

The Rate of CSF Formation, Resistance to Reabsorption of CSF, and Aperiodic Analysis of the EEG Following Administration of Flumazenil to Dogs

Alan A. Artru, M.D.*

The effects of flumazenil, a benzodiazepine antagonist, on the rate of cerebrospinal fluid (CSF) formation (\dot{V}_f), resistance to reabsorption of CSF (R_a) and the electroencephalogram (EEG) was determined in 12 dogs anesthetized with halothane (0.4%, end-expired) and nitrous oxide (66%, inspired) in oxygen. In six dogs the responses to flumazenil were measured during administration of midazolam (1.6 mg/kg followed by $1.25 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$, intravenously) given along with inhalational anesthesia, whereas in the other six dogs the responses to flumazenil were measured during inhalational anesthesia without midazolam. \dot{V}_f and R_a were determined using ventriculocisternal perfusion, and EEG activity was evaluated using aperiodic analysis. Flumazenil, 0.0025 and 0.16 mg/kg, was administered both when CSF pressure was normal and when CSF pressure was increased to 36–38 cmH₂O by continuous infusion of mock CSF. Flumazenil produced no statistically significant change in \dot{V}_f . Flumazenil did produce inconsistent and relatively small changes in R_a . Quantitative aperiodic analysis indicated changes in EEG activity only when the larger dose of flumazenil was given to dogs receiving midazolam. At normal CSF pressure the changes were consistent and were comprised of decreases in θ , α , and total hemispheric power. At elevated CSF pressure the changes were less consistent. It is concluded that smaller doses of flumazenil (which cause no EEG changes with the present method of analysis) and larger doses of flumazenil (which reverse midazolam-induced increase of θ and α activity) produce no change of \dot{V}_f and no consistent change of R_a . Although flumazenil given in the presence of midazolam may increase R_a , thereby increasing CSF pressure and impairing contraction of CSF volume, this effect is not likely to be clinically important. (Key words: Anesthetics, intravenous: midazolam. Antagonists, benzodiazepine: flumazenil. Brain: electroencephalogram; intracranial pressure. Cerebrospinal fluid: formation; pressure; reabsorption. Hypnotics: midazolam.)

FLUMAZENIL is an imidazodiazepine that binds to benzodiazepine receptor sites in the brain. Studies on the cerebral effects of flumazenil reported that doses of 0.0025–1.0 mg/kg increased cerebrospinal fluid (CSF) pressure in patients and dogs receiving midazolam.^{1†‡}

* Professor of Anesthesiology.

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Address reprint requests to Dr. Artru: Department of Anesthesiology, RN-10, University of Washington School of Medicine, Seattle, Washington 98195.

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One cause for flumazenil-induced increase of CSF pressure appears to be cerebral vasodilation. Flumazenil doses of 0.01–1.0 mg/kg were reported to produce a significant decrease of cerebral vascular resistance and increase of cerebral blood flow (CBF). Other causes for flumazenil-induced increase of CSF pressure may be alteration of the rate of CSF formation (\dot{V}_f), resistance to reabsorption of CSF (R_a), or intracranial volume–pressure relationships. One aim of the present study was to determine whether flumazenil produces significant changes in \dot{V}_f and/or R_a in dogs receiving midazolam.

Flumazenil also was previously reported to cause activation of the electroencephalogram (EEG) when given to patients or dogs receiving midazolam.^{1,§} The method of EEG analysis was spectral analysis of the EEG based on fast Fourier transform, or visual inspection of “raw” EEG activity. However, the significance of flumazenil-induced changes in EEG activity is not certain because the method of EEG analysis used in the previous studies may not be optimal for statistical treatment. Fast Fourier transform collects data only in repeated windows of finite time, and it has been proposed by some authors that the technique is not ideally suited for the stochastic nature of the EEG signal.² Visual inspection of “raw” EEG does not provide numerical data. A second aim of the present study was to examine the effects of flumazenil on the EEG using aperiodic analysis, a technique designed to continuously analyze waveforms not characterized by consistent periods.

A third aim of this study was to examine the effects of flumazenil on \dot{V}_f , R_a , and EEG at elevated CSF pressure where CSF pressure was increased by volume expansion of the CSF space. The technique used for CSF volume expansion was continuous infusion of mock CSF.³ This technique and the level of increase of CSF pressure were designed to avoid focal cerebral ischemia and distortion of intracranial contents.³

If in the present study flumazenil was found to alter \dot{V}_f , R_a , or the EEG in dogs receiving midazolam, it would not be certain whether the changes resulted from reversal of benzodiazepine effect or a direct action of flumazenil. A fourth aim of the present study was to determine whether a preexisting benzodiazepine effect altered the

§ Schwilden H, Stoeckel H, Lauren PM, Schüttler J: Action of a benzodiazepine antagonist during midazolam infusion in steady-state: Quantitative EEG studies (abstract). *ANESTHESIOLOGY* 57:A326, 1982

response of \dot{V}_f , R_a , or the EEG to flumazenil. For that purpose, two groups of dogs were studied—one receiving midazolam and one not receiving midazolam.

Methods

ANIMAL PREPARATION

This study was approved by the Animal Care Committee of the University of Washington. Twelve unmedicated mongrel dogs (weighing 15–22 kg) were studied. Anesthesia was induced with halothane (>1.5% inspired concentration) and nitrous oxide (N_2O , 66%, inspired) in oxygen. The trachea was intubated, expired CO_2 was continuously monitored (Beckman Medical Gas Analyzer, Model LB2, Beckman Instruments, Inc., Fullerton, California), and ventilation was regulated by a servocontroller to maintain expired CO_2 at normocapnia. The right femoral artery was cannulated to permit arterial blood sampling for blood–gas analysis and to permit continuous monitoring of systemic arterial blood pressure and heart rate. Mean arterial blood pressure (MAP) was determined by electronic integration. A urinary catheter was inserted, the right femoral vein was cannulated for saline and drug administration, and temperature was monitored by a nasopharyngeal thermistor probe. Intravenous infusion of pancuronium 2–4 mg/h maintained muscle relaxation. Temperature was maintained at $37.0 \pm 0.5^\circ C$ by heat lamps. Depletion of vascular volume was minimized by continuous infusion of saline at $4\text{--}6 \text{ ml} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$.

The EEG was recorded using bilateral frontoparietal electrodes and a Lifescan™ Brain Activity Monitor System (Diatek Medical™ Technology Incorporated, San Diego, California) with a bandpass of 0.5 to 29.9 Hz. Electrodes were adjusted to maintain impedance between electrode pairs at $<7 \text{ K}\Omega$. This system uses aperiodic analysis to convert the analog EEG signal into a set of digital parameters.^{2,4} With aperiodic analysis local extremes in the unprocessed EEG waveforms are identified. The height of each peak and valley and the time of occurrence are measured. Amplitude is defined as the average of the difference in voltage between a peak and the preceding and following valleys. Frequency is defined as the inverse of the difference in the time of occurrence of the two valleys. Computer (Zenith Data Systems, Glenview, Illinois) analysis of the EEG was performed using a Lifescan™ Research System program. For each 60 s of EEG activity the following values were determined for both the right and left cerebral hemisphere: the power (calculated as amplitude squared) and number of waves in each of the standard frequency bins (δ , 0.5–3 Hz; θ , 3–8 Hz; α , 8–12 Hz; and β , 12–30 Hz), total hemispheric power, and the activity edge (the frequency below which 80% of hemispheric power was present).

A burr hole was placed over the left hemisphere and a catheter was directed into the underlying lateral ventricle. The posterior neck muscles were surgically separated to expose the atlantooccipital membrane, and a catheter was directed into the cisterna magna. A 0.3-ml sample of CSF was obtained from the cisternal cannula for measurement of osmolality using a Wescor Model 5100 B Vapor Pressure Osmometer (Wescor, Inc., Logan, Utah). Mock CSF of matching osmolality was prepared by mixing standard solutions⁵ (osmolality 290, 300, or 310 mOsm/kg) labeled with blue dextran (1 mg/ml) (Sigma Chemical Co., St. Louis, Missouri). Details of this animal preparation have been previously reported.^{6,7}

Mock CSF was infused into the ventricle and the mixture of mock and native CSF was permitted to flow passively out of the cisterna magna through a short length of tubing attached to the cisternal cannula. Details for the technique of ventriculocisternal perfusion have been previously reported.^{8,9} The infusion rate (\dot{V}_i) was controlled with a roller pump and gradually increased to 0.6 ml/min, while ventricular CSF pressure was continuously monitored. The open end of the cisternal outflow tubing was placed at the same height as the CSF level present in the cisternal and ventricular cannula before perfusion was begun. This arrangement did not alter CSF pressure from the value normally occurring in the intact animal because the height of the open end of the outflow tubing determines CSF pressure during open ventriculocisternal perfusion. Successful ventriculocisternal perfusion was indicated by outflow of labeled CSF from the cisternal cannula with no increase in ventricular CSF pressure above preperfusion values. This initial period of open ventriculocisternal perfusion allowed for equilibrium of labeled mock CSF with native CSF in the intracranial and cisternal CSF spaces of the dog. Details of this modified open ventriculocisternal perfusion technique have been previously reported.^{6,10}

After 1 h of ventriculocisternal perfusion, \dot{V}_i was gradually reduced to 0.3 ml/min. The concentration of blue dextran in cisternal outflow samples was determined intermittently using light absorbance at 620 nm on a Beckman DU-2 Spectrophotometer (Beckman Instruments, Inc., Fullerton, California) (fitted with a Gilford absorbance indicator, Gilford Instrument Laboratories, Inc., Oberlin, Ohio). Equilibration of the tracer was considered complete when measured blue dextran concentrations in three consecutive samples of cisternal outflow (collection time = 4–6 min per sample) agreed within 2%. Total equilibrium time was 90 min.

Prior to the completion of the surgical preparation six dogs (group 1) received midazolam 1.6 mg/kg intravenously followed by continuous iv infusion of midazolam at $1.25 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$. Intravenous infusion of midazolam continued until the end of the study. The other six dogs

(group 2) received no midazolam. In both groups wounds were infiltrated with bupivacaine (0.5%) during the surgical preparations, and the expired concentration of halothane was decreased to 0.4% (end-expired concentration determined by gas chromatography, N₂O continued) after completion of the surgical preparation.

EXPERIMENTAL PERIOD

The experimental period began once tracer equilibration was complete. \dot{V}_f and R_a were determined at each of six experimental conditions: three during normal CSF pressure and the same three during increased CSF pressure. The three experimental conditions were as follows: 1) no flumazenil (control), 2) flumazenil 0.0025 mg/kg, and 3) flumazenil 0.16 mg/kg. These doses of flumazenil were selected because the smaller dose previously was reported to reverse the reduction of cerebral metabolic rate for oxygen and alteration of EEG activity caused by midazolam and the larger dose previously was reported to also decrease cerebral vascular resistance, increase cerebral blood flow, and increase CSF pressure compared with dogs not receiving midazolam.[†] Each dose of flumazenil was given intravenously over 60 s. Three dogs were examined first at normal CSF pressure and then at increased CSF pressure, and three dogs were examined first at elevated CSF pressure and then at normal CSF pressure in both group 1 (midazolam) and group 2 (no midazolam). Assignment to these two sequences was randomized. At each experimental condition \dot{V}_f and the rate of reabsorption of CSF (\dot{V}_a) were determined twice—once at the target CSF pressure (whether normal or increased) and once with the CSF pressure increased 10 cmH₂O above the target value. It was necessary to determine \dot{V}_a twice at each experimental condition (*i.e.*, at two CSF pressures) to permit calculation of R_a . Normal CSF pressure was defined as that when the open end of the cisternal outflow tubing was placed at the same height as the CSF level present in the cisternal and ventricular cannulae before perfusion was begun. Increased CSF pressure was defined as CSF pressure when the open end of the cisternal outflow tubing was placed approximately 30 cm above the level used for normal CSF pressure.

Sixty minutes was allowed for restabilization when CSF pressure was changed from normal to increased or from increased to normal. At least 27 min (the first 17 min with $\dot{V}_i = 0.6$ ml/min followed by at least 10 min with $\dot{V}_i = 0.3$ ml/min) was allowed between each of the six experimental conditions to permit reequilibration of the tracer. At each condition three consecutive samples of cisternal outflow were collected (collection time = 3–6 min per sample) for determinations of \dot{V}_f and \dot{V}_a . The duration of the experimental period was 7 h.

\dot{V}_f and \dot{V}_a were calculated as previously described¹⁰

according to the formulas of Heisey *et al.*⁸ For these calculations concentrations of blue dextran in samples of the labeled mock CSF being perfused into the ventricle also were determined using light absorbance at 620 nm. For the normal CSF pressure conditions, R_a was calculated from the two \dot{V}_a values determined at normal CSF pressure and 10 cmH₂O above normal CSF pressure. For the elevated CSF pressure conditions, R_a was calculated from the two \dot{V}_a values determined at elevated CSF pressure and 10 cmH₂O above elevated CSF pressure. By definition, R_a is a reciprocal measure of the slope relating \dot{V}_a (expressed as ml/min) to CSF pressure (expressed as cmH₂O).⁹ Systemic values also were recorded at each experimental condition. EEG data were recorded for 4 min prior to each dose of flumazenil and for 10 min following each dose of flumazenil.

STATISTICAL ANALYSIS

Statistical comparisons within groups were made using repeated-measures analysis of variance (ANOVA), and comparisons between groups were made using one-way and multiple ANOVA.¹¹ Multiple ANOVA indicated that in both group 1 (midazolam) and group 2 (no midazolam), dogs studied first at increased and then at normal CSF pressure responded to both flumazenil and the increase of CSF pressure similarly to dogs studied first at normal and then at increased CSF pressure. Thus, the data from each group ($n = 6$) were treated as a set and were not subdivided according to the sequence of increase of CSF pressure. Comparisons between the present groups and previously reported “normal” values for \dot{V}_f and R_a for dogs^{12–14} were made using one-way ANOVA. Where the calculated F value exceeded the critical value for the 0.05 probability level, the Student-Newman-Keuls’ test was used to determine which differences were significant at $P < 0.05$.¹⁵ Values are tabulated as mean \pm SD.

Results

Mean control \dot{V}_f values ranged from 0.035 ± 0.009 to 0.040 ± 0.012 ml/min in dogs receiving midazolam, and from 0.028 ± 0.011 to 0.037 ± 0.011 ml/min in dogs not receiving midazolam (tables 1 and 2). Flumazenil produced no significant change of \dot{V}_f in either group whether CSF pressure was normal or elevated. Within groups, control \dot{V}_f values at normal CSF pressure were not different from those at increased CSF pressure. At either CSF pressure, control \dot{V}_f values were not different between groups.

At normal CSF pressure, R_a in dogs receiving midazolam (218 ± 32 cmH₂O \cdot ml⁻¹ \cdot min⁻¹) was not different than R_a in dogs not receiving midazolam (268 ± 24 cmH₂O \cdot ml⁻¹ \cdot min⁻¹). The smaller dose of flumazenil increased R_a by 77% in dogs receiving midazolam, but

TABLE 1. Cerebral Values and Mean Arterial Pressure in Dogs Receiving Midazolam (n = 6; mean \pm SD)

	Control		Flumazenil 0.0025 mg/kg		Flumazenil 0.16 mg/kg	
	CSF State 1	CSF State 2	CSF State 1	CSF State 2	CSF State 1	CSF State 2
Normal CSF pressure						
\dot{V}_f (ml/min)	0.035 \pm 0.009	0.036 \pm 0.009	0.033 \pm 0.009	0.029 \pm 0.008	0.031 \pm 0.008	0.030 \pm 0.007
R_a (cmH ₂ O \cdot ml ⁻¹ \cdot min ⁻¹)	218 \pm 32		385 \pm 18*		251 \pm 26	
CSF pressure, intraventricular (cmH ₂ O)	10 \pm 2	20 \pm 2†	10 \pm 1	20 \pm 1†	12 \pm 3	22 \pm 3†
Mean arterial pressure (mmHg)	102 \pm 15	106 \pm 15	104 \pm 7	105 \pm 6	108 \pm 8	107 \pm 9
Increased CSF pressure						
\dot{V}_f (ml/min)	0.040 \pm 0.012	0.040 \pm 0.012	0.039 \pm 0.010	0.039 \pm 0.007	0.038 \pm 0.005	0.034 \pm 0.004
R_a (cmH ₂ O \cdot ml ⁻¹ \cdot min ⁻¹)	104 \pm 14†		116 \pm 11		130 \pm 10*	
CSF pressure, intraventricular (cmH ₂ O)	38 \pm 2†	48 \pm 2†	38 \pm 2	48 \pm 2†	38 \pm 2	48 \pm 2†
Mean arterial pressure (mmHg)	109 \pm 7	110 \pm 13	111 \pm 12	112 \pm 14	115 \pm 10	114 \pm 11

* Significantly different from values at the other experimental conditions for that CSF pressure, $P < 0.05$.

† Significantly different from value at target CSF pressure, $P < 0.05$.

‡ Significantly different from corresponding control value at normal CSF pressure, $P < 0.05$.

the larger dose produced no significant change in R_a . The larger dose of flumazenil decreased R_a by 10% in dogs not receiving midazolam, whereas the smaller dose produced no significant change in R_a . In both groups increasing CSF pressure to 36–38 \pm 2 cmH₂O decreased R_a by 47–52%. At this CSF pressure the larger dose of flumazenil increased R_a by 25% in the dogs receiving midazolam, but the smaller dose caused no significant change in R_a . Neither flumazenil dose changed R_a in dogs not receiving midazolam at increased CSF pressure.

At both normal and increased CSF pressure, one-way ANOVA indicated that the EEG in dogs receiving midazolam was comprised of fewer β -waves than the EEG of dogs not receiving midazolam (table 3). In addition, mul-

tipole ANOVA indicated that the combination of θ and α power and number of waves was greater in dogs receiving midazolam than in dogs not receiving midazolam. Within each of the two groups of dogs, elevation of CSF pressure caused no significant change in number of waves or power in any of the frequency bins, and no change in total hemispheric power or activity edge. In dogs receiving midazolam the larger dose of flumazenil decreased α , θ , and total hemispheric power at normal CSF pressure, both on the visual display of processed EEG activity (fig. 1) and by quantitative analysis of the EEG (fig. 2). These decreases attained statistical significance at 2–3 min postflumazenil and remained significant through 10 min postflumazenil. At increased CSF pressure the larger dose of

TABLE 2. Cerebral Values and Mean Arterial Pressure in Dogs Not Receiving Midazolam (n = 6; mean \pm SD)

	Control		Flumazenil 0.0025 mg/kg		Flumazenil 0.16 mg/kg	
	CSF State 1	CSF State 2	CSF State 1	CSF State 2	CSF State 1	CSF State 2
Normal CSF pressure						
\dot{V}_f (ml/min)	0.032 \pm 0.011	0.028 \pm 0.011	0.029 \pm 0.009	0.031 \pm 0.009	0.029 \pm 0.006	0.028 \pm 0.007
R_a (cmH ₂ O \cdot ml ⁻¹ \cdot min ⁻¹)	268 \pm 24		233 \pm 30		215 \pm 16*	
CSF pressure, intraventricular (cmH ₂ O)	11 \pm 1	21 \pm 1†	11 \pm 2	21 \pm 2†	11 \pm 2	21 \pm 2†
Mean arterial pressure (mmHg)	111 \pm 7	113 \pm 10	109 \pm 11	112 \pm 12	107 \pm 11	107 \pm 11
Increased CSF pressure						
\dot{V}_f (ml/min)	0.037 \pm 0.011	0.037 \pm 0.011	0.037 \pm 0.008	0.037 \pm 0.011	0.033 \pm 0.010	0.034 \pm 0.009
R_a (cmH ₂ O \cdot ml ⁻¹ \cdot min ⁻¹)	143 \pm 18†		101 \pm 23		117 \pm 20	
CSF pressure, intraventricular (cmH ₂ O)	36 \pm 2†	46 \pm 2†	36 \pm 2	46 \pm 2†	36 \pm 2	46 \pm 2†
Mean arterial pressure (mmHg)	113 \pm 9	115 \pm 9	110 \pm 10	110 \pm 12	112 \pm 8	109 \pm 10

* Significantly different from values at the other experimental conditions for that CSF pressure, $P < 0.05$.

† Significantly different from value at target CSF pressure, $P < 0.05$.

‡ Significantly different from corresponding control value at normal CSF pressure, $P < 0.05$.

TABLE 3. Aperiodic Analysis of EEG Activity Prior to Administration of Flumazenil (mean \pm SD)

	Dogs Receiving Midazolam (n = 6)		Dogs Not Receiving Midazolam (n = 6)	
	Normal CSF Pressure	Increased CSF Pressure	Normal CSF Pressure	Increased CSF Pressure
δ				
Power ($\mu V^2 \cdot 10^{-3} \cdot \text{min}^{-1}$)	41.3 \pm 17.3	62.2 \pm 39.6	74.4 \pm 39.7	42.7 \pm 17.9
Waves (number/min)	53 \pm 3	64 \pm 30	73 \pm 35	63 \pm 12
θ				
Power ($\mu V^2 \cdot 10^{-3} \cdot \text{min}^{-1}$)	63.7 \pm 33.7*	84.4 \pm 48.4*	44.6 \pm 31.4	42.8 \pm 24.2
Waves (number/min)	125 \pm 50*	120 \pm 50*	105 \pm 51	91 \pm 45
α				
Power ($\mu V^2 \cdot 10^{-3} \cdot \text{min}^{-1}$)	97.8 \pm 53.8*	70.5 \pm 42.8*	60.7 \pm 39.6	58.4 \pm 31.8
Waves (number/min)	177 \pm 59*	167 \pm 68*	165 \pm 62	156 \pm 45
β				
Power ($\mu V^2 \cdot 10^{-3} \cdot \text{min}^{-1}$)	82.8 \pm 39.3	74.6 \pm 41.5	86.0 \pm 39.6	74.9 \pm 41.9
Waves (number/min)	462 \pm 185†	448 \pm 196†	656 \pm 243	617 \pm 180
Total power ($\mu V^2 \cdot 10^{-3} \cdot \text{min}^{-1}$)	285.6 \pm 136.7	294.6 \pm 149.2	265.8 \pm 111.1	228.8 \pm 121.9
Activity edge (Hz)	14 \pm 3	13 \pm 5	15 \pm 5	17 \pm 4

* The combination of θ and α power and number of waves was significantly different from dogs not receiving midazolam by multiple ANOVA, $P < 0.05$.

† Significantly different from dogs not receiving midazolam as determined by one-way ANOVA, $P < 0.05$.

flumazenil decreased α and total hemispheric power by quantitative analysis but not on the visual display. These decreases were less consistent than at normal CSF pressure and attained statistical significance only at 6, 9, and 10 min postflumazenil for α power and at 5–7, 9, and 10 min postflumazenil for total power.

Raising the distal end of the cisternal outflow tubing by 10 or 30 cm increased CSF pressure by 10 or 30 cmH₂O, respectively. Systemic values did not differ between any experimental conditions at normal or elevated CSF pressure within each group and were therefore combined (table 4). Combined systemic values did not differ between groups.

Discussion

The results of this study indicate that flumazenil does not alter \dot{V}_f , whether it is given at normal or increased CSF pressure. Following administration of flumazenil 0.0025 or 0.16 mg/kg, \dot{V}_f was unchanged both in dogs receiving midazolam and in dogs not receiving midazolam. In contrast, flumazenil did produce some changes in R_a . These were inconsistent, with statistical increases occurring following two of the four flumazenil doses in dogs receiving midazolam and a decrease occurring following one of the four doses in dogs not receiving midazolam. It should be noted that with one of the two increases in R_a (at increased CSF pressure in dogs receiving midazolam), the value was greater than the control value for that group but was not different from the control value (at increased CSF pressure) for the group not receiving midazolam. With the decrease in R_a (at normal CSF pressure in dogs not receiving midazolam), the value was less than

the control value for that group but was not different from the control value (at normal CSF pressure) for the group receiving midazolam.

The results of this study also indicate that flumazenil produced changes in EEG activity in dogs receiving midazolam but not in dogs not receiving midazolam. The visual display of these EEG changes on the present aperiodic analysis program were similar to those previously reported by Fleischer *et al.*¹ on the visual display of a power spectral analysis program. In the previous study flumazenil 1.0 mg/kg produced a dramatic decrease in amplitude and an increase in frequency to primarily β activity, with EEG activity returning to a preflumazenil pattern over the next 15 min. In the present study the visual data were similar to those of the previous study. Quantitative analysis of the EEG indicated that flumazenil-induced EEG changes were comprised of decrease in power but not number of waves or activity edge in dogs receiving midazolam at normal CSF pressure. There were also significant EEG changes in dogs receiving midazolam at increased CSF pressure, although these occurred in fewer frequency bins and at fewer time intervals following flumazenil than when flumazenil was administered at normal CSF pressure.

In the present study certain findings support the suitability of the methodology and experimental design for the purposes of this study. Control values for \dot{V}_f were similar to previously reported control values for dogs not receiving midazolam^{13,14} and dogs receiving midazolam.¹² Values for \dot{V}_f did not change when CSF pressure was increased by 10 cmH₂O, consistent with previous reports that small increases in CSF pressure do not alter \dot{V}_f .¹⁶ Values for \dot{V}_f also did not change when CSF pressure was

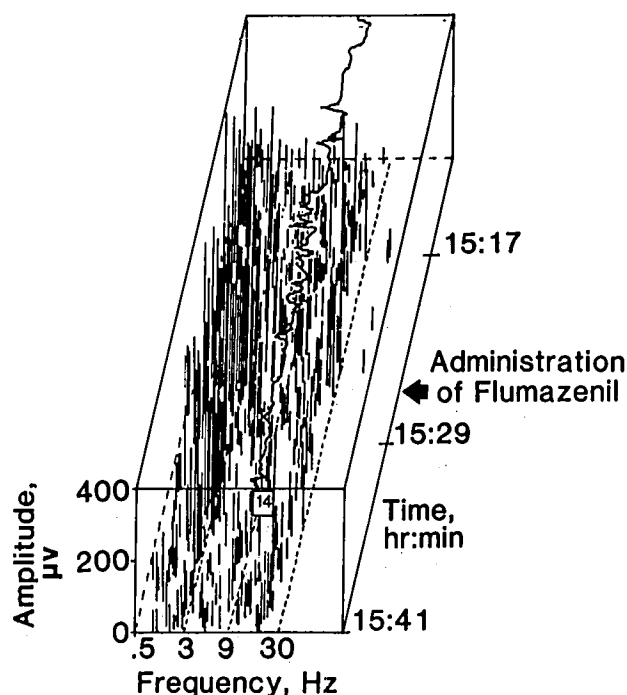


FIG. 1. Thirty minutes of aperiodic analysis of the EEG is shown for a dog receiving midazolam ($1.25 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$) and halothane (0.4% end-expired) at normal CSF pressure. On this tracing EEG frequency (0.5–30 Hz) is displayed on the horizontal axis, EEG amplitude (0–400 μV) is displayed on the vertical axis, and actual time (h:min) is displayed on the diagonal axis with past events at the “back” of the tracing and current events at the “front” of the tracing. The vertical lines within this three-axis “box” represent the amplitude of EEG activity at each frequency and time period. The line on the top of the box and the numerical value in the attached small square represent the activity edge. Prior to administration of flumazenil (above the arrow) EEG activity is present across the entire frequency distribution. At the arrow flumazenil 0.16 mg/kg was given intravenously over 60 s. The first 10 min following administration of flumazenil (below the arrow) shows a decrease of θ and α EEG activity. Thereafter, the pattern of EEG activity returns toward the preflumazenil tracing.

increased to 46–48 cmH_2O , consistent with previous reports that \dot{V}_f does not decrease as long as cerebral perfusion pressure remains greater than $\sim 70 \text{ mmHg}$.^{17,18} Control values for R_a were similar to previously reported control values at normal CSF pressure both in dogs not receiving midazolam^{13,14} and in dogs receiving midazolam.¹² R_a decreased when CSF pressure was increased to 36–38 cmH_2O , consistent with previous reports that R_a is relatively stable at CSF pressure $< 25\text{--}30 \text{ cmH}_2\text{O}$ and decreases at CSF pressures $> 25\text{--}30 \text{ cmH}_2\text{O}$.^{3,19,20} The EEG of dogs receiving midazolam was comprised of less high frequency activity than in dogs not receiving midazolam, as previously reported.¹² Increase of CSF pressure to 36–38 cmH_2O produced no significant change in EEG power, number of waves, or the activity edge, in accordance with reports that EEG activity does not change sig-

nificantly as long as cerebral perfusion pressure remains $> 30\text{--}40 \text{ mmHg}$.²¹ Values in dogs examined first at increased CSF pressure and then at normal CSF pressure were not different from values examined first at normal CSF pressure and then at increased CSF pressure, indicating that the magnitude of increase and technique for increase of CSF pressure in this study did not produce irreversible changes in CSF dynamics, the EEG, or systemic values. When CSF pressure was increased to approximately 21, 37, and 47 cmH_2O , the increase of CSF pressure equalled the distance in centimeters by which the distal end of the cisternal outflow tubing was raised, indicating that the integrity of the perfusion system was intact.

The results of the present study indicate that flumazenil exerts little direct action on \dot{V}_f , R_a , or EEG activity in the absence of a preexisting benzodiazepine effect. In dogs receiving midazolam, both doses of flumazenil produced inconsistent increases in R_a and the larger dose reversed the increase of θ and α EEG activity seen with midazolam. The potential clinical relevance of the present findings relates to the effects of flumazenil on both CSF pressure and accommodation by CSF volume to changes in intracranial volume when flumazenil is used to reverse ben-

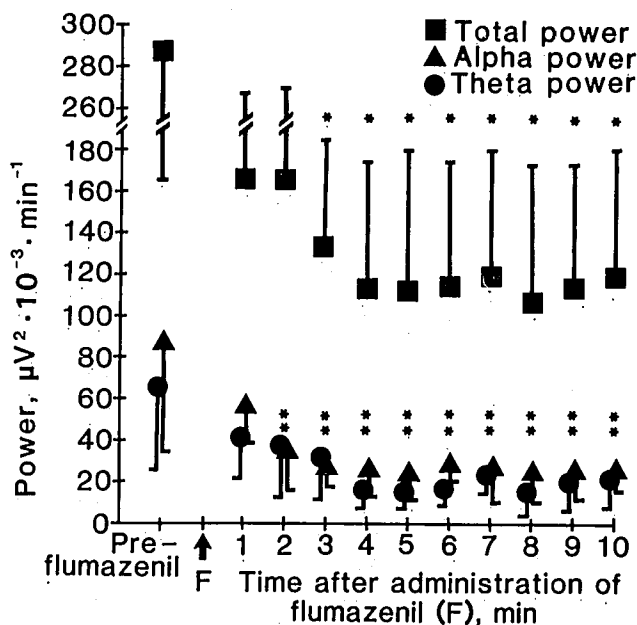


FIG. 2. Flumazenil 0.16 mg/kg decreased the activity of the EEG in the group of dogs receiving midazolam ($1.25 \text{ mg} \cdot \text{kg}^{-1} \cdot \text{h}^{-1}$) and halothane (0.4% end-expired) at normal CSF pressure. Preflumazenil, administration of flumazenil, and the time after administration of flumazenil (minutes) is displayed on the horizontal axis and EEG power ($\mu\text{V}^2 \cdot 10^{-3} \cdot \text{min}^{-1}$) is displayed on the vertical axis. F represents the time of administration of flumazenil, squares represent total EEG power, triangles represent power of the α frequencies, circles represent power of the θ frequencies, vertical bars represent SD, and asterisks mark values that differ significantly from preflumazenil values.

TABLE 4. Systemic Values* and CSF Osmolality (mean \pm SD)

	Dogs Receiving Midazolam (n = 6)		Dogs Not Receiving Midazolam (n = 6)	
	Normal CSF Pressure	Increased CSF Pressure	Normal CSF Pressure	Increased CSF Pressure
CSF osmolality (mOsm/kg)	318 \pm 8	—	316 \pm 15	—
P _{aCO₂} (mmHg)	38 \pm 2	38 \pm 4	36 \pm 2	37 \pm 2
pH	7.32 \pm 0.02	7.32 \pm 0.03	7.32 \pm 0.02	7.32 \pm 0.01
Bicarbonate (mEq/l)	19.0 \pm 1.0	18.8 \pm 1.4	18.6 \pm 1.2	18.8 \pm 1.4
P _{aO₂} (mmHg)	137 \pm 11	136 \pm 8	140 \pm 12	142 \pm 16
Hemoglobin (g/dl)	14.6 \pm 1.9	14.5 \pm 1.5	14.8 \pm 1.8	15.1 \pm 1.2
Heart rate (beats/min)	111 \pm 17	109 \pm 14	111 \pm 11	114 \pm 15
Temperature, nasopharyngeal ($^{\circ}$ C)	37.2 \pm 0.4	37.2 \pm 0.4	37.4 \pm 0.4	37.3 \pm 0.4

* Values did not differ between experimental conditions and were therefore combined.

zodiazepine effects. Both CSF pressure and contraction of CSF volume are regulated by \dot{V}_f and R_a . Because flumazenil produces no change in \dot{V}_f and inconsistent increases in R_a , CSF pressure will rise when increased R_a causes \dot{V}_a to decrease relative to \dot{V}_f . In addition, when R_a is increased, the magnitude of contraction of CSF volume (in response to increase of cerebral blood volume or brain tissue volume) decreases, thereby augmenting the effect of increase of cerebral blood volume or brain tissue volume on CSF pressure. Because the increase in R_a produced by flumazenil is inconsistent and relatively small, it is not likely to be clinically important.

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