Discontinuous Monitoring of Propofol Concentrations in Expired Alveolar Gas and in Arterial and Venous Plasma during Artificial Ventilation

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**Background:** Analyzing propofol concentration in expired alveolar gas (cPA) may be considered as a convenient, noninvasive method to follow the propofol concentration in plasma (cPPL). In the current study, the authors established procedures to measure cPA and cPPL for the assessment of their relation in two animal models during anesthesia.

**Methods:** Expired alveolar gas and mixed venous and arterial blood were simultaneously sampled during continuous application of propofol for general anesthesia to three goats and three pigs. Propofol infusion rates were varied to modify plasma concentrations. cPA, sampled cumulatively over several respiratory cycles, was quantified by thermal desorption gas chromatography–mass spectrometry. cPPL was determined using reversed phase high-performance liquid chromatography with fluorescence detection.

**Results:** cPA ranged from 0 to 1.4 and from 0 to 22 parts per billion in goats and pigs, respectively, at cPPL of 0–8 μg/mL. The relation between cPA and cPPL was linear; however, the slopes of the regression lines varied between animals.

**Conclusion:** Propofol can be quantified in expired alveolar gas. The results stress the role of marked species-specific variability.

THE inspiratory and expiratory measurement of gas concentrations of volatile anesthetics is a suitable procedure for monitoring anesthetic uptake and measuring minimal alveolar concentration. The definitions of minimal alveolar concentrations for the patient conditions “awake” or “incision” are used to quantify inhaled anesthetic potency. The continuous end-tidal measurement of anesthetic gases represents a valuable aid for the assessment of sufficiently high anesthetic levels. This approach for monitoring anesthesia, however, fails with intravenously applied anesthetics.

A major representative of the latter group is the intravenous narcotic propofol (2,6-diisopropyl-phenol), which is currently used in a wide range of anesthetic procedures. Because the anesthetic potency of propofol is related to its effect site concentration and—depending on the blood/tissue partition coefficient—to the plasma concentration (cPPL), the determination of cPPL might provide an effective tool for monitoring and controlling anesthesia. However, a noninvasive method for continuously monitoring cPPL is not yet available, and therefore, alternative procedures such as the technique of target-controlled infusion of propofol or the control of propofol infusion by monitoring electroencephalographically derived indices (e.g., Bispectral Index) have been developed.

The analysis of propofol concentration in the expired alveolar gas (cPA) may be a promising noninvasive method for monitoring cPPL. However, several conditions must be fulfilled before introducing such a method into clinical routine: (1) an animal model has to be established in which, as with humans, the concentration of an anesthetic agent can be controlled and determined, and even minor changes in anesthetic concentration must be clearly registered in both breath and plasma samples; (2) because propofol is hardly volatile, its presence in exhaled air must be proven and quantified unequivocally; (3) because the relation between breath and plasma concentrations of propofol must be characterized, samples of blood and breathing gas must be collected simultaneously; (4) taking into consideration that the lung may participate in the extrahepatic clearance or metabolism of propofol, the effects of pulmonary metabolism and distribution of propofol must be studied, e.g., by measuring its plasma concentrations in both the mixed venous and the arterial compartment of the vascular system; and (5) a reliable, simple and robust routine procedure must be developed to monitor cPA.

The aim of the current study was to prove conditions 1–4, namely that propofol can be measured qualitatively and quantitatively in expired alveolar gas, and to characterize the relation between cPA and cPPL before and after lung passage in two well-established animal models.

**Materials and Methods**

**Animal Protocol**

This investigation was approved by the regional authority for animal research (Ministerium für Umwelt, Naturschutz und Landwirtschaft, Kiel, Schleswig-Holstein, Germany) and studied both goats and pigs. After intramuscular premedication, three goats were endotracheally intubated, and three pigs received a tracheotomy during adequate anesthesia. An artery and a pulmonary artery catheter were inserted for hemodynamic monitoring and for drawing the plasma samples. The ventilation...
was adjusted in a normocapnic range, and volume deficits after fasting were corrected. Further deficits due to bleeding remained insignificant.

After obtaining the blank samples for venous plasma and breath gas analysis, propofol (2% Disoprivan®, AstraZeneca, Wedel, Germany) was infused via a separate peripheral venous access. Propofol was administered in seven different levels as indicated in the profiles (fig. 1). These changes in propofol dosage were designed to reflect the infusion of propofol during a routine anesthetic procedure. At the end of a 10-min period of constant propofol dosing, further blood samples for measuring mixed venous and arterial $c_{PL}$ were collected.

**Gas Sampling Procedure**

Samples of exhaled breath from mechanically ventilated animals were collected using a method essentially described by Schubert et al.\textsuperscript{12} Alveolar gas was detected using a mainstream carbon dioxide sensor connected to the endotracheal tube. From the gas mainstream samples were drawn through a T-piece by a pump (fig. 2), which was automatically activated and deactivated when a preset carbon dioxide concentration of 25 mmHg was passed. End-tidal gas, 7–10 ml, was obtained from several breaths, revealing a total gas volume of 150 ml for goats and 50 ml for pigs drawn through the adsorption tubes.

**Analysis of $c_{PA}$**

Alveolar breath samples were adsorbed in duplicates onto Tenax TA tubes (Supelco, Bellefonte, PA), which were thermally desorbed and transferred using a cryo-trap (Thermodesorption system; Gerstel, Muelheim, Germany) to a gas chromatograph–mass spectrometer (6890 Plus and 5973; Hewlett-Packard Co., Waldbronn, Germany). The chromatography conditions were as follows: carrier gas: 1 ml/min helium; column: Optima 5 MS, 60 m × 0.25 mm × 0.25 μm (Macherey & Nagel, Düren, Germany); temperature injection/thermodesorption: 260°C; temperature detection: 260°C; temperature oven: 40°–290°C. The retention time for propofol was 22.6 min. For the calibration procedure, known amounts of 2,6-diisopropylphenol (0.096–19.2 ng/tube in six lev-
Results

Propofol concentration in expired alveolar gas was successfully measured in all animals by the experimental setup (fig. 2). The data from the individual animals reflect the time course of propofol concentrations measured in corresponding plasma samples as well as the varying profile of propofol dosing (fig. 1).

The measured range of 0–8 \( \mu g/ml \) for propofol in the various animals was reflected by ranges of 0–1.4 ppb for propofol in goats and 0–22 ppb for propofol in pigs (fig. 3).

The close relation between propofol concentrations measured in plasma and in expired gas in each animal was indicated by regression analysis. The \( R^2 \) values in the individual measurements ranged from 0.760 to 0.985 in the goats and 0.804 to 0.906 in the pigs (fig. 3). There was also an individual variability expressed by the slopes of the regression lines (fig. 3).

Calculating the difference between each data pair (mixed venous and arterial \( c_PPL \)), the goats demonstrated a higher mixed venous to arterial difference of propofol as compared with the pigs: These differences were 0.81 ± 0.50 \( \mu g/ml \) in the goats and 0.11 ± 0.34 \( \mu g/ml \) in the pigs.

Discussion

The current communication reports the initial results from our efforts toward breath analysis of propofol as a noninvasive procedure to monitor plasma concentrations. Our findings clearly indicate that during propofol anesthesia, propofol is present in mammalian breath gas at concentrations that seem to be linearly related to the respective plasma concentrations.

As indicated by its melting (18°C) and boiling (256°C) points, the intravenous anesthetic propofol is not a typical volatile organic compound. The identification of propofol in the expired alveolar gas was assisted by several means: (1) comparison of the retention time of the measured substance in a sample with that of pure 2,6-diisopropylphenol in the gas chromatography system, (2) identification of the fragment ion pattern using a library of mass spectra, (3) selection of the three highest peaks within this pattern for quantitative measurement, and (4) demonstration of a constant ratio between these peaks.

One other study on propofol breath analysis has been published that reported preliminary results from the monitoring of propofol in human breath by proton transfer reaction mass spectrometry.14 In this investigation, Harrison et al.14 reported qualitative results revealed with a sidestream sampling mode during the first 10 min after induction of anesthesia in one intubated woman. They presented an operationally defined concentration unit (counts per second) without a calibration procedure, assuming that these values corresponded to a

Analysis of \( c_PPL \)

The \( c_PPL \) samples were measured in triplicate by reversed phase high-performance liquid chromatography with fluorometric detection essentially according to Kita et al.15 The measurable range of this method extended from 0.001 to 6.0 \( \mu g/ml \); samples with higher propofol concentrations were appropriately diluted with physiologic saline before analysis. Pooled plasma samples repeatedly (n = 21) tested for propofol yielded intraassay coefficients of variation of 8.7% throughout the study period with the same amount of 2,6-diisopropylphenol (1.92 ng/tube) yielded an interassay coefficient of variation of 12.1%. The mean recovery of the propofol added onto the Tenax polymer for these repeated measurements was 106% (range, 84–136%). As indicated by the calibration procedure, a propofol concentration of 1 part per billion (ppb) corresponded to 7.3 ng/l in gas (see appendix).

Statistical Analysis

For each sampling point, \( c_PPL \) and mixed venous and arterial \( c_PPL \) were compared by linear regression analysis.

\[ \text{Fig. 2. Schematic setup for the gas sampling system: A mainstream carbon dioxide sensor triggered the pump, which was automatically activated and deactivated when a preset carbon dioxide (CO}_2\text{) concentration of 25 mmHg was passed.} \]
range that was approximately between 5 and 50 ppb. Because Harrison et al. followed with his method the first passage of propofol through the lung, this might be in a similar range when compared with the data of our animal models with concentrations of 0–2 ppb in the goats and 0–20 ppb in the pigs. However, major differences were evident between the two investigations regarding the study subjects, the mode of breath sampling, and the way in which the propofol concentration data were collected. Harrison et al. applied an unheated tube of 4 m length for breath sampling and analyzed expired air from mechanically ventilated patients in a quasi-continuous way that was also prone to drawbacks (e.g., sampling of mixed air instead of expired alveolar gas, or condensation of humidity on the inner wall of the tube). In our study, the discontinuously inserted Tenax tube
combined with the gas chromatography–mass spectrometry method offered certain advantages: (1) For calibration, we applied defined amounts of 2,6-diisopropylphenol to the Tenax polymer in the tube; and (2) by passing expired alveolar gas from several breaths through the adsorption tube, the substance was accumulated on the polymer according to the analytical sensitivity required. Using this method, we were able to quantify propofol in the lower parts per billion range in breath samples (1 ppb corresponding to 7.3 ng/l).

Comparing propofol concentrations in expired gas and plasma, one has to consider to what extent the lung itself participates in the metabolism and thereby extrahepatic elimination of propofol. From the discrepancy between the mixed venous and arterial plasma concentrations as well as the concurring appearance of metabolites, Dawidowics et al.\textsuperscript{10} concluded that propofol is indeed eliminated by the lungs. He et al.\textsuperscript{15} found not only that propofol is stored in the lung by a first-pass effect, but that it is also rereleased. Under steady state conditions, no difference between mixed venous and arterial propofol concentration was found in this study in humans.

Our results reveal major differences between the two investigated animal species. All of the goats examined here revealed a consistent difference between mixed venous and arterial propofol concentration over the entire monitoring phase. No marked difference was observed among the pigs. We also found clear differences between the two species regarding propofol concentrations measured in both plasma and expired gas. The concentration in the plasma of the goat was in the same range as that seen in the pig, whereas the concentration in expired gas was lower by a factor of approximately 10.

The extrahepatic metabolism of propofol has not been definitively deduced in humans or various other animal species.\textsuperscript{16} Kuipers et al.\textsuperscript{17} first investigated uptake and clearance of propofol in sheep lungs and found a clearance of at least approximately 30% during this phase. They suggested that the relatively volatile substance propofol is removed from the lung by exhalation and stated that this phenomenon should be evaluated. Our results clearly confirmed that propofol is exhaled through the lungs.

From the data acquired in this study, we can demonstrate that noninvasive measurement of propofol concentration in expired gas allows individual biologic variability to be revealed and assessed.

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References


Appendix

Based on the molecular weight of propofol (178.27 g/mol) and the volume of 1 mol of an ideal gas at 25°C (24.47 l/mol), mass concentrations of propofol (ng/l) applied for calibration of the gas chromatography–mass spectrometry system were transformed into the widely used unit parts per billion (ppb). As is demonstrated by the following example, 1 ppb corresponds to 7.3 ng/l propofol:

1. 1 l expired alveolar gas consists of 1/24.47 mol or 0.041 mol
2. 1 ng propofol per 1 l expired alveolar gas reflects 1/178.27 nmol/l or 5.61 pmol/l
3. 5.61 pmol propofol per 0.041 mol expired alveolar gas is equivalent to 0.137 × 10\textsuperscript{-7} mol/mol or 0.137 ppb
4. 1 ng/l = 0.137 ppb; 1/0.137 ng/l = 7.3 ng/l = 1 ppb

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