Pharmacology Biochemistry & Behavior, Vol. 21, pp. 675–677, 1984. © Ankho International Inc. Printed in the U.S.A. 0091-3057/84 $3.00 + .00

BRIEF COMMUNICATION

Effect of Sex and Castration on Nicotine-Induced Activity Responses

TERRY CRONAN, REBECCA BRYSON AND EILEEN McNAIR

Department of Psychology, San Diego State University, San Diego, CA 92182-0350

Received 15 December 1983

CRONAN, T., R. BRYSON AND E. McNAIR. *Effect of sex and castration on nicotine-induced activity responses.* PHARMACOL BIOCHEM BEHAV **21**(4) 675–677, 1984.—The nature of the short-term interactions between nicotine and sex hormones in affecting activity remain unclear. The present study was an investigation of these effects. Three levels of nicotine injection—0.0 (saline control), 0.2 mg/kg body weight, and 0.4 mg/kg body weight—were given to male, female, and castrated Sprague-Dawley albino rats. Activity was measured when the animals were 6 weeks and 12 weeks old. Nicotine produced an initial depression of activity relative to the saline control levels, and a later activation which peaked at 40 to 60 minutes. The biphasic effect was most striking in female animals, but the *forms* of the curves relating activity to time in all groups were quite similar.

Psychopharmacology Nicotine Activity Sex differences

NICOTINE tends to reach high levels in the brains of female rats earlier than in males [7], and mature females are more active in response to nicotine than are mature males [1,6]. There is a tendency for young animals, regardless of sex, to show more pronounced nicotine-related increases in activity than do mature males [1].

An age-related decrease in the rate of nicotine metabolism in males has also been demonstrated [3]. Castrated males had liver nicotine levels similar to those in intact *females*, while ovariectomy had no effect on liver nicotine in females. Androgen levels may affect nicotine metabolism, and hence underlie the sex-related dosage effects mentioned above [1].

Activity levels depend upon the dosage of nicotine, sex, and time since the dose was administered [4, 5, 8]. High levels of nicotine may depress activity in both sexes for as long as 2 hours post-injection. Moderate and low dosage levels (0.2 to 0.4 mg/kg bw) may produce a slight initial depression, followed by heightened activity. Male and female dose-response curves may differ [3], and females become more like males after an injection of testosterone.

Castrated mature males should have less testosterone and hence metabolize nicotine faster than unaltered males. This difference should be apparent at sexual maturity, around 12 weeks of age. If females metabolize nicotine more rapidly, they should manifest an earlier rise in activity following injection, followed by a more rapid decline. Castrated males' response curves should more nearly parallel the females'.

The present study was a test of these suppositions, and further addressed the question of how long injections of nicotine would affect activity.

METHOD

Subjects

The subjects were 33 experimentally naive Sprague-Dawley-derived rats: 11 males, 11 females, and 11 castrated males. Each rat was anesthetized with 0.05 cc of ketamine hydrochloride, and subjected to sham or to real surgery at 21 days.

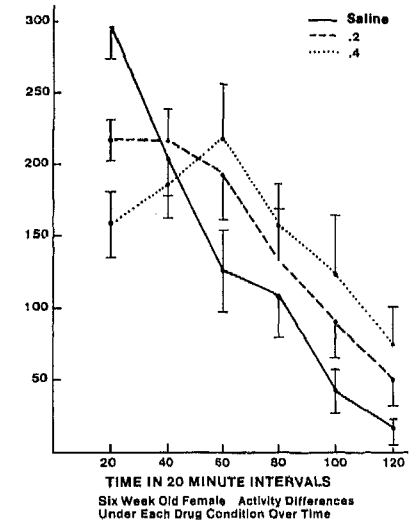
Apparatus

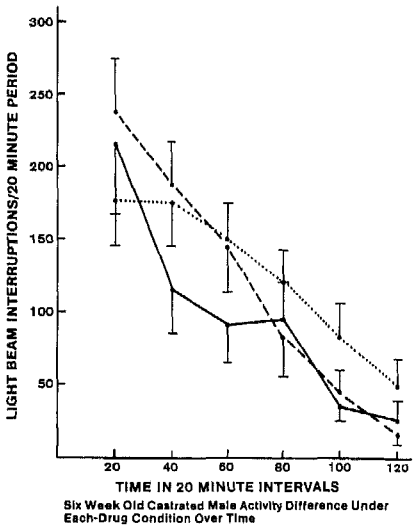
Twenty-four electric photocell chambers (40×20×25 cm high) were used. Walls and floors were unfinished wood approximately 1 cm thick, with a wire mesh floor raised 2.4 cm above the base platform, and a hinged wire mesh lid. One photocell and two mirrors were used to create two horizontal parallel light beams 7 cm from either side and 5 cm above the mesh floor. All of the photocell chambers were interfaced with an Apple II computer, which recorded the number of light beam interruptions within each 20-minute period for each of the 2-hour observation periods.

Procedure

Observations were made when the animals were 6 weeks old and again when they were 12 weeks old. Lights in the home cage room remained on between 7 a.m. and 7 p.m. The temperature was maintained at 21 degrees Celsius.

Activity was measured on each group of rats for a 120-min post-injection time for three consecutive days at 6 weeks, and again at 12 weeks, of age. Within the 3-day period, each





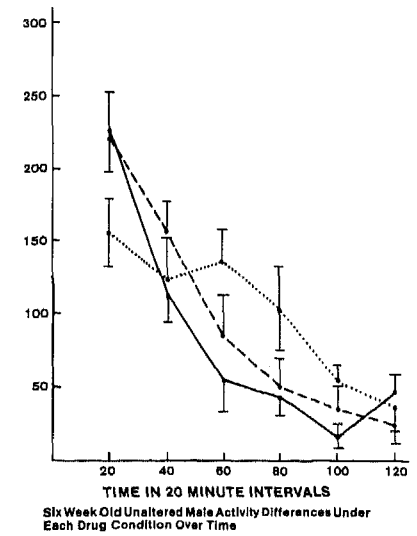
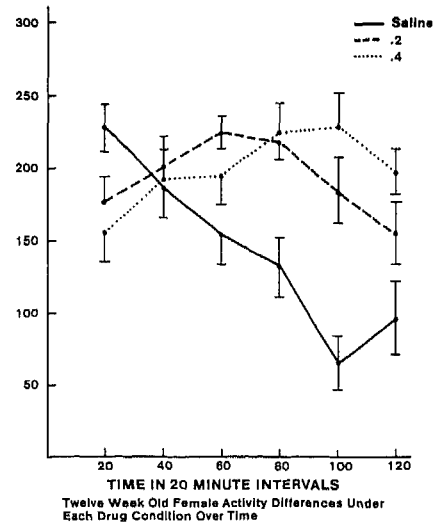
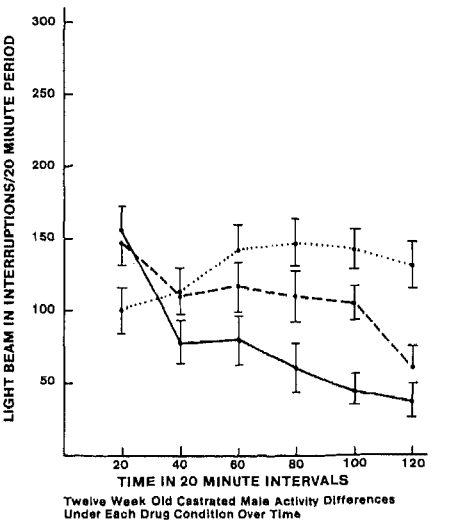


FIG. 1. Mean number of light beam interruptions per 20 minute period for six week old female, castrated, and unaltered males. Each animal received one injection of saline, 0.2 mg/kg of nicotine, and 0.4 mg/kg of nicotine. Activity was measured for two-hour periods.





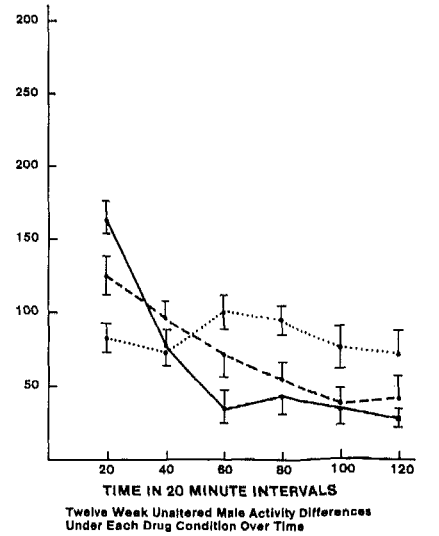


FIG. 2. Mean number of light beam interruptions per 20 minute period for twelve week old female, castrated, and unaltered males. Each animal received one injection of saline, 0.2 mg/kg of nicotine, and 0.4 mg/kg of nicotine. Activity was measured for two-hour periods.

rat received each subcutaneous injection once: 0.4 mg/kg bw of 98 to 100% of pure liquid nicotine, distributed by Sigma Chemicals, dissolved in physiological saline, 0.2 mg/kg bw of nicotine dissolved in physiological saline, and 0.9% physiological saline. Order of dosage amounts was counterbalanced within each group. Injection volume was always 0.1 ml/250 g of body weight, calculated as the base.

After injection, all animals were immediately placed in a photocell chamber. General activity was measured for six consecutive 20-minute intervals. Each subject was tested in the same photocell chamber each day under dark conditions. An electronic device which emitted white noise at a constant decibel level was used to control for ambient noise. Testing was begun at 11:00 a.m. each day, and was finished by shortly after 1:00 p.m.

RESULTS

A sex (S, female vs. castrated male vs. unaltered male) × age (A, 6 weeks vs. 12 weeks) × dosage (D, saline vs. 0.2 mg/kg bw vs. 0.4 mg/kg bw) × interval (I, time in six 20-minute increments) mixed analysis of variance was performed on the mean activity levels. The S by D by I interaction was significant, $F(20,300)=2.150$, $p<0.01$. This interaction can be portrayed by combining Figs. 1 and 2 by collapsing over age. The S by I interaction was also significant $F(10,150)=2.807$, $p<0.01$, as was the main effect of sex $F(2,30)=20.723$, $p<0.01$. Females were more active than castrated or unaltered males, and castrated males were more active than unaltered males during each 20-minute interval; however, these latter differences were significant only at the 60-, 80-, and 100-minute readings. The D by I interaction was significant, $F(10,300)=20.120$, $p<0.01$, as was the main effect for D, $F(2,6)=12.075$, $p<0.01$. Overall, the rate of decrease in activity as a function of time was attenuated in a dose-related fashion by nicotine. In addition, at the high dose (0.4 mg), activity at some time intervals actually increased compared to previous intervals.

Collapsing over sex in Figs. 1 and 2 produces the significant D by A by I interaction, $F(10,300)=2.080$, $p<0.05$. Nicotine initially depressed activity relative to saline at both six and twelve weeks, and increased it during later intervals. There were no differences at the 60-minute interval between animals at six and twelve weeks, but six-week readings were higher during the first two intervals, 12-week readings were higher during the last three intervals, and there were clearer differences as a function of injection condition at 12 weeks. This A by I interaction, $F(5,150)=33.316$, $p<0.01$, and I main effect, $F(5,150)=102.91$, $p<0.01$, were significant. Differences in activity from each interval to the subsequent one were significant during the first hour, but not during the second.

Finally, the S by A interaction was significant, $F(2,30)=6.181$, $p<0.01$. Females were more active than males at both ages; castrated males were more active than unaltered males at twelve, but not at six, weeks. Females were more active at 12 weeks than they had been at six weeks.

DISCUSSION

As expected, females were more active than males, and castrated males were intermediate in activity. Activity decreased more over a session in 6-week-old animals, starting higher and ending lower during each session for the younger animals.

Nicotine had a stable initial depressive effect on activity, which occurred in every case except that of the castrated males injected with 0.2 mg/kg bw. In all cases the initial depression was followed by an increase in activity relative to controls, continuing until the 120-minute reading. These results are similar to those found previously; however, the previous studies employed only male animals [2,5]. In the present study the consistent biphasic effect was independent of sex, but, particularly in animals injected with 0.4 mg/kg bw of nicotine, its strength depended on sex. It was strongest in females and weakest in males. The castrated male curves did not differ markedly from those of the unaltered males.

It has been suggested [3] that testosterone decreases the rate of nicotine metabolism. If this were true, we would expect males to show a longer-lasting increase in activity than females. In the present study, the activity peaks for males and females occurred at the same time, and the general shapes of their curves were not markedly different. Although blood levels of nicotine were not measured in this study, the activity results constitute presumptive evidence against the hypothesis that testosterone inhibits the metabolism of nicotine.

Nicotine had a longer-lasting effect in 12-week-old animals than in 6-week-old animals. This could be produced either by changes in nicotine metabolism with age, or by greater emotionality in younger animals as a result of being injected and placed in the boxes. In any event, activity levels decreased more rapidly in younger animals, and older animals more nearly maintained their activity level throughout the 2-hour period.

REFERENCES

1. Bryson, R., P. M. Biner, E. McNair, M. Bergondy and O. Abrams. Effects of nicotine on two types of motor activity in rats. *Psychopharmacology (Berlin)* **73:** 168–170, 1981.
2. Clarke, P. B. S. and R. Kumar. The effects of nicotine on locomotor activity in non-tolerant and tolerant rats. *Br J Pharmacol* **78:** 329–337, 1983.
3. Hatchell, P. C. The pharmacological and behavioral effects of nicotine in inbred mice (Doctoral dissertation, University of Colorado, 1976). *Diss Abstr Inter* **37:** 1521B–1522B, 1976.
4. Meliska, C. J., K. W. Fitzpatrick and J. E. Rosine. Effects of nicotine on spontaneous motor activity in rats having high and low basal activity levels. *Physiol Psychol* **2:** 487–489, 1974.
5. Morrison, C. F. and P. N. A. Lee. A comparison of the effects of nicotine and physostigmine on a measure of activity in the rat. *Psychopharmacologia* **13:** 210–221, 1968.
6. Pereboom, A. C. Systematic-representative study of spontaneous activity in the rat. *Psychol Rep* **22:** 717–732, 1968.
7. Rosecrans, J. A. Brain area nicotine levels in male and female rats with different levels of spontaneous activity. *Neuropharmacology* **11:** 863–870, 1972.
8. Stolerman, I. P., R. Fink and M. E. Jarvik. Acute and chronic tolerance to nicotine measured by activity in rats. *Psychopharmacologia* **30:** 329–342, 1973.