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# *Fertility, Reproduction and Postnatal Survival in Mice Chronically Exposed to Halothane*

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**Reproductive studies were performed in Swiss/ICR mice chronically exposed to subanesthetic and anesthetic concentrations of halothane. Male and female mice were treated five or seven days a week for nine weeks prior to mating; exposure of females was continued daily throughout pregnancy. Halothane exposures were 0.025, 0.1, 0.4, 1.2, and 4.0 MAC hours per day. No adverse effect on reproduction was observed at the lowest two exposure levels studied. Exposures to 0.4 MAC hour per day or more were associated with decreased maternal weight gain, fetal length and weight, and early postnatal weight gain. Pregnancy rate, implantation rate, and number of live fetuses per litter were significantly decreased at 1.2 MAC hours per day. The percentage of resorptions or fetuses dead *in utero* was not increased, and postnatal survival of offspring was unaltered. Subsequent matings between untreated females and males exposed to halothane, 1.2 MAC hours per day for 17 weeks, resulted in normal reproductive performance; this suggests that the adverse reproductive changes observed when both males and females were exposed represented a primary effect on females. The least exposure at which effects were seen is approximately 40 times greater than the level of human occupational exposure in unscavenged operating rooms. (Key words: Anesthetics, volatile, halothane; Anesthetics, volatile, trace concentrations; Toxicity, fetal; Toxicity, reproductive.)**

THE INCIDENCE of spontaneous abortion among operating room personnel is reported to be higher than incidences in various control populations.[1–6] In addition, it has been suggested that female infertility occurs more frequently in physician anesthetists than in physician controls.[4] Occupational exposure of operating room personnel to trace concentrations of waste anesthetic gases has been well documented,[7,8] and may be responsible for these epidemiologic findings.

The embryotoxic and teratogenic effects of gestational exposure to nitrous oxide and halothane have been studied in rodent species.[9–16] The results have been inconclusive, owing to differences in concentrations of anesthetic agents tested, durations and frequencies of exposures, timings of exposures relative to the period of fetal development, and specific reproductive characteristics evaluated. Moreover, the effects of chronic exposure to anesthetic agents prior to mating have barely been explored.[13,15] The present study was designed to detect possible adverse effects of halothane on many phases of reproduction, including spermatogenesis in the male. Various levels of halothane exposure were studied in order to define a dose–response relationship for any observed effect. The exposures employed ranged from approximately two and a half to 400 times the daily occupational exposure in an unscavenged operating room.¶

## Methods and Materials

Five-week-old virgin male and female Swiss/ICR mice** were individually marked with metal ear tags, observed for seven days for signs of illness, then randomly divided into experimental groups. Mice were caged by sex and treatment group, four animals per cage, and bedded on ground corncob.†† Mice were maintained at a temperature of 21 ± 1 C in a room provided with artificial lighting on a regular cycle, 13 hours each day. No other animal species or mouse strain was housed in the same room during the experiment. No germicides or pesticides were used in the facility. Mice were allowed continuous access to small laboratory animal chow‡‡ and water, except during inhalational exposures, and were weighed weekly.

### INHALATIONAL EXPOSURES

Inhalational exposures were performed in two stainless steel and plexiglass gas-tight chambers, each approximately 1,500 liters in capacity. Halothane was vaporized in a Copper Kettle with medical-grade

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¶ Anesthetic exposure in MAC hours is calculated by multiplying anesthetic concentration, expressed as a fraction of MAC, by the duration of administration. Occupational exposure to halothane in an unscavenged room is approximately 0.0013 MAC (10 ppm) × 8 hours/day = 0.01 MAC hour/day.[7,8]

** Hilltop Lab Animals, Inc., Scottsdale, Pennsylvania 15683.
†† Tab Litter, Paxton Processing Co., Paxton, Illinois 60957.
‡‡ Wayne Lab-Blox, Allied Mills, Inc., Chicago, Illinois 60606.

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TABLE 1. Exposure Schedule for Mouse Reproductive Studies (Males and Females Exposed)

| | Exposure | | | | Weeks of Exposure Prior to Mating | Number of Mice Starting Study | |
|---|---|---|---|---|---|---|---|
| | Agent | Hours/Day | MAC Hours/Day | Days/Week | | M | F |
| Experiment A Group 1 | No treatment (colony control) | — | — | — | — | 20 | 40 |
| Group 2 | Compressed air (treatment control) | 4 | — | 7 | 9 | 20 | 40 |
| Group 3 | 0.05 per cent halothane | 0.5 | 0.025 | 7 | 9 | 16 | 32 |
| Group 4 | 0.05 per cent halothane | 2 | 0.10 | 7 | 9 | 16 | 32 |
| Group 5 | 0.10 per cent halothane | 4 | 0.40 | 7 | 9 | 17 | 34 |
| Experiment B Group 6 | No treatment (colony controls) | — | — | — | — | 26 | 52 |
| Group 7 | Compressed air (treatment control) | 4 | — | 5 | 9 | 26 | 52 |
| Group 8 | 0.10 per cent halothane | 4 | 0.40 | 5 | 9 | 21 | 42 |
| Group 9 | 0.30 per cent halothane | 4 | 1.2 | 5 | 9 | 21 | 42 |
| Group 10 | 1.0 per cent halothane | 4 | 4.0 | 5 | 9 | 21 | 42 |

compressed air and delivered at a total air flow rate of 3–6 l/min to the chambers through latex rubber tubing. Soda lime was placed on the floor of the chambers to absorb carbon dioxide. A high-volume fan recirculated the atmosphere within each chamber in order to maintain a uniform anesthetic vapor concentration. Concentrations of halothane were monitored every 5 to 15 minutes using a Varian 1440 gas chromatograph, and were maintained within 10 per cent of the desired concentrations.

## EXPERIMENT A—LOW-DOSE HALOTHANE EXPOSURES IN MALE AND FEMALE MICE

One hundred-seventy-eight female and 89 male mice were randomly divided into five groups (table 1): two groups were controls; three groups were treated with halothane in air. Control animals were either untreated (colony controls), Group 1; or were exposed to compressed air daily, for four hours in an inhalation chamber (treatment controls), Group 2. Halothane exposures were 0.05 per cent for ½ hour daily (0.025 MAC hour/day),§§ Group 3; 0.05 per cent for 2 hours daily (0.1 MAC hour/day), Group 4; 0.1 per cent for 4 hours daily (0.4 MAC hour/day), Group 5. Following the ninth week of treatment, females were recaged in pairs and one male from the same treatment group was placed with each pair of females, nightly, for seven nights. Each morning following mating, males were returned to their original cages and females were examined for vaginal copulatory plugs. The day a copulatory plug was observed was considered day "0" of pregnancy. Females that did not show copulatory plugs after seven nights were remated with a new male for seven additional nights. Daily halothane treatments were continued throughout mating and pregnancy. On day 18 of pregnancy, one day prior to expected parturition, each female was weighed, then sacrificed by cervical dislocation. The uterus was immediately exposed and examined for the number and position of live and dead fetuses, resorptions (equivalent to abortions in humans), and total implantations; the crown–rump length, weight, and sex of each fetus also were determined. Pregnant females in which no copulatory plug was observed were sacrificed late in pregnancy as judged by serial weight determinations and physical appearance; fetuses were similarly examined. All non-plugged females that did not show signs of pregnancy were sacrificed 14 to 18 days following the last day of mating and examined for implantations. The prior treatment history of each animal was unknown to the examiner.

## EXPERIMENT B—HIGH-DOSE HALOTHANE EXPOSURES IN MALE AND FEMALE MICE

Because the previous experiment failed to establish a threshold level for reproductive toxicity, a second experiment was performed employing higher halo-

§§ One per cent halothane equals 1 MAC in the mouse.[17]

thane dosages (table 1). An additional 230 female and 115 male mice were randomly divided into five groups: Groups 6 and 7 were colony and treatment control groups, respectively, corresponding to Groups 1 and 2 of experiment A. The remaining animals were exposed to halothane, 0.1 per cent (Group 8), 0.3 per cent (Group 9) and 1 per cent (Group 10) vaporized in compressed air, for 4 hours daily, 5 days per week, for 9 weeks prior to mating; anesthetic exposures were 0.4, 1.2, and 4.0 MAC hours/day for Groups 8–10, respectively. Mating was performed as in Experiment A. All treatments were carried out seven days per week during mating and pregnancy. Two thirds of the pregnant dams were sacrificed on day 18 of pregnancy and the uterine contents examined as in Experiment A. In order to determine the effect of maternal administration of halothane on postnatal survival, the remaining third were allowed to deliver and nurse their offspring; no animal was exposed to halothane after delivery. The total number of live pups at birth was determined, as were the numbers and mean weights of pups surviving on days 1 and 4. After 4 days, litters were randomly culled to a maximum of eight pups; survivals and mean weights of pups were then determined on days 7, 14 and 21 after birth. Pups were weaned, and weighed individually at 4 weeks of age.

### Experiment C—High-dose Halothane Exposure in Male Mice

After a total of 17 weeks of uninterrupted treatment, each male mouse from Groups 7 (treatment control) and 9 (halothane, 1.2 MAC hours/day) was mated, nightly, for seven nights with two untreated, 10-week-old, virgin, female Swiss/ICR mice. Treatment of males was continued throughout mating; females were never exposed to halothane or compressed air. Females that did not show a copulatory plug were remated with a different male for seven additional nights. Sacrifices of female mice and uterine examinations were performed on day 18 of pregnancy as in Experiments A and B.

### Analysis of Data

Copulatory rate (females with copulatory plugs/number mated), pregnancy rate among females with copulatory plugs, and overall pregnancy rate (number of pregnant dams/number mated) were determined for each group, as was the percentage of males siring litters. Mean weight gain during pregnancy, mean fetal length, and mean fetal weight were determined for all pregnant dams with copulatory plugs sacrificed on day 18 of pregnancy. Mean number of implantations (total of live and dead fetuses plus resorptions), fetuses per litter (live plus dead fetuses), and live fetuses per litter were determined for all pregnant dams sacrificed prior to spontaneous delivery. Indices of reproductive wastage, *i.e.*, resorptions plus fetuses dead *in utero*, equivalent to human abortions and stillbirths, respectively, were calculated for each female as percentages of the total number of implantations, and the means for each group were then determined. For each female allowed to deliver in Experiment B, the number of live offspring on the day of delivery, the percentage of liveborn pups that survived until day 4 (viability index), the percentage of pups alive at 4 days that survived until weaning (lactation index), and the mean weights of surviving pups were determined, and group means calculated. In all cases, halothane-treated groups were compared with the treated control group in the same experiment. Additionally, the colony control and treated control groups in each experiment were compared with each other and with the corresponding control group in the other experiment. Chi-square analysis using the Yates correction was employed when comparing the distribution of a variable, *e.g.*, number of females with copulatory plugs, in one group with the distribution of the same variable in another group. When one or more of the expected frequencies was 5 or less, Fisher's exact test was used.[18] When intergroup comparisons of means were made, Student's t test was employed.

Data from pregnant dams sacrificed prior to delivery for which the precise day of copulation was not known (*i.e.*, not copulatory plug was observed) were included in comparisons of implantation rates, litter sizes, and reproductive wastage, but not in comparisons of maternal weight gains or fetal sizes. Ten dams in Experiment A delivered spontaneously prior to the intended date of cesarean section, making accurate counts of implantations, resorptions, and live and dead fetuses impossible; data for these litters were omitted from all such comparisons.

## Results

### Experiments A and B

*Fertility.* During the nine-week pre-mating treatment period there was no adverse effect on weight gain or survival among any of the groups treated with subanesthetic concentrations of halothane, *i.e.*, as much as 1.2 MAC hours per day. However, 35 of 42 females and 19 of 21 males in Group 10 (4.0 MAC hours per day) died prior to mating. Deaths were distributed approximately equally throughout

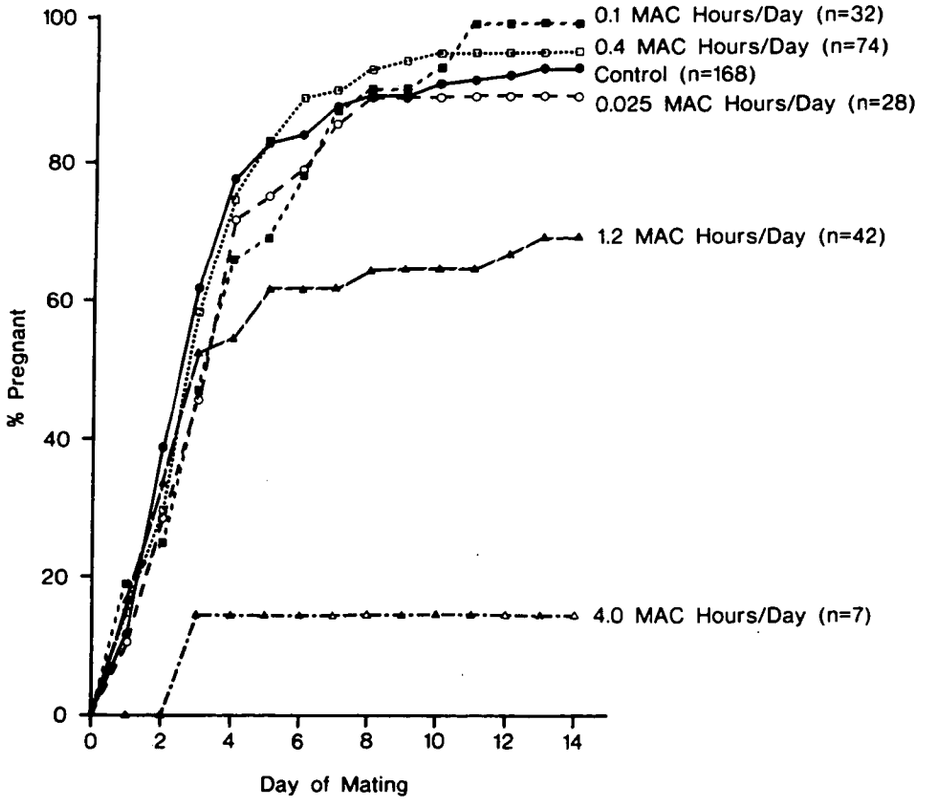


FIG. 1. Experiments A and B. Cumulative pregnancy rate. Pregnancy rate decreased in dose-related fashion beginning at the 1.2 MAC hour per day exposure. Control groups and 0.4 MAC hour per day groups were combined for purposes of graphic presentation.

the nine-week premating period, nearly all occurring during the inhalational exposures. The cause of death appeared to be hypothermia, since surviving mice had core temperatures as low as 24 C following treatments.

Of the 351 female mice mated in Experiments A and B, 313 became pregnant. Cumulative pregnancy rate as a function of the day of mating is shown in figure 1. Table 2 presents the copulatory rate, pregnancy rate in females with copulatory plugs, overall pregnancy rate, and percentage of males siring litters. There was no difference between the untreated groups (colony controls) and those exposed to compressed air only (treatment controls) in either experiment. Similarly, no difference between corresponding control groups was seen in Experiments A and B. No effect on copulation or fertility was found at the three lowest halothane exposures. However, the preg-

TABLE 2. Experiments A and B, Copulatory and Fertility Rates in Mice after Chronic Exposure of Males and Females to Halothane

| | Halothane Exposure (MAC Hours per Day) | Number of Females Mated | Copulatory Rate (Per Cent) | Pregnancy Rate in Copulated Females (Per Cent) | Overall Pregnancy Rate (Per Cent) | Males Siring Litters (Per Cent) |
|---|---|---|---|---|---|---|
| Experiment A | | | | | | |
| Group 1 | Colony control | 38 | 76 | 97 | 95 | 100 |
| Group 2 | Treatment control | 36 | 94 | 94 | 92 | 100 |
| Group 3 | 0.025 | 28 | 89 | 96 | 89 | 100 |
| Group 4 | 0.10 | 32 | 84 | 100 | 100 | 100 |
| Group 5 | 0.40 | 34 | 88 | 97 | 94 | 94 |
| Experiment B | | | | | | |
| Group 6 | Colony control | 48 | 90 | 93 | 92 | 100 |
| Group 7 | Treatment control | 46 | 94 | 93 | 94 | 100 |
| Group 8 | 0.40 | 40 | 95 | 100 | 98 | 100 |
| Group 9 | 1.2 | 42 | 86 | 78‡ | 67† | 90 |
| Group 10 | 4.0 | 7 | 43† | 33* | 14†§ | 0§ |

* $P < 0.05$ *vs.* Group 7 (Fisher's exact test).
† $P < 0.01$ *vs.* Group 7 ($X^2$ or Fisher's exact test).
‡ $P < 0.05$ *vs.* Groups 6 and 7 combined ($X^2$); $P = 0.12$ *vs.* Group 7 alone ($X^2$).
§ Litter sired by non-treated male.

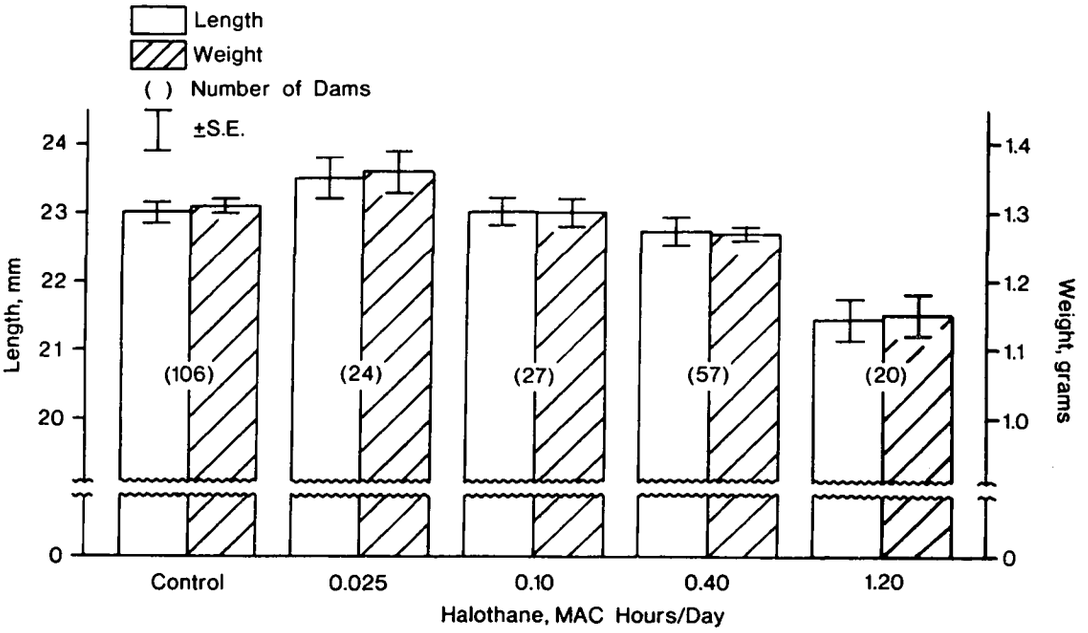


FIG. 2. Experiments A and B. Mean fetal length and weight. Mean fetal weight decreased at the 0.4 and 1.2 MAC hour per day exposures. Mean fetal length also decreased in dose-related fashion. Control groups and 0.4 MAC hour per day groups were combined for purposes of graphic presentation.

nancy rate was significantly decreased in females exposed to 1.2 MAC hours daily ($P < 0.01$), although copulatory rate was unaffected. Three of seven surviving females in the 4.0 MAC hour per day group (Group 10) copulated; only one pregnancy resulted. This occurred in a female mated with an untreated male, as only two treated males survived the experiment. Both copulatory and pregnancy rates in this group were significantly lower than in controls. However, in view of the high maternal lethality and marked physiologic disturbances, the majority of reproductive toxicity at this exposure level is probably an indirect anesthetic effect, rather than directly related to halothane exposure. There were insufficient data from this group for inclusion in subsequent statistical comparisons.

Table 3 shows the mean number of implantations and the number of live fetuses per litter as a function of halothane exposure. Implantation rate was decreased at the 0.4 MAC hour per day exposure in both experiments, approaching statistical significance ($P = 0.09$) in Experiment B. Among females exposed to 1.2 MAC hours per day of halothane, Group 9, implantation rate and number of live fetuses per litter were significantly lower than those in the treatment control, Group 7 ($P < 0.001$ and $P < 0.05$, respectively).

*Reproductive Performance.* Post-implantation re-

TABLE 3. Experiments A and B, Implantations and Live Fetuses per Dam, and Reproductive Wastage as a Percentage of Total Number of Implantations, Mean ± SE

| | Exposure (MAC Hours per Day) | Number of Litters | Implantations/ Dam | Live Fetuses/ Litter | Reproductive Wastage | | |
|---|---|---|---|---|---|---|---|
| | | | | | Resorbed (Per Cent) | Dead *in Utero* (Per Cent) | Resorbed plus Dead *in Utero* (Per Cent) |
| Experiment A | | | | | | | |
| Group 1 | Colony control | 29 | 13.07 ± 0.34 | 11.24 ± 0.49 | 14.0 ± 2.6 | 0.50 ± 0.35 | 14.5 ± 2.6 |
| Group 2 | Treatment control | 26 | 12.27 ± 0.60 | 10.62 ± 0.56 | 12.4 ± 2.5 | 0.74 ± 0.41 | 13.1 ± 2.6 |
| Group 3 | 0.025 | 25 | 11.64 ± 0.60 | 10.08 ± 0.59 | 14.3 ± 2.3 | 0.44 ± 0.44 | 14.8 ± 2.3 |
| Group 4 | 0.10 | 29 | 12.72 ± 0.41 | 11.31 ± 0.37 | 9.9 ± 1.4 | 0.98 ± 0.55 | 10.9 ± 1.5 |
| Group 5 | 0.40 | 30 | 11.97 ± 0.31 | 10.90 ± 0.39 | 8.6 ± 1.6 | 0.84 ± 0.47 | 9.4 ± 1.7 |
| Experiment B | | | | | | | |
| Group 6 | Colony control | 31 | 12.65 ± 0.50 | 11.06 ± 0.51 | 10.4 ± 1.5 | 2.31 ± 0.79§ | 12.7 ± 1.7 |
| Group 7 | Treatment control | 29 | 12.52 ± 0.34 | 10.62 ± 0.52 | 13.8 ± 3.7 | 0.74 ± 0.41 | 14.5 ± 3.7 |
| Group 8 | 0.40 | 29 | 11.69 ± 0.32* | 10.83 ± 0.40 | 6.8 ± 1.7 | 0.94 ± 0.52 | 7.7 ± 1.8 |
| Group 9 | 1.2 | 20 | 10.10 ± 0.62† | 9.00 ± 0.55‡ | 7.8 ± 1.7 | 2.60 ± 1.07 | 10.4 ± 1.6 |

* $P = 0.09$ *vs.* Group 7.
† $P < 0.001$ *vs.* Group 7.
‡ $P < 0.05$ *vs.* Group 7.
§ $P < 0.05$ *vs.* Group 1.

TABLE 4. Experiment B, Survival of Offspring and Postnatal Weight Gains, Mean ± SE

| Group | Exposure (MAC Hours per Day) | Number of Litters | Number of Live Pups/Dam | Viability Index (Per Cent) | Lactation Index (Per Cent) | Mean Pup Weight (g) | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | 24 Hours | 4 Days | 7 Days | 14 Days | 21 Days | 28 Days |
| 6 | Colony control | 13 | 10.46 ± 0.64 | 98 ± 1 | 96 ± 4 | 1.77 ± 0.04 | 2.55 ± 0.08 | 4.01 ± 0.11 | 6.91 ± 0.16 | 9.38 ± 0.28 | 16.04 ± 0.68 |
| 7 | Treatment control | 14 | 10.57 ± 0.71 | 98 ± 1 | 96 ± 2 | 1.79 ± 0.04 | 2.52 ± 0.10 | 4.11 ± 0.13 | 7.05 ± 0.26 | 9.59 ± 0.43 | 16.40 ± 0.86 |
| 8 | 0.4 | 10 | 9.70 ± 0.92 | 84 ± 10 | 99 ± 1 | 1.70 ± 0.05 | 2.30 ± 0.09† | 3.69 ± 0.10* | 6.66 ± 0.23 | 9.04 ± 0.46 | 14.74 ± 1.24 |
| 9 | 1.2 | 8 | 9.38 ± 1.05 | 95 ± 4 | 98 ± 2 | 1.66 ± 0.04* | 2.35 ± 0.13 | 3.76 ± 0.17 | 6.87 ± 0.42 | 9.40 ± 0.61 | 16.84 ± 1.26 |

* $P < 0.05$ *vs.* Group 7.

† $P < 0.05$ *vs.* Groups 6 and 7 combined.

productive wastage as a percentage of the total number of implantations is shown in table 3. The number of fetuses dead *in utero* was greater in the colony control animals of Experiment B (Group 6) than in the corresponding group of Experiment A. Exposure to as much as 1.2 MAC hours per day halothane did not increase the percentage of resorptions or fetuses dead *in utero*.

Mean fetal length and weight are graphed in figure 2. Mean fetal weights were decreased approximately 5 per cent in Experiments A and B at the 0.4 MAC hour per day exposure ($P = 0.07$ and $P < 0.01$, respectively), and 15 per cent at the 1.2 MAC hour per day exposure ($P < 0.001$). Mean fetal length also was decreased in dose-related fashion: 4 per cent at 0.4 MAC hours per day, $P < 0.01$, Experiment B; 10 per cent at 1.2 MAC hours per day, $P < 0.001$. Similarly, maternal weight gain during pregnancy decreased with increasing halothane exposure: 12 per cent at 0.4 MAC hours per day, $P < 0.05$, Experiment A; 28 per cent at 1.2 MAC hours per day, $P < 0.001$. Sex ratio of the offspring was not affected by exposure to halothane.

*Postnatal Survival and Weight Gain.* There was no difference in survivals among any of the groups (table 4). However, there was a tendency towards lower mean pup weight during the first week of life in halothane-exposed animals. At 14 days and thereafter there was no difference in weights among any of the groups.

### HIGH-DOSE HALOTHANE EXPOSURE IN MALE MICE, EXPERIMENT C

The results of this experiment are shown in table 5. There was no difference in reproductive indices between the control group (treatment control males and untreated females) and the halothane-treated group (males exposed to 1.2 MAC hours/day of halothane for 17 weeks and untreated females) except an increase in the overall pregnancy rate in the latter group.

## Discussion

The results of the present study define a threshold level of halothane exposure at which reproductive toxicity occurs in Swiss/ICR mice. No adverse reproductive effect was observed at halothane exposures of as much as 0.1 MAC hour per day. Exposures to 0.4 MAC hour per day or more were associated with decreases in maternal weight gain during pregnancy, mean weight and length of live fetuses, and early postnatal weight gain. Additionally, at 1.2 MAC

TABLE 5. Experiment C, Reproductive Indices Following Halothane Exposure in Male Mice, Mean ± SE

| Exposure (MAC Hours per Day) | Number of Females Mated | Copulation Rate (Per Cent) | Pregnancy Rate in Copulated Females (Per Cent) | Overall Pregnancy Rate (Per Cent) | Males Siring Litters (Per Cent) | Implantations per Dam | Reproductive Wastage | | | Fetal Weight (g) | Fetal Length (mm) | Maternal Weight Gain (g) |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | | | | | | | Resorbed (Per Cent) | Dead *in Utero* (Per Cent) | Dead *in Utero* plus Resorbed (Per Cent) | | | |
| Treatment control | 40 | 88 | 89 | 88 | 95 | 12.61 ± 0.34 | 7.84 ± 1.57 | 0.76 ± 0.43 | 8.61 ± 1.54 | 1.34 ± 0.02 | 23.21 ± 0.25 | 27.52 ± 0.67 |
| 1.2 | 36 | 92 | 100 | 100* | 94 | 13.03 ± 0.32 | 9.57 ± 1.54 | 0.92 ± 0.55 | 10.49 ± 1.70 | 1.33 ± 0.01 | 23.39 ± 0.16 | 27.45 ± 0.68 |

* $P < 0.05$ *vs.* control (Fisher's exact test).

hours per day, decreases in pregnancy rate, implantation rate, and number of live fetuses per litter were found. The lower pregnancy rate appeared to result from decreased female fertility, since the percentage of males siring litters was not affected, and the reproductive indices in matings between treated males and untreated females were normal. There was no increase in post-implantation reproductive wastage, and postnatal survival was not affected.

These data have several implications. The least exposure at which effects were seen was 0.4 MAC hour daily, *i.e.*, 0.1 per cent halothane for 4 hours; this is approximately 40 times greater than the exposure to halothane in an unscavenged operating room. Thus, in mice, the reproductive hazard of human occupational levels of halothane exposure is negligible. The absence of increased fetal wastage suggests that, once implantation has been established, chronic exposure to halothane does not prevent maturation into a viable fetus. The postnatal survival data indicate that prenatal exposure to halothane does not result in lethal developmental abnormalities.

There are several possible explanations for the lowered pregnancy rates and decreased numbers of implantations observed in the present study: failure of ovulation, production of nonviable ova, failure of fertilization or implantation, or very early post-implantation embryonic wastage. The decreases in mean fetal weight and length at term may have resulted from reduced maternal caloric intake, interference with placental transport of nutrients, or depressed fetal intermediary metabolism. The changes were not great and were no longer present 14 days after birth.

Two prior studies examined the reproductive effects of pre-mating exposure to halothane. Bruce[13] demonstrated no effect on fertility or reproduction when mice were exposed to 0.0016 per cent halothane, seven hours daily (0.011 MAC hour/day), five days per week for six weeks prior to mating. However, the low pregnancy rate in the control animals (38 per cent) precluded the detection of all but major effects. Furthermore, animals were not examined for copulatory plugs, and all pregnant dams were not sacrificed on the same day of pregnancy; thus, several important reproductive modalities could not be evaluated. In addition, a six-week period of treatment is not sufficient to detect adverse effects on early spermatogenesis, which, in mice, requires 56 days.[19] In the other study, Kennedy *et al.*[15] found no adverse effect on reproduction following one hour of exposure of either male or female rats to 1.4 per cent halothane on five consecutive days, at various intervals preceding mating. The extremely short period of treatment in this study did not test the effects of halothane on all phases of spermatogenesis in male rats. Additionally, in females, it might not have detected adverse effects of halothane on maturation of ova from primary oocytes.

In other studies of the reproductive effects of halothane, Basford and Fink[11] exposed Sprague-Dawley rats to 0.8 per cent halothane in 25 per cent oxygen for 12 hours at various times during pregnancy: no increase in fetal wastage was observed; fertility was not evaluated. Kennedy *et al.*[15] reported no adverse reproductive effect in rats and rabbits exposed to 1.4 per cent halothane in oxygen-enriched air for one hour daily, on five consecutive days, during various stages of pregnancy. Most recently, Lansdown *et al.*[16] concluded that as much as 0.32 per cent halothane administered to Sprague-Dawley rats for eight hours on days 8–12 of pregnancy was not fetotoxic.

The present study provides a systematic approach to the evaluation of the reproductive effects of inhalational anesthetics, and will allow future comparative studies of other anesthetic agents. Direct human experimentation in this area is impossible, and epidemiologic studies do not allow for the examination of isolated drug effects. Therefore, comparative animal studies are essential to further the understanding of occupational hazards due to anesthetic exposure. Ultimately it will be possible to develop a scale of relative reproductive toxicities of anesthetic agents in a common animal strain. Direct extrapolation of such data to man, however, will be the subject to the limitations occasioned by species differences in drug metabolism and excretion, as well as differences in susceptibilities to the toxic phenomena.

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