TITLE: EFFECTS OF HALOTHANE ON THE FUNCTION OF SARCOPLASMIC RETICULUM IN SKINNED MYOCARDIAL FIBERS OF NEWBORN AND ADULT RABBIT

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Introduction. Newborns may be more susceptible to halothane-induced hypotension than adults,[1] and isolated cardiac muscle from newborn rabbit myocardium is more sensitive than that of adult rabbits to halothane-induced depression of contractility.[2] Therefore, clinical hypotension in the newborn is due at least in part to a direct action of halothane upon the myocardium. The site of halothane-induced depression of newborn myocardium is undefined. The purpose of this study was to compare the effects of halothane on Ca^{2+} uptake and release by the sarcoplasmic reticulum (SR) of newborn and adult myocardium.

Method.[3] Newborn (1-3 day old) and young adult (2-3 kg) New Zealand white rabbits were sacrificed, and the hearts were rapidly isolated. Right ventricular strips were gently homogenized in relaxing solution (pCa > 9) in order to disrupt the sarcolemma. Fiber bundles (1-2 mm long, 100 μm wide, and 10-20 μm thick) were mounted between clips; one end was attached to a photodiode tension transducer. Isometric tension was continuously recorded with a Gould™ 2400S 4 channel recorder. Newborn and adult preparations were studied simultaneously.

The bathing solutions contained (in mM): Mg^{2+},0.1; $MgATP^{2-}$,2; K^{+},35; Na^{+},35; creatine phosphate,15; EGTA,7 or 0.05; and caffeine,25. Ionic strength was 0.15 and pH was 7.00 ± 0.02 at 20 ± 2°C. $[Ca^{2+}]$ varied from less than 10^{-9}M (pCa > 9, relaxing solution) to $10^{-6.5}$M (pCa = 6.5). $[Ca^{2+}]$ was measured by atomic absorption spectrophotometry (Perkin-Elmer™ 303). Control solutions were saturated with 100% N_2. Halothane-containing solutions were equilibrated with N_2 plus halothane, which was regulated with a Verni-Trol® vaporizer.

For each experiment, fiber bundles were sequentially immersed in five solutions to load, than release Ca^{2+} from the SR. Solution 1 (caffeine, high EGTA, pCa > 9) emptied the SR and relaxed the fiber bundle. Solution 2 (no caffeine, high EGTA) washed caffeine from the fiber bundle. Solution 3 (high EGTA, pCa = 6.5) loaded Ca^{2+} into the SR. Solution 4 (low EGTA, pCa = 6.5) removed EGTA. Finally, Solution 5 was identical to Solution 4, but contained 25 mM caffeine, which resulted in tension development ("tension transient") caused by release of Ca^{2+} from the SR. The area under the tension transient was used as a measure of the amount of Ca^{2+} stored in the SR.[4]

Three sets of experiments were made for each fiber bundle for each concentration of halothane, (0.3, 0.5, 1.0, and 1.7%). In one, the fiber bundle was exposed to halothane during the SR Ca^{2+} loading phase only (Solutions 2-4). In the second, the fiber bundles were exposed to halothane during the SR Ca^{2+} release phase only (Solution 5). And in the third, exposure to halothane was during the entire loading and release cycle (Solutions 2-5). Each halothane experiment was bracketed by two control experiments (no halothane); the result of the effect of halothane on tension transients was expressed as percent of the mean of the two bracketing controls. Data were converted to a normal distribution with the arc-sine transformation prior to statistical comparison using Student's t-test for paired and unpaired data. $P < 0.05$ was regarded as statistically significant.

Results. Halothane exposure of newborn and adult myocardial fibers during the uptake phase resulted in reversible, dose-dependent depression of the area under the tension transient. Halothane exposure during the release phase did not change the areas under the tension transient. Continuous halothane exposure during both SR Ca^{2+} uptake and release phases yielded results similar to exposure to halothane during Ca^{2+} uptake phase only.

SR Ca^{2+} loading of newborn myocardial fibers was less depressed than adult fibers bundles by 0.33 and 0.5% halothane (Fig. 1A). With 1.0 and 1.7% halothane, newborn SR tended to be less depressed than adult SR. There were no differences between newborn and adult myocardium to exposure to halothane during only the Ca^{2+} release phase (Fig. 1B).

Discussion. Halothane exerts a potent depressant effect on SR Ca^{2+} accumulation in both newborn and adult myocardium which is of the same order of magnitude as depression of contractility seen in isolated intact myocardial preparations.[2] Newborn SR is less sensitive to halothane than adult SR which is in contrast to observations in isolated intact myocardial preparations.[2] We conclude that depletion of intracellular SR storage of Ca^{2+} in newborns, as in adults, is an important mechanism of halothane's negative inotropic effect, but that halothane's effects on the SR do not account for the greater depression of myocardial contractility observed in isolated newborn myocardium.[2] We postulate that other mechanisms lead to the greater sensitivity of the newborn myocardium to halothane.

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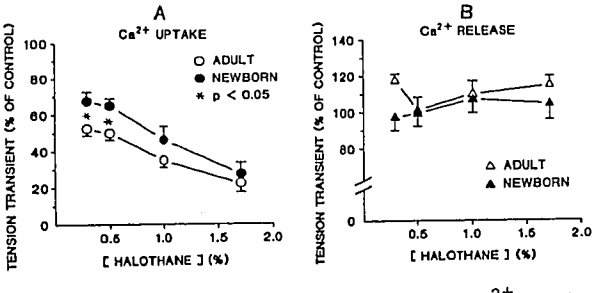

Fig. 1. The effect of halothane during SR Ca^{2+} uptake phase (A) and release phase (B) on tension transients.