Sex Differences in Pentobarbital Sensitivity in Mice.* (28953)

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It has been shown that male rats are considerably less susceptible to barbiturate anesthesia (hypnosis) than female rats(1-3). As some of our observations in other research studies on sleeping time in mice receiving pentobarbital did not seem to follow the pattern reported in rats, a series of experiments was planned to establish whether there is a species difference between mice and rats in relation to sensitivity to pentobarbital.

Methods. Adult male and female mice of Swiss-Webster Strain (from the same breeding farm), age 9 to 12 months and weighing 24 to 40 g were used in this study. The average weight of all male mice was 34.5 g and that of all female mice was 28.1 g.

After preliminary tests, a dose of 5 mg/100 g was selected as optimum to promote anesthesia (hypnosis) in the mice used. Sleeping time was defined as time from injection until the mouse was able to right itself. Every 5 minutes the mouse was stimulated by pinching the tail with forceps to obtain a more accurate and meaningful end point. Forty-four males and 50 females were used under identical conditions. Since male mice slept much longer than females, additional experiments were performed. A comparison was made of sleeping times between control male mice and male mice (controls and experimental purchased as a group) receiving stilbestrol 12 days earlier. Sleeping times were measured for both groups a few days before administering the hormone and there was no significant difference between them (control mice 56.4 minutes, mice to receive stilbestrol 53.8 minutes, $p > 0.45$). Then the control group received saline injections and the experimental group received sodium diethylstilbestrol diphosphate (0.289 mg/g) 12 days prior to the next sleeping time determination.

In similar fashion 20 females were divided into 2 groups of 10 each (control mice 21.6 minutes, mice to receive testosterone 20.9 minutes, $p > 0.80$). One group received saline injections and the other received testosterone enanthate in sesame oil (U.S.P.) (1.476 mg/g).

Average weight changes in both treated and control mice during the 12 days differed by less than 0.5 g.

Statistical analyses were done using student's "t" test.

Results. Fig. 1 shows a summary of the data. The 44 male mice slept an average of 70.5 minutes whereas, the group of 50 female mice slept only 24.9 minutes ($p<0.0005$). Fig. 2 shows that the male mice receiving stilbestrol slept only 31.6 minutes as compared to the control group of male mice which slept 53.8 minutes ($p<0.008$). The group of female mice which received testosterone slept 42 minutes as compared to 20.9 minutes ($p<0.002$) for the control group receiving no testosterone.

This indicates that male mice of Swiss-Webster strain are more susceptible to the effects of pentobarbital sodium than are female mice of that strain. Furthermore, female mice receiving testosterone are depressed longer than control groups and males receiving stilbestrol sleep for shorter periods than control male mice.

Discussion. Although no work was done during this investigation on the mechanism of action of the injected hormones in influencing the sleeping time in mice, it is interesting that the results were the reverse of those found in rats. This was especially noteworthy since some investigators have suggested that the testosterone which decreased sleeping time in castrated male rats presumably did so by increasing the liver detoxification rate of barbiturate. If that should be true in the rat, it leaves some

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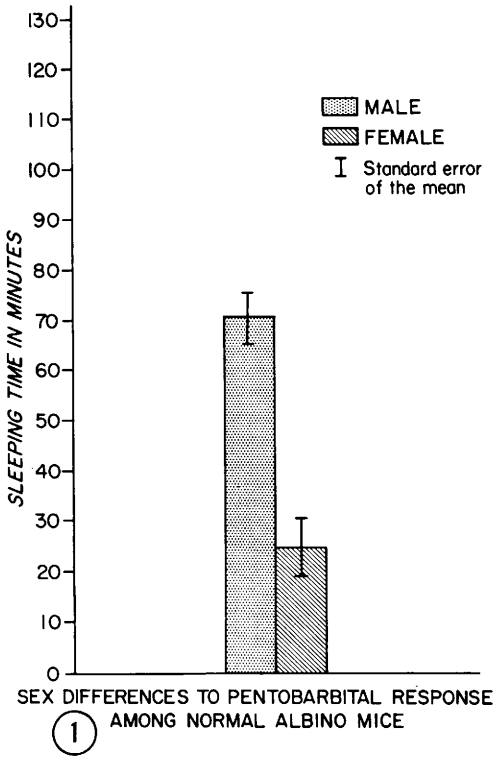

FIG. 1. Sex differences in pentobarbital sensitivity indicated by longer sleeping time in male mice (44 animals) compared to females (50 animals).

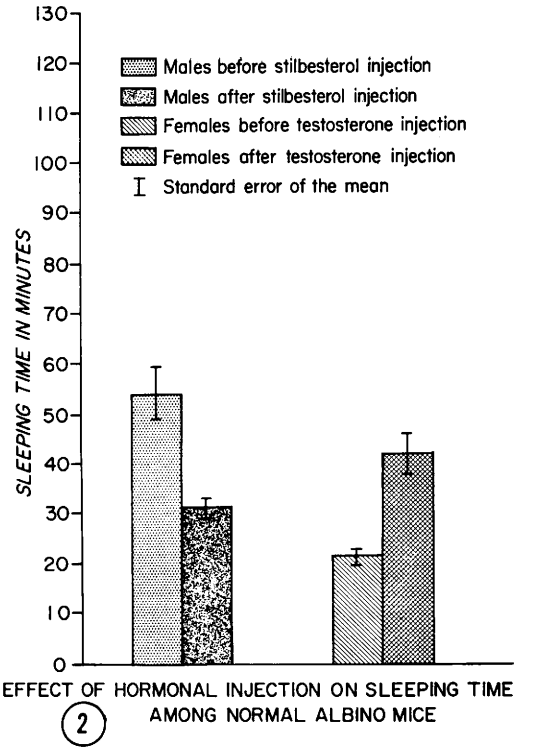

FIG. 2. Male mice (10 animals) pretreated with stilbestrol slept less than control males (10 animals). Female mice (10 animals) pretreated with testosterone slept longer than control females (10 animals).

pertinent questions. For example, is the fate of barbiturates different in mice and rats? One problem posed by our findings concerns the mechanism whereby the injected hormones modify the sleeping times. There are several possibilities. They may act directly on the central nervous system, altering the sensitivity to barbiturates. They may act on the liver, modifying the patterns of detoxification. Another possibility is that they may alter the degree to which barbiturates are absorbed by cells and fluids outside the central nervous system.

Another important aspect of these findings is the demonstration again of the need for extreme caution in extrapolation from one species to another, even when the 2 species are so close as rats and mice.

Conclusion. 1. Male albino mice sleep longer than female mice after identical dosage of pentobarbital sodium. 2. Injection of stilbestrol into male mice 12 days prior to pentobarbital hypnosis, shortened sleeping time significantly. 3. Injection of testosterone into female mice 12 days prior to pentobarbital hypnosis, prolonged sleeping time significantly.

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