.*, verapamil with saline as treatment) died within 85 min after verapamil infusion (fig. 1). Four of six EPI-treated animals survived to 240 min, and only two of the remaining four survived the 3.0-mg/kg verapamil bolus despite increases in the EPI infusion rate to 10.0 μg/kg/min. Three of six GLC-treated animals survived to 240 min, and none of these three survived the verapamil bolus. GLC infusion rate was not increased as with EPI. All six HIE animals survived to 240 min and subsequently survived the verapamil bolus. HIE achieved statistically significant difference in mortality rate compared to the other three groups ($P < .05$ by log rank statistic).

Hemodynamics and cardiodynamics. Control animals (saline infusion with 1 mg/kg/hr of verapamil) underwent progressive reductions in HR, MABP, LVP, $+dP/dt$, CO and RBF until death (table 3). E_{max} decreased and LVEDP increased, and these trends continued until death (figs. 2 and 3). CABF (fig. 4) also decreased progressively until death.

Hemodynamic responses to EPI and HIE were similar throughout the experimental protocol, except for RBF, which was decreased significantly during EPI treatment compared to HIE. CABF was elevated with HIE compared to other treatments. In all EPI-treated animals, to maintain LVP at basal level, the initial infusion rate (1.0 μg/kg/min) required continual increases (mean TX_{240} EPI infusion rate in four survivors = 1.47 ± 0.18 μg/kg/min). HIE significantly increased E_{max} compared to EPI, and maintained similar LVP and $+dP/dt$ values throughout the experimental protocol (fig. 2; table 3). Neither EPI nor HIE restored sinus rhythm in any animal; however, frequent episodes of junctional and ventricular tachycardia occurred with EPI, but not with HIE treatment.

GLC increased HR significantly compared to other groups, and was the only treatment which restored sinus rhythm at any time. This effect occurred transiently in four animals, degenerating to sustained ventricular tachycardia in two animals and A-V dissociation in the other two. GLC produced significantly smaller increases in LVP, $+dP/dt$ (table 3) and E_{max} (fig. 2) compared to EPI or HIE.

Metabolic effects (table 4). Control animals demonstrated significant increases in both glucose and lactate compared to basal measurements. EPI caused immediate and persistent hyperglycemia and increased lactate compared to the other groups. GLC induced a biphasic glucose response: initial hyperglycemia, followed by hypoglycemia. Coincident with hypoglycemia, GLC treatment demonstrated decreasing lactate concentrations. Lactate with HIE treatment was similar to GLC.

Electrolytes (table 4). Control animals demonstrated no change in $[Na^+]$, $[K^+]$ or $[Ca^{++}]$. $[K^+]$ decreased to similar concentrations during treatment with both EPI and HIE. $[K^+]$ remained unchanged for all surviving animals treated with GLC. All animals that died during treatment demonstrated an increase in potassium (mean = 5.5 ± 0.16 mEq/l during agonal stages, immediately before death) compared to basal. $[Ca^{++}]$ increased during treatment with HIE compared to the other three groups.

Discussion

Our data demonstrate that HIE is superior to EPI and GLC as treatment for lethal verapamil toxicity in anesthetized canines. This was evident from the survival data: no deaths occurred when HIE was used as our treatment in contrast to 67 and 100% lethality with EPI and GLC, respectively. HIE

TABLE 1
Mean hemodynamic data during BC and after 30 min of verapamil $TITR_{30}$

	HR	MABP	LVSP	LVEDP	$+dp/dt$	E_{max}	CO	CABF	RBF
	bpm	*mm Hg*	*mm Hg*	*mm Hg*	*mm Hg/sec*	*mm Hg/mm*	*ml/min*	*ml/min*	*ml/min*
Control									
BC	142 ± 11.1	94 ± 8	108 ± 8	6 ± 0.7	1370 ± 138	32 ± 8	1340 ± 124	40 ± 5	91 ± 11
$TITR_{30}$*	68 ± 10	44 ± 4	66 ± 5	10 ± 1.5	462 ± 72	11 ± 4	705 ± 78	46 ± 6	31 ± 5
EPI									
BC	131 ± 11	94 ± 6	117 ± 6	8 ± 0.8	1560 ± 146	23 ± 3	1399 ± 180	42 ± 5	74 ± 11
$TITR_{30}$*	67 ± 6	42 ± 3	74 ± 9	10 ± 1.0	608 ± 161	12 ± 3	708 ± 112	48 ± 8	27 ± 2
HIE									
BC	129 ± 8	91 ± 5	108 ± 6	5 ± 0.8	1340 ± 101	22 ± 5	1443 ± 136	40 ± 5	71 ± 4
$TITR_{30}$*	70 ± 10	41 ± 4	57 ± 5	9 ± 1.2	566 ± 162	9 ± 3	728 ± 129	50 ± 10	27 ± 4
GLC									
BC	132 ± 14	91 ± 6	109 ± 5	6 ± 0.9	1420 ± 86	26 ± 2	1523 ± 118	44 ± 7	70 ± 14
$TITR_{30}$*	73 ± 8	43 ± 1	68 ± 3	10 ± 1.7	465 ± 42	9 ± 3	921 ± 110	52 ± 8	38 ± 6

* $P < .05$ for all data at $TITR_{30}$ *vs.* BC except for CABF (by ANOVA).

TABLE 2

Arterial electrolyte, glucose and lactate concentrations during BC and after 30 min $TITR_{30}$

	$[Na^+]$	$[K^+]$	$[Ca^{++}]$	Glucose	Lactate
	mEq/l	*mEq/l*	*mEq/l*	*mg/dl*	*mM*
Control					
BC	149.3 ± 0.7	3.33 ± 0.16	1.10 ± 0.06	79.1 ± 2.3	0.96 ± 0.08
$TITR_{30}$	148.5 ± 1.2	4.00 ± 1.4	1.16 ± 0.05	89.5 ± 10.7	1.70 ± 0.35*
EPI					
BC	148.5 ± 0.5	3.40 ± 0.72	1.15 ± 0.02	72.6 ± 2.6	0.83 ± 0.09
$TITR_{30}$	146.7 ± 1.1	3.81 ± 0.29	1.10 ± 0.01	97.0 ± 16.0	1.95 ± 0.46*
HIE					
BC	148.3 ± 0.4	3.29 ± 0.11	1.24 ± 0.03	74.8 ± 6.6	1.16 ± 0.13
$TITR_{30}$	147.3 ± 1.0	3.54 ± 0.18	1.22 ± 0.02	96.8 ± 12.1	1.92 ± 0.31*
GLC					
BC	148.5 ± 0.6	3.25 ± 0.15	1.07 ± 0.04	72.3 ± 4.1	0.94 ± 0.11
$TITR_{30}$	143.7 ± 1.6	3.76 ± 0.16	1.00 ± 0.02	73.4 ± 2.9	1.34 ± 0.17

* $P < .05$ compared to BC (by ANOVA).

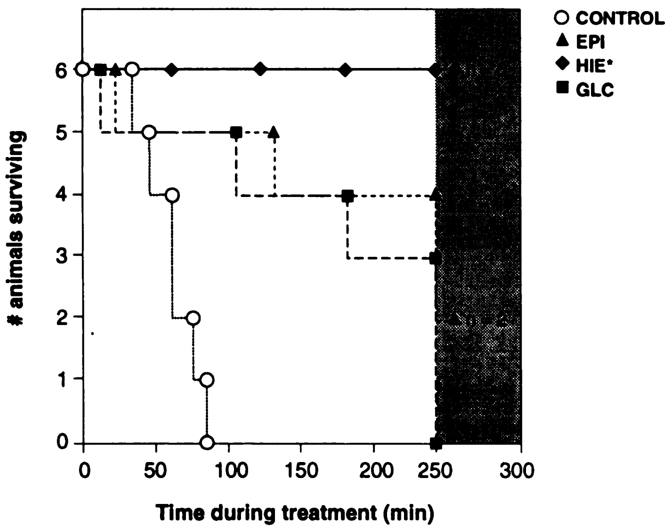

Fig. 1. Kaplan-Meier survival curve during treatment. All control animals (○) died within 85 min. Four of six EPI-treated animals (▲) survived to 240 min; however, two of these four died after the 3.0-mg/kg verapamil bolus (represented as shaded area). Six of six HIE-treated animals (◆) survived to 240 min; in addition, all six survived the bolus (*$P < .05$ *vs.* other groups by log-rank statistic). Three of six GLC-treated animals (■) survived to 240 min, all three remaining animals died during bolus.

increased E_{max} within 15 min, as did EPI and GLC, and produced steady increases in contractility with fewer episodes of tachydysrhythmias than EPI or GLC. At the end of the 4-hr experimental protocol, HIE-treated animals survived the additional verapamil bolus (3.0 mg/kg i.v.) with only transient reductions in hemodynamic indices; however, in two of the four EPI-treated animals that survived the bolus, maximal EPI infusion rate (approximately 10.0 μg/kg/min) did not reverse the profound cardiodepression as did HIE. All GLC-treated animals died as a result of the bolus verapamil injection.

The primary antidotal effect of all treatments was mediated through increases in contractility. HIE maintained adequate organ perfusion, and improved survival, by increasing E_{max} throughout the experimental protocol. EPI and GLC were unable to sustain contractility to a similar degree and thus demonstrated lower coronary and renal perfusion during the treatment protocol. No treatment produced significant increase in MABP. The fact that insulin did not increase blood pressure is consistent with data from previous studies that have demonstrated insulin to cause vasodilatation *in vivo* (Liang *et al.*, 1982; Krukenkamp *et al.*, 1986; Downing and Lee, 1979).

The mechanism by which insulin increases contractility is unknown. Although infusion of pharmacological doses of insulin, while maintaining arterial glucose concentrations constant, does produce small increases in plasma catecholamines in human and canine studies (Liang *et al.*, 1982; Rowe *et al.*, 1981), it is doubtful that these small increases contribute significantly to insulin's antidotal effect in the present study. Plasma catecholamines with glucose "clamped" increase only slightly, and are unlikely to be equivalent to those achieved in the EPI treatment group (Liang *et al.*, 1982). Moreover, insulin increases contractility during anoxic and *beta* adrenergic blockade-induced heart failure *in vivo* (Lucchesi *et al.*, 1972; Krukenkamp *et al.*, 1986; Reikeras *et al.*, 1985b), and *ex vivo* in isolated hearts and papillary muscle perfused with buffers devoid of catecholamines (Lucchesi *et al.*, 1972; Weissler *et al.*, 1973; Farah and Alousi, 1981; Downing and Lee, 1976). Additionally, alterations in metabolism induced by insulin are unlikely the mechanism responsible for enhanced contractility, because insulin has been shown to increase contractility in propranolol-depressed isolated hearts perfused with buffers lacking carbohydrates or fatty acids (Lucchesi *et al.*, 1972; Downing and Lee, 1976).

Insulin may increase contractility during verapamil toxicity by altering ion homeostasis. In the present study, HIE increased plasma ionized $[Ca^{++}]$ compared to control. However, to reverse severe verapamil toxicity in animal models requires infusion of calcium salts in amounts that should produce total $[Ca^{++}]$ well above those demonstrated with HIE (Hariman *et al.*, 1979). The relationship of increased plasma $[Ca^{++}]$ during HIE remains unclear, and its relation to antidotal effect of HIE during verapamil toxicity is currently under investigation in our laboratory. Insulin does increase cytoplasmic ionized calcium in developing skeletal muscle (Schudt *et al.*, 1976) and adipose tissue (Peshadsingh *et al.*, 1987; Kissenbah *et al.*, 1974).

Additionally, in these tissues insulin increases inositol polyphosphate concentrations (Fox *et al.*, 1987; Rosen, 1987), which may augment calcium release from the sarcoplasmic reticulum during systole and reuptake during diastole (Rasmussen, 1986b; Lewis *et al.*, 1990). Insulin also hyperpolarizes the heart as a result of increasing the ratio of intracellular/extracellular $[K^+]$, and increasing efflux of $[Na^+]$ from myocardial cells (Zierler *et al.*, 1966; Regan *et al.*, 1963; Zierler, 1957). Other investigators have found increased plasma $[K^+]$ to potentiate

TABLE 3

Hemodynamic measurements during early (Tx_{15}), mid (Tx_{120}) and late (Tx_{240}) treatment

	HR	MABP	LVP	+dp/dt	CO	RBF
	bpm	*mm Hg*	*mm Hg*	*mm Hg/sec*	*ml/min*	*ml/min*
Control						
Tx_{15}	58 ± 12	48 ± 6	61 ± 5	488 ± 125	621 ± 71	20 ± 5
Tx_{120}						
Tx_{240}						
EPI						
Tx_{15}	59 ± 6	51 ± 2	123 ± 13	1635 ± 223	1037 ± 240	21 ± 3
Tx_{120}	68 ± 15	45 ± 7	124 ± 12	1625 ± 102	1043 ± 599	13 ± 5
Tx_{240}	58 ± 8	39 ± 2	112 ± 6	1400 ± 85	1012 ± 148	10 ± 4
HIE						
Tx_{15}	46 ± 5	49 ± 4	101 ± 9	1350 ± 144	950 ± 121	38 ± 5
Tx_{120}	52 ± 11	40 ± 4	102 ± 7	1437 ± 244	1043 ± 167	25 ± 4
Tx_{240}	48 ± 6	41 ± 3	103 ± 9	1407 ± 207	1016 ± 118	28 ± 3*
GLC						
Tx_{15}	94 ± 12*	40 ± 4	76 ± 15	1007 ± 215	962 ± 183	32 ± 7
Tx_{120}	91 ± 17	43 ± 23	77 ± 4*	664 ± 73*	973 ± 146	31 ± 8
Tx_{240}	67 ± 20	36 ± 5	62 ± 5*	325 ± 125*	784 ± 218	13 ± 1

* $P < .01$ *vs.* all other groups at corresponding times by contrast analysis.

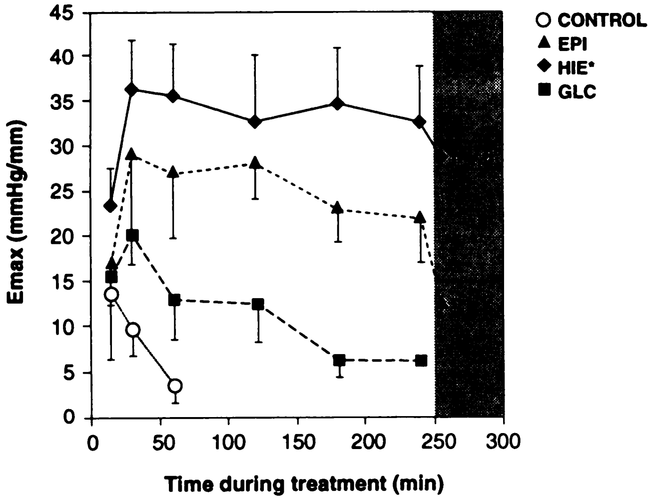

Fig. 2. Contractility (E_{max}) during treatment protocol. E_{max} was significantly elevated for HIE (*$P < .05$ *vs.* other treatment groups by repeated measures ANOVA).

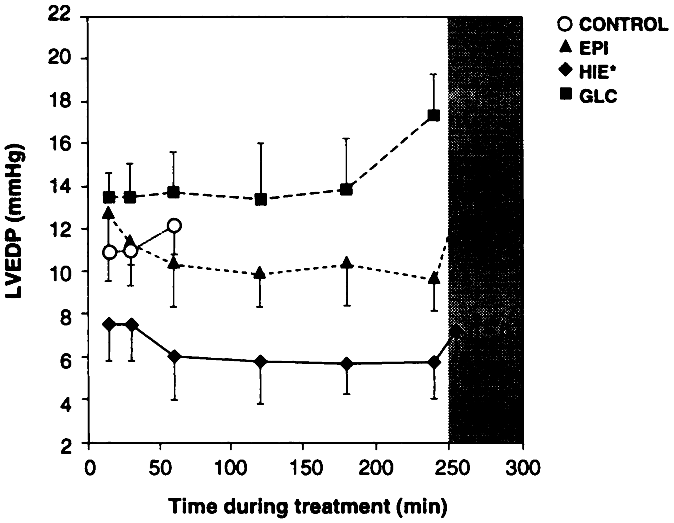

Fig. 3. LVEDP during treatment protocol. LVEDP was significantly reduced by HIE treatment (*$P < .05$ *vs.* other treatment groups by repeated measures ANOVA).

verapamil toxicity in animals and humans (Jolly *et al.*, 1991). Because plasma [K^+] decreased the same amount (approximately 1 mEq/l) with both HIE and EPI treatment, it is unlikely that reduction in plasma [K^+], *per se*, mitigated verapamil's toxicity in this model. Thus, it appears plasma [K^+] affects verapamil's ability to block the L-type channel, but the relevance of this relationship to treatment for verapamil toxicity is uncertain and warrants further study.

HIE provided sustained increases in contractility despite severe verapamil toxicity, suggesting that insulin increases intracellular calcium availability independently of the L-type channel. In contrast, with EPI treatment, E_{max} increased initially, but declined in all animals over time despite continual increases in EPI infusion rate. Catecholamines increase contractility *via* increases in cAMP. cAMP increases phosphorylation of the L-type channel, which increases the probability of Ca^{++} entry through this channel (Cachelin *et al.*, 1983). If insulin increases contractility *via* mechanisms that are independent of cAMP formation, then these increases in E_{max} may not be susceptible to tachyphylaxis as is reported with EPI (Keely *et al.*, 1974).

The positive chronotropic effect of GLC during verapamil toxicity was rapid and clearly different than the other treatments. As with EPI, the positive effects of GLC decreased with time. This is not surprising because GLC and catecholamines are reported to produce identical postreceptor responses (Murad and Vaughan, 1969), suggesting that GLC should demonstrate cardiac tachyphylaxis in a time frame similar to EPI. GLC is purported to differ from catecholamines by its ability to retain chronotropic and inotropic effects in the presence of *beta*-blockade (Murad and Vaughan, 1969; Lucchesi, 1968; Lucchesi *et al.*, 1969), which has lead investigators to postulate the existence of a specific GLC receptor (Jolly *et al.*, 1987; Murad and Vaughan, 1969; Levey *et al.*, 1974), although biochemical evidence for its existence is inferential (Murad and Vaughan, 1969; Levey et al., 1974). In this study, GLC provided unique ability to briefly restore sino-atrial automaticity, thus increasing HR, an effect clearly missing during EPI and HIE treatments. However, this effect degenerated to either sustained ventricular tachycardia in two cases (immediately in one case and at 45 min in another), or reversion to complete A-V block

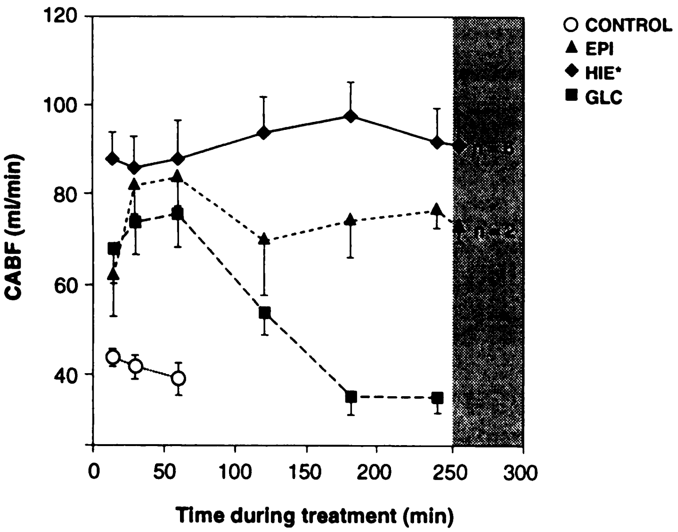

Fig. 4. CABF during treatment protocol. CABF was significantly increased by HIE treatment (*$P < .05$ *vs.* other treatment groups by repeated measures ANOVA).

with intermittent episodes of junctional or ventricular tachycardia in the remaining four animals.

Our objective was to compare treatments administered in doses that would best improve survival and hemodynamics during severe verapamil toxicity. Inasmuch as no cardiac inotropic concentration-response data are available for canines treated with HIE, we reasoned that maximal myocardial glucose uptake corresponded with maximal inotropic effect. Four units per minute of insulin infusion was chosen because this dose has been shown previously to maximally increase glucose uptake in endotoxin-shocked, mongrel canines (Raymond *et al.*, 1988). The EPI infusion rate was adjusted carefully to maintain LVP and +*dP*/*dt* equal to, or slightly above, basal levels. Despite diligent titration of the EPI infusion rate to maintain LVP and +*dP*/*dt* at basal levels, four animals died while receiving EPI. In these animals, decreases in cardiac function were rapid, and were not reversed by prompt increases in EPI infusion rate. We used doses of GLC that we felt were large compared to single-bolus doses used by others (Jolly *et al.*, 1987). Because prolonged GLC infusion during sustained verapamil toxicity has not been documented previously *in vivo*, we chose the highest GLC infusion rates recommended for severe *beta*-blocker toxicity in humans (Rumack and Spoerke, 1993). We did not titrate the GLC infusion rate because, in preliminary studies, increasing GLC infusion rate above 150 μg/kg/hr led to more pronounced hypoglycemia late in the protocol, with more ventricular ectopy, without increase in HR (data not shown). It is thus unlikely that higher doses of GLC would have significantly improved myocardial function. However, the present study was not designed to determine the optimal doses of standard treatments, rather to provide the current clinical standard in care to compare with HIE. It remains a possibility that if higher doses of either EPI or GLC were used, survival may have been affected for these treatments.

The clinical significance of HIE as a treatment for verapamil toxicity is best related to the ability of HIE to safely "prophylax" against worsened toxicity in hemodynamically stable patients with unknown quantities of verapamil ingestion and vague medical histories. Patients who overdose on verapamil, often initially present for care before onset of maximal clinical toxicity, most commonly in cases involving sustained-release preparations and patients may be unwilling or unable to relate their past medical histories (Ramoska *et al.*, 1993). Treatment with calcium salts, catecholamines or GLC requires vigilant cardiovascular monitoring to balance infusion rate against incipient cardiodepression, *vs.* undesirable increases in cardiac work, especially in patients with coronary artery disease. In our model, HIE maintained increases in contractility and cardiac output despite worsened toxicity, and insulin improved CABF and LVEDP to a greater extent than EPI or GLC. Therefore, HIE may be a safer treatment for cardiac failure precipitated by accidental or intentional overdose of verapamil in patients with coronary artery disease. Insulin has been used clinically as an adjunct treatment in a patient with hyperglycemia after verapamil overdose (Enyeart *et al.*, 1983), and HIE is recognized as safe in humans (Rowe *et al.*, 1981).

Several aspects of the acute animal model limit the application of our data to the clinical setting. First, HIE was not studied during toxicity from other classes of CCBs (dihydropyridines and benzothiazepines). These classes are implicated in about one-third of fatal CCB overdoses (Litovitz *et al.*, 1991). Second, although insulin significantly increased RBF late in

TABLE 4

Arterial electrolyte, glucose and lactate concentrations during early (Tx_{15}), mid (Tx_{120}) and late (Tx_{240}) treatment

	[Na^+]	[K^+]	[Ca^{++}]	Glucose	Lactate
	mEq/l	*mEq/l*	*mEq/l*	*mg/dl*	*mM*
Control					
Tx_{15}	148.8 ± 1.0	3.99 ± 0.13	1.13 ± 0.04	118.8 ± 18.5	2.42 ± 0.53
Tx_{120}					
Tx_{240}					
EPI					
Tx_{15}	146.7 ± 0.4	2.94 ± 0.62	1.07 ± 0.02	168.4 ± 13.2	2.39 ± 0.31
Tx_{120}	149.9 ± 1.1	2.40 ± 0.08	0.99 ± 0.06	136.5 ± 5.0	4.63 ± 0.38**
Tx_{240}	150.9 ± 0.2	2.36 ± 0.07	1.06 ± 0.02	118.0 ± 11.0	4.49 ± 0.44**
HIE					
Tx_{15}	148.5 ± 1.4	2.83 ± 0.27	1.23 ± 0.02*	112.3 ± 23.5	2.40 ± 0.51
Tx_{120}	147.7 ± 0.4	2.51 ± 0.08	1.46 ± 0.04*	84.7 ± 15.0	2.47 ± 0.77
Tx_{240}	146.6 ± 0.8	2.52 ± 0.08	1.41 ± 0.03*	77.3 ± 6.7	2.32 ± 0.66
GLC					
Tx_{15}	142.1 ± 1.1	4.20 ± 0.55	0.97 ± 0.04	161.7 ± 10.4	1.51 ± 0.25
Tx_{120}	143.3 ± 1.5	3.68 ± 0.14	0.85 ± 0.13	68.4 ± 16.8	0.84 ± 0.18
Tx_{240}	145.7 ± 2.9	4.19 ± 0.24	0.99 ± 0.10	34.5 ± 3.5**	0.60 ± 0.05

* $P < .05$ *vs.* other treatment groups (repeated measures ANOVA); ** $P < .01$ *vs.* corresponding times for all other groups (contrast analysis).

the protocol compared to EPI and GLC, RBF was still 70% below normal. The effect of these reductions on long-term renal function cannot be assessed in an acute animal model. Furthermore, it is not possible to determine if HIE, in fact, only delayed death from verapamil toxicity. To answer these questions, we are currently undertaking a study using chronically instrumented canines, in which data are collected 48 hr after verapamil overdose.

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