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Propofol Modification of Evoked Hippocampal Dentate Inhibition in Urethane-anesthetized Rats

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To learn more about the site and mode of action of propofol, the authors anesthetized rats with urethane, and implanted a stimulating electrode in the perforant pathway and a recording electrode in the hippocampal dentate gyrus to measure evoked field responses. Catheters were placed in the jugular vein and carotid artery to measure blood pressure, take blood samples for measurement of blood gases, and inject drugs. Rat lungs were ventilated, and increasing doses of propofol or its 10% fat emulsion vehicle were administered. Over a dose range of 2.5–40 mg/kg, propofol had little effect on the field excitatory postsynaptic potential (EPSP) or the field population spike (PS), suggesting no important actions on perforant pathway fibers or intrinsic granule cell excitability. With the use of various paired-pulse stimulation paradigms, it was demonstrated that propofol administration led to a dramatic decrease in granule cell excitability over interpulse intervals of 10–100 ms. The magnitude of this effect clearly was related to dose, and the effect was reversible with discontinuance of drug. The vehicle had marginal effects on granule cell excitability. These were significantly less than those produced by propofol. Transient reductions in blood pressure followed by transient increases to above baseline were observed after administration of propofol at most doses. Increased heart rates accompanied the slight increase in blood pressure. Except at the largest dose of propofol tested, cardiovascular measures had returned to near control levels by 8 min, when the evoked potentials were evaluated. Arterial blood gases showed little change throughout the study. The action of propofol on dentate gyrus evoked responses is very similar to that produced by drugs known to enhance GABA-mediated inhibition. Although indirect, these data suggest that propofol also may produce a significant part of its anesthetic actions by augmenting GABA-mediated inhibition in the central nervous system. (Key words: Anesthetics, intravenous: propofol. Brain, hippocampus: evoked potentials; GABA-mediated inhibition; perforant pathway; recurrent inhibition.)

LITTLE IS KNOWN about the mechanism(s) of action of the new intravenous anesthetic agent propofol (2,6-diisopropylphenol, ICI 35858). As outlined in a recent review on propofol,[1] few neurophysiologic or biochemical studies in animals have been performed to date. In this article, we evaluate the effects of propofol and its fat-emulsion vehicle (Intralipid®) on field responses evoked in the dentate gyrus of the hippocampus of the rat by stimulation of the perforant pathway. This approach has been used widely to assess drug effects on neuronal excitability and on stimulus-evoked inhibition and excitation *in vivo*.

The dentate gyrus in the rat is a highly laminated limbic structure with well-defined afferents, efferents, and neurotransmitters.[2–4] It yields evoked field potentials to stimulation of the perforant pathway that are monosynaptic and mediated by glutamate. These field potentials have been studied extensively, and their components have been well characterized[5–8] (see fig. 1).

The amplitude of the evoked field population spike (PS) reflects the number and synchrony of granule cell discharges in the dentate gyrus.[5,9] Changes in the field excitatory postsynaptic potential (EPSP) amplitude or slope reflect changes in the number of perforant pathway fibers activated by the stimulus or changes in the efficacy of the synaptic process through altered transmitter release or receptor properties.[7,9] Stimulation of the perforant pathway with the use of a classic paired-pulse paradigm leads to a series of changes in granule cell response to the second pulse. The source of these changes in response to the second stimulation depend on the paired-pulse interval. At short interpulse intervals (10–40 ms), granule cell responsiveness to the second stimulus is decreased because of both feed forward and recurrent collateral activation of interneurons subserving γ-aminobutyric acid ($GABA_A$) –mediated inhibition[10–16] (period of early inhibition). At intermediate intervals (40–200 ms), granule cell responsiveness to the second stimulus is enhanced (period of facilitation), whereas at longer paired-pulse intervals (400–1,000 ms) granule cell responsiveness is depressed again (period of late inhibition). The mechanisms responsible for these latter phases are more complex, involve both presynaptic and postsynaptic components, and have been discussed previously.[17]

The drug-induced changes in these field potentials after single or paired stimulation have been analyzed for many drugs, particularly drugs affecting $GABA_A$-mediated inhibition. Drugs enhancing $GABA_A$-mediated inhibition prolong the period of early inhibition,[14,18–22] whereas drugs antagonizing GABA reduce it.[14,17] Drugs that do not affect the GABA system usually produce little or no effect on this measure. These background data provide valuable assistance in interpreting changes produced by propofol in these potentials.[23] In this article, we report that propofol affects dentate field potentials in a manner

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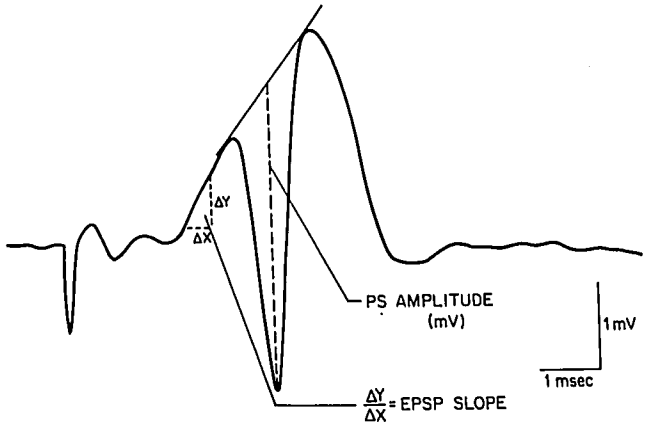

FIG. 1. Measurement of dentate gyrus field response components. Response recorded from the dentate gyrus after perforant pathway stimulation showing stimulus artifact (leftmost deflection) followed by a small fiber response and a large excitatory postsynaptic potential (EPSP) (positive, upward wave) and population spike (PS) (downward, negative wave). EPSP amplitudes were determined as the maximum rate of rise of the EPSP between its onset and the onset of the PS. PS amplitudes were measured from the extrapolated EPSP baseline to the peak of the PS waveform. EPSP and PS thresholds were defined as the lowest stimulus intensities producing an identifiable response. Latencies were measured from the stimulus artifact to the point of onset of the EPSP or PS.

characteristic of drugs that enhance $GABA_A$-mediated inhibition. This may be an important mechanism by which propofol acts in the central nervous system.

Materials and Methods

Subjects and Surgical Preparation

This study was approved by the University of California, Davis, animal studies committee. Eight male Sprague-Dawley rats (250–350 g) were anesthetized with intraperitoneal urethane (1.2 g/kg). Under a dissecting microscope, PE-50 polyethylene catheters containing heparinized (10 units/ml) saline were inserted into the right jugular vein and carotid artery. Through the same incision, a 16-G Teflon intravenous catheter trimmed to 1.5 cm length was placed into the trachea and secured with ties before the wound was closed. The animals were placed into a stereotaxic device (incisor 5 mm above the intraaural line). A Propaq® ultraportable vital signs monitor (model 106) was used to monitor constant rectal body-temperature, electrocardiogram (lead II), and blood pressure (COBE® disposable pressure transducer) throughout the experiment. An Edco rodent ventilator (model 802) was used to give tidal volume in milliliters equal to 0.0075 times the body weight in grams.[24] The respiratory rate was fixed at approximately 120 breaths per min. A Harvard pump (model 22) was used to infuse drugs.

The skull was exposed, and 1-mm-diameter holes were drilled for electrode placement. The concentric stimulating electrode consisted of a tip made of sharpened 34-G nichrome wire and a barrel made of 21-G stainless-steel tubing with a tip-to-hub spacing of 1 mm. The stimulating electrode was placed into the right medial entorhinal cortex (near the angular bundle), using coordinates of 7.0 mm posterior to bregma, 4.1 mm lateral to midline, and 3.2 mm below the cortical surface. The recording electrode, a tungsten microelectrode with tip resistance of 1–2 Mohms, was lowered into the dentate gyrus of the hippocampus using surface coordinates of 2.0 mm posterior to bregma and 1.6 mm lateral to the midline. The final placement depth of the recording electrode was made while the perforant pathway input was stimulated to obtain the largest recordable dentate response (approximately 3.5 mm below the surface of the exposed cortex). A stainless-steel screw electrode was placed over the frontal sinus and served as a ground. Body temperature was kept at 38 ± 1° C with constant-temperature heating pads and wraps.

Recording and Stimulation

Stimuli were square-wave pulses formed by a WPI model 830 stimulator and delivered through a model 850 isolation unit. All stimuli consisted of pulse pairs separated by differing interpulse intervals. Pulses were 0.05 ms in duration and varied in constant current, depending on the experiment. Repetition frequency was always 0.16 Hz.

Dentate responses were amplified using Grass model 7P511 amplifiers with low- and high-frequency cutoffs at 1 Hz and 10 kHz. Responses were displayed on a Nicolet 3091 oscilloscope. For analysis, responses were digitized with the use of a Keithly DAS Series 500 analog-to-digital converter coupled to an IBM PC XT computer. The conversion rate used was 10 kHz.

Analysis of Response

For data analysis, responses always were gathered in groups of four, averaged, and stored as a single record. The analysis of waveforms closely followed those we have described.[15,17,18,25] EPSP amplitudes and PS amplitudes were determined (fig. 1). EPSP amplitudes were determined as the maximum rate of increase of the EPSP between its onset and the onset of the PS. PS amplitudes were measured from the extrapolated EPSP baseline to the peak of the PS waveform. Thresholds were defined as the lowest stimulus intensities producing a visible response on the oscilloscope.

Experimental Procedures

After final placement of electrodes, subjects were given at least 60 min for stabilization of responses. Data were

collected continuously during this time to assure that responses had stabilized. Then each subject underwent six equivalent data collection periods. These included a control and five periods in which propofol or its vehicle was administered in increasing amounts. Each period lasted for 25 min. Drug administration consisted of a bolus injection of propofol during the first minute, followed by a constant infusion for the remainder of the period. The doses of propofol included the following: 1) a 2.5 mg/kg bolus, 2.5 $mg \cdot kg^{-1} \cdot h^{-1}$ infusion; 2) additional 2.5 mg/kg bolus, 5 $mg \cdot kg^{-1} \cdot h^{-1}$ infusion; 3) additional 5 mg/kg bolus, 10 $mg \cdot kg^{-1} \cdot h^{-1}$ infusion; 4) additional 10 mg/kg bolus, 20 $mg \cdot kg^{-1} \cdot h^{-1}$ infusion; and 5) additional 20 mg/kg bolus, 40 $mg \cdot kg^{-1} \cdot h^{-1}$ infusion. Vehicle control dosing with Intralipid used the same volumes and rates as with propofol. The drug doses listed in the Results section represent the cumulative bolus dose given at the time of data collection.

Blood pressure and heart rate were monitored continuously throughout the experiment. For quantification, measurements were taken just before the bolus administrations of propofol or vehicle and each minute thereafter. Three arterial blood samples were taken from three rats in each group to measure blood gases. The first sample (0.2 ml) was withdrawn after control data collection and before the first propofol or vehicle dose. The second sample was withdrawn at the end of the third dosing period of propofol or vehicle, and the third sample was obtained after the fifth dosing period. An equal amount of normal saline was given intraarterially after the collection of each sample.

A number of different approaches were taken to evaluate the effects of propofol on dentate gyrus granule cells, which were necessitated by the fact that the intensity chosen for perforant pathway stimulation has proved to be a significant variable itself in previous drug studies.[17,25] The following electrophysiologic measurements were made, starting 8 min after the bolus injection for each period: 1) determining the EPSP and PS thresholds; 2) recording a series of responses (n = 8) to pairs of perforant path stimuli (interpulse interval = 20 ms) using stimulus intensities from just over threshold to that producing a maximal response (fig. 2); 3) recording a similar series of responses at an interpulse interval of 60 ms; and 4) recording pairs of responses at a fixed stimulus intensity (that producing a PS 80% of maximal) at interpulse intervals of 10, 15, 20, 25, 30, 40, 60, 100, 200, 400, and 1,000 ms (fig. 3). Paired responses were computer subtraction corrected when they overlapped in time.[25] Stimulus intensities for measurement 4 were adjusted, if needed, to maintain the size of the response equal to that recorded during the control period. These responses have been shown to remain stable over the time periods used in this study.[15,17–19,25] The time intervals chosen for measurements 2 and 3 were selected because, in the control situation, 20 ms is a time point at which early inhibition can be measured conveniently, whereas by 60 ms it has terminated effectively.

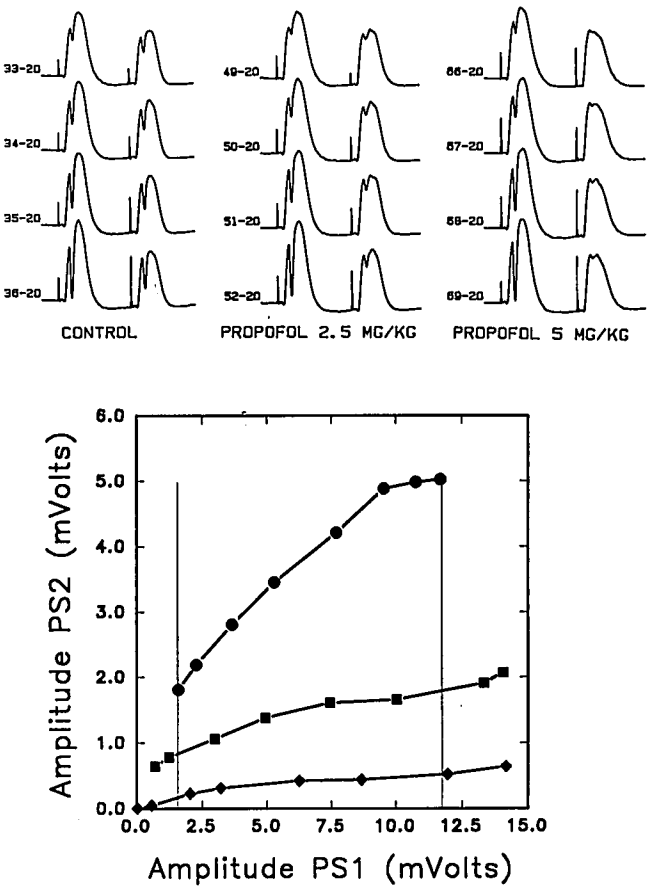

FIG. 2. Quantitative evaluations of changes in granule cell excitability. *Top:* Twelve pairs of stimuli using an interpulse interval of 20 ms and intensities ranging from threshold to maximal are applied to the perforant pathway. Responses to four pairs of stimuli are shown for control data (*left*), after 2.5 mg/kg propofol (*center*), and after 5 mg/kg propofol (*right*). The number before each of the response pairs gives the sample number and the interpulse interval. EPSP and PS amplitudes and latencies are determined for each first response. These then are averaged to provide the data in table 1. To quantitate stimulus evoked changes in excitability, the first (PS1) and second PS (PS2) amplitudes are measured and plotted as shown at the bottom. *Bottom:* Curves show PS2 amplitudes as a function of PS1 amplitudes for all eight responses in each series. Where PS1 amplitudes overlap between series (perpendicular lines) the area of PS1 × PS2 is determined. Changes in area reflect changes in excitability of granule cells to equivalent ranges of perforant pathway input.

Data Analysis

Data from each period were quantified in the following manner. Threshold data from measurement 1 were recorded as μA of current, and all values within an experiment were normalized to the value obtained during the

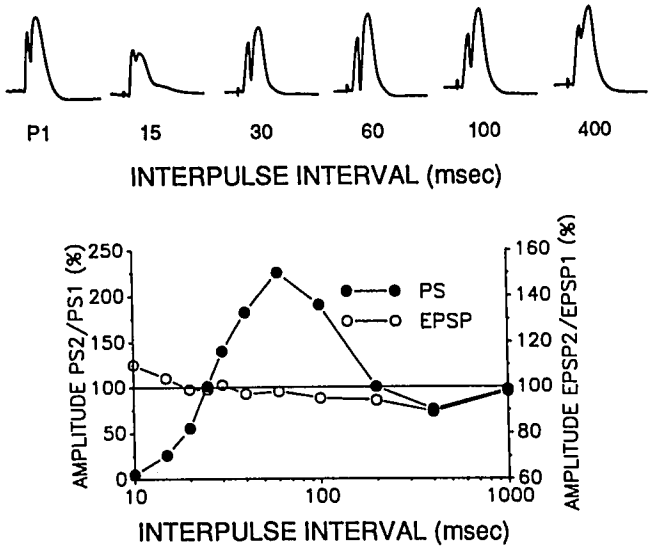

FIG. 3. Excitability curves generated for the amplitude components of the dentate gyrus field response using paired pulse stimulation of the perforant pathway. *Top:* Representative responses produced by the first stimulus (P1) and by the second stimulus at various interpulse intervals. *Bottom:* Curves generated for the EPSP and PS components of the response over a range of interpulse intervals. To avoid possible confounding effects of conductance changes induced in granule cells by their discharge, EPSP amplitudes (slopes) were measured using stimuli below threshold for evoking a PS. PS amplitudes were measured using stimuli producing a PS of 80% of maximal. Amplitude axis for the PS (*left*) and EPSP (*right*) have been adjusted so as to represent approximately equivalent degrees of depression or facilitation, so that stimulus-evoked changes in EPSPs under control conditions would be expected to produce the indicated changes in PS amplitudes. When the perforant pathway is stimulated at an intensity sufficient to produce granule cell discharge and a PS, a triphasic sequence of changes in granule cell responsiveness occurs over the next 10–1,000 ms. Both presynaptic and postsynaptic mechanisms can be involved[9,30,*] (see reference 17 for a discussion of mechanisms). As this figure indicates, in our preparations the EPSP shows some early facilitation during paired-pulse stimulation and a late depression between 100 and 1,000 ms. These clearly are not responsible for the changes observed in PS amplitudes through the first 200 ms, and we must assume that postsynaptic changes are more important.

control period. EPSP and PS amplitudes and latencies were obtained from the first responses of the pairs recorded in measurement 3 and averaged (fig. 2). The data from measurements 2 and 3 also were used to generate graphs of the amplitude of the second response as a function of the first for all stimulus pairs (fig. 2). Changes in the areas under these curves provide a measure of the relative excitability of the granule cell population at the time intervals of 20 ms (measurement 2) and 60 ms (measurement 3) after perforant pathway stimulation. Increased areas indicate increased excitability and decreased areas indicate decreased excitability. Data from measurement 4 were used to generate graphs of changes in the ratio of PS2/PS1 amplitudes (fig. 3) as a function of interpulse interval over the range of 10–1,000 ms. These provide additional information regarding drug-induced changes in excitability. As a third measure of changes in excitability, we determined the time required for granule cell excitability (as measured by the second PS amplitude) to return to 75% of the first PS amplitude, using the data from measurement 4. The 75% point was chosen rather than 100% because, after certain treatments, 100% recovery did not occur over the 1,000-ms interval tested.

DRUGS

Urethane was purchased from Sigma Chemical Company. Fat emulsion (Intralipid) was a 10% solution purchased from KabiVitrum, Inc. It contained soybean oil (100 mg/ml), phospholipids (from powdered egg yolks, 12 mg/ml), and glycerin (USP, 22.5 mg/ml). Propofol (10 mg/ml; Diprivan®) was purchased from Stuart Pharmaceuticals (ICI Americas), Inc. In addition to propofol, it contained soybean oil (100 mg/ml), glycerol (22.5 mg/ml), egg lecithin (12 mg/ml), and sodium hydroxide to adjust *p*H.

Results

EFFECTS OF PROPOFOL ON EVOKED DENTATE GYRUS POTENTIALS

The effects of propofol and the vehicle on EPSP and PS responses are given in table 1. The vehicle had practically no effect. The only significant change was a modest increase in PS threshold after the last dose. Propofol produced dose-related increases in EPSP thresholds, PS thresholds, and the latency to onset of the PS. The magnitude of the changes was generally less than 10%. No effects were found on EPSP slopes or PS amplitudes at any dose.

EFFECTS ON PAIRED-PULSE PARADIGMS

Examples of the effects of propofol on granule cell responses using a paired-pulse interval of 20 ms are shown in the top portion of figure 2. Although first-response amplitudes were changed little after propofol administration, second-response amplitudes were reduced dramatically. The effects of propofol and its vehicle on excitability, as measured at 20 ms and 60 ms by changes in areas generated from graphing the amplitude of the first PS as a function of the second PS, are seen in figure 4. The vehicle control significantly reduced excitability at 20 ms and 60 ms. Excitability was reduced by 20–30% at the 20-ms time point at doses of 0.5 ml/kg and greater. Smaller effects were observed at 60 ms. Propofol produced much greater reductions in excitability at both time points. The second PS amplitudes at 20 ms were reduced by 90% or more at propofol doses of 5 mg/kg and greater.

TABLE 1. Effect of Drug Administration on Dentate Gyrus Responses

Drugs and Dose	EPSP		PS		
	Threshold (%)	Amplitude (%)	Threshold (%)	Amplitude (%)	Latency to PS (%)
Intralipid (ml/kg) (n = 4)					
Control	16.8 ± 0.5 μamp	5.2 ± 0.3 mv/ms	33.0 ± 4.9 μamp	3.2 ± 0.7 mV	3.1 ± 0.2 ms
0.25	104.4 ± 2.4	98.4 ± 1.3	100.6 ± 1.2	84.6 ± 5.4	98.3 ± 0.3
0.50	103.0 ± 2.6	95.1 ± 2.2	103.1 ± 2.7	92.1 ± 12.3	98.4 ± 0.4
1.0	106.0 ± 2.1	98.8 ± 2.9	104.1 ± 2.1	86.3 ± 10.2	99.0 ± 0.5
2.0	106.1 ± 2.1	93.7 ± 3.3	105.5 ± 2.2	79.2 ± 6.0	98.6 ± 1.1
4.0	107.8 ± 2.6	93.9 ± 4.7	106.2 ± 2.1*	75.8 ± 7.8	99.3 ± 1.2
	F(5,15) = 2.25, NS	F(5,15) = 1.24, NS	F(5,15) = 3.43, $P \leq 0.05$	F(5,15) = 2.82, NS	F(5,15) = 0.64, NS
Propofol (mg/kg) (n = 4)					
Control	16.3 ± 1.0 μamp	5.4 ± 0.8 mV/ms	26.0 ± 3.7 μamp	4.6 ± 0.4 mV	3.2 ± .2 ms
2.5	104.9 ± 1.4	101.2 ± 2.2	102.9 ± 2.5	84.7 ± 4.6	98.7 ± 0.9
5.0	106.9 ± 3.5	101.2 ± 4.3	104.0 ± 2.2	77.6 ± 4.2	98.5 ± 1.1
10	107.9 ± 1.6*	105.4 ± 3.4	107.6 ± 2.6	82.1 ± 6.1	98.9 ± 1.7
20	107.9 ± 1.6*	108.2 ± 6.3	111.5 ± 2.3†	88.5 ± 6.7	101.6 ± 2.3
40	109.7 ± 2.1†	104.4 ± 8.0	113.7 ± 2.2†	98.7 ± 11.8	107.1 ± 2.9†
	F(5,15) = 4.96, $P \leq 0.01$	F(5,15) = 0.76, NS	F(5,15) = 10.69, $P \leq 0.01$	F(5,15) = 2.86, NS	F(5,15) = 6.52, $P \leq 0.01$

Data, except where noted by specific units, are the means of the percent of control trial ± SEM; dose reflects constant infusion.
EPSP = field excititatory postsnyaptic potential; PS = field population spike; NS = not significant.
* $P \leq 0.05$.
† $P \leq 0.01$.

Responses at 60 ms were less affected, but doses greater than 10 mg/kg reduced the amplitude of the second by more than 70%. The reduction in excitability was significantly greater after administration of propofol than after administration of Intralipid alone (at 20 ms, F[1,6] = 731.7, $P \leq 0.001$; at 60 ms, F[1,6] = 40.9, $P \leq 0.01$).

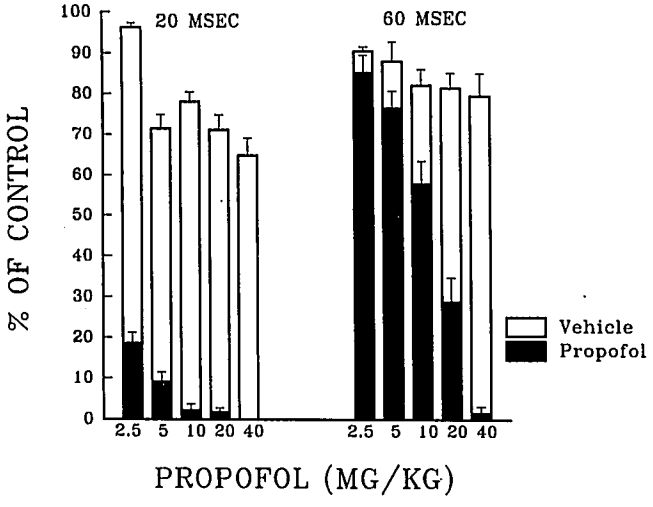

FIG. 4. Effects of propofol and vehicle control on the excitability of dentate gyrus granule cells 20 and 60 ms after perforant pathway stimulation. Open bars indicate effects of vehicle, and filled bars the effects of propofol. Excitability is depicted as percent of control excitability. Data are means ± SEM; n = 4. Significant dose effects were seen for both compounds at 20 and 60 ms. For propofol at 20 ms, F(5,15) = 15.4 ($P < 0.001$) and at 60 ms, F(5,15) = 9.3 ($P < 0.01$). For Intralipid at 20 ms, F(5,15) = 7.7 ($P < 0.01$) and at 60 ms F(5,15) = 3.9 ($P < 0.05$). At both time points propofol was significantly more effective in reducing excitability than was the vehicle (see text for statistical values).

Effects on excitability using the fixed-amplitude, variable-interval approach (measurement 4) are shown in figure 5 . At the highest dose, the vehicle caused a marginal reduction in excitability but did not significantly affect the duration of the early period of inhibition. In contrast, propofol produced a definite increase in both the intensity and the duration of this phase. At the higher doses of propofol, significant changes could be detected to 100 ms.

Figure 6 shows the effects of these drugs on the time required for the second PS amplitude to recover to 75% of the first PS amplitude with paired stimulation of the perforant pathway. This measure was not affected by Intralipid at any dose. Propofol produced dose-related changes that became significant at doses of 20 mg/kg and greater.

Two of the four rats given propofol were evaluated for 60 min after the infusion was discontinued. A partial reversal of the neurophysiologic effects was observed in both rats.

EFFECTS ON BLOOD PRESSURE, HEART RATE, AND ARTERIAL BLOOD GASES

Because of the relatively large variability in baseline heart rates and blood pressures, subtle changes may not be obvious. With the first and subsequent boluses of pro-

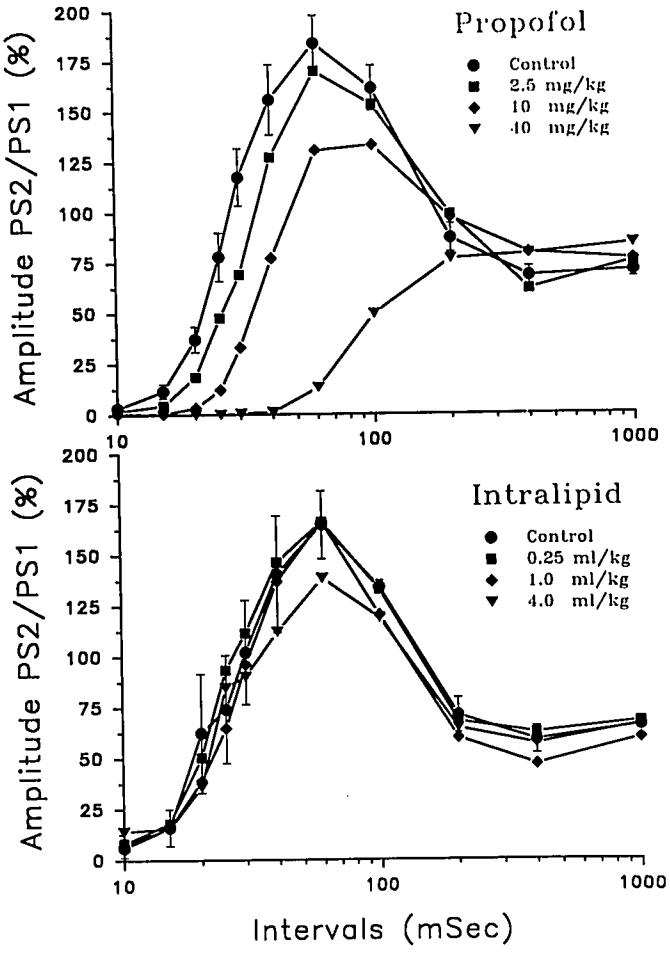

FIG. 5. Effects of propofol and vehicle on granule cell excitability using fixed amplitude stimuli at variable paired-pulse intervals. Data are means ± SEM for control curves and means for others; n = 4. Propofol markedly increased the duration of the early depression of granule cell excitability. The extent of effect was clearly dose-dependent. Marginal changes were observed after administration of the vehicle. For clarity, only selected doses are shown.

pofol, a small decrease (10–15 mmHg) in systolic and diastolic blood pressure tended to occur by the first minute. Except after the last dose of propofol, blood pressure increased, within 3–4 min after administration of the bolus dose, to exceed baseline blood pressures by 10–15 mmHg (fig. 7). The reductions and elevations in blood pressure after propofol administration were associated with heart rate reductions and elevations. The Intralipid bolus appeared to have no major effect on blood pressure or heart rate in this study. Except for the decrease in blood pressure and heart rate seen with the largest dose of propofol tested (20 mg/kg bolus, 40 $mg \cdot kg^{-1} \cdot h^{-1}$ infusion), blood pressures and heart rates were at or near baseline values before neurophysiologic data were collected at 8 min after administration of the bolus dose, despite constant infusions. Table 2 demonstrates that arterial blood gases changed little after administration of bolus and maintenance doses of propofol and Intralipid. A small but statistically significant ($P \leq 0.05$) reduction in arterial oxygen tension (Pa_{O_2}) occurred after administration of the largest dose of propofol (table 2).

After the final data collection and continued constant infusion, propofol-treated animals did not breathe spontaneously after discontinuation of mechanical ventilation, whereas Intralipid-treated animals did breathe spontaneously. In the two of four animals evaluated for an additional 30 min after termination of propofol administration so that recovery could be observed, spontaneous breathing occurred by 15–20 min.

Discussion

These data demonstrate that propofol increases the duration of the early depression in excitability observed in dentate gyrus granule cells after stimulation of the perforant pathway. The effect is dose related and reversible

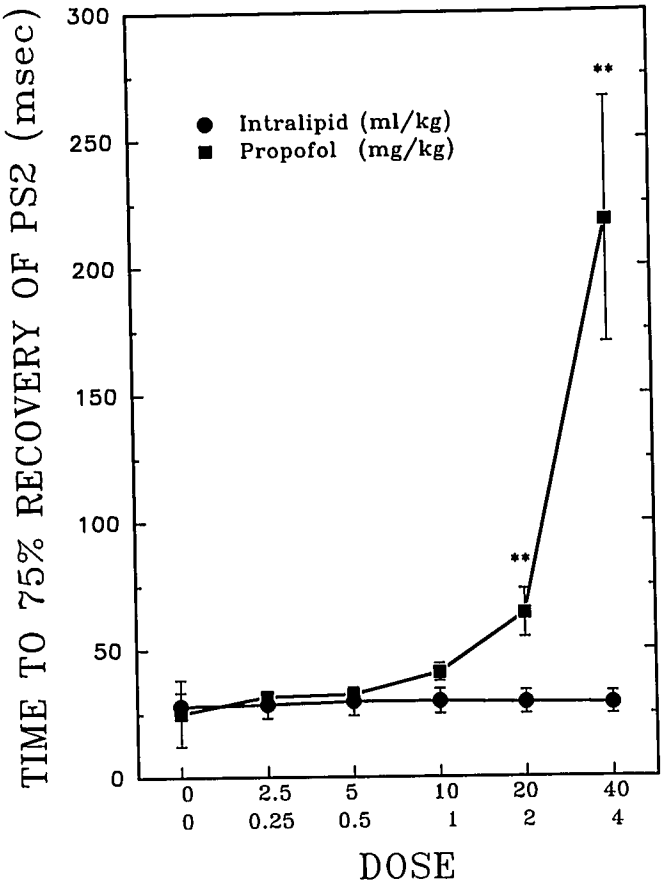

FIG. 6. Time required for PS2 to recover to 75% of PS1 amplitude using a fixed amplitude stimulus is shown for propofol and its vehicle. Data are means ± SEM; n = 4. Propofol increased this measure at the higher dose levels. Its effects were significantly different from those of the vehicle: $F(1,6) = 9.9$ ($P < 0.05$).

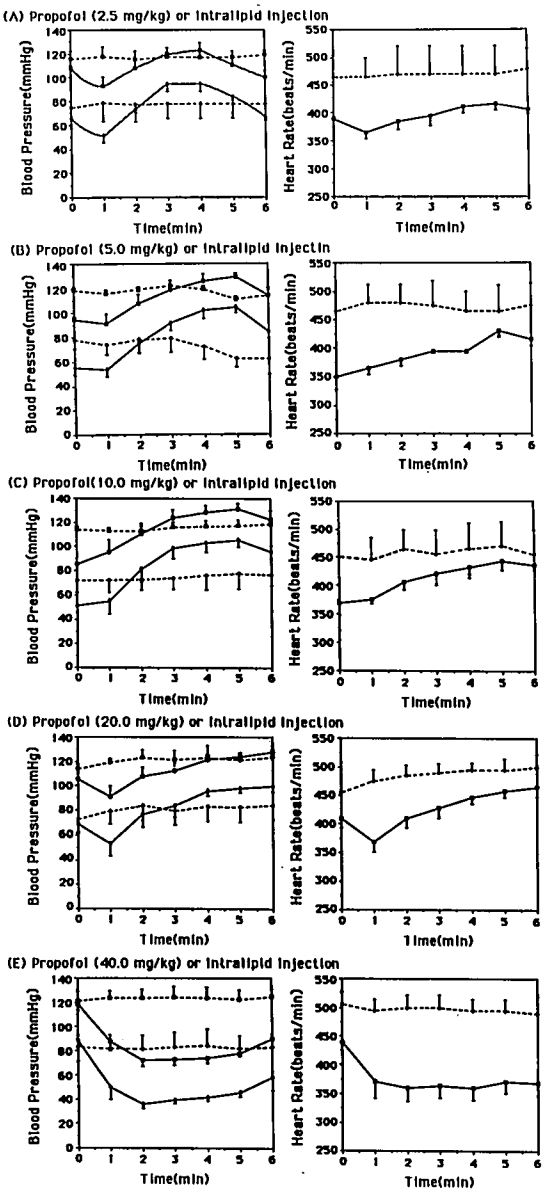

FIG. 7. The effect of propofol (solid line) and its vehicle (dashed line) on blood pressure and heart rate. Data are means ± SEM; n = 4. Except at the highest dose of propofol, heart rates and blood pressures had returned to near baseline levels prior to neurophysiologic testing.

when administration is discontinued. The duration of depression increased sufficiently to alter the period of paired-pulse facilitation (fig. 5), which peaks under control conditions at approximately 60 ms. From the dose-response characteristics of propofol (figs. 4 and 5), it appears that this primarily represents an extension of early inhibition. It is possible, however, that propofol also may reduce paired-pulse facilitation directly. All of these effects occurred without evidence of significant depression of perforant pathway terminals or granule cells. Although the exact mechanisms responsible for this remain to be determined, these effects are similar to those observed with drugs known to enhance $GABA_A$-mediated inhibition, including diazepam[14,19,20,22]; the anesthetic agents pentobarbital,[21,22,26] thiopental, and Althesin (currently under investigation); and SKF100330A,[18] a GABA uptake blocker. Although data are limited, propofol exhibits other actions expected of agents enhancing GABA-mediated inhibition. Propofol provides anticonvulsant actions against electroconvulsive shock and pentylenetetrazol.[23] As is true for benzodiazepines and barbiturates, it affords a greater degree of protection against the $GABA_A$–chloride channel blocker pentylenetetrazol than against electroconvulsive shock.[23]

In these studies, urethane anesthesia was used to enable accurate placement and maintenance of electrodes for quantitative measurements. The use of urethane produces its own effects, and it may have altered the effects of propofol. However, urethane often is used in studies of the hippocampus and appears to affect excitation and inhibition in the hippocampus and cortex to a lesser extent than other anesthetic agents.[27–29] It has been reported that its depressive actions primarily affect small neurons in the reticular formation.[30] The duration of early inhibition, as measured by the paired-pulse technique, is similar in awake and urethanee-anesthetized rats. We previously reported the time to 50% recovery to be 27 ± 5 ms in awake rats (n = 5).§ In this study, controls anesthetized with urethane took 24.6 ± 1.4 ms (n = 4) to reach 50% recovery. Thus, early $GABA_A$-mediated inhibition does not seem to be affected significantly by urethane. Many studies on other agents also have been done under urethane anesthesia, which at least allows comparisons under equivalent conditions.

The serial dosing approach used to provide dose-response data on just a few subjects has an impact on quantitative estimates. Thus, the degree of changes occurring after the second through the fifth cumulative administration is unlikely to be exactly the same as would have occurred if a single higher dose had been used. Nevertheless, the significant finding in this study is the presence of a dose-related, reversible increase in the duration of early inhibition after administration of propofol. With serial dosing, the effect was significant at exposures below, through, and above the rat anesthetic dose range of 10–24 mg/kg.

In the current study, the transient changes in blood pressure and heart rate had resolved before neurophysiologic testing, except after the largest dose of propofol. These findings are consistent with previous reports in hu-

§ Joy RM, Albertson TE: Effects of lindane on excitation and inhibition evoked in the dentate gyrus by perforant path stimulation. Neurobehav Toxicol Teratol 7:1–8, 1985.

TABLE 2. Arterial Blood Gas Data

Treatment	Pa_{O_2} (mmHg)	Sp_{O_2} (%)	Pa_{CO_2} (mmHg)	pH	HCO_3 (mEq)
Control					
Intralipid	109.3 ± 2.2	98.0 ± 0.0	22.7 ± 3.0	7.475 ± 0.026	16.3 ± 1.7
Propofol	118.7 ± 10.2	98.3 ± 0.5	23.7 ± 2.8	7.447 ± 0.038	16.0 ± 0.9
After second dose of propofol (5 mg/kg) or intralipid					
Intralipid	113.0 ± 3.1	98.0 ± 0.0	21.7 ± 2.6	7.477 ± 0.014	16.7 ± 1.4
Propofol	112.7 ± 6.5	98.0 ± 0.5	23.0 ± 2.1	7.470 ± 0.046	16.7 ± 0.7
After fifth dose of propofol (40 mg/kg) or intralipid					
Intralipid	107.0 ± 2.8	98.0 ± 0.0	24.0 ± 2.4	7.467 ± 0.010	17.0 ± 1.3
Propofol	88.7 ± 12.8*	95.3 ± 1.9*	29.7 ± 5.0	7.407 ± 0.064	17.7 ± 0.7

All data are means ± SEM.
Pa_{O_2} = arterial oxygen tension; Sp_{O_2} = oxygen hemoglobin saturation; Pa_{CO_2} = arterial carbon dioxide tension; and HCO_3 = calculated bicarbonate ion.
* $P \leq 0.05$ compared to Intralipid group.

mans and rats[31–34] and cannot be the basis of the neurophysiologic changes. Similarly, the small reduction in Pa_{O_2} seen after the last dose of propofol is unlikely to be responsible for the dramatic increase in inhibition. Propofol can reduce cerebral blood flow, cerebral perfusion pressure, and cerebral glucose utilization (including the hippocampus).[33,35] If any of these events were critical, one would anticipate that basal granule cell excitability, as measured by the first PS amplitude, also would be reduced greatly. The fact that the effects on early inhibition were dose related and reversible argue against the possibility that nonspecific damage of neurons is the basis for this effect.

These data support the recent hypothesis that propofol increases $GABA_A$-mediated inhibition in the dentate gyrus of the hippocampus.[36] This occurs at doses that do not alter either initial neurotransmitter release, as measured by the EPSP slope, or basal granule cell excitability, as measured by PS amplitude. This effect of propofol may be important in its action as an anesthetic agent.

References

1. Sebel PS, Lowdon JD: Propofol: A new intravenous anesthetic. ANESTHESIOLOGY 71:260–277, 1989
2. Cowan WM, Stanfield BB, Kishi K: The development of the dentate gyrus, Current Topics in Developmental Biology. Volume 2. Edited by Hunt RD. New York, Academic Press, 1980, pp 103–157.
3. Fonnum F: Glutamine: A neurotransmitter in mammalian brain. J Neurochem 42:1–11, 1984.
4. Storm-Mathisen J: Localization of transmitter candidates in the brain. The hippocampal formation as a model. Prog Neurobiol 8:19–181, 1977
5. Andersen P, Bliss TVP, Skrede KK: Unit analysis of hippocampal population spikes. Exp Brain Res 13:208–221, 1971
6. Andersen P, Holmquist B, Voorheave PE: Entorhinal activation of dentate granule cells. Acta Physiol Scand 66:448–460, 1966
7. Bliss TVP, Lomo T: Long lasting potentiation of synaptic transmission in the dentate area of the anesthetized rabbit following stimulation of the perforant path. J Physiol (Lond) 232:357–374, 1973
8. McNaughton BL, Barnes CA: Physiological identification and analysis of dentate granule cell responses to stimulation of the medial and lateral perforant pathway in the rat. J Comp Neurol 175:439–454, 1977
9. Lömo T: Patterns of activation in a monosynaptic cortical pathway. The perforant path input to the dentate area of the hippocampal formation. Exp Brain Res 12:18–45, 1971
10. Andersen P, Eccles JC, Loyning V: Pathway of postsynaptic inhibition in the hippocampus. J Neurophysiol 27:608–619, 1964
11. Matthews WO, McCafferty GP, Setler PE: An electrophysiological model of GABA-mediated neurotransmission. Neuropharmacology 20:561–565, 1981
12. Oliver MW, Miller JJ: Alterations of inhibitory processes in dentate gyrus following kindling-induced epilepsy. Exp Brain Res 57: 443–447, 1985
13. Thalman RH, Ayala GF: A late increase in potassium conductance follows synaptic stimulation of granule neurons in the dentate gyrus. Neurosci Lett 29:243–248, 1982
14. Tuff LP, Racine RJ, Adamec R: Effects of kindling on GABA-mediated inhibition in the dentate gyrus of the rat. I. Paired-pulse depression. Brain Res 277:79–90, 1983
15. Joy RM, Albertson TE: Interactions of lindane with synaptically mediated inhibition and facilitation in the dentate gyrus. Neurotoxicology 8:529–542, 1987
16. Peet MJ, Melennan H: Pre and postsynaptic actions of baclofen: Blockade of the late synaptically evoked hyperpolarization of CAI hippocampal neurones. Exp Brain Res 61:567–574, 1986
17. Joy RM, Albertson TE: Convulsant-induced changes in perforant path–dentate gyrus excitability in urethane-anesthetized rats. J Pharmacol Exp Ther 246:887–895, 1988
18. Albertson TE, Joy RM: Increased inhibition in dentate gyrus granule cells following exposure to GABA-uptake blockers. Brain Res 435:283–292, 1987
19. Albertson TE, Joy RM: Modification of evoked hippocampal den-

tate inhibition by diazepam and three antagonists in urethane-anesthetized rats. Exp Neurol 106:142–149, 1989
20. Adamec RE, McNaughton B, Racine R, Livingston KE: Effects of diazepam on hippocampal excitability in the rat: Action in the dentate area. Epilepsia 22:205–215, 1981
21. Tsuchiya T, Fukushima H: Effects of benzodiazepines and pentobarbitone on the GABA-ergic recurrent inhibition of hippocampal neurons. Eur J Pharmacol 48:421–424, 1978
22. Wolf P, Haas HL: Effects of diazepines and barbiturates on hippocampal recurrent inhibition. Naunyn Schmiedebergs Arch Pharmacol 299:211–218, 1977
23. Lowson S, Gent JP, Goodchild CS: Anticonvulsant properties of propofol and thiopentone: Comparison using two tests in laboratory mice. Br J Anaesth 64:110–112, 1990
24. Dejours P: Respiratory characteristics and animal size, Principles of Comparative Respiratory Physiology. Edited by Dejours P. New York, American Elsevier Publishing, 1975, pp 164
25. Joy RM, Albertson TE: Factors responsible for increased excitability of dentate gyrus granule cells during exposure to lindane. Neurotoxicology 8:517–528, 1987
26. Joy RM, Albertson TE: *In vivo* assessment of the importance of GABA in convulsant and anticonvulsant drug action, Neurotransmitters in Epilepsy. Edited by Avanzini G, Engel J Jr, Fariello RG, Heinmann U. New York, Demos Publications, 1991, pp 91–108
27. Pirch JH, Corbus MJ, Ridgon GC: Conditioning-related single unit activity in the frontal cortex of urethanee anesthetized rats. Int J Neurosci 25:263–271, 1985
28. Rogers RC, Butcher LL, Noven D: Effects of urethanee and pentobarbital anesthesia on the demonstration of retrograde and anterograde transport of horseradish peroxidase. Brain Res 187: 197–200, 1980
29. Scholfield CN: Potentiation of inhibition by general anesthetics in neurones of the olfactory cortex *in vitro*. Pflugers Arch 383: 249–255, 1980
30. McClung R, Reilly E, Dafny N: Urethane modification of EEG-like activity and acoustically evoked field potentials recorded from deep nuclei. Appl Neurophysiol 39:11–26, 1976–1977
31. Rocchiccioli C, Saad MAA, Elghozi J-L: Attentuation of the baro receptor reflex by propofol anesthesia in the rat. J Cardiovasc Pharmacol 14:631–635, 1989
32. Bentley GN, Gent JP, Goodchild CS: Vascular effects of propofol: Smooth muscle relaxation in isolated veins and arteries. J Pharm Pharmacol 41:797–798, 1989
33. Dam M, Ori C, Pizzolato G, Ricchieri GL, Pellegrini A, Giron GP, Battistin L: The effects of propofol anesthesia on local cerebral glucose utilization in the rat. ANESTHESIOLOGY 73:499–505, 1990
34. Van Hemelrijck JV, Van Aken H, Plets C, Goffin J, Vermant G: The effects of propofol on intracranial pressure and cerebral pressure in patients with brain tumors. Acta Anaesthesiol Belg 40:95–100, 1989
35. Vandesteene A, Trempont V, Engelman E, Deloof T, Focroul M, Schontens A, DeRood M: Effect of propofol on cerebral blood flow and metabolism in man. Anaesthesia 43(Suppl):42–43, 1988
36. Concas A, Santoro G, Mascia M, Serra M, Sanna E, Biggio G: The general anesthetic propofol enhances the function of γ-aminobutyric acid–coupled chloride channel in the rat cerebral cortex. J Neurochem 55:2135–2138, 1990