Variability of the Respiratory Response to Diazepam

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The authors investigated the respiratory effects of diazepam in 24 healthy volunteers using a modified Read rebreathing circuit. Resting end-tidal CO₂ (PETCO₂) and the slopes of the ventilatory (VE/ PETCO₂) and occlusion pressure (P0.1/PETCO₂) response to CO₂ were measured just prior to and 5, 20, 40, and 60 min after diazepam, 0.1 mg/kg iv. The slope of VE/PETCO₂ for all 24 subjects analyzed as a single group was never significantly depressed. The slope of P0.1/PETCO₂ for all 24 subjects analyzed as a single group was significantly depressed only at 5 min after diazepam. The resting PETCO₂, however, had small but statistically significant increases throughout the 1 h of study. Group or cluster analysis of the slope of P0.1/PETCO₂ clearly divided subjects into one group of five subjects, whose P0.1/PETCO₂ slope was significantly and consistently augmented for 1 h and a second group of 19 subjects whose P0.1/PETCO₂ slope was always less than control for the entire hour. Diazepam may, through effects on pulmonary mechanics and/or the central nervous system, sometimes enhance respiratory responses to CO₂ rebreathing. Failure to select for such group effects when studying drug effects by CO₂ rebreathing may obscure the severity and duration of respiratory depression that occur in the majority of individuals. Resting PETCO₂ indicated consistent depression of resting minute ventilation by diazepam and may be a more appropriate or sensitive measure of mild or subtle drug-induced respiratory effects. (Key words: Anesthetics, intravenous: diazepam. Carbon dioxide: occlusion pressure response; ventilatory response. Ventilation: drug effects.)

DIAZEPAM is a COMMONLY used sedative–hypnotic in anesthetic practice. Although widely reported to produce respiratory depression,1–6 some studies have shown no significant or highly variable respiratory effects from diazepam.7–11 Occasional enhancement of respiratory drive has been noted,9 and it has been suggested that diazepam may counteract meperidine-induced respiratory depression.8 Recently, Bourke et al.11 have postulated that responders to diazepam may be of two distinctly different types. We therefore decided to investigate the possibility that diazepam might enhance respiration in some individuals while depressing it in others.

Materials and Methods

Permission to perform the study was obtained from the Institutional Review Board at the University of Utah Medical Center Hospital. Subjects were healthy volunteers ranging from 20 to 32 yr of age and weighing 50 to 84 kg. Informed consent was obtained, and subjects refrained from alcohol, caffeine, and aspirin consumption for 24 h prior to their participation in the study. No subjects ate for 8 h prior to the study session, and all studies started at 0800 h.

Protocol

Twenty-four subjects (6 women and 18 men) were studied for ½ h before and for 1 h after the administration of diazepam. Upon arrival in the morning, subjects reclined in a dimly lit, quiet area for 15 min. After local infiltration with 0.5 ml of 1% lidocaine, an intravenous cannula was inserted into an antecubital vein and a 0.9% NaCl solution was infused at 100 ml/h.

After blood pressure and heart rate were recorded, an initial CO₂ rebreathing challenge was performed by subjects to familiarize themselves with the test. Subjects wore comfortable head phones emitting white noise to standardize auditory stimulus level and soft nose clips to prevent nasal breathing during each CO₂ challenge. Subjects were instructed to keep their eyes closed during each test session. Fifteen minutes later, resting end-tidal carbon dioxide tension (PETCO₂) was measured during quiet breathing followed by a control CO₂ rebreathing test. Following another 15-min rest period, diazepam (0.1 mg/kg) was given iv. The next resting (PETCO₂) determination and CO₂ challenge was done 5 min after diazepam. Additional PETCO₂'s were measured and CO₂ rebreathing tests were performed at 20, 40, and 60 min in all 24 subjects.

Rebreathing Circuit and Measurement

We used a modified Read rebreathing circuit (fig. 1). The rebreathing apparatus has a 7.5 l neoprene rebreathing bag enclosed in a lucite box, with a Validyne® differential pressure transducer measuring the pressure drop across a Fleisch® pneumotachograph at the outlet of the box to measure ventilatory flow. Flow was directed either into the bag or through the pneumotachograph by a three-way valve located at the mouth of the box, permitting the subject to breathe directly into the room when not rebreathing CO₂. Inspiratory and expiratory limbs of the
circuit were separated by a Collins® J-Valve. A solenoid-operated occlusion valve was located at the proximal end of the inspiratory airway. CO₂ concentration was measured by a Beckman® LB-2 Infrared CO₂ Analyzer, which sampled gas at the mouthpiece at a rate of 200 ml/min and returned it to the central chamber of the Collins® valve. Airway occlusion pressure was measured by a Microswitch® pressure transducer in the central chamber of the Collins® valve. The pressure signal was low-pass-filtered at 30 Hz. Total circuit volume was 9 l. Inspiratory circuit resistance was 1.9 cmH₂O · 1 · s⁻¹. Expiratory circuit resistance was 1.7 cmH₂O · 1 · s⁻¹ and constant between flow rates of 15 and 135 l/min.

Flow, CO₂, and pressure signals were sampled by a microcomputer (Motorola® Exerciser II) 12-bit analog to digital (A/D) convertor (Burr-Brown® MP7208 Data Acquisition System) with resolution of 4.8 mV per A/D unit and range of ±10 V. Occlusion pressure (P₀) measurements were made at the start of every inspiration. The inspiratory occlusion valve was closed 300 ms after the start of expiration. If the shape of the occluded pressure waveform was satisfactory, inspiration pressure was sampled and stored. A signal to reopen the valve was sent 120 ms after the onset of inspiration.

REBREATHING DATA COLLECTION AND ANALYSIS

After allowing the subject to breath quietly through the mouth piece with the nose clip in place, the resting (PETO₂) was recorded and the three-way valve was switched to the rebreathing bag previously filled with 7.8% CO₂ and 92.2% O₂. For each breath, the following data were displayed on the video terminal and stored in memory: Inspiratory time (Tᵢ), breath duration (T_TOB), fractional inspired concentration (%INCO₂) and end-tidal CO₂ concentration (%ETCO₂), tidal volume (VT), minute ventilation (VE), inspiratory mouth occlusion pressure at 100 ms (P₀.1), and elapsed time since start of CO₂ rebreathing. All gas volumes were corrected to BTPS. Subjects were encouraged to rebreathe as long as possible, but could stop at any time; the desired goal was to reach a PETO₂ of 65 mmHg. The change in PETO₂ during rebreathing challenges was always an increase of at least 20 mmHg.

After completion of each CO₂ challenge, plots of VE versus (PETO₂) and P₀.₁ versus PETO₂ were displayed on the video display terminal. To ensure that the regression line reflected only data from the linear portion of ventilatory response, data from the first ten breaths were excluded from analysis. Data from all other breaths were used for least-squares linear regression. The resting PETO₂, the slope of VE versus PETO₂, and the slope of P₀.₁ versus (PETO₂) were saved for later analysis.

STATISTICAL ANALYSIS

Possible group changes in resting PETO₂, slope of VE/PETO₂, and slope of P₀.₁/(PETO₂) at 5, 20, 40, and 60 min after diazepam injection were determined by Hotelling T-square analysis, a form of multivariate analysis of variance for repeated measures. If joint hypotheses were rejected, individual comparisons were performed by Bonferroni adjusted paired t tests. Statistical analyses were performed on the logarithm of the actual value divided by the control value. For purposes of tabular listing, mean values were expressed as per cent of control values. Statistical software was the P1D and P3D programs from the BMDP statistical package.* The null hypothesis was rejected for P < 0.05.

Cluster analysis, a descriptive statistical approach, was used to ascertain whether there were qualitative differences among subjects' responses to diazepam. Cluster analysis can be constrained to construct the two smallest clusters or groups for all points. The $P$ value is a translation of the $F$ value, which describes the ratio of intercluster (intergroup) variance to intracluster (intragroup) variance. The $P$ value denotes the "tightness" of each group and the "separateness" of the groups from each other. The smaller the $P$ value, the greater the chance that two separate groups exist. Results were expressed as per cent of control for clarity and ease of understanding. Statistical software came from the PKM program of BMDP.**

**Results**

Control respiratory measurements were within normal limits for all 24 subjects. The control slopes of the ventilatory and occlusion pressure responses to CO$_2$ were 2.43 ± 0.18 l·min$^{-1}$·mmHg$^{-1}$ and 0.61 ± 0.05 cmH$_2$O/mmHg, respectively (mean ± SEM). The control (PET$_{CO_2}$) was 36.5 ± 0.5 mmHg. Analysis of the slope of the ventilatory response to carbon dioxide rebreathing ($V_E$/PET$_{CO_2}$) for all 24 subjects as a single group for 1 h following diazepam revealed that at no time did diazepam produce statistically significant decreases in the CO$_2$ response. The maximum decrease was to 85% of control ($P$ = 0.0508) at the 5-min test (table 1). Analysis of the slope of the occlusion pressure response to carbon dioxide rebreathing (P0.1/PET$_{CO_2}$) for all 24 subjects revealed that diazepam produced a decrease in respiratory occlusion pressures that reached statistical significance only at 5 min after injection (63% of control; $P$ = 0.0006) (table 1).

Resting end-tidal CO$_2$, on the other hand, was increased by a small but statistically significant degree for the entire hour of study after diazepam injection (table 2).

Cluster analysis of the occlusion pressure responses to CO$_2$ rebreathing clearly divided subjects into one group of five, who showed consistently greater occlusion pressures following diazepam and a second group of 19 whose P0.1/(PET$_{CO_2}$) slope was always less than control (table 3). These differences remained statistically significant throughout the entire study period. No subject crossed over into the other group for that hour. There were no differences between the two groups with respect to gender, age, weight, alcohol consumption, or cigarette smoking.

Cluster analysis of the ventilatory response to carbon dioxide rebreathing also yielded two groups of responders to diazepam. However, the intergroup $P$ value was statistically significant (0.016) only at 20 min after diazepam. In addition, two subjects demonstrated an inconsistent effect from diazepam, an increase of the slope of their $V_E$/PET$_{CO_2}$ after an initial decrease. One of the five subjects who demonstrated increased slopes of the P0.1/(PET$_{CO_2}$) after diazepam showed an increased slope of the ventilatory response to CO$_2$. Cluster analysis of the resting (PET$_{CO_2}$) after diazepam failed to show more than one subject group.

**Discussion**

The results of this study demonstrate that diazepam, as a 0.1 mg/kg iv bolus, did not depress the ventilatory response to CO$_2$ and only briefly depressed the occlusion pressure response to CO$_2$ in healthy young adult volunteers considered as a group. In contrast, individual (cluster) analysis revealed that although a minority of individuals demonstrated an increase in their responses to CO$_2$, the majority of subjects sustained a significant decrease of their ventilatory and occlusion pressure responses to CO$_2$. When these differing effects were taken into account, a decrease in the response to CO$_2$, when it oc-

![Table 2](https://www.anesthesiology.org/doi/pdf/10.1097/00000542-198504000-00002)
curred, lasted for at least 1 h. Furthermore, to the extent that changes in resting PETCO\textsubscript{2} may be taken as an index of changes in resting ventilation, it is clear that the entire group showed a significant decrease of resting ventilation over the full hour following iv injection of diazepam.

Ventilatory and, more recently, occlusion pressure responses to CO\textsubscript{2} rebreathing have become standard methods for assessing respiratory-center function and drug effects.\textsuperscript{12,15} It is recognized that testing and interpreting respiratory responses to CO\textsubscript{2} is fraught with difficulties, not the least of which is the considerable interindividual and intraindividual variability of the ventilatory response to CO\textsubscript{2}. The latter has been reported to be as high as 15-fold.\textsuperscript{14} Although the ventilatory response to CO\textsubscript{2} is believed to be mediated primarily through intracranial chemoreceptors that are highly sensitive to local changes in hydrogen ion concentration, a multitude of other factors, including gender, race, and personality, appear to influence respiratory responses to CO\textsubscript{2}.\textsuperscript{15} Minimizing extraneous stimuli has also been noted to be an important methodologic component in studying the response to CO\textsubscript{2}.\textsuperscript{16}

In our study, we tried to standardize auditory stimuli by applying white noise through soft, comfortable head phones. Subjects were also instructed to keep their eyes closed during all testing in order to reduce visual distractions. Whether or not these attempts to standardize extraneous stimuli (and thus reduce the variability of the ventilatory response to CO\textsubscript{2}) were successful, numerous other factors may have affected our subjects' response to CO\textsubscript{2}.

Pain is one factor known to increase respiration, and diazepam often produces pain when injected into a peripheral vein. We found that no subject reported any discomfort after diazepam infusion, and we attribute this to the use of a large antecubital fossa vein as our iv site, along with a free-flowing infusion for injection. Headache, too, is frequently induced by CO\textsubscript{2} rebreathing tests. Although when it occurs, the headache is usually transient (5-min duration), one subject reported a headache for the entire 1 h of testing and for 3 days after her CO\textsubscript{2} challenge.

The perception of the "need to breathe," sensed as shortness of breath, is another possibly important factor influencing how individuals respond to CO\textsubscript{2} rebreathing. Although a trial CO\textsubscript{2} challenge was experienced by every subject to familiarize them with CO\textsubscript{2} rebreathing and hopefully allay anxiety about the experiment, certain individuals may, in an anticipatory fashion, have altered their breathing as a result of various perceptions, e.g., the "need to breathe." Also, different individuals may perceive a CO\textsubscript{2} challenge as a typical "test," and certain attitudes about performance may influence the response to CO\textsubscript{2}. The extraversion score (Eysenek Personality Inven-

**Table 3. Slope of the Occlusion Pressure Response to Carbon Dioxide Rebreathing (P0.1/PETCO\textsubscript{2}) as a Per Cent (±SEM) of the Slope Prior to Diazepam**

<table>
<thead>
<tr>
<th>Group</th>
<th>Time after Diazepam (min)</th>
<th>0</th>
<th>20</th>
<th>40</th>
<th>60</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (n = 5)</td>
<td>102</td>
<td>192</td>
<td>203</td>
<td>219</td>
<td></td>
</tr>
<tr>
<td>2 (n = 19)</td>
<td>55</td>
<td>64</td>
<td>72</td>
<td>69</td>
<td></td>
</tr>
<tr>
<td>P value (intergroup difference)</td>
<td>0.012</td>
<td>0.000</td>
<td>0.000</td>
<td>0.000</td>
<td></td>
</tr>
</tbody>
</table>

It is clear from other studies\textsuperscript{5-6} that diazepam will often produce a significant decrease of the ventilatory and/or occlusion pressure response to CO\textsubscript{2}. However, as suggested by others\textsuperscript{5,6} and as shown in our study, diazepam will, in a minority of individuals, augment the ventilatory and/or occlusion pressure response to CO\textsubscript{2}. There are several possible explanations for this observation. Diazepam is a potent anxiolytic as well as a muscle-relaxing drug. In our experience, it is difficult to guarantee that all subjects are relaxed and free of anxiety about being the "guinea pig." Anxiety might result in a chronic state of increased inspiratory tone, associated with a shorter diaphragm (with a longer radius of curvature) at the end of expiration. This places the diaphragm at a mechanical disadvantage based on length–tension and LaPlacian relationships. Diazepam, by allaying anxiety and/or by muscle relaxation (and thus decreasing or eliminating inspiratory muscle tone during expiration), may return the diaphragm to a position where its mechanical advantage is superior to its condition prior to diazepam. Under these conditions the diaphragm would be able to generate more force for a given neural drive after diazepam. However, even without diazepam, the position and mechanical advantage that the diaphragm assumes during CO\textsubscript{2} rebreathing may vary or be dichotomous.\textsuperscript{17}

Diazepam has also been noted to increase airway resistance\textsuperscript{19} and this, in a parallel fashion, could increase neural respiratory drive.\textsuperscript{20} Although the ventilatory response to CO\textsubscript{2} is affected by peripheral components of the respiratory system, such as resistance, as well as central neural drive, the occlusion pressure response is relatively uninfluenced by changes in mechanical factors such as airway resistance.\textsuperscript{21} Thus, increased inspiratory resistance could lead to increased central respiratory drive and an increased P0.1 that might not be evident in the ventilatory response to CO\textsubscript{2}. This may account for the more significant grouping of responders to diazepam by the P0.1.

Two recent reviews on benzodiazepines consider their major properties to be sedation, anxiolysis, antiseizure...
activity, and muscle relaxation. It is interesting to note that respiratory depression is not specifically mentioned. Several investigators, however, have found a positive correlation between respiratory depression and decreasing levels of unconsciousness or sedation. We did not perform psychomotor function tests during this study and cannot relate degree of sedation to respiratory depression. All diazepam effects are, however, thought to be receptor-mediated. Although some benzodiazepine receptors are present in the medulla, other brain areas, such as the cerebral cortex, are significantly richer in these receptors. Diazepam’s respiratory effects, therefore, may be related to nonspecific actions at other than medullary sites. However, although not conclusive, a recent review on gamma-aminobutyric acid (GABA) and respiratory function does cite several reports of GABA or GABA-related compounds stimulating respiration. It is possible that differing effects (e.g., stimulation or depression) may be dose- and receptor- (GABA$_B$ vs. GABA$_A$) dependent.

We did not study subjects with repeated CO$_2$ challenges in the absence of drug intervention. Indeed, there is a paucity of such data in the literature, and the possibility exists that repeated CO$_2$ challenges over time may have revealed significant differences in responses among some of our subjects in this study. However, our own experience in pilot studies done during the development of our rebreathing system indicated no systematic differences in the CO$_2$ response over time in a given subject. Furthermore, Cherniak and co-workers†† in their experience, find no systematic differences in CO$_2$ responses in normal subjects over time.

In contrast to our finding that some individuals demonstrated an enhanced response to CO$_2$ rebreathing following diazepam, all subjects in the present study manifested an increased resting end-tidal CO$_2$ after diazepam. This could be due to either an increase in metabolic rate with the same alveolar ventilation or a decrease in alveolar ventilation in the absence of any increase in metabolic rate. We believe that it is unlikely that intravenous diazepam would result in a significant increase in metabolism, and accordingly we conclude that all subjects in the present study manifested a decrease in resting alveolar ventilation following diazepam. Although this may be due to a nonspecific sedative effect, it does represent significant respiratory depression. The absolute changes in the PET$_{CO_2}$ induced by diazepam are small (table 2). That these small changes are statistically significant is due to the scant variability of this respiratory parameter.

The responses to a CO$_2$ challenge indicate, among other neurochemical respiratory control actions, the gain or drive of the chemical respiratory control mechanism.

To date, however, what the implications of the slopes and changes in slopes of the ventilatory and occlusion pressure responses to CO$_2$ are in health and disease is not clear. Rebreathing CO$_2$, up to PET$_{CO_2}$ of 55 to 65 mmHg is a substantial stress. While the ventilatory and occlusion pressure responses to CO$_2$ have proved useful in documenting the severity and duration of effect of potent respiratory depressants, e.g., opioids, the nature and outcome of these tests may be subject to considerable influences from other environmental or physiologic stimuli, and such overriding effects may obscure the more subtle respiratory actions of less potent drugs or of small to moderate doses of respiratory depressants. We suggest, therefore, that although the response to a CO$_2$ challenge has been used as a standard index of respiratory drive and in the assessment of drug effects, changes in resting ventilation and/or resting CO$_2$ levels may be a more useful tool for the evaluation of subtle physiologic and pharmacologic interventions.

In summary, we have found that some individuals manifest an augmentation of the occlusion pressure response to CO$_2$ following diazepam. If one does not select for this group of individuals when studying diazepam’s and perhaps other drugs’ respiratory effects by CO$_2$ rebreathing, the severity and duration of respiratory depression that occurs in the majority of individuals may be masked or obscured. This may account for some of the discrepancies concerning the respiratory effects of diazepam found in the literature.

References

9. Catchlove RFH, Kafer ER: The effects of diazepam on the ventilatory response to carbon dioxide and on steady-state gas exchange. ANESTHESIOLOGY 34:9–13, 1971

†† Cherniak NS: Personal communication.


