tion for this discrepancy is that the CO_2 pressure in the sediment is higher than that in equilibrium with the atmosphere, and it is this that keeps the *p*H down. A calculation of the CO_2 pressure based on the *p*H change indicates that CO_2 pressure in the sediment is almost ten times that of the atmosphere. Measurements with a glass electrode, with a saturometer technique (*4*), indicated that the *in situ* sediment water is saturated with respect to $CaCO_3$ (calcite). The increased CO_2 is most likely the result of bacterial oxidation of organic matter. If this is so, CO_2 apparently diffuses upward through the sediment too slowly to allow the sediment to equilibrate with the overlying sea water, which, at this location, is almost certainly close to equilibrium with the atmosphere. Saturation with respect to calcite indicates that the solid carbonate shells are quick to equilibrate with the increased CO_2 by dissolving slightly (*5*).

RAYMOND SIEVER, ROBERT M. GARRELS
JOHN KANWISHER, ROBERT A. BERNER
Woods Hole Oceanographic Institution, Woods Hole, Massachusetts, and
Harvard University, Cambridge, Massachusetts

References and Notes

1. R. Siever, *J. Sediment. Petrol.*, in press.
2. G. Eisenman, D. O. Rudin, J. U. Casby, *Science* **126**, 831 (1957).
3. B. Kullenberg, *Medd. Oceanog. Inst. Göteborg* **6**, (1952).
4. P. K. Weyl, *J. Geol.* **69**, 32 (1961).
5. This research was supported by various grants from the Petroleum Research Fund, American Chemical Society; the Committee on Experimental Geology, Harvard University; The Office of Naval Research, under Contract NONR 2196 (00), NR 083 (004); and the Woods Hole Oceanographic Institution. This paper is Contribution No. 1168 from the Woods Hole Oceanographic Institution. Chlorinity analyses were made by D. R. Waldbaum.

3 May 1961

Spontaneous Ejaculation in Rat

Abstract. Daily seminal discharges were observed when individually caged male rats were prevented from grooming the genital region orally.

Seminal ejaculation in the rat is believed to depend upon genital stimulation derived from a series of intromissions (*1*). Few observations have been made of ejaculation in the absence of penile stimulation. Aronson (*2*) observed erection and ejaculation during sleep in the domestic cat. But these events were often accompanied by pelvic movement. We have recently observed spontaneous ejaculations in hooded and albino rats after bilateral damage to amygdala and hippocampus primarily, but also after damage to septum, olfactory bulb, hypothalamus, and cortex (*3*).

Table 1. Daily ejaculations and penile smears of ten albino rats.

Animal	Seminal plugs/days of observation		Penile smears		
	Rats unrestrained	Rats girdled	Percent of 11 smears, taken during daylight, containing sperm; rats unrestrained	Presence of sperm in one smear taken nocturnally; rats unrestrained	Presence of sperm in one smear taken during daylight; rats girdled
5	2/20*	3/1	9	Yes	Yes
11	0/20	1/1	18	Yes	Yes
14	1/20	1/1	36	Yes	Yes
16†	27/20‡	2/1‡	0	No	No
17	1/20*‡	2/1	82	Yes	Yes
18	0/19	4/2	45	Yes	Yes
21	1/19‡	4/2	45	Yes	Yes
27	5/20	2/1	0	Yes	Yes
30	0/20	1/1	73	Yes	Yes
31	0/18	3/3	82	Yes	Yes

* Plugs were partly visible at the penis and were removed with forceps. † In this rat, each testis was about one-quarter the normal weight. Microscopic examination revealed no evidence of spermatogenesis. ‡ No sperm were found on microscopic examination.

The unoperated rat reveals a similar though lower incidence of spontaneous ejaculation. The data are based upon daily counts of seminal plugs deposited on the service pan which was lined with paper toweling. These plugs, consisting of coagulated semen, were verified microscopically, and the presence or absence of sperm was noted. Ten albino rats obtained from the National Institutes of Health were observed over a 21-day period. These rats were sexually mature and weighed 220 to 260 g. Four were caged in pairs, and the remaining six were caged individually. The differences in caging had no differentiating effects, and the data for all ten rats are presented in Table 1.

Six of the ten rats deposited at least one seminal plug during the period of observation. One rat, No. 16, deposited no less than one plug each day. This rat had inordinately small testes, each weighing 0.35 g, and there was no histological evidence of spermatogenesis. Microscopic examination of the plugs revealed that four rats had little or no sperm in their plugs (including No. 16), while the other six had plugs which were full of sperm.

Penile smears of the ten rats, taken on 11 successive days during daylight hours, were examined microscopically. The smears were prepared by passing a glass slide across the end of the exposed penis. In eight rats, sperm was indentified in at least one smear. Nocturnal smears, however, all contained sperm with the exception of the smear from rat No. 16. This led us to believe that all rats must have spermatic discharges quite frequently and that the lack of positive penile smears during daylight hours must be due to oral grooming of the genital region. Coupled with this, the more frequent deposition of plugs by the rats with brain lesions suggested that a seminal discharge might be occurring periodically in all rats, unoperated as well as operated, and that the more fastidious rats, the unoperated ones, tend to remove the evidence by oral grooming.

To test this hypothesis, we restrained the ten rats from grooming. Each rat was encased in a plastic tube which served as a stiff girdle around the thorax and abdomen, thus preventing the rat from bending to groom the genital region orally. Table 1 shows that the girdled rats deposited at least one seminal plug each day. All plugs contained sperm except those of No. 16. Under similar restraint, a single penile smear taken during daylight revealed a positive smear in all rats, again excepting No. 16. In order to rule out mechanical stimulation by the girdle as a factor in ejaculation we muzzled rats with adhesive tape and found a similar frequency of plugs. Our hypothesis that all rats have spontaneous daily seminal discharges, and that they frequently groom away the evidence, was thus confirmed (*4*).

We have made similar observations on two additional colonies of rats under unrestrained conditions. Seventy-six Sprague-Dawley rats were observed over a 10-day period with confirmatory results. Sixteen rats deposited at least

one plug each, all containing sperm, and 70 showed positive penile smears. A number of hooded Long-Evans rats, observed in a less systematic way, gave similar results.

These figures for the incidence of seminal plugs are probably conservative. Many plugs must go unrecognized because of their small size. Some are no bigger than the head of a pin, and are yellow and translucent, while others are as large as a kernel of corn. Sometimes a plug is missed because of its flat shape (probably fashioned by lying against the side of the ensheathed penis).

Spontaneous ejaculations first appear in hooded rats when they are between 50 and 60 days old. Sperm is often found in these first plugs and in the penile smears at the same age. Rats 16 months old, our oldest, continue to ejaculate spontaneously.

Occasionally a rat will deposit seminal plugs repeatedly, day after day for weeks. When paired with a female, such a male will continue to ejaculate without intromission (the vaginal smears of the cage mate reveal an absence of cornified cells and sperm, thus ruling out intromission).

After castration of rats, the frequency of ejaculation tapers and ceases within 7 to 10 days. (Sperm continues to appear in the initial postcastration plugs.) Conversely, a daily injection of testosterone propionate (0.5 mg) seems to reduce the incidence of seminal plugs. These results are contrary to the known androgenic control of male sexuality. However, there is little evidence on the hormonal control of grooming, and I am inclined to argue that the level of testosterone may determine the extent to which the rat will groom the genital region orally. This may explain why the rat with abnormally small testes (No. 16) seemed to produce the most semen, when unrestrained.

As regards autonomic involvement (*5*), seminal plugs were found within 30 min after the injection of epinephrine (0.07 mg subcutaneously), and within 4 min after the injection of mecholyl (5 to 8 mg subcutaneously). Under neither condition was an erection seen.

In order to rule out visual and olfactory cues in eliciting the seminal flow, we have redistributed rats in cages and rearranged cages in the rack with no effect on spontaneous ejaculation.

We have investigated other species but have not indentified sperm in the smears of any of them. Our samples included mice, rabbits, cats, and monkeys. Only the mice, when restrained from grooming the genital region orally, deposited seminal plugs.

We cannot distinguish the spontaneously produced plugs from those produced during copulation. This raises the issue whether fertile copulation need involve a *triggered* ejaculation following a series of intromissions. The multiple intromissions may serve the function of maintaining genital contact until a *spontaneous* ejaculation takes place. Alternatively, the spontaneous seminal discharges may represent an overflow of seminal fluid. In this sense, it may be possible to distinguish between "ejaculation" and "discharge" (*6*).

J. ORBACH

Institute for Psychosomatic and Psychiatric Research and Training, Michael Reese Hospital, Chicago, Illinois

References and Notes

1. F. A. Beach, "Characteristics of masculine 'sex drive,'" in *Nebraska Symposium on Motivation, 1956*, M. R. Jones, Ed. (Univ. of Nebraska Press, Lincoln, 1956), vol. 4, pp. 1–32.
2. L. R. Aronson, *J. Comp. and Physiol. Psychol.* **42**, 226 (1949).
3. N. B. Schwartz and A. Kling, *Federation Proc.* **20**, 335 (1961).
4. Oral grooming was not affected by topical application of t. valerian to the genitals and by anesthesia (10 mg Nembutal intraperitoneally).
5. J. H. Semans, and O. R. Langworthy, *J. Urol.* **40**, 836 (1938).
6. This study was supported by research grant No. M-3830 from the National Institute of Mental Health, U.S. Public Health Service. I am indebted to Dr. Neena B. Schwartz for advice, to Dr. Theodore Schafer for permission to make observations on his Sprague-Dawley rats, and to Anita F. Block for technical assistance.

14 August 1961

Immunization to Schistosoma mansoni in Mice Inoculated with Radiated Cercariae

Abstract. Preliminary experiments indicate that mice inoculated with cercariae of *Schistosoma mansoni* that have been exposed to cobalt-60 radiation in the range of 2500 to 3000 rep develop immunity to reinfection with nonradiated cercariae.

Resistance in mice and monkeys to reinfection with *Schistosoma mansoni* has been demonstrated by several investigators (*1*). Other reported studies include attempts to induce immunity by the inoculation of animals with immune serum, use of killed worms, and injection with extracts or metabolites of the worms (*2*). Instead of employing killed worms or metabolic products of *S. mansoni,* we thought that immunity might be induced by inoculation of cercariae exposed to an amount of radiation which would produce sexual sterilization and yet allow the cercariae to migrate in the tissues of the host in order to exert an antigenic stimulus.

Previous investigators have demonstrated that animals will develop immunity to reinfection with certain helminths after a primary infection with radiated larvae [*Trichinella spiralis* (*3*), *Dictyocaulus viviparus* (*4*), *Haemonchus contortus* (*4*), *Ascaris suum* (*5*)].

In the present investigation, cercariae from the Puerto Rican strain of the snail, *Australorbis glabratus,* were employed. In the initial experiment, 22 Swiss mice were divided into five groups: four groups of four mice each and one group of six mice. Each animal was inoculated intraperitoneally with 300 cercariae of mixed sexes. The first group received nonradiated cercariae. The second group received cercariae exposed to 1000 rep (roentgen equivalent physical); the third group, cercariae exposed to 2500 rep; the fourth group, cercariae exposed to 5000 rep; and the fifth group, cercariae exposed to 7500 rep. Eight weeks after the inoculation all the mice were killed and their livers and mesenteric veins were examined for schistosomes according to the method of Yolles *et al.* (*6*).

In a second experiment, five like groups of mice were similarly inoculated with nonradiated and radiated cercariae. After 8 weeks, each animal was given a challenging infection of 300 nonradiated cercariae of mixed sexes. The animals were then killed and examined for schistosomes 4 weeks after the challenging infection. At the time the inoculated mice were challenged, members of a sixth group of uninoculated mice each received 300 of the nonradiated cercariae.

In the first experiment, the mean (geometric) number of schistosomes per mouse recovered from each group receiving cercariae radiated with 0, 1000, 2500, 5000, and 7500 rep was 51.5, 29.0, 1.5, 0, and 0, respectively (Fig. 1, top). In the second experiment, the mean number of schistosomes recovered per mouse, after challenge, from groups first inoculated with cercariae radiated with 0, 1000, 2500, 5000, or 7500 rep was 46.4, 33.8, 6.6, 27.3, and 38.2, respectively. The mean