Coating of extracorporeal circuit with heparin does not prevent sequestration of propofol \textit{in vitro}

E. Hammarén$^{1,*}$, P. H. Rosenberg$^1$ and M. Hynynen$^2$

$^1$Department of Anaesthesia, Helsinki University Hospital, Haartmaninkatu 4, FIN-00290 Helsinki, Finland.
$^2$Departments of Anaesthesia and Intensive Care, Jorvi Hospital, Turuntie 150, FIN-02740 Espoo, Finland

*To whom correspondence should be addressed

Propofol is sequestered in extracorporeal circuits, but the factors responsible for the phenomenon are mostly unknown. We have compared two extracorporeal circuits (oxygenators, reservoirs and tubings) coated with heparin with two corresponding uncoated circuits for their capacity to sequester propofol \textit{in vitro}. Three experiments were conducted with each circuit. The circuit was primed with a mixture of Ringer’s acetate solution and whole blood, and the study conditions (pump flow, temperature, pH) were standardized. Propofol was added to the solution to achieve a concentration of 2 $\mu$g ml$^{-1}$. These studies were followed with concentrations of 10- and 100-fold to assess possible saturation of propofol binding. Serial samples were obtained from the circulating solution for measurement of propofol concentration. Propofol concentrations decreased to 22–32% of the initial predicted concentration of 2 $\mu$g ml$^{-1}$ in the circuits (no significant difference between circuits). With greater concentrations, the circuits did not become saturated with propofol, even with the highest predicted concentration of 200 $\mu$g ml$^{-1}$. We conclude that propofol was sequestered in extracorporeal circuits \textit{in vitro}, irrespective of coating the circuit with heparin.

Br J Anaesth 1999; 82: 38–40

Keywords: anaesthetics i.v., propofol; equipment, cardiopulmonary bypass circuits

Accepted for publication: August 18, 1998

Sequestration, that is binding or adsorption, of various drugs (e.g. fentanyl, thiopental (thiopentone), nimodipine, nitroglycerin and propofol) to extracorporeal circuits \textit{in vitro} has been described.$^{1-7}$ Sequestration can change the pharmacokinetic behaviour of drugs during cardiopulmonary bypass (CPB) and complicate adequate dosing of drugs in the patient undergoing CPB.$^8$ Factors affecting sequestration of various drugs to extracorporeal circuits are, for the most part, unknown. However, composition and design of the extracorporeal circuit may alter adsorption of a drug to the circuit, as has been shown with fentanyl and nitroglycerin.$^5$ In addition, the use of blood in the priming solution has been shown to decrease sequestration of nimodipine and fentanyl within the extracorporeal circuit.$^4$ It was speculated that protein coating of the extracorporeal circuit by plasma proteins in blood contributed, at least in part, to attenuation in sequestration of nimodipine.$^4$

With recent advances in extracorporeal circuit technology, it has become possible to have heparin bound to the inside of the circuit to form a biocompatible surface during CPB compared with conventional (uncoated) circuits.$^{10}$ Whether or not coating of extracorporeal circuit with heparin affects the capacity of the circuits to sequester drugs is not known. Therefore, we have compared the capacity of extracorporeal circuits, either coated with heparin or uncoated, to sequester propofol \textit{in vitro}. Two currently available coated circuits, with different methods of binding heparin, and corresponding uncoated circuits were compared.

Materials and methods

We compared four different extracorporeal circuits. Two of the circuits were heparin-coated and the other two were corresponding uncoated circuits from the same manufacturers. The circuits included a hollow fibre membrane oxygenator, venous reservoir, and polyvinylchloride and silicone tubings. Oxygenators and reservoirs were as follows: (1) Spiralgold and BMR-1900 Gold (Baxter Healthcare Corporation, Irvine, CA, USA). (2) Maxima with Carmeda Bio-Active Surface (Medtronic Cardiopulmonary Division, Kerkrade, The Netherlands), (3) Spiraloxy and BMR-1900 (Baxter, Irvine, CA, USA) and (4) Maxima (Medtronic Cardiopulmonary Division, Kerkrade, The Netherlands). The two first circuits are heparin-coated. Three experiments were performed with each circuit.

The circuit was primed with a mixture of Ringer’s acetate solution and just out-dated blood (stored at +2 to +8 °C
Propofol sequestration in extracorporeal circuit

for 36–42 days); the total amount of priming solution was 2000 ml. Heparin 5000 u. was added to the circuit and the temperature of the solution was maintained at 28°C with a heat exchanger. Pump flow was set at 4 litre min⁻¹, and oxygen 2 litre min⁻¹ was added to the circuit. Carbon dioxide was added to maintain $PCO_2$ at 4.6–6.0 kPa and pH was adjusted to 7.35–7.45 (α-stat principle) with tromethamol (Addex-THAM, Kabi Vitrum AB, Stockholm, Sweden).

The experiments were performed in three stages. In the first stage of each experiment, a bolus dose of propofol 4 mg (Diprivan 10 mg ml⁻¹, Zeneca Pharmaceuticals, Cheshire, UK) was injected via a stopcock into the venous site of the circuit to achieve a calculated concentration of $2\mu g\text{ml}^{-1}$ in the solution. In the second and third stages, immediately after the last sample of the previous stage, bolus doses of propofol 40 mg and 356 mg, respectively, were injected into the circuit to achieve calculated concentrations of 22 and $200\mu g\text{ml}^{-1}$, respectively, in the solution. Each stage lasted 60 min. In stages one and two, 5-ml samples for measurement of propofol concentrations, pH and packed cell volume were obtained from the arterial site of the circuit via the arterial–venous sampling stopcock. Samples were obtained before, and at 5, 10, 20, 40 and 60 min after addition of the drug. In stage three, samples were obtained at 10, 30 and 60 min after addition of the drug. Samples for measurement of propofol concentrations were obtained into pre-chilled heparinized tubes and centrifuged at $+4°C$, 3000 rpm, for 10 min, and plasma was stored at $-70°C$ until assayed. Plasma samples were buffered and propofol was extracted with cyclohexane and concentrated and assayed using a HPLC method. The signal was detected with a fluorescence detector with wavelengths of excitation and emission of 276 and 310 nm, respectively. The lower limit of detection was 5 ng ml⁻¹ and the intra-assay coefficient of variation was 4–5%.

Statistical analysis
Analysis of variance (ANOVA) for repeated measurements was used to detect significant changes over time in the variables and to detect differences between circuits in all three stages of the study. $P<0.05$ was considered statistically significant. Data are expressed as mean (SD).

Results
The study conditions were stable during the experiments and comparable between circuits. Propofol concentrations were lower than predicted during all stages of the study for all extracorporeal circuits (Fig. 1). There were no significant differences between circuits. At the end of stage one, Baxter’s non-coated and coated circuits and Medtronic’s non-coated and coated circuits contained, on average, 32%, 22%, 28% and 29%, respectively, of the initial predicted concentration of propofol in the circulating solution. At the end of stage two, corresponding values were 38%, 42%, 30% and 37%, respectively, and at the end of stage three, 57%, 54%, 41% and 46%, respectively.

Discussion
We have confirmed our previous observation that propofol was sequestered in vitro by an extracorporeal circuit uncoated with heparin. Coating of the circuit with heparin did not affect sequestration in two currently available circuits (i.e. Baxter and Medtronic circuits). We used three different concentrations in the circuit: the two lower concentrations were within or near the range of propofol concentrations measured in anaesthetized patients and the highest concentration was clearly above the concentrations measured in clinical practice. The highest concentration was used to estimate the capacity of the circuits to bind...
propofol and to assess possible saturation of propofol binding when very high concentrations are used.

Our finding with the lowest concentration of propofol was similar to our previous observation with the Compactflo System oxygenator and reservoir (Dideco, Mirandola, Italy), that is 60 min after adding propofol, approximately 30% of the initially predicted concentration was detected in the circuits.7

Rosen and colleagues have reported that the capacity of a CPB oxygenator to sequester fentanyl is considerable and that the oxygenator becomes saturated when high concentrations are used in vitro.12 The same workers showed that different oxygenators vary considerably in their capacity to bind fentanyl in vitro.5 In our study, different circuits had similar capacities to bind large amounts of propofol without becoming saturated, even with very high concentrations.

The heparin-coated circuits we studied used different methods for binding heparin to the plastic surface. In Baxter’s circuit, ionic linkages were used and in Medtronic’s circuit, heparin was covalently attached.10 The surface of Baxter’s circuit was less negatively charged than that of Medtronic’s surface. Theoretically, electrostatic attraction of charged molecules would be possible but it is improbable that the propofol molecule, with a pH of approximately 7.4. Indeed, no difference in the binding of propofol between the circuits was found.

In summary, our study showed that two other uncoated, clinically used, extracorporeal circuits also sequester propofol in vitro. Coating of the circuit with heparin using either of the two different methods available did not seem to change binding of propofol to the circuit. With regard to propofol concentrations in patients, it does not seem to matter whether the circuit is coated with heparin or uncoated.

Acknowledgements
We thank Baxter Medical AB and Medtronic Inc. for supplying the extracorporeal circuits. We also thank Kaisa Salminen.

References
2 Hynynen M. Binding of fentanyl and alfentanil to the extracorporeal circuit. Acta Anaesthesiol Scand 1987; 31: 706–10
9 Rosen KR, Rosen DA. Factors which affect fentanyl uptake by the membrane oxygenator. Anesthesiology 1986; 65: A225