Among all the MLAEP parameters, Pa and Nb latencies are the best predictors of increasing sedation and loss of consciousness. (Key words: Anesthesia; hypnotic; monitoring; opioid.)

MIDLATENCY auditory evoked potentials (MLAEP) have been proposed as monitors of the state of anesthesia. They have been shown to be profoundly affected by hypnotic drugs in a graded, reversible, and non–agent-specific manner. The bispectral index (BIS) is another electroencephalogram (EEG)-derived parameter that has been optimized to correlate with level of sedation and loss of consciousness. The purpose of the present study was to examine the effect of the combination of an opioid, alfentanil, and a hypnotic, propofol, on the MLAEP and compare the changes in MLAEP with increasing sedation and loss of consciousness to that produced in the BIS. In addition, we tested the hypothesis that the administration of alfentanil with propofol should not affect those MLAEP parameters, which measure the hypnotic state of anesthesia, even though alfentanil decreases the propofol concentration required for loss of consciousness.
Methods

This was a prospective study conducted in 20 paid healthy male or female volunteers. Institutional review board approval was obtained, and all volunteers gave written informed consent. Those with known neurologic disorders, including current use of anticonvulsant or other psychoactive medications, long-term drug or alcohol use, hypertension, or other serious medical conditions that would interfere with response analysis were excluded. The volunteers were divided in two groups of 10 subjects according to which drug they received: Propofol alone or propofol and alfentanil. These volunteers were also part of two other studies, the results of which have already been published.17,18

All volunteers underwent a history and physical examination before being enrolled in the study. All were instructed to take nothing by mouth from midnight the night before the study. On the morning of the study, volunteers had an intravenous catheter inserted for fluid and drug administration. A radial arterial catheter was also inserted to monitor blood pressure and to obtain blood samples for subsequent measurement of drug concentration. The volunteers were also monitored using a standard three-lead electrocardiograph and a peripheral pulse oximeter for SpO2. They had nasal prongs to receive oxygen and to monitor end-tidal CO2 and respiratory rate. An observer monitored respiratory and cardiovascular function and determined the need for interventions such as jaw support to maintain an adequate airway.

EEG signals were acquired using gold cup electrodes applied to the scalp with collodion or cream. Skin impedance was maintained at less than 5 kΩ. The following leads were recorded: Fp1 and Fp2 using Cz as the reference (channels 1 and 2), two additional frontal locations between the preauricular point and the lateral canthus using “Fpz” as the reference (channels 3 and 4), and a ground electrode. The EEG was recorded continuously using an Aspect A-1000 EEG monitor (Aspect Medical Systems, Natick, MA), which also computed the BIS in real time. This instrument has an amplifier bandwidth of 0.5–400 Hz, sampled at 1 kHz, and digitally filtered with a high-pass filter of 25 Hz. Recording began simultaneously with stimulation and continued for 125 ms. Responses to 512 stimuli were averaged together to form each evoked potential tracing. Before measuring the waveforms, additional filtering was applied. A high-pass filter of 25 Hz and three-point smoothing were applied to reduce variability due to noise. Four evoked potentials were collected at baseline (before the administration of any drug) and at each drug infusion level. The amplitudes (µV) and latencies (ms) of the waves V, Pa, and Nb were measured manually using a ruler on printed tracings of AEP by an individual (I.C.) who was blinded to which group the volunteer had been randomized. We measured the amplitude as the height of the peak of the wave from the prestimulus baseline (time zero) of the x-axis and the latencies as its distance from the stimulus artifact. Peak V belongs to the brainstem auditory evoked potentials (BAEP), which demonstrates that auditory stimuli are correctly transduced.20 If peak V could not clearly be identified, the evoked potential tracing was rejected.

After all monitoring had been instituted, the volunteers were given a 15-min resting period. Thereafter, baseline readings were obtained. (These readings were also performed following drug administration at each target steady state drug level.) These consisted of a BIS score,
AEP, measure of sedation, and two arterial blood samples for drug concentration before and after each assessment.

Sedation was measured using the responsiveness component of the Observer Assessment of Alertness and Sedation rating scale (table 1). This assessment procedure involves presentation of progressively more intense stimulation, ranging from a moderate speaking voice to physical shaking or moderate noxious stimuli (trapezius squeeze) until a response is observed.

The effect of propofol and propofol plus alfentanil on the BIS score, AEP, and level of sedation were evaluated. The intravenous drugs were administered via a computer-assisted continuous infusion device to a target effect site concentration.\textsuperscript{21} The pharmacokinetic parameters of Gepts \textit{et al.}\textsuperscript{22} were used for propofol administration and those of Scott and Stanski\textsuperscript{23} were used for alfentanil. The accuracy of these pharmacokinetic parameters has previously been validated.\textsuperscript{16,24,25} In every subject, propofol was administered in increasing steps to a target effect site concentration of 1, 2, 4, and 6 \textmu g/ml until volunteers lost consciousness as defined by loss of response to a verbal command. The propofol concentra-

tion was decreased by the same steps until consciousness occurred and then increased and decreased so that at least two crossovers of consciousness–unconsciousness occurred. Alfentanil or placebo (saline) was administered in a double blind fashion to a preselected effect site concentration of 100 ng/ml. At each step, the target concentration was maintained for a minimum of 10 min to permit equilibration with the effect site, as modeled by the propofol and alfentanil \textit{ke}\textsuperscript{0}.\textsuperscript{23,26} (fig. 1).

Plasma drug concentrations were determined from the arterial blood samples collected at the beginning and end of each steady state assessment period. The measured concentration at the beginning of each assessment period was used in correlating concentration to MLAEP or BIS.

Propofol determinations were made using high-pressure liquid chromatography analysis performed at the Anesthesia Research Laboratory at Duke University Medical Center (Durham, North Carolina). The separation and quantification procedures were conducted with a C-18, 15-cm \times 4.6-mm column (Supelcosil LC-18; Supelco, Bellefonte, PA), and detection was fluorometric. Spiked standards (0.5 and 5 \textmu g/ml) were required to be within 20% of the true value with a coefficient of variability of 12%. The duplicate assays of the participant samples were required to be within 20% of each other.

Plasma samples were measured for alfentanil in duplicate by radioimmunoassay using kits from Janssen Pharmaceutical (Olen, Belgium). The coefficient of variability averaged 11.2% over the range of concentrations encountered in this study.\textsuperscript{28}

\begin{table}[!h]
\centering
\begin{tabular}{|l|l|}
\hline
Response & Score Level \\
\hline
Responds readily to name spoken in normal tone & 5 (Alert) \\
Lethargic response to name spoken in normal tone & 4 \\
Responds only after name is called loudly or repeatedly & 3 \\
Responds only after mild prodding or shaking & 2 \\
Does not respond to mild prodding or shaking & 1 \\
Does not respond to noxious stimulus & 0 \\
\hline
\end{tabular}
\caption{Responsiveness Scores of the Modified Observer’s Assessment of Alertness/Sedation Scale}
\end{table}

\begin{figure}[!h]
\centering
\includegraphics[width=\textwidth]{Fig1.pdf}
\caption{Study design showing the time of arterial blood sampling (BS) for drug concentration determination; measurement of bispectral index (BIS), auditory evoked potentials (AEP), and sedation score (SS); and the adjustment of propofol target effect site concentration according to the sedation level.}
\end{figure}
**Statistical Analysis**

The relation among the different evoked potential variables or BIS, measured drug concentration, and sedation level was analyzed using generalized estimating equations. Generalized estimating equations were introduced by Liang and Zeger in 1986 as an extension of generalized linear models to accommodate correlated data using the same link function and linear predictor setup. Generalized estimating equations allow in addition the ability to model the covariance structure of the response measures. It models the first two moments without specifying the complete distribution of the responses. This statistical approach was chosen because it accounts for the within-subject correlation in the face of repeated measurements in the same subject. The effect of the presence of alfentanil was determined by including an indicator (group) in each of the generalized estimating equation models.

Pearson correlations of BIS and AEP variables with sedation and propofol concentration were calculated. Tests for the equality of these correlations for each AEP variable and BIS between the two treatment groups were performed using the Fisher Z transformation. Tests for the equality of these correlations within each group was also done using the single sample test of Olkin. Bonferroni correction was done to account for the multiple testing.

All volunteers who responded to any verbal command (sedation scores 3, 4, and 5) were classified as conscious. Those who did not respond (sedation scores of 0, 1, and 2) were considered to be unresponsive and unconscious. Logistic regression techniques were used to analyze these relations for the quantal endpoints of loss of consciousness. We determined for the plasma concentration of propofol and for the MLAEP parameters (Pa and Nb latencies and amplitudes), propofol measured, and target concentration and compared within each group using the usual Gaussian test statistic. To enable comparison of $P_k$ values, we used $1 - P_k$ when the $P_k$ value was less than 0.5. Bonferroni corrections were done to account for multiple tests.

**Table 2. Demographics**

<table>
<thead>
<tr>
<th>Groups</th>
<th>Propofol</th>
<th>Propofol/Alfentanil (100 ng/ml)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of subjects</td>
<td>10</td>
<td>10</td>
</tr>
<tr>
<td>Mean age ±SD (yr)</td>
<td>31 ± 6</td>
<td>31 ± 7</td>
</tr>
<tr>
<td>Mean weight ±SD (kg)</td>
<td>72 ± 20</td>
<td>81 ± 17</td>
</tr>
<tr>
<td>Mean height ±SD (cm)</td>
<td>171 ± 9</td>
<td>174 ± 10</td>
</tr>
<tr>
<td>Male</td>
<td>5</td>
<td>8</td>
</tr>
<tr>
<td>Female</td>
<td>5</td>
<td>2</td>
</tr>
</tbody>
</table>

SD = standard deviation.

Descriptive statistics were used to compare demographic variables between the study groups. Probability values less than 0.05 were considered significant. Statistical analyses were done using the SAS software of SAS Institute (Cary, NC).

**Results**

There were no statistically significant differences between the two groups with regard to age, weight, height, and gender (table 2). Typical changes in MLAEP with increasing propofol concentration are illustrated in figure 2. Peak V amplitude of the BAEP did not change significantly either with the increase of the plasma propofol concentration ($P = 0.6$) or with increasing level of sedation ($P = 0.7$). Peak V latency was slightly prolonged by both conditions ($P = 0.03$ with propofol concentration; $P = 0.015$ with sedation level). All other MLAEP parameters showed highly significant changes (decrease of BIS index and Pa and Nb amplitudes and increase of Pa and Nb latencies) with increasing plasma propofol concentrations ($P < 0.0001$) and increasing sedation levels ($P < 0.0001$) with and without alfentanil (figs. 3 and 4). These changes were comparable between groups for all AEP parameters analyzed except for Pa and Nb amplitudes, which were more pronounced in the group receiving propofol alone.

The BIS index showed a better correlation with propofol concentration ($P < 0.00005$) and sedation level ($P = 0.0000005$) than did the MLAEP. This is illustrated in figures 3 and 4, where the relation between change in sedation score and the BIS or MLAEP parameter values are plotted. Among all waveform parameters measured, Pa and Nb latency showed the best correlation with propofol concentration and sedation level ($P < 0.005$). MLAEP correlations were comparable between the propofol and propofol-alfentanil groups ($P > 0.7$; tables 3 and 4).
The probability of consciousness versus BIS, Pa or Nb latency, and PA or Nb amplitude is plotted in figure 5. BIS and latency provided better predictors of consciousness, as reflected by the slope of the probability curves. The addition of alfentanil did not significantly affect the values of Pa and Nb amplitudes and latencies associated with a 50% probability of loss of consciousness (table 5; fig. 5; \( P > 0.15 \)). Alfentanil did, however, lower the propofol concentration required for loss of consciousness in 50% of subjects (table 5; fig. 5; \( P < 0.04 \)).

The \( P_k \) for BIS, MLAEP parameters, propofol target, and measured concentration are listed in table 6. The BIS was a better predictor of loss of consciousness than was Pa and Nb amplitude (\( P < 0.002 \)). The MLAEP parameters were comparable as predictors of loss of consciousness. The \( P_k \) values calculated for the MLAEP parameters were not affected by the presence of alfentanil (\( P > 0.1 \)).

**Discussion**

In this volunteer trial, increasing doses of propofol resulted in graded changes on the MLAEP which correlated well with the level of sedation. The addition of alfentanil decreased the propofol \( C_P 50 \) for loss of consciousness but did not significantly affect the MLAEP wave amplitudes and latencies, \( s_{50} \), for the same endpoint. The BIS correlated better with the level of sedation than any of the MLAEP parameters. Among all the MLAEP parameters, Pa and Nb latencies showed the best correlation with the sedation level and were comparable to the BIS as a predictor of loss of consciousness.

Increasing doses of propofol associated with increasing sedation scores affected wave V of BAEP only slightly (no change in peak V amplitude and only a slight increase in peak V latency). Conversely, propofol caused major changes in MLAEP (significant decrease in Pa and Nb amplitudes and increase in Pa and Nb latencies). The MLAEP has been shown to be suppressed by many hypnotic drugs in a dose-dependent manner. General anesthetics alter the MLAEP and, by inference, the processing of auditory stimuli. The effects of general anesthetics can in part be reversed by surgical stimulation, in regard to amplitude more than latency. In patients given a light and stable anesthesia, Thornton et al. demonstrated an increase in the amplitudes of the MLAEP following surgical stimulation without reversing their latencies. In these studies, the MLAEP changes were analyzed in relation to variable doses or concentrations of hypnotic. General anesthetics alter the MLAEP and, by inference, the processing of auditory stimuli. The effects of general anesthetics can in part be reversed by surgical stimulation, in regard to amplitude more than latency. In patients given a light and stable anesthesia, Thornton et al. demonstrated an increase in the amplitudes of the MLAEP following surgical stimulation without reversing their latencies. In patients administered continuous epidural analgesia with general anesthesia, Schwender et al. showed a decrease in latencies and an increase in amplitudes of the MLAEP before and during spontaneous movements observed intraoperatively or during emergence of anesthesia. It is thus important to establish, in the absence of any stimulus, the relation between changing levels of sedation and changes in MLAEP. In our study, we demonstrated not only a good correlation between the MLAEP changes and propofol concentration but also a good correlation between MLAEP parameters (especially latency) and sedation score.

Our study showed that the MLAEP parameters analyzed were comparable as predictors of loss of consciousness, but Pa and Nb latency had the best correlation with propofol concentration and sedation level. Previous studies have tried to extract from the AEP an index able to predict intraoperative wakefulness, awareness, and recall. Thornton et al. analyzed a characteristic MLAEP pattern reflecting the change between wakefulness (response to command detected by isolated arm technique) and light anesthesia (no response) and identified a threshold Nb latency of 44.5 ms as the best...
feature distinguishing these two states. Similar to our study, Newton et al. demonstrated that the largest changes accompanying the transition from partial to no response to verbal command (consciousness to unconsciousness) were in Pa and Nb latencies. Schwender et al. showed that a threshold of Pa latency increase of greater than or less than 12 ms provided a sensitivity of 100% and a specificity of 77% in distinguishing patients with implicit memory from patient without implicit memory postoperatively. In contrast, Heneghan et al.
compared the slopes of the changes of the MLAEP parameters against minimum alveolar concentration multiples for isoflurane, enflurane, and halothane. They demonstrated that Pa and Nb amplitude were more promising indices of the depth of anesthesia than were Pa and Nb latencies because they were not quantitatively differently affected by the three volatile agents and were qualitatively similarly affected by etomidate and althesin. Mantzaridis and Kenny derived mathematically from the AEP a single numerical parameter reflecting both amplitudes and frequencies known as the auditory evoked potential index (AEPidx). This index proved to be reli-

Fig. 4. Scatter diagrams of the relation between bispectral index (BIS), Nb and Pb amplitude (μV), and increasing sedation score as listed in table 1, when volunteers were administered increasing concentrations of propofol or propofol combined with a target alfentanil concentration of 100 μg/ml.
The MLAEP changes with the administration of propofol were not affected by the addition of alfentanil. Our results agree with other studies showing a preservation of the BAEP and slight changes in the early cortical waves of the MLAEP during administration of high doses of opioids. The MLAEP are the AEP components occurring 10–100 ms after stimulus presentation and originate (mainly Pa) from the primary auditory cortex of the temporal lobe. In the presence of high doses of opioids, the auditory stimuli can be transduced and conducted in the primary auditory cortex. The auditory input is often described as the last sensory modality to be abolished and the first to return at light level of anesthesia. As there is a parallel between perception of auditory stimuli and consciousness, the absence of effect of opioids on MLAEP can be related to reports of higher incidence of intraoperative awareness when opioids are the primary anesthetic.

At present, MLAEP have four inconveniences which may detract from their use in clinical practice. First, they need considerable time to produce a response (0.5–5 min to build up an average for the AEP compared with continuous display of an index for the BIS). A moving time averaging technique described by Davies et al. has overcome this problem, allowing a faster response time (every 3 s). Second, difficulties exist in interpreting the waveforms and on-line measurement of Pa and Nb latencies. The AEPIdx mentioned previously can now be calculated as an index for the BIS, and has been optimized to correlate with calculated blood propofol concentration during anesthesia.

### Table 3. Pearson Correlation Coefficients between BIS, Auditory Evoked Potentials Parameters, and Sedation in Both Groups

<table>
<thead>
<tr>
<th></th>
<th>Propofol</th>
<th>Propofol/alfentanil</th>
</tr>
</thead>
<tbody>
<tr>
<td>BIS</td>
<td>0.884*</td>
<td>0.848*</td>
</tr>
<tr>
<td>V amplitude</td>
<td>-0.035</td>
<td>0.068</td>
</tr>
<tr>
<td>Pa amplitude</td>
<td>0.385</td>
<td>0.486</td>
</tr>
<tr>
<td>Nb amplitude</td>
<td>0.328</td>
<td>0.39</td>
</tr>
<tr>
<td>V latency</td>
<td>-0.164</td>
<td>-0.361</td>
</tr>
<tr>
<td>Pa latency</td>
<td>-0.685†</td>
<td>-0.673§</td>
</tr>
<tr>
<td>Nb latency</td>
<td>-0.658‡</td>
<td>-0.725</td>
</tr>
</tbody>
</table>

BIS = bispectral index.

* Significantly different from all the auditory evoked potentials parameters ($P < 0.0000005$, at $\alpha = 0.002$).

† Significantly different from V amplitude and latency, Pa and Nb amplitudes ($P < 0.001$ at $\alpha = 0.002$).

‡ Significantly different from V amplitude and latency ($P < 0.001$ at $\alpha = 0.002$).

§ Significantly different from V amplitude ($P = 0.0003$ at $\alpha = 0.002$).

### Table 4. Pearson Correlation Coefficients between BIS, Auditory Evoked Potentials, and Propofol Measured Concentration in Both Groups

<table>
<thead>
<tr>
<th></th>
<th>Propofol</th>
<th>Propofol/alfentanil</th>
</tr>
</thead>
<tbody>
<tr>
<td>BIS</td>
<td>-0.8*</td>
<td>-0.74*</td>
</tr>
<tr>
<td>V amplitude</td>
<td>-0.111</td>
<td>-0.02</td>
</tr>
<tr>
<td>Pa amplitude</td>
<td>-0.533†</td>
<td>-0.438</td>
</tr>
<tr>
<td>Nb amplitude</td>
<td>-0.483</td>
<td>-0.337</td>
</tr>
<tr>
<td>V latency</td>
<td>0.088</td>
<td>0.102</td>
</tr>
<tr>
<td>Pa latency</td>
<td>0.615§</td>
<td>0.397§</td>
</tr>
<tr>
<td>Nb latency</td>
<td>0.827</td>
<td></td>
</tr>
</tbody>
</table>

BIS = bispectral index.

* Significantly different from all the auditory evoked potentials parameters ($P < 0.000005$ at $\alpha = 0.002$).

† Significantly different from V and Nb amplitude ($P < 0.0002$ at $\alpha = 0.002$).

‡ Significantly different from V latency ($P < 0.001$ at $\alpha = 0.002$).

§ Significantly different from Nb latency ($P < 0.0005$ at $\alpha = 0.002$).

|| Significantly different from V amplitude and latency ($P < 0.0005$ at $\alpha = 0.002$).

# Significantly different from Pa latency ($P < 0.000001$ at $\alpha = 0.002$).

Anesthesiology, V. 92, No 5, May 2000
Fig. 5. The relation between measured propofol concentration, bispectral index (BIS), Pa amplitude, Nb amplitude, Pa latency, Nb latency, and probability of a positive response (0–100% probability) to verbal command were determined using logistic regression analysis of a quantal endpoint (conscious–unconscious) for all volunteers receiving either propofol alone (solid line) or propofol and alfentanil 100 ng/ml (dotted line).
Table 5. MLAEP50 and 95, Cp50, and 95 for Loss of Consciousness and Limits Corresponding to the 95% Confidence Intervals for Both Groups

<table>
<thead>
<tr>
<th></th>
<th>Propofol</th>
<th>Propofol/Alfentanil</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pa amplitude 50</td>
<td>0.45 (0.31–0.60)</td>
<td>0.39 (0.26–0.52)</td>
</tr>
<tr>
<td>Pa amplitude 95</td>
<td>*</td>
<td>*</td>
</tr>
<tr>
<td>Pa latency 50</td>
<td>36.4 (34.55–38.23)</td>
<td>36 (34.18–37.82)</td>
</tr>
<tr>
<td>Pa latency 95</td>
<td>45.83 (42.23–49.42)</td>
<td>43.99 (40.84–47.15)</td>
</tr>
<tr>
<td>Nb amplitude 50</td>
<td>0.38 (0.19–0.57)</td>
<td>0.3 (0.11–0.49)</td>
</tr>
<tr>
<td>Nb amplitude 95</td>
<td>*</td>
<td>*</td>
</tr>
<tr>
<td>Nb latency 50</td>
<td>54.5 (51.68–57.64)</td>
<td>56.49 (53.58–59.4)</td>
</tr>
<tr>
<td>Nb latency 95</td>
<td>73.57 (66.55–80.71)</td>
<td>69.99 (64.63–75.37)</td>
</tr>
<tr>
<td>Cp50</td>
<td>2.25 (1.80–2.69)‡</td>
<td>1.63 (1.24–2.02)</td>
</tr>
<tr>
<td>Cp95</td>
<td>6.47 (4.77–8.23)†</td>
<td>4.13 (3.09–5.16)</td>
</tr>
</tbody>
</table>

* These values are not reported because these parameters did not reach the 95% probability of unconsciousness.
† Significantly different between propofol and propofol–alfentanil (P = 0.04).
‡ Significantly different from propofol measured concentration (P = 0.04).

Table 6. Prediction Probability (P ± SE) Scores for Correctly Predicting Whether Subjects Were Conscious (OAAS Scores 3–5) or Unconscious (OAAS scores 1–2) in Both Groups

<table>
<thead>
<tr>
<th></th>
<th>Propofol</th>
<th>Propofol/Alfentanil</th>
</tr>
</thead>
<tbody>
<tr>
<td>BIS</td>
<td>0.952 ± 0.023*</td>
<td>0.937 ± 0.031†</td>
</tr>
<tr>
<td>Pa amplitude</td>
<td>0.729 ± 0.067</td>
<td>0.780 ± 0.056</td>
</tr>
<tr>
<td>Nb amplitude</td>
<td>0.699 ± 0.07†</td>
<td>0.673 ± 0.067</td>
</tr>
<tr>
<td>Pa latency</td>
<td>0.869 ± 0.044</td>
<td>0.860 ± 0.046</td>
</tr>
<tr>
<td>Nb latency</td>
<td>0.873 ± 0.045</td>
<td>0.885 ± 0.039</td>
</tr>
<tr>
<td>Propofol measured</td>
<td>0.932 ± 0.03</td>
<td>0.834 ± 0.052</td>
</tr>
<tr>
<td>Propofol target</td>
<td>0.854 ± 0.041</td>
<td>0.864 ± 0.037</td>
</tr>
</tbody>
</table>

BIS = bispectral index.
* Significantly different from Pa and Nb amplitude (P < 0.002 at α = 0.0025),
† Significantly different from Nb amplitude (P < 0.0005 at α = 0.0025),
‡ Significantly different from propofol measured concentration (P < 0.0025 at α = 0.0025).

The authors thank Kevin Weatherwax, Department of Anesthesiology, Duke University Medical Center, for his help with the data and graphs.

References


Anesthesiology, V 92, No 5, May 2000


38. Doi M, Gajraj RJ, Mantzaridis H, Kenny GNC: Relationship between calculated blood concentration of propofol and electrophysiological variables during emergence from anaesthesia: Comparison of bispectral index, spectral edge frequency, median frequency and auditory evoked potential index. Br J Anaesth 1997; 78:180–4


