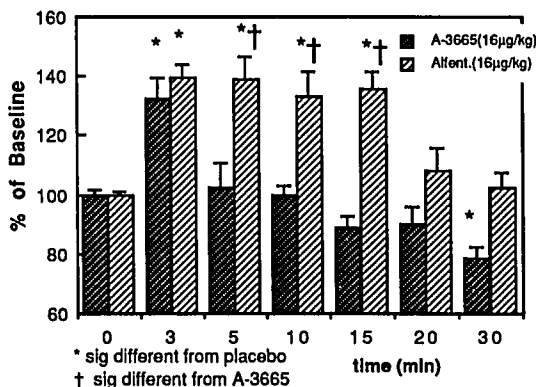


**Title:** COMPARISON OF ANALGESIA AFTER A-3665 AND ALFENTANIL**Authors:** J.J. Cambareri, MD, M.S. Afifi, MD, B.F. Esposito, RN, P.S.A. Glass, MD and E.M. Camporesi, MD**Affiliation:** Dept. of Anesthesiology, SUNY Health Science Center, Syracuse NY 13210

A-3665 is a new short-acting synthetic opioid of the piperidine class currently undergoing phase-1 trials. After obtaining informed consent, we compared the safety and efficacy of A-3665 with that of Alfentanil at three equipotent doses in human volunteers. The research protocol was approved by the Institutional Review Board. Analgesic efficacy was assessed by measuring maximum pressure tolerated by subjects using a modification of the spring loaded rod.<sup>1</sup> Pain tolerance measurements were obtained (2 replications) on the tibia and sternum prior to and at 3, 5, 10, 20, 30, 45, and 60 minutes following drug administration. Heart rate and blood pressure were monitored using ECG trace and radial arterial catheter. Respiratory parameters were monitored using pulse oximetry, capnography and arterial blood gases at 1, 3, 5, 10, 20, 40 and 60 minutes after injection. A-3665 was administered in a double blind manner in increasing doses (0.25, 0.5, 1, 2, 4, 8, 16, 32, and 64 µg/kg) to 34 volunteers. Alfentanil was administered in doses of 4, 8, and 16 µg/kg to 12 volunteers. Placebo was given to 8 subjects. Data were analyzed by ANOVA and F test with significance at  $p < 0.05$ . Both drugs displayed potent analgesia with peak effect occurring at approximately three minutes after injection. There were no detectable differences in hemodynamic parameters measured even at the highest doses of narcotic. There was no significant difference in analgesic potency of A-3665 and Alfentanil as measured by tolerance to tibial pressure at 3 min. At

the dose of 16 µg/kg (see figure, 4 subjects per drug) both drugs significantly increased pain tolerance to tibial pressure compared with placebo at three minutes but Alfentanil continued to display significant analgesic effect vs. placebo and vs. A-3665 up to 20 minutes. A-3665 exhibited significant respiratory depression as measured by PaO<sub>2</sub> and PaCO<sub>2</sub> at 1 and 3 minutes after injection but not at 5 minutes. Alfentanil did not show significant respiratory depression at the doses tested. In parallel to the short lived respiratory depression, A-3665 displays profound analgesia of very short duration. Reference: 1) Anaesthesia, 16:80, 1961.

**Change in Tibial Pressure Tolerance****A350****TITLE:** PRODUCTION AND DETECTION OF VOLATILE ANESTHETIC-ASSOCIATED NEOANTIGENS IN VITRO**AUTHORS:** A.P. Brown, B.S., K.L. Hastings, Dr. P.H., A.J. Gandolfi, Ph.D.**AFFILIATION:** Dept. of Anesthesiology, University of Arizona, Tucson, AZ 85724

Halothane can be biotransformed by the liver to produce a reactive intermediate, which reacts with lysine groups on liver proteins to form trifluoroacetyl lysine. It is proposed that this moiety can act as an epitope to alter the immunogenicity of the native protein and elicit a hypersensitivity leading to fulminant hepatitis.<sup>1</sup> Although halothane-associated neoantigens and their elicited antibodies have been reported to occur in both humans and guinea pigs<sup>2</sup>, an *in vitro* system provides a method for experimental manipulation not possible *in vivo*. Thus liver slices were used to study the conditions for halothane associated neoantigen formation *in vitro*.

Liver slices, (1 cm; 300 µm) from male Hartley guinea pigs were exposed to 1.0 or 1.7 mM halothane (media concentration) in 95% O<sub>2</sub>/5% CO<sub>2</sub> for 12 hr. Covalent binding of a halothane adduct was determined using <sup>14</sup>C-halothane. Neoantigens were detected by Western immunoblot analysis using rabbit anti-trifluoroacetylated albumin antiserum. Deuterated halothane, which is not metabolized to the same extent as halothane, was used as a negative control in the immunoblot analysis. Covalent binding of a halothane intermediate was detected by 1 hr of incubation, increased linearly

through 12 hr, and was concentration-dependent (Table). Covalent binding preceded and correlated with the appearance of liver neoantigens. By 12 hr of incubation, 5 neoantigens were seen with molecular weights ranging from 51 - 97 Kd. These neoantigens have molecular weights similar to those seen in previous animal studies and in liver biopsy samples from patients with halothane hepatitis.<sup>3</sup> Slices incubated with deuterated halothane did not produce detectable neoantigen, demonstrating that oxidative metabolism is a prerequisite to neoantigen formation.

This novel *in vitro* system can be used as a model for mechanistic studies of anesthetic derived neoantigen production in the liver. This system also allows for the potential use of human liver samples to be used in elucidating the mechanisms for anesthetic-induced hypersensitivity reactions.

Supported by NIH GM 34788.

**References**

1. Ann Rev Pharmacol 28:367-387, 1988.
2. Drug Metab Rev 20(2-4):203-217, 1989.
3. Hepatology 8(6):1635-1641, 1988.

**Table** Covalently Bound Halothane Adducts To Liver Slices<sup>a</sup>

Halothane (mM)	Time of Incubation (hr)			
	1	3	6	12
1.0	11.9 ± 1.4	16.5 ± 2.3	19.9 ± 2.0	27.2 ± 2.2
1.7	20.7 ± 2.5	36.1 ± 3.8	39.1 ± 3.5	48.5 ± 4.7

Values are X ± SEM of nanoequivalents bound/mg protein.  
N=24-30 slices from 3 animals.