Anesthesiology
78:155–162, 1993

J. B. Lippincott Company, Philadelphia

Effect of Midazolam Infusion and Flumazenil Administration on Epinephrine Arrhythmogenicity in Dogs Anesthetized with Halothane

Michael H. Court, B.V.Sc., Nicholas H. Dodman, B.V.M.S.,† David J. Greenblatt, M.D.,‡ Rajeev K. Agarwal, Ph.D.,§ M. S. A. Kumar, D.V.M., Ph.D.‖*

Background: **Midazolam is being selected increasingly for use in patients with cardiovascular compromise. Although clinical doses of midazolam have minimal effects on cardiac function, the influence of midazolam (and other benzodiazepine sedatives) on cardiac arrhythmogenesis has yet to be elucidated fully.**

Methods: **In this study, we investigated the effect of midazolam, with and without flumazenil, on the arrhythmogenic serum concentration of epinephrine (ACE) in six halothane-anesthetized dogs. Midazolam was administered as a loading dose (1.5 mg/kg over 5 min) followed by a 4.5-h infusion at two rates (10 and 40 $\mu g \cdot kg^{-1} \cdot min^{-1}$) to achieve and maintain predetermined clinical and supraclinical plasma midazolam concentrations. The arrhythmogenic serum concentration of epinephrine determinations were made prior to midazolam infusion, following 2 h of midazolam infusion and following 3.5 h of midazolam infusion and 1 mg flumazenil/kg iv. Saline control studies were also performed in four of the six dogs.**

Results: **Plasma midazolam concentrations ranged from 363 to 855 ng/ml in the low-dose infusion study, essentially spanning the clinically effective range for humans. In the high-dose infusion study, plasma midazolam concentrations were up to four times greater, ranging from 1168 to 3563 ng/ml. The arrhythmogenic serum concentration of epinephrine values were unchanged following low-dose midazolam infusion and saline. In the high-dose study, ACE increased from baseline values of 68 ± 13 (SEM) ng/ml to 112 ± 25 ng/ml (P = .03) following midazolam infusion and decreased to 79 ± 13 ng/ml with flumazenil administration. Plasma midazolam concentrations, however, were poorly correlated with ACE values normalized for control ACE (ACE ratio). Diastolic arterial pressure was significantly depressed following both low-dose (−14%) and high-dose (−19%) midazolam infusion. This decrease in blood pressure was unaffected by flumazenil administration. Other hemodynamic parameters were unaffected by drug treatment.**

Conclusions: **This study has demonstrated that midazolam infusion results in either no effect (with clinical plasma midazolam concentrations) or flumazenil-reversible suppression (with supraclinical concentrations) of halothane-epinephrine arrhythmogenesis. (Key words: Anesthetics, intravenous: midazolam. Anesthetics, volatile: halothane. Antagonists, benzodiazepine: flumazenil. Heart: arrhythmias. Sympathetic nervous system, catecholamines: epinephrine.)**

CONTINUOUS intravenous infusion of midazolam is being evaluated for anesthesia and prolonged sedation of critically ill patients in the intensive care setting. Many of these patients have cardiovascular disturbances and are prone to developing life-threatening cardiac arrhythmias. One of the major advantages of using midazolam over other hypnotic agents is that this drug, like other benzodiazepines, when administered at clinical dosages minimally affects cardiac contractile function.[1–3] The influence of midazolam on cardiac arrhythmogenesis, however, is somewhat unclear.

Muir *et al*[4] demonstrated a decrease in the frequency of ventricular ectopic beats after diazepam administration in conscious dogs following coronary artery occlusion. This observation in research animals was substantiated in the clinical setting by observing a reduced incidence of ventricular arrhythmias in patients with myocardial infarction following administration of diazepam when compared to a control group.[5] This apparent antiarrhythmic effect was attributed to anxiolysis and associated reduction in endogenous catecholamine secretion. A similar theory was proposed to explain the lower incidence of arrhythmias in dental patients ad-

* Assistant Professor, Anesthesia Section, Department of Surgery.

† Professor, Anesthesia Section, Department of Surgery.

‡ Professor of Pharmacology, Psychiatry, and Medicine, Division of Clinical Pharmacology, Tufts University School of Medicine, and New England Medical Center, Boston, Massachusetts.

§ Postgraduate Research Fellow, Department of Anatomy and Cellular Biology.

‖ Associate Professor, Department of Anatomy and Cellular Biology.

Received from Tufts University School of Veterinary Medicine, North Grafton, Massachusetts. Accepted for publication September 13, 1992. Supported in part by NIH Grant S07-RR05852 and by Grant MH-34223 from the United States Department of Health and Human Services.

Address reprint requests to Dr. Court: Department of Surgery, Tufts University School of Veterinary Medicine, 200 Westboro Road, North Grafton, Massachusetts 01536.

ministered diazepam with local anesthetic block compared to the incidence of arrhythmias in patients who received local anesthetic alone.[6]

In contrast, it has been suggested that midazolam may have arrhythmogenic properties. Arcos[7] anecdotally reported tachyarrhythmias and ventricular irritability in three healthy patients following midazolam premedication. In a recent prospective study, a relatively high incidence (25%) of ventricular extrasystoles was reported prior to surgery in midazolam premedicated dental patients compared with an absence of arrhythmias in patients who received no premedication.#

The effect of thiopental and midazolam on epinephrine-aminophylline-induced arrhythmias in halothane-nitrous oxide- anesthetized dogs was recently reported.[8] Induction of anesthesia with 10 mg/kg midazolam was less arrhythmogenic than induction with 30 mg/kg thiopental. Both drugs, however, were administered only once, as a single dose at the beginning of the procedure, and the arrhythmogenic threshold was not determined until more than 2 h into the study. Since blood concentrations of midazolam were not determined and control (non-drug) arrhythmogenic threshold measurements were not made, it is impossible to ascertain from this study whether midazolam administration had an effect on cardiac arrhythmogenesis.

The purpose of this study was to evaluate the effect of midazolam and a benzodiazepine antagonist, flumazenil, in a canine halothane-epinephrine arrhythmia model.

Methods and Materials

The study protocol was approved by the Institutional Animal Care and Use Committee of the authors' institution. Six healthy, mature cross-bred dogs (four intact males, two intact females) with an average body weight of 20.4 ± 0.67 (SEM) kg were used in this study.

Drugs

Midazolam and flumazenil were supplied as the dry free base (Hoffman-La Roche, Nutley, NJ). The hydrochloride salt of midazolam was obtained by addition of 0.9% saline and hydrochloric acid to the free base to reach a stock concentration of 5 mg base/ml of solution at a pH value of 3.0. Solutions of midazolam used for infusion were diluted in the intravenous maintenance fluids (0.9% saline). Consequently, the final concentration of midazolam infused in the low-dose (10 $\mu g \cdot kg^{-1} \cdot min^{-1}$) midazolam study was 120 μg/ml, and in the high-dose (40 $\mu g \cdot kg^{-1} \cdot min^{-1}$) midazolam study was 480 μg/ml. Flumazenil was dissolved in 0.9% saline and administered at a concentration of 0.2 mg flumazenil/ml of solution.

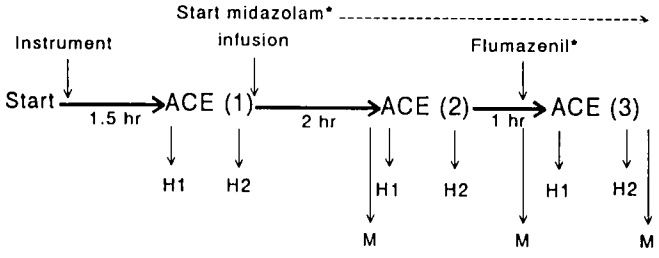
Instrumentation

After inhalation induction, the trachea of the animal was intubated and anesthesia was maintained with halothane in oxygen. End-tidal halothane concentration (Beckman LB-2, Schiller Park, IL) was maintained at 1.3%. The lungs were ventilated and sodium bicarbonate was administered intravenously as needed to maintain normal arterial blood gas values (pH 7.35–7.45; P_{CO_2} 35–45 mmHg). Catheters were inserted percutaneously into each cephalic vein and a jugular vein for drug and maintenance fluid administration. Normal saline (0.9%) was administered throughout the procedure at the rate of 5 $ml \cdot kg^{-1} \cdot h^{-1}$ to replace insensible fluid losses. A catheter also was inserted percutaneously into a femoral artery for direct arterial pressure measurement and collection of samples for blood gas analysis and drug assay. Esophageal temperature was monitored and maintained between 38° and 39° C by use of a recirculating warm-water blanket, hot-water bottles, and a heat lamp. The surface electrocardiogram and direct arterial pressure (Microtrans, Sorensen, Salt Lake City, UT) were monitored continuously and recorded using a two-channel paper recorder (Model 7702B, Hewlett Packard, Waltham, MA). Averaged values for heart rate (HR; from R-R interval), systolic arterial pressure (SAP), and diastolic arterial pressure (DAP) were obtained from the recorded tracings of five consecutive heart beats.

Experimental Protocol

Figure 1 shows a flow chart illustrating the timing of experimental procedures. Following instrumentation, at 1.5 h after induction of anesthesia, the arrhythmogenic serum concentration of epinephrine (ACE) was determined. The technique used was a modification of that described by Weiskopf *et al.*[9] Briefly, this involved infusing epinephrine solution by a calibrated infusion pump (Model 1500, IVAC Corporation, San Diego, CA) through the jugular catheter, initially at a rate of 0.25 $\mu g \cdot kg^{-1} \cdot min^{-1}$. This infusion rate was maintained for

#Rodrigo CR, Rosenquist JB, Cheng CH: Cardiac dysrhythmias with midazolam sedation. Anesthesia Progress 37(1):20–23, 1990.



ACE - determination of arrhythmogenic concentration of epinephrine (0.75 hours)

H1 - hemodynamic parameters prior to ACE

H2 - hemodynamic parameters prior to arrhythmias

M - arterial blood sample for [midazolam]

* - saline controls received equivalent volume saline

Fig. 1. A schematic illustration of the experimental protocol.

5 min, and then the rate was doubled every 5 min (*i.e.*, next rate 0.5 $\mu g \cdot kg^{-1} \cdot min^{-1}$) until four premature ventricular contractions were recorded within 15 s). Immediately following the fourth premature ventricular contraction, the epinephrine infusion was discontinued and a 10-ml sample of blood was withdrawn from the femoral arterial catheter for determination of epinephrine concentration.

After determination of baseline ACE [ACE(1)], a loading infusion of midazolam to a total dose of 1.5 mg/kg was administered over 5 min and immediately followed by a maintenance infusion of midazolam at a rate of either 10 or 40 $\mu g \cdot kg^{-1} \cdot min^{-1}$. The arrhythmogenic serum concentration of epinephrine was redetermined after 2 h of midazolam infusion [ACE(2)] and again 15 min after a slow (over 5 min) intravenous bolus of flumazenil at a dose of 1 mg/kg, which was given after 3.5 h of midazolam infusion [ACE(3)]. After the final ACE determination, the vascular access catheters and electrocardiogram electrodes were removed, lung ventilation and anesthetic administration were discontinued, and the animal was allowed to recover.

Blood samples (10 ml) were collected from the arterial catheter for measurement of midazolam concentration immediately before ACE(2), before flumazenil administration, and after ACE(3). These collection times corresponded to 2, 3.5, and 4.5 h of midazolam infusion.

To ascertain whether any aspects of the procedure, apart from drug administration, affected ACE determination, a saline control study also was performed on four dogs (dogs A–D). Equivalent volumes of 0.9% saline were substituted for the midazolam infusion and flumazenil bolus.

The order of treatments (midazolam infusions and saline control) was randomized with not less than 1 week between experiments.

Sample Analyses

Arterial blood gas samples were withdrawn anaerobically into heparin-containing syringes and analyzed immediately (System 1301, Instrumentation Laboratory, Lexington, MA). Blood samples for midazolam assay were collected into heparin-containing tubes and centrifuged for 20 min at 3,000 RPM and the plasma stored in polypropylene tubes at −70° C until the time of assay (within 6 months). Plasma midazolam concentrations were determined by electron-capture gas-liquid chromatography.[10,11] The lower limit of sensitivity for this assay was approximately 1–3 ng midazolam/ml of plasma. Intra- and interassay coefficients of variation were less than 8%. Samples for epinephrine determination were collected into plain tubes, allowed to clot at 4° C (20 min) and centrifuged for 20 min at 2,000 RPM and 4° C. Serum was then transferred into polypropylene tubes and stored at −70° C until assayed (less than 6 months). Serum epinephrine assays were performed using high-performance liquid chromatography.[12] The lower limit of sensitivity of this assay was approximately 50 pg/ml of serum. Coefficients of variation were less than 12% within assays and less than 18% between assays.

Data Analysis

Analysis of variance corrected for repeated measures was used to determine the effects of infusate (saline and two infusion rates of midazolam) and flumazenil treatment on ACE, SAP, DAP, and HR data; and the effect of infusion time on plasma midazolam concentration. The Newman-Kuels method of multiple comparisons was used as the *post hoc* test where indicated by the ANOVA. The significance level was set at $P < .05$. Data, unless otherwise indicated, are expressed as mean ± SEM.

Linear regression analysis was used to determine whether a relationship could be detected between plasma midazolam concentration and ACE values normalized to baseline ACE values (ACE ratio) and whether flumazenil affected this relationship. The arrhythmogenic serum concentration of epinephrine ratio values were calculated by dividing ACE values collected during midazolam infusion by the baseline ACE value. Thus, two sets of ACE ratio values were generated using this technique: ACE ratio values from dogs that had

received only midazolam [ACE(2)/ACE(1)] and ACE ratio values from dogs that also received flumazenil [ACE(3)/ACE(1)]. Regression lines were fitted to each of these sets of data. Midazolam concentration was considered the independent variable and ACE ratio the dependent variable. The slopes of each of these fitted lines were compared using a *t* test to determine whether there was a significant difference indicating a treatment effect. Correlation coefficients also were calculated for each set of data.

Results

Arterial midazolam concentrations that were measured in plasma samples collected during both high- and low-dose midazolam infusion studies are given in table 1. Although the mean plasma concentrations appeared to decrease between the 2- and 3-h measurements and again between the 3- and 4.5-h measurements for both infusion rates, these changes did not achieve statistical significance ($P > .05$).

Figure 2 shows ACE values associated with each treatment group. During high-dose midazolam infusion (40 $\mu g \cdot kg^{-1} \cdot min^{-1}$), ACE values increased significantly from a mean baseline value of 68 ± 13 ng/ml to 112 ± 25 ng/ml ($P = .03$). Subsequent administration of flumazenil to these animals resulted in a return to values not significantly different from baseline (79 ± 13 ng/ml). There was no change in ACE in dogs following low-dose infusion of midazolam (10 $\mu g \cdot kg^{-1} \cdot min^{-1}$) or following flumazenil administration (56 ± 9, 54 ± 12, and 44 ± 12 ng/ml, respectively). The saline infusion study also was associated with no change in ACE values (49 ± 14, 60 ± 12, and 59 ± 16 ng/ml, respectively).

The relationship between plasma midazolam concentration and ACE ratio is illustrated in figure 3. Al-

Table 1. Plasma Arterial Midazolam Concentrations in Halothane-Anesthetized Dogs

Infusion Rate	N	Midazolam Concentration (ng/ml) +2 h Infusion	+3.5 h Infusion	+4.5 h Infusion
10 $\mu g \cdot kg^{-1} \cdot min^{-1}$	6	600 ± 53 (505–855)	468 ± 59 (391–574)	438 ± 73 (363–591)
40 $\mu g \cdot kg^{-1} \cdot min^{-1}$	6	2463 ± 278 (1626–3563)	2188 ± 299 (1168–2213)	1837 ± 162 (1426–2444)

Data are mean ± SEM, with range in parentheses. Samples were collected at 2, 3.5, and 4.5 h after a 1.5 mg/kg IV bolus of midazolam and constant infusion of midazolam at either 10 or 40 $\mu g \cdot kg^{-1} \cdot min^{-1}$.

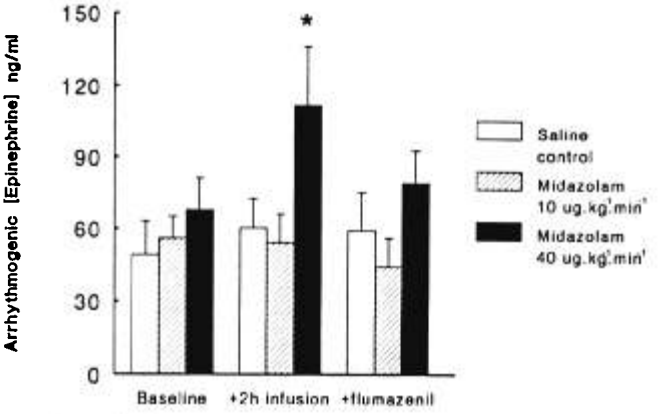


Fig. 2. Arrhythmogenic serum concentrations of epinephrine (ACE) from dogs that received saline, low-dose midazolam (1.5 mg/kg bolus then 10 $\mu g \cdot kg^{-1} \cdot min^{-1}$) or high-dose midazolam (1.5 mg/kg bolus then 40 $\mu g \cdot kg^{-1} \cdot min^{-1}$). Determinations of ACE were made prior to commencement of infusion (baseline), after 2 h of infusion (+2-h infusion), and after 4 h of infusion with bolus intravenous administration of 1 mg flumazenil/kg body weight (+flumazenil). Dogs receiving saline infusion received an equivalent volume bolus of saline instead of flumazenil. Error bars indicate the SEM.

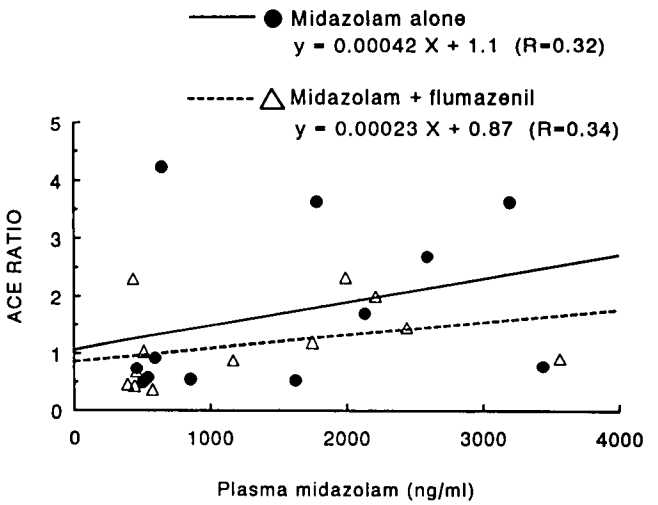


Fig. 3. The relationship between plasma midazolam concentration and arrhythmogenic serum concentrations of epinephrine (ACE) ratio for dogs that received midazolam by infusion (closed circles) and dogs that received midazolam and 1 mg/kg flumazenil intravenously (triangles). The arrhythmogenic serum concentration of epinephrine ratio was calculated by dividing ACE values obtained after initiation of midazolam infusion by baseline ACE value for each study. Two sets of data were produced using this technique: ACE ratio values with midazolam alone [ACE(2)/ACE(1)] and ACE ratio values with midazolam and flumazenil [ACE(3)/ACE(1)]. Also shown are the formulas for the regression lines and corresponding correlation coefficients for each set of data.

Table 2. Initial Hemodynamic Parameters Measured before Initiation of Epinephrine Infusion for Determination of the Arrhythmogenic Serum Concentration of Epinephrine

Infusate	Measurements	N	SAP (mmHg)	(%Δ)	DAP (mmHg)	(%Δ)	HR (beats/min)	(%Δ)
Saline (0.9%)	Baseline	4	96 ± 9	—	52 ± 1	—	86 ± 10	—
	+2-h infusion	4	94 ± 6	(−1)	50 ± 2	(−3)	93 ± 9	(+6)
	+Saline bolus	4	88 ± 9	(−6)	47 ± 1	(−10)	95 ± 9	(+13)
Midazolam 10 $\mu g \cdot kg^{-1} \cdot min^{-1}$	Baseline	6	98 ± 7	—	60 ± 3	—	84 ± 4	—
	+2-h infusion	6	98 ± 8	(−1)	52 ± 4*	(−14*)	86 ± 3	(+2)
	+Flumazenil	6	96 ± 5	(−1)	51 ± 2*	(−15*)	98 ± 5	(+17)
Midazolam 40 $\mu g \cdot kg^{-1} \cdot min^{-1}$	Baseline	6	89 ± 7	—	50 ± 2	—	90 ± 7	—
	+2-h infusion	6	78 ± 6	(−11)	40 ± 2*	(−19*)	91 ± 6	(0)
	+Flumazenil	6	80 ± 6	(−9)	40 ± 3*	(−21*)	96 ± 8	(+7)

Raw data are expressed as mean ± SEM. Percent change (%Δ) data are expressed as percentage change of mean values from baseline mean values. SAP = systolic arterial pressure; DAP = diastolic arterial pressure; HR = heart rate.

* $P < .05$ *versus* baseline data.

though ACE ratio, both with and without flumazenil treatment, was correlated with midazolam concentration, the correlations were considered poor (R = 0.32 midazolam alone, R = 0.34 midazolam + flumazenil). Flumazenil appeared to decrease the slope of the fitted lines (from 0.00042 to 0.00023), however the magnitude of this change was not statistically significant ($P > .05$).

Changes from baseline values of initial hemodynamic parameters measured immediately prior to epinephrine infusion for ACE determination are given in table 2. Mean DAP decreased from baseline values following both low- (−14%) and high-dose (−19%) midazolam infusion and remained decreased after flumazenil administration (−15% and −21%, respectively). There were no statistically significant changes in mean initial HR or SAP with any treatment.

Hemodynamic parameters (HR, SAP, and DAP) measured immediately before arrhythmia detection during epinephrine infusion for ACE determination were unchanged following midazolam infusion or flumazenil administration (table 3).

Discussion

This study has demonstrated that infusion of midazolam results in either no change (with low-dose in-

Table 3. Hemodynamic Parameters Measured during Epinephrine Infusion Immediately Before Arrhythmias during Determination of the Arrhythmogenic Serum Concentration of Epinephrine

Infusate	Measurement	N	SAP (mmHg)	(%Δ)	DAP (mmHg)	(%Δ)	HR (beats/min)	(%Δ)
Saline (0.9%)	Baseline	4	290 ± 13	—	185 ± 2	—	96 ± 5	—
	+2-h infusion	4	288 ± 15	(−1)	186 ± 3	(+1)	93 ± 8	(−3)
	+Saline bolus	4	293 ± 18	(−1)	191 ± 4	(+4)	98 ± 8	(+1)
Midazolam 10 $\mu g \cdot kg^{-1} \cdot min^{-1}$	Baseline	6	279 ± 6	—	158 ± 7	—	77 ± 4	—
	+2-h infusion	6	278 ± 8	(0)	164 ± 5	(+4)	82 ± 6	(+8)
	+Flumazenil	6	289 ± 9	(+4)	168 ± 4	(+7)	92 ± 9	(+24)
Midazolam 40 $\mu g \cdot kg^{-1} \cdot min^{-1}$	Baseline	6	269 ± 13	—	168 ± 6	—	93 ± 8	—
	+2-h infusion	6	269 ± 14	(0)	167 ± 9	(−1)	94 ± 8	(+2)
	+Flumazenil	6	271 ± 18	(0)	174 ± 11	(+3)	99 ± 9	(+8)

Raw data are expressed as mean ± SEM. Percent change (%Δ) data are expressed as percentage change of mean values from baseline mean values. SAP = systolic arterial pressure; DAP = diastolic arterial pressure; HR = heart rate.

* $P < .05$ *versus* baseline data.

fusion) or a flumazenil-reversible decrease (with high-dose infusion) in the propensity for epinephrine-induced arrhythmias in dogs anesthetized with halothane.

The arrhythmogenic threshold for epinephrine has been shown to be dose- and/or blood-concentration dependent for a number of intravenous drugs, including thiopental,[13] propofol,[14] dexmedetomidine,[15] and aminophylline.[8] Midazolam has a rapid elimination profile in both humans and dogs.[11,16,17] Midazolam was therefore administered as a loading dose followed by an infusion in this study to achieve and maintain both clinical and supraclinical plasma midazolam concentrations. Plasma concentrations of midazolam in humans range from 100 to 300 ng/ml for sedation[18] and up to 800 ng/ml as a component of total intravenous anesthesia.[19] Values for midazolam concentration that were obtained during the low-infusion rate study ranged from 363 to 855 ng/ml. In the high-infusion rate study, midazolam concentrations were up to four times greater, varying from 1,168 to 3,563 ng/ml.

To extrapolate the findings of this research study to the clinical situation (for humans), we would need to know the relative potency of midazolam in humans compared with dogs. To our knowledge, there have been no studies that would provide such a direct comparison; however, it may be possible to draw some conclusions from available pharmacodynamic studies. Studies by Hall *et al.*[20,21] investigated the interaction of midazolam and enflurane in dogs. They reported a 50% reduction in enflurane EC_{50} (MAC) at a corresponding plasma midazolam concentration of 610 ng/ml. Although a similar study using enflurane could not be found for humans, Inagaki *et al.*[22] recently reported a 51% reduction in halothane MAC in human subjects with a serum midazolam concentration of 250 ng/ml. This suggests that the plasma concentration-effect relationship is at least within the same order of magnitude for humans and dogs for one pharmacodynamic property of midazolam, anesthetic potency.

The method used in the current study for determination of the arrhythmogenic threshold to epinephrine is similar to methods previously described by Sohn *et al.*[23] and Weiskopf *et al.*[9] It differs from other reported techniques in that epinephrine is administered as a continuous infusion at increasing rates until the termination criterion (four premature ventricular contractions within 15 s) is reached, at which time arterial blood samples are collected for later determination of epinephrine concentration. Other techniques commonly use multiple short (5 min or less) epinephrine infusions with at least 10 min between individual infusions. These techniques can take up to 2 h for a single threshold determination. We chose the current method to enable rapid (within 30 min) determination of the arrhythmogenic state. We could therefore easily perform three ACE measurements (baseline, after 2-h infusion, and after flumazenil) in the same anesthetic period and maintain total study duration at less than 5 h.

Several criteria are used for reporting and comparing the arrhythmogenic threshold of epinephrine, including the total dose of epinephrine administered, epinephrine infusion rate, and ACE. We used the latter criterion since this measure appears to produce the most consistent results, especially when attempting to compare data from experiments that have used different epinephrine infusion protocols.[24]

Control ACE determinations in this study compare well with, although are slightly higher than, data from other studies.[13–15,25] Reported values for ACE in halothane-anesthetized dogs range from mean values of 36 ng/ml[14] to 55 ng/ml[15] compared with baseline values of 49 ± 14, 56 ± 9, and 68 ± 13 ng/ml, which were obtained in this study. Such minor differences between our data and other reported values could be attributable to variations in the epinephrine infusion protocol and the severity of infusion termination criteria.

Several potential weaknesses in the design of this study are worthy of discussion. Since a control study that included midazolam infusion without flumazenil was not performed, the return to baseline arrhythmia threshold following flumazenil administration possibly could be attributable to transient effects of midazolam rather than pharmacologic antagonism by flumazenil. While acute "tolerance" to the central respiratory depressant effects of repeated single intravenous doses of midazolam to dogs has been described,[26] a study by Hall *et al.*[21] showed consistent depression of enflurane MAC when plasma levels of midazolam were maintained constant over 5 h.

Baseline endogenous epinephrine concentrations were not measured during this study. Benzodiazepine sedatives have been shown to depress basal and stress-induced increases in plasma catecholamines.[27–29] Although depression of endogenous plasma epinephrine levels by midazolam might be expected to increase the absolute requirement for exogenously administered epinephrine to produce arrhythmias, the arrhythmogenic serum concentration of epinephrine (the threshold criterion used in this study) is unlikely to be affected.

The relatively poor relationship between plasma midazolam concentration and ACE ratio is not surprising. Benzodiazepines usually have a gradual slope to their dose/plasma concentration-response curves.[21] It is therefore likely that we have only elucidated a small portion of the complete curve. Investigation of lower (<300 ng/ml) and, particularly, higher (>3,000 ng/ml) plasma concentrations of midazolam would probably help to clarify this relationship.

The precise etiology of halothane-epinephrine arrhythmias has yet to be determined fully. It is therefore difficult to speculate as to the exact mechanism responsible for the increase in the arrhythmogenic threshold following high-dose midazolam infusion. One possible contributing factor is the arterial hypotension observed following midazolam administration (14–21% reductions in diastolic arterial pressure; table 2). Midazolam causes dose-related hypotension, presumably by venodilation of the splanchnic vasculature.[30] Arterial pressure is thought to be an important determinant of the epinephrine-arrhythmia threshold.[31,32] Arterial hypotension may have a protective effect, whereas hypertension may promote halothane-epinephrine arrhythmia formation. Although an elevation in ACE was associated with a significant decrease in initial diastolic pressure following high-dose midazolam administration (mean − 19%), the ACE was unchanged following low-dose midazolam infusion with associated similar, albeit lesser, reductions in initial diastolic pressure (mean − 14%). In addition, although flumazenil administration appeared to antagonize the antiarrhythmic effect of midazolam, initial diastolic pressure was still decreased, suggesting that other mechanisms, either distinct from or in addition to systemic hypotension, may be responsible for the observed alteration in arrhythmia threshold.

Pharmacologic antagonism by flumazenil, a specific benzodiazepine antagonist, would suggest that the antiarrhythmic effect of midazolam is at least in part mediated *via* benzodiazepine receptor interaction. A large proportion of the effect of benzodiazepines is exerted by binding to specific receptor sites and enhancing the actions of gamma-aminobutyric acid (GABA), a major inhibitory neurotransmitter within the central nervous system.[5] Recent studies suggest that GABA-ergic neurons exert a tonic inhibitory influence over central autonomic nervous system outflow.[33–35] In urethan-anesthetized rats, for instance, DiMicco[33] showed that benzodiazepines, including midazolam, have a primarily parasympatholytic effect. In other species, the autonomic effects of benzodiazepines is predominantly sympatholytic. Benzodiazepines have been shown to antagonize the hypertension, tachycardia, and arrhythmias associated with toxic doses of local anesthetics in the dog,[36] pig,[37] and rabbit,[38] presumably by enhancement of sympathetic nervous system inhibition. In the rat, however, these toxic effects were enhanced by diazepam administration,[39] thereby supporting DiMicco's observations.[33] In humans, it has been suggested that benzodiazepines are effective in treating bupivacaine-induced arrhythmias, although this has yet to be substantiated experimentally.[40]

In the current study, it is conceivable that midazolam administration resulted in no change (with low-dose infusion) or a decrease (with high-dose infusion) in sympathetic nervous system outflow, thereby increasing the halothane-epinephrine arrhythmia threshold. The practical implications of these findings are that clinical dosages of midazolam may be safe to administer to patients who are prone to sympathetically mediated arrhythmias, while higher dosages may have an antiarrhythmic effect under similar circumstances. Further studies to investigate this hypothesis are indicated.

The authors would like to acknowledge the generous donation of midazolam and flumazenil by Dr. Peter Sorter, of Hoffman-La Roche Inc., Nutley, New Jersey.

References

1. Amrein R, Hetzel W, Bonetti EP, Gerecke M: Clinical pharmacology of Dormicum (midazolam) and Anexate (flumazenil). Resuscitation 16(suppl):5–27, 1988

2. Jones DJ, Stehling LC, Zauder HL: Cardiovascular responses to diazepam and midazolam maleate in the dog. ANESTHESIOLOGY 51: 430–434, 1979

3. Reves JG, Fragen RJ, Vinik HR, Greenblatt DJ: Midazolam: Pharmacology and uses. ANESTHESIOLOGY 62:310–324, 1985

4. Muir WW, Werner LL, Hamlin RL: Antiarrhythmic effects of diazepam during coronary artery occlusion in dogs. Am J Vet Res 36: 1203–1206, 1975

5. Melsom M, Anreassen P, Melsom H, Hansen T, Grendahl H, Hillestad LK: Diazepam in acute myocardial infarction: Clinical effects and effects on catecholamines, free fatty acids, and cortisol. Br Heart J 38:804–810, 1976

6. Driscol EJ, Smilack ZH, Lightbody PM, Florucci RD: Sedation with intravenous diazepam. J Oral Surg 30:332–343, 1972

7. Arcos GJ: Midazolam-induced ventricular irritability (letter). ANESTHESIOLOGY 67:612, 1987

8. Lina AA, Dauchot PJ, Anton AH: Epinephrine-aminophylline-induced arrhythmias after midazolam or thiopentone in halothane-anaesthetized dogs. Can J Anaesth 38:1037–1042, 1991

9. Weiskopf RB, Eger EI II, Holmes MA, Rampil IJ, Johnson B, Brown JG, Yasuda N, Targ AG: Epinephrine-induced premature ventricular contractions and changes in arterial blood pressure and heart

rate during I-653, isoflurane, halothane anesthesia in swine. ANESTHESIOLOGY 70:293–298, 1989

10. Arendt RM, Greenblatt DJ, Garland WA: Quantitation by gas chromatography of the 1- and 4-hydroxy metabolites of midazolam in human plasma. Pharmacology 29:158–164, 1984

11. Greenblatt DJ, Locniskar A, Ochs HR, Lauven PM: Automated gas chromatography for studies of midazolam pharmacokinetics. ANESTHESIOLOGY 55:176–179, 1981

12. Patel V, Borysenko OM, Kumar MSA: Effect of Δ^9-THC on brain and plasma catecholamine levels as measured by HPLC. Brain Res Bull 14:85–90, 1985

13. Hayashi Y, Sumikawa K, Yamatodani A, Tashiro C, Wada H, Yoshiya I: Myocardial sensitization by thiopental to arrhythmogenic action of epinephrine in dogs. ANESTHESIOLOGY 71:929–935, 1989

14. Kamibayashi T, Hayashi Y, Sumikawa K, Yamatodani A, Kawabata K, Yoshiya I: Enhancement by propofol of epinephrine-induced arrhythmias in dogs. ANESTHESIOLOGY 75:1035–1040, 1991

15. Hayashi Y, Sumikawa K, Maze M, Yamatodani A, Kamibayashi T, Kuro M, Yoshiya I: Dexmedetomidine prevents epinephrine-induced arrhythmias through stimulation of central α-2 adrenoceptors in halothane-anesthetized dogs. ANESTHESIOLOGY 75:113–117, 1991

16. Court MH, Greenblatt DJ: Pharmacokinetics and preliminary observations of behavioral changes following administration of midazolam to dogs. J Vet Pharmacol Ther in press

17. Greenblatt DJ, Abernethy DR, Locniskar A, Harmatz JS, Limjuco RA, Shader RI: Effect of age, gender and obesity on midazolam kinetics. ANESTHESIOLOGY 61:27–35, 1984

18. Persson MP, Nilsson A, Hartvig P: Relation of sedation and amnesia to plasma concentrations of midazolam in surgical patients. Clin Pharmacol Ther 43:325–331, 1988

19. Nilsson A, Tamsen A, Persson P: Midazolam-fentanyl anesthesia for major surgery: Plasma levels of midazolam during prolonged total intravenous anesthesia. Acta Anaesthesiol Scand 30:66–69, 1986

20. Hall RI, Schwieger IM, Hug CC Jr: The anesthetic efficacy of midazolam in the enflurane-anesthetized dog. ANESTHESIOLOGY 68: 862–866, 1988

21. Hall RI, Szlam F, Hug CC Jr: Pharmacokinetics and pharmacodynamics of midazolam in the enflurane-anesthetized dog. J Pharmacokinet Biopharm 16:251–262, 1988

22. Inagaki Y, Sumikawa K, Fukumitsu K, Yoshikawa K, Yoshiya I: Serum concentrations of midazolam and the anesthetic efficacy (abstract). ANESTHESIOLOGY 73:A323, 1990

23. Sohn YZ, Hong JC, Katz RL: Antiarrhythmic and hemodynamic responses to adenosine triphosphate during infusion of epinephrine in dogs anesthetized with halothane. Anesth Analg 61:423–429, 1982

24. Woehlck HJ, Rusy BF, Atlee JL III: Comparison of logdose and bracket protocols for determination of epinephrine arrhythmia thresholds in dogs anesthetized with thiopental-halothane. ANESTHESIOLOGY 75:884–892, 1991

25. Sumikawa K, Ishizaka N, Suzaki M: Arrhythmogenic plasma levels of epinephrine during halothane, enflurane and pentobarbital anesthesia in the dog. ANESTHESIOLOGY 58:322–325, 1983

26. Al-Khudairi D, Askitopoulu H, Whitwam JG: Acute "tolerance" to the central respiratory effects of midazolam in the dog. Br J Anaesth 54:953–958, 1982

27. Gaumann DM, Yaksh TL, Tyce GM: Effects of intrathecal morphine, clonidine, and midazolam on the somatosympathoadrenal reflex response in halothane-anesthetized cats. ANESTHESIOLOGY 73:425–432, 1990

28. Marty J, Gauzit R, Lefevre P, Coudere E, Farinotti R, Henzel C, Desmonts JM: Effects of diazepam and midazolam on baroreflex control of heart rate and on sympathetic activity in humans. Anesth Analg 65:113–119, 1986

29. Pieri L: Preclinical pharmacology of midazolam. Br J Clin Pharmac 16(suppl):17–27, 1983

30. Gelman S, Reves JG, Harris D: Circulatory responses to midazolam anesthesia: Emphasis on canine splanchnic circulation. Anesth Analg 62:135–139, 1983

31. Raynold AK: On the mechanism of myocardial sensitization to catecholamines by hydrocarbon anaesthetics. Can J Physiol Pharmacol 62:183–198, 1984

32. Zink J, Sasyniuk BI, Dressel PE: Halothane-epinephrine-induced cardiac arrhythmias and the role of heart rate. ANESTHESIOLOGY 43:548–555, 1975

33. DiMicco JA: Evidence for control of cardiac vagal tone by benzodiazepine receptors. Neuropharmacology 26:553–559, 1987

34. Schmidt B, DiMicco JA: Blockade of GABA receptors in periventricular forebrain of anesthetized cats: Effects of heart rate, arterial pressure, and hindlimb vascular resistance. Brain Res 301:111–119, 1984

35. Wible JH Jr, Luft FC, DiMicco JA: Hypothalamic GABA suppresses sympathetic outflow to the cardiovascular system. Am J Physiol 254:R680–R687, 1988

36. de Jong RH, Heavner JE: Diazepam and lidocaine-induced cardiovascular changes. ANESTHESIOLOGY 39:633–638, 1973

37. Bernards CM, Carpenter RL, Rupp SM, Brown DL, Morse BV, Morell RC, Thompson GE: Effect of midazolam and diazepam premedication on central nervous system and cardiovascular toxicity of bupivacaine in pigs. ANESTHESIOLOGY 70:318–323, 1989

38. Bernards CM, Artru AA: Hexamethonium and midazolam terminate dysrhythmias and hypertension caused by intracerebroventricular bupivacaine in rabbits. ANESTHESIOLOGY 74:89–96, 1991

39. Gregg RV, Turner PA, Denson DD, Stuebing RC, Schlhorst CS, Forsberg T: Does diazepam really reduce the cardiotoxic effects of intravenous bupivacaine. Anesth Analg 67:9–14, 1988

40. DeJong RH, Davis NL: Treating bupivacaine arrhythmias: Preliminary report. Reg Anesth 6:99–103, 1981