Vascular properties of isoflurane: comparison between normal and cirrhotic rats

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Summary

Isoflurane is known to dilate blood vessels and to modulate nitric oxide production. Because cirrhosis is characterized by over production of endothelial nitric oxide, isoflurane-induced vasodilatation may be altered in this situation. We have compared the vasodilator effects of isoflurane in normal rats and rats with secondary biliary cirrhosis. Aortic rings (intact or endothelium denuded) from normal and cirrhotic rats were suspended in HEPES solution and preconstricted with KCl 40 mmol litre⁻¹. Isoflurane dose-dependently relaxed vessels in both groups. Maximal relaxation was comparable between normal and cirrhotic rats in intact (mean 80 (SEM 4)% vs 81 (6)%; ns) and in denuded (100 (4) vs 95 (5)%; ns) vessels. Intact vessels relaxed more than denuded vessels in both groups (100 (4) vs 80 (4)% (P = 0.0008) in normal rats and 95 (5) vs 80 (6)% (P = 0.0008) in cirrhotic rats). We conclude that cirrhosis did not modify isoflurane-induced vasodilatation and that the modulator effect of endothelium was conserved. (Br. J. Anaesth. 1998; 81: 968–969).

Keywords: anaesthetics volatile, isoflurane; blood, vessels; liver, cirrhosis; rat

Cirrhosis is characterized by systemic and splanchnic vasodilatation involving over production of nitric oxide by the endothelium. Isoflurane is a volatile anaesthetic known to induce vasodilatation and to modulate endothelium-derived nitric oxide production. Therefore, the pharmacological properties of isoflurane may be modified in cirrhosis. The aim of this study was to compare the response to increasing concentrations of isoflurane in normal rats and in rats with secondary biliary cirrhosis.

Methods and results

The study was approved by the French Agriculture Office, in accordance with European legislation involving animals. We studied male Sprague–Dawley rats (weighing 300–350 g) (Iffa-Credo, France). Induction of secondary biliary cirrhosis was performed as described previously. Briefly, under ether anaesthesia, the common bile duct was exposed by median laparotomy and occluded by a double ligature with non-resorbable suture (7–0 silk, Peters Laboratories, Bobigny, France). The common bile duct was resected between the two ligatures. A control group using the same weight sham-operated rats was studied in parallel. Animals were studied 4 weeks after operation.

For the preparation of aortic rings, animals were anaesthetized with pentobarbital 50 mg kg⁻¹ i.p. The thoracic aorta was removed and placed in a modified HEPES buffer aerated with a mixture of 95% oxygen and 5% carbon dioxide. Aortae were dissected clean of fat and extraneous tissues and divided in 3.5–4.0-mm ring segments. In some rings, endothelium was removed mechanically by gently rubbing the intimal surface with a stainless steel rod. The rings were suspended between two L-shaped stainless steel hooks and placed in a 10-ml water-jacketed organ chamber containing a solution composed of (mmol litre⁻¹): NaCl 118.2, KCl 4.7, NaHCO₃ 25.0, KHPO₄ 1.2, MgSO₄ 1.2, CaCl₂ 2.5, glucose 11.1 and HEPES 5. Organ chambers were aerated continuously with 95% oxygen and 5% carbon dioxide with pH maintained at 7.35–7.40. Temperature was maintained constant at 37°C. Isometric tension was measured (g) using a force displacement transducer (UFI Pioden Controls, Paris, France) connected to the upper hook. Tension was recorded on a Servo trace recorder (Sefram, Paris). During the equilibration period of 60 min, the buffer was changed every 15 min and resting tension was adjusted periodically to 1.5 g. Active contraction was induced with KCl 40 mmol litre⁻¹ and the integrity of the endothelium was assessed by testing relaxation produced by addition of acetylcholine 10⁻⁶ mol litre⁻¹. Endothelium was considered intact when acetylcholine elicited a minimum of 30% vasodilatation. Endothelium was considered removed when acetylcholine induced no vasodilatation. After acetylcholine testing, the rings were washed and re-equilibrated to baseline tension for 60 min. After this new equilibration period, concentration–responses curves were constructed with norepinephrine (noradrenaline), acetylcholine and isoflurane.

Increasing cumulative doses of norepinephrine 10⁻⁸–10⁻⁵ mol litre⁻¹ were administrated in rings from control (n = 12) and cirrhotic (n = 12) rats. The same...
procedure was performed in rings without endothelium from control (n = 12) and cirrhotic (n = 12) rats.

Rings were preconstricted with norepinephrine 10⁻⁵ mol litre⁻¹ (maximal contraction). When a plateau of maximal contraction was reached, increasing cumulative doses of acetylcholine 10⁻⁸–10⁻⁵ mol litre⁻¹ were then administered in rings from control (n = 6) and cirrhotic (n = 6) rats.

Aortic rings were preconstricted (KCl 40 mmol litre⁻¹) and exposed to cumulative concentrations of isoflurane (1–5%). Isoflurane was delivered using a calibrated agent-specific vaporizer (Abbott, France). Aeration concentrations of isoflurane were monitored and adjusted using an anaesthetic agent monitor. Each concentration was maintained until the effect on the aortic ring reached a plateau (i.e. an average of 10 min). Isoflurane was administered to intact rings from normal (n = 12) and cirrhotic (n = 8) rats. Isoflurane was also administered in denuded rings from normal (n = 12) and cirrhotic (n = 6) rats.

Data are expressed as mean (SEM). Contractions to norepinephrine are expressed in grams. Relaxation to acetylcholine and isoflurane is expressed as percentage of precontraction obtained with norepinephrine 10⁻⁵ mol litre⁻¹ or KCl 40 mmol litre⁻¹. Data were analysed statistically using ANOVA. The statistical package was Statview SE (Abacus Concept, Inc, Berkley, USA, CA). Differences were considered significant at P < 0.05.

In intact rings, the maximal response to norepinephrine was significantly lower in rings from cirrhotic rats compared with controls (1.95 (0.18) vs 2.91 (0.42) g; P = 0.0007). In contrast, contraction to norepinephrine was comparable between denuded rings from cirrhotic and control rats (1.48 (0.24) vs 1.29 (0.15) g; P = 0.49). Acetylcholine-induced relaxation was significantly greater in cirrhotic than in control rats (47.5 (5.6) vs 22.6 (2.8)%; P = 0.003).

After isoflurane, maximal relaxation was comparable between normal and cirrhotic rats in intact (80 (4) vs 81 (6)%; ns) and denuded (100 (4) vs 95 (5)%; ns) vessels. In contrast, intact vessels relaxed more than denuded vessels in both groups of rats (100 (4) vs 80 (4)% (P = 0.0008) in normal rats and 95 (5) vs 80 (6)% in cirrhotic rats (fig. 1).

Comment

We have confirmed that vascular reactivity to an agonist, norepinephrine, was decreased in isolated vessels from cirrhotic rats compared with controls. Among the hypotheses to explain this result, over production of nitric oxide by the endothelium of vessels from cirrhotic animals has been proposed.

In normal rats, it has been shown previously that isoflurane-induced vasodilatation was more important in vessels without endothelium than in intact vessels. To explain this result, we hypothesized that isoflurane inhibits nitric oxide production and at the same time dilates vessels, leading to relative constriction in vessels with endothelium. In our study, this result was also observed in rats with cirrhosis. As a result, modulation of isoflurane-induced vasodilatation by endothelium was conserved in vessels of cirrhotic rats in which over production of nitric oxide took place in the endothelium.

As endothelial production of nitric oxide is increased in cirrhosis and as isoflurane modulates this production, the effect of isoflurane in intact vessels of cirrhotic rats should be different from the effect observed in intact vessels from controls. In fact, we showed that isoflurane-induced vasodilatation was comparable between normal and cirrhotic rats, suggesting that endothelial over production of nitric oxide did not alter vascular reactivity to isoflurane. This result strongly suggests that vasodilatation produced by isoflurane is independent of nitric oxide.

In summary, we conclude that isoflurane-induced vasodilatation was not modified by cirrhosis, a condition involving over production of nitric oxide by the endothelium, and that the modulator effect of endothelium was conserved.

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References