Pergamon

Pharmacology Biochemistry and Behavior, Vol. 50, No. 2, pp. 191–198, 1995

0091-3057/95 $9.50 + .00

0091-3057(94)00285-1

Strain and Age Differences in Acoustic Startle Responses and Effects of Nicotine in Rats[1]

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Received 23 February 1994

ACRI, J. B., K. J. BROWN, M. I. SAAH AND N. E. GRUNBERG. *Strain and age differences in acoustic startle responses and effects of nicotine in rats.* PHARMACOL BIOCHEM BEHAV **50**(2) 191–198, 1995. – Two experiments examined the effects of age, genetic strain, and nicotine on acoustic startle response (ASR) amplitude and prepulse inhibition (PPI) in rats. ASR amplitude measures reactivity to external stimulation, and PPI is used as an index of sensory gating related to attention. Both ASR amplitude and PPI have been previously reported to be increased by nicotine in adult rats. Experiment 1 examined effects of chronically administered nicotine and saline on ASR and PPI in Wistar, Long-Evans, and Sprague-Dawley rats (40 days of age). Experiment 2 examined the effects of chronically administered nicotine and saline in Sprague-Dawley rats of two age groups: 40 and 70 days of age at the beginning of the study. ASR amplitude differed significantly across strains with the values for Wistar > Sprague-Dawley > Long-Evans, and there were no differences in percent of PPI among the three strains. In addition, results of Experiment 2 indicated that older rats had significantly greater ASR amplitudes and PPI than younger rats. Consistent with previous reports, nicotine increased ASR and PPI in the older rats; however, there were no significant differences in the younger rats. Therefore, age and genetic strain are important variables in the analysis of nicotine's effects on startle behaviors in rats.

Acoustic startle Sensory gating Sensorimotor reactivity Genetic strain Rat Age Nicotine

THE ACOUSTIC startle response (ASR) is an unconditioned behavioral measure of reactivity to external stimulation in mammals (12) that has been reported to be sensitive to changes in attentional processes in humans (4). In rats, nicotine has been reported to modulate the ASR (2,3), and to increase prepulse inhibition (PPI) of the ASR (1,3). PPI occurs when a less intense stimulus precedes a startle-eliciting stimulus by a brief interval, and the ASR amplitude is reduced. PPI has been used as a model for neural mechanisms underlying processes of sensory gating related to attention (31). Because nicotine has effects on attention [see (21) for review], it has been suggested that the ASR may provide a useful model for further study of nicotine effects on attentional processes (1-3).

Age and genetic strain of experimental subjects are typically reported in studies of drug effects, but the rationale for use of a particular age or strain is rarely specified. There is evidence that these parameters may influence the degree and direction of drug effects on certain behavioral and physiological measures. Strain differences have been reported in effects of nicotine on measures of locomotor activity in rats (13,28). In mice, effects of nicotine on ASR may differ in direction for different genetic strains (9,10,22). Further, different strains of rat differ in response to acoustic startle measures of amplitude, prepulse inhibition, and habituation (11,14,23,26,27, 32). For some mouse and rat strains, there are known biochemical and behavioral characteristics to account for experimental differences in various behaviors or in response to particular drugs, such as an absence of a particular receptor subtype (33), selective breeding for specific behaviors (5), or supersensitivity to specific neurotransmitters (25), but for other strains, the mechanisms accounting for behavioral differences remain unknown.

[1] This research was supported by USUHS protocol CO 7223. The views contained herein are the private ones of the authors and do not reflect those of the Uniformed Services University of the Health Sciences, the Department of Defense, the University of Virginia, or the National Institute on Drug Abuse.

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TABLE 1

ASR AMPLITUDES (GRAMS, MEANS AND SEM) FOR THREE GENETIC STRAINS ADMINISTERED NICOTINE OR SALINE

		Genetic Strain		
Time	Drug	Wistar (n = 20)	Long-Evans (n = 20)	Sprague-Dawley (n = 20)
115 dB				
Baseline	Saline	31.1 ± 7.0	17.1 ± 2.1	22.1 ± 2.6
	Nicotine	32.0 ± 5.6	15.8 ± 2.5	21.7 ± 2.1
Early drug	Saline	60.7 ± 8.3	27.5 ± 4.9	43.3 ± 6.7
	Nicotine	66.8 ± 4.7	20.9 ± 3.0	35.0 ± 6.0
Late drug	Saline	92.15 ± 13.2	35.8 ± 8.2	68.1 ± 10.0
	Nicotine	97.0 ± 14.9	42.9 ± 5.6	58.2 ± 12.6
Cessation	Saline	106.55 ± 14.8	36.8 ± 8.3	90.1 ± 11.7
	Nicotine	111.3 ± 15.0	50.1 ± 9.9	58.6 ± 9.7
122 dB				
Baseline	Saline	36.8 ± 6.0	18.9 ± 2.2	41.2 ± 4.9
	Nicotine	52.8 ± 8.3	20.7 ± 2.1	43.1 ± 539
Early drug	Saline	76.5 ± 7.7	35.6 ± 6.4	73.5 ± 9.5
	Nicotine	104.6 ± 13.2	37.3 ± 5.5	84.9 ± 18.7
Late drug	Saline	123.4 ± 14.4	46.4 ± 10.4	115.8 ± 8.9
	Nicotine	152.6 ± 18.0	65.4 ± 10.1	105.9 ± 23.3
Cessation	Saline	128.5 ± 14.9	52.6 ± 9.9	135.6 ± 19.3
	Nicotine	165.1 ± 16.7	66.6 ± 6.1	90.4 ± 20.3

Documentation of age differences in response to drugs in adult animals has been largely anecdotal, but it has been reported that drug effects are greater in older guinea pigs administered apomorphine (8) and older rats administered cocaine (19). Acoustic startle measures without drug have been reported to differ in humans (4,15,17,24) and rats (7,20,27) according to age, but there are no reports in which drug effects on ASR are examined in rats of different age ranges.

We have previously reported that chronic or acute nicotine increases ASR amplitude and PPI in Sprague-Dawley rats over 70 days of age (1-3), but there have been other reports that nicotine has no effect on ASR amplitude (16,30). The present experiments assessed the effects of nicotine in young rats of three different strains, and in young and mature rats of one strain. These ages and strains were selected to resolve inconsistencies in reports of nicotine's effects, to determine whether rats of different ages and strains respond differently to nicotine, and to establish optimal subject characteristics for further study of the effects of nicotine on acoustic startle measures.

EXPERIMENT 1

This experiment was designed to assess effects of nicotine in young rats of three genetic strains commonly used in behavioral pharmacology: Wistar, Long-Evans hooded, and Sprague-Dawley rats. Dependent measures were ASR amplitude, amount, and percent of PPI, which we have previously reported to be increased by acute and chronic nicotine in Sprague-Dawley rats (1-3).

METHOD

Subjects

Subjects were 60 experimentally naive male rats of approximately 40 days of age at the beginning of the experiment. There were 20 rats of each of three strains: Wistar (mean weight 195 g), Long-Evans hooded (mean weight 185 g), and Sprague-Dawley (mean weight 187 g). Rats were singly housed in standard plastic cages (35.6 × 15.2 × 20.3 cm) over Pine-Dri bedding with rat chow and water continuously available throughout the experiment, resulting in slightly higher weights per age than published norms. Lights were on a 12L : 12D cycle with light on 0700 to 1900 h, and temperature was maintained at ~22°C and 50% relative humidity.

Drug

Saline (0.9% NaCl) or nicotine (12 mg/kg/day) was administered by Alzet minipumps (Model 2002, Alza Corporation, Palo Alto, CA), implanted between the shoulders under sterile conditions with methoxyflurane anesthesia. These procedures have been reported in detail elsewhere (1,2). Nicotine dihydrochloride was dissolved in saline and delivered by minipump at the rate of ~5 μl/h. The dosage is expressed as nicotine base, and was selected on the basis of previous experiments in which this dosage resulted in significant increases in ASR amplitude (1,2). Minipumps were removed by the same surgical and anesthetic procedures after 13 days to ensure drug cessation.

Startle Response Testing

ASR amplitudes and PPI were measured in a four-station startle chamber (Coulbourn Instruments, Lehigh Valley, PA). Each animal was individually placed in a 8 × 8 × 16 cm open air cage that restricted locomotion, and was placed on one of four sensor platforms within a sound-attenuating chamber. Platforms were arranged radially around speakers in the floor and ceiling of the chamber. A ventilating fan provided an ambient noise of 50 dB SPL (re: 0.0002 dynes/cm^2). There was a 3-min quiet adaptation period following placement of rats within the chamber. Startle pulses consisted of 115 dB

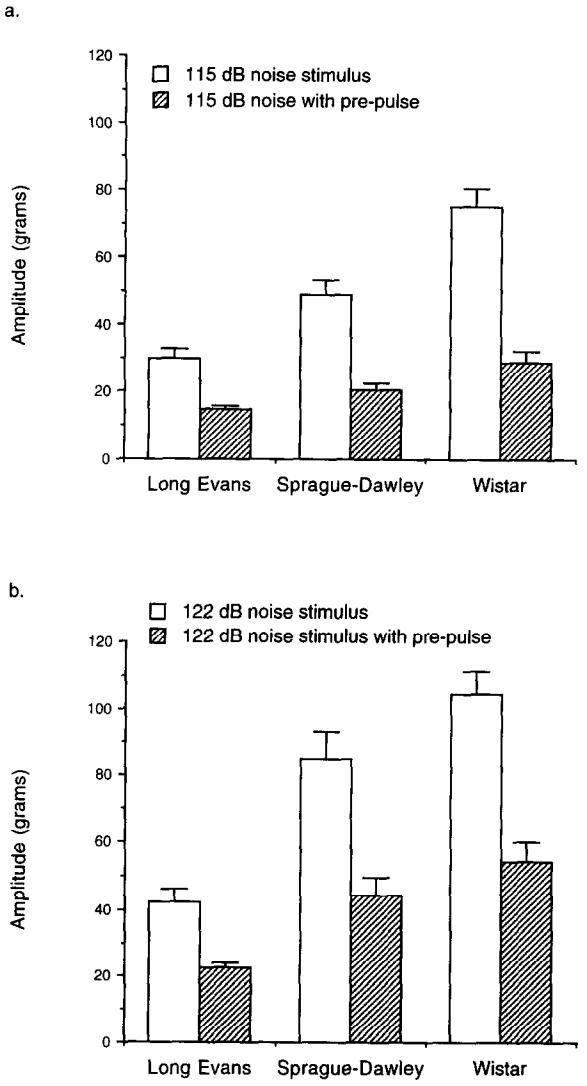

FIG. 1. Acoustic startle response amplitudes (means and SEMs) for Wistar, Long-Evans, and Sprague-Dawley rats collapsed across drug and days of drug administration and cessation (open bars). Response amplitudes for PPI trials consisting of startle stimuli preceded by 70-dB prepulse (means and SEMs) for three strains collapsed across drug and days of drug administration (hatched bars). Upper panel illustrates responses to 115-dB stimuli; lower panel illustrates responses to 122-dB stimuli.

and 122 dB SPL noise bursts sometimes preceded 100 ms by 70 dB 1 kHz prepulses. These intensity levels were verified with a GenRad Type 1982 sound level meter with microphone placement in the position of a subject's head. There were 10 of each of four types of stimulus trials, presented in a randomized block design. The trial types consisted of 115-dB noise bursts alone and with prepulses, and 122-dB noise bursts alone and with prepulses. There were also 10 prepulse-alone trials and five no-stimulus trials for a total session of 55 trials. Interstimulus intervals ranged randomly between 10 and 20 s.

Following presentation of each stimulus, an animal's movement was measured for a period of 200 ms by coupling through a sensor pin connected to a strain gauge within the platform under each animal. Four platforms were measured simultaneously by a microcomputer that controlled both the stimulus presentation schedule and recorded the digitized responses. Amplitudes were recorded as the maximum response occurring within the 200-ms window following stimulus presentation, and were converted to grams. The no-stimulus control value was subtracted from the maximum response recorded for each trial type, and each animal's responses were then averaged within trial types. Platforms were calibrated for accuracy and linearity with a series of weights (50–500 g) prior to each use.

Procedure

Animals were handled and weighed during the baseline period and were tested for startle response amplitudes. Within each strain, animals were quasirandomly assigned to drug condition according to body weight and baseline startle amplitudes so that drug conditions within each strain had equal means and variance for both body weight and ASR amplitude. Minipumps were implanted as above, and ASR was tested on days 1, 5, 8, and 12 of drug administration. Pumps were explanted on day 13, and ASR was tested on days 1 and 5 of drug cessation.

Data Analysis

Amount of prepulse inhibition was calculated as the difference in amplitude between trials with and without prepulses, within a given stimulus intensity. Percent of inhibition was calculated as [(amount of inhibition/amplitude of response without prepulse) × 100] for each stimulus intensity. Although there is no consensus on the appropriateness of either measure of PPI, it is reasonable that when there are differences in ASR amplitude, differences in percent of PPI should be calculated to yield results that are not confounded by the change in amplitude itself. Thus, both measures are presented. Results were analyzed separately by trial types and amount and percent of inhibition in a three-factor (drug, strain, time) repeated-measures analysis of variance (ANOVA) and analysis of covariance (ANCOVA) to control for baseline differences. Results for all trial types were collapsed into blocks consisting of baseline (days 1 and 3 of baseline), early drug (days 1 and 5 of drug administration), late drug (days 8 and 12 of drug administration), and cessation (days 1 and 5 after explantation). Post hoc Student-Newman-Keuls comparisons were used to determine which groups differed at specific time points, and all analyses utilized two-tailed distributions and a significance level of 0.05.

RESULTS

Table 1 presents the mean ASR amplitude using 115- and 122-dB stimulus intensities for the three rat strains. Repeated-measures ANOVA for amplitude using 115-dB stimuli indicated a significant main effect of strain, $F(2, 53) = 22.23$, $p < 0.05$, with significant differences between Wistars and Sprague-Dawley rats, and between Wistars and Long-Evans hooded rats at baseline, and significant differences among all groups at all other time points ($p < 0.05$). With baseline differences controlled by ANCOVA, significant strain differences remained, $F(2, 52) = 12.37$, $p < 0.05$, with Wistar > Sprague-Dawley > Long-Evans. There were no significant effects of drug, and there were no interactions. Figure 1a includes mean ASR amplitude in response to 115 dB for each strain collapsed across drug and time points.

TABLE 2

AMOUNT AND PERCENT OF PREPULSE INHIBITION IN THREE GENETIC STRAINS ADMINISTERED NICOTINE OR SALINE

		Genetic Strain		
Time	Drug	Wistar (n = 20)	Long-Evans (n = 20)	Sprague-Dawley (n = 20)
Baseline	Saline	7.0 ± 2.4	5.4 ± 1.8	4.9 ± 1.5
		(10.0 ± 13.4)	(16.2 ± 10.5)	(9.9 ± 8.4)
	Nicotine	11.4 ± 3.3	3.6 ± 1.5	4.2 ± 0.9
		(8.0 ± 8.2)	(8.0 ± 9.9)	(7.1 ± 6.9)
Early drug	Saline	27.8 ± 5.7	11.5 ± 3.3	24.3 ± 5.8
		(17.9 ± 7.9)	(17.9 ± 8.7)	(42.2 ± 5.9)
	Nicotine	36.7 ± 6.9	7.9 ± 1.6	11.9 ± 2.7
		(43.5 ± 8.4)	(65.7 ± 44.1)	(30.9 ± 6.9)
Late drug	Saline	59.3 ± 7.9	16.3 ± 6.4	37.0 ± 8.0
		(70.7 ± 2.7)	(28.6 ± 13.5)	(49.0 ± 11.5)
	Nicotine	46.0 ± 10.8	19.6 ± 4.1	31.3 ± 6.8
		(55.2 ± 8.6)	(42.2 ± 15.5)	(66.8 ± 11.9)
Cessation	Saline	75.1 ± 11.4	22.6 ± 7.4	68.0 ± 10.8
		(71.7 ± 3.0)	(39.4 ± 11.3)	(71.6 ± 5.4)
	Nicotine	80.0 ± 11.1	28.6 ± 6.0	38.6 ± 7.9
		(66.0 ± 7.0)	(58.6 ± 11.6)	(66.6 ± 3.3)

Stimuli were 115-dB noise bursts preceded by 70-dB prepulses. Means and SEMs for percent of inhibition are shown in parentheses directly below means and SEMs for amount of inhibition.

Similar results were obtained using the 122-dB stimulus (Table 1). Repeated-measures ANOVA for this stimulus revealed a significant main effect of strain, $F(2, 53) = 20.72$, $p < 0.05$, with significant differences at all time points. These results are presented in Fig. 1b. There were no significant effects of drug on startle amplitude using either 115 or 122 dB, and there were no interactions.

Table 2 presents mean amount and percent of PPI across days of drug administration using the stimulus intensity of 115 dB preceded by 70-dB prepulses. Repeated-measures ANOVA for amount of inhibition revealed a significant effect of strain, $F(2, 53) = 17.33$, $p < 0.05$, with significant differences between Wistars and Sprague-Dawley rats, and between Wistars and Long-Evans hooded rats at baseline, and between Sprague-Dawley and Long-Evans hooded rats at early drug and cessation ($p < 0.05$). With baseline differences in amount of inhibition controlled by ANCOVA, a significant effect of strain on amount of inhibition remained (Wistar > Sprague-Dawley > Long-Evans).

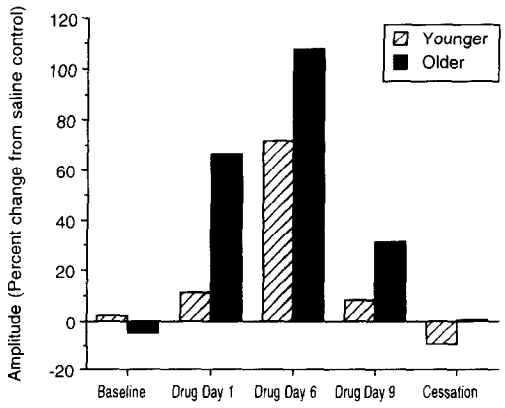

FIG. 2. Effects of nicotine on ASR amplitude in response to 115-dB noise bursts are shown across days of drug administration and cessation (days 1, 3, and 6 of cessation were combined) for younger (hatched bars) and older (solid bars) rats. Significant effects of nicotine occurred on days 1 and 6 of drug administration. For clarity, response amplitudes of nicotine-treated rats are expressed as percent change from saline control value for each age group at each time point (0 = saline control value).

When calculated as percent of inhibition to control for differences in startle amplitude, there were no significant effects of strain. Raw amplitudes of PPI trials (without calculation of amount or percent of PPI) for the three strains, collapsed across drug conditions, are presented in Fig. 1a.

Similar results were obtained using the 122 stimulus preceded by 70-dB prepulses (Fig. 1b). Repeated-measures ANOVA for this stimulus revealed a significant main effect of strain on amount of inhibition, $F(2, 53) = 20.72$, $p < 0.05$, with significant differences among strains at all time points. Differences in percent of inhibition were not significant. Raw amplitude of PPI trials for 122 dB are presented in Fig. 1b.

In contrast to previous studies that used older rats (1–3), there were no significant differences in effects of drug or interactions between drug and strain on ASR or PPI. Therefore, Experiment 2 was designed to compare the effects of nicotine on ASR and PPI in rats that were a similar age and strain to those used in previous reports with effects of nicotine in rats that were of a similar age to those in Experiment 1.

EXPERIMENT 2

This experiment assessed the effects of nicotine in Sprague-Dawley rats of two different age categories. This was done to explore the possibility that nicotine did not produce changes

TABLE 3

ASR AMPLITUDES (GRAMS, MEANS AND SEM) FOR YOUNGER AND OLDER RATS ADMINISTERED NICOTINE OR SALINE

		115 dB		122 dB	
Time	Drug	Younger (n = 16)	Older (n = 16)	Younger (n = 16)	Older (n = 16)
Baseline	Saline	24.4 ± 3.3	44.3 ± 10.0	85.3 ± 9.8	148.1 ± 11.7
	Nicotine	25.0 ± 3.6	42.3 ± 7.3	67.0 ± 12.8	132.8 ± 18.6
Drug day 1	Saline	14.6 ± 2.9	21.2 ± 7.1	56.6 ± 27.9	79.7 ± 29.3
	Nicotine	16.3 ± 2.6	35.3 ± 14.3	65.3 ± 18.8	92.4 ± 27.4
Drug day 6	Saline	22.4 ± 3.2	27.0 ± 5.5	113.8 ± 23.1	128.0 ± 17.9
	Nicotine	38.4 ± 10.5	56.0 ± 16.3	105.5 ± 14.6	142.1 ± 22.7
Drug day 9	Saline	31.6 ± 6.3	38.6 ± 6.4	101.9 ± 21.1	155.6 ± 20.6
	Nicotine	34.3 ± 8.0	50.6 ± 13.4	119.4 ± 18.5	181.9 ± 14.4
Cessation	Saline	35.6 ± 5.5	51.5 ± 7.3	139.0 ± 29.3	145.6 ± 16.3
	Nicotine	32.2 ± 4.8	51.6 ± 9.8	119.0 ± 12.7	128.6 ± 18.2

in ASR and PPI in Experiment 1 because the rats used were younger than those used in previous experiments (1-3). Dependent measures in Experiment 2 were ASR amplitude, amount, and percent of PPI.

METHOD

Subjects

Subjects were 32 experimentally naive male Sprague-Dawley rats of two different age categories. At the beginning of this experiment, 16 rats were 39–42 days of age (mean weight 187 g) as in Experiment 1, and 16 rats were 67–74 days of age (mean weight 350 g) as in previous reports (1-3). Individual housing conditions were identical to those of Experiment 1, and resulted in slightly greater body weights than published norms.

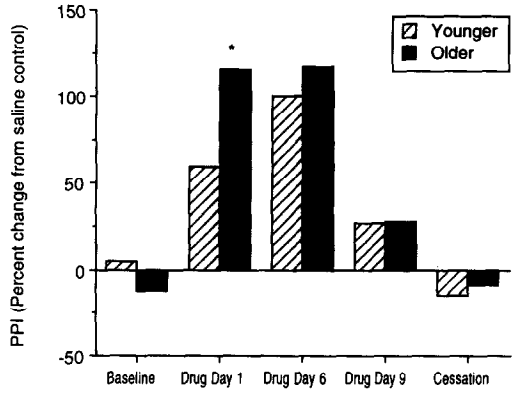

FIG. 3. Effects of nicotine on amount of PPI in response to 115-dB noise bursts preceded by 70-dB prepulses are shown across days of drug administration and cessation for younger (hatched bars) and older (solid bars) rats. Significant effects of nicotine occurred on day 1 of drug administration, and the asterisk indicates older animals administered nicotine had a significant increase in percent of PPI compared to saline-treated animals of the same age. For clarity, data of PPI amount are expressed as percent change from saline control value for each age group at each time point (0 = saline control value).

Drug

Saline (0.9% NaCl) or nicotine (12 mg/kg/day) was administered by Alzet minipumps (Model 2002, Alza Corporation), as in Experiment 1. The minipumps were removed after 10 days to ensure drug cessation.

Startle Response Testing

ASR amplitudes and PPI were measured using methods identical to those used in Experiment 1.

Procedure

Animals were handled and weighed during the baseline period and were tested for startle response amplitudes. Within each age group, animals were quasirandomly assigned to drug condition according to body weight and baseline startle amplitudes so that drug conditions within each age had similar means and variance for both ASR amplitude and body weight. Minipumps containing saline or nicotine were implanted, and ASR was tested on days 1, 6, and 9 of drug administration. Minipumps were explanted on day 10, and ASR was tested on days 1,3, and 6 of drug cessation.

Data Analysis

Amount and percent of PPI were calculated as in Experiment 1. Results from each trial type and amount and percent of inhibition were analyzed separately by three-factor (drug, age, time) repeated-measures ANOVA and ANCOVA to control for baseline differences. Subsequent factorial ANOVAs were done at each time point. Cessation data were combined because results from days 1,3, and 6 of cessation did not differ. All analyses utilized two-tailed distributions and a significance level of 0.05. Percent of saline control was calculated for graphic presentation only, and was not included in the statistical analyses.

RESULTS

Figure 2 presents ASR amplitudes as percent of saline control using the 115-dB stimulus. Repeated-measures ANOVA revealed a significant effect of age, $F(1, 21) = 9.31$, $p < 0.05$, with ASR amplitudes for older rats > younger rats.

TABLE 4

AMOUNT AND PERCENT OF PREPULSE INHIBITION FOR YOUNGER AND OLDER RATS ADMINISTERED NICOTINE OR SALINE

		115 dB		122 dB	
Time	Drug	Younger (n = 16)	Older (n = 16)	Younger (n = 16)	Older (n = 16)
Baseline	Saline	15.0 ± 3.2	34.9 ± 10.6	63.5 ± 8.8	99.4 ± 14.5
		(58.5 ± 5.7)	(69.6 ± 10.5)	(73.6 ± 3.5)	(65.2 ± 5.5)
	Nicotine	15.8 ± 3.4	30.6 ± 7.0	44.1 ± 10.4	93.3 ± 12.7
		(59.3 ± 4.5)	(65.4 ± 7.9)	(61.0 ± 5.4)	(71.5 ± 4.9)
Drug day 1	Saline	4.6 ± 2.5	9.7 ± 5.2	38.3 ± 23.8	59.7 ± 23.8
		(19.8 ± 10.6)	(34.7 ± 14.5)	(45.0 ± 9.1)	(61.0 + 11.2)
	Nicotine	7.4 ± 1.8	24.5 ± 13.8	48.5 ± 17.2	65.9 ± 20.8
		(43.1 ± 6.7)	(29.1 ± 21.6)	(52.3 ± 13.3)	(58.7 ± 10.4)
Drug day 6	Saline	13.4 ± 3.6	17.4 ± 3.9	91.8 ± 19.6	105.0 ± 16.9
		(49.0 ± 14.7)	(61.3 ± 7.5)	(76.6 ± 4.9)	(80.8 ± 4.3)
	Nicotine	26.0 ± 10.4	38.0 ± 13.6	83.9 ± 11.5	101.8 ± 19.4
		(59.4 ± 6.4)	(64.5 ± 4.8)	(79.7 ± 2.6)	(69.8 ± 5.2)
Drug day 9	Saline	19.9 ± 7.2	31.0 ± 6.8	72.3 ± 16.1	117.7 ± 21.1
		(51.1 ± 10.6)	(71.3 ± 9.8)	(68.2 ± 5.8)	(73.2 ± 5.9)
	Nicotine	25.4 ± 8.0	39.6 ± 13.2	90.6 ± 14.8	137.6 ± 18.3
		(60.6 ± 9.9)	(70.3 ± 5.8)	(75.8 ± 3.8)	(74.0 ± 5.6)
Cessation	Saline	23.8 ± 5.4	40.5 ± 6.8	69.3 ± 19.3	79.5 ± 10.2
		(62.8 ± 6.6)	(79.0 ± 3.2)	(63.5 ± 4.6)	(73.0 ± 4.2)
	Nicotine	20.1 ± 3.3	38.5 ± 10.2	67.8 ± 9.3	62.5 ± 10.6
		(59.8 ± 5.5)	(65.4 ± 10.1)	(73.8 ± 1.4)	(63.3 ± 3.7)

Stimuli were preceded by 70-dB prepulses. Means and SEMs for percent of inhibition are shown in parentheses directly below means and SEMs for amount of inhibition.

When baseline (predrug) differences were controlled by ANCOVA, there were significant effects of nicotine on day 1, $F(1, 20) = 4.23, p < 0.05$, and day 6, $F(1, 20) = 4.43, p < 0.05$, with nicotine producing greater amplitude increases in older animals (Table 3). Using a stimulus intensity of 122 dB, results were similar to those obtained with 115 dB, with a significant effect of age (older > younger) on repeated-measures ANOVA, $F(1, 23) = 11.57, p < 0.05$.

Figure 3 presents mean amount of PPI as percent of saline control values. Repeated-measures ANOVA revealed a significant effect of age on amount of PPI, $F(1, 21) = 10.69, p < 0.05$, and percent of PPI, $F(1, 21) = 10.23, p < 0.05$, with significant differences between younger and older rats (older > younger). With baseline differences controlled by ANCOVA, there were significant effects of nicotine on day 1 for amount of inhibition, $F(1, 20) = 5.42, p < 0.05$, and percent of inhibition approached significance, $F(1, 20) = 4.13, p = 0.06$, with nicotine producing significant increases in percent of PPI in older animals, $F(1, 10) = 5.47, p < 0.05$. There was also a significant effect of age using the 122-dB stimulus to measure amount of PPI, $F(1, 23) = 4.72, p < 0.05$, with no differences in percent of inhibition for this stimulus (Table 4).

There were no significant effects of prior drug treatment during cessation, and there were no interactions.

DISCUSSION

The results of Experiment 1 revealed that different genetic strains of rat have different ASR amplitudes and amount, but not percent of PPI (Wistar > Sprague-Dawley > Long-Evans). This finding is consistent with genetic studies of mice, in which different strains vary in startle amplitude (22), although differences in PPI have not been studied in mice. The mechanism underlying these strain differences in startle reactivity is unknown.

Although nicotine has robust effects on startle measures in rats (1-3) and in mice (22), such differences were not observed in the present study of young rats from three genetic strains despite baseline (predrug) differences in startle reactivity. Unlike previous studies, nicotine administration appeared to have no effect on ASR amplitude or PPI at any time during chronic administration for the three rat strains. However, the major methodological difference between this experiment and those in which significant effects of nicotine on ASR were reported was the age of the subjects. Experiment 2 was subsequently conducted to evaluate effects of age in one of the strains used in Experiment 1.

The results of Experiment 2, in which Sprague-Dawley rats of 40 and 70 days of age were compared, confirmed that age is an important variable that also results in significant differences in ASR and PPI. Older rats consistently had greater ASR amplitudes and both amount and percent of PPI than younger animals. Consistent with previous reports, nicotine significantly increased ASR and PPI in the older rats in this experiment.

The mechanism underlying age differences in ASR and PPI does not appear to be related only to differences in body weight. Age and body weight are generally correlated variables in free-feeding rats, so that any effect of age also will result in a significant correlation with body weight when age categories are disparate. When the two age ranges are taken together, body weight and ASR amplitude were correlated as expected,

$r(29) = +0.49$, $p < 0.05$; however, within a single age group, body weight and ASR amplitude are not associated [$r(14) = -0.42$; $r(13) = +0.28$, NS, for younger and older rats, respectively], suggesting that correlations between amplitude and body weight cannot explain the differences in this experiment. It is likely that maturational factors in addition to differences in body weight were involved.

Although male rats of approximately 40 days of age are generally considered to be adult, they have not yet reached sexual maturity (29). Horlington (18) reported age differences according to the onset of sexual maturity in female Wistar rats, with ASR amplitude increases from 50 to 100 days of age. However, in that experiment, an age effect was measured only when rats were tested during the dark portion of the activity cycle, so again, effects of weight could not have been the cause of differences. In the present experiment, male rats were tested during the light portion of the activity cycle and older male rats had greater response amplitudes than younger males, consistent with Horlington (18) despite methodological differences. Therefore, it seems likely that maturational effects other than weight are responsible for age differences in ASR and PPI.

One maturational factor that may explain age differences in drug effects in this experiment involves drug metabolism. Male rats that are not sexually mature have lower levels of androgenic hormones and, consequently, younger rats metabolize drugs at a reduced rate relative to adult males (6,29). Reduced metabolism can result in potentiation or prolongation of drug effects, and given the inverted U-shaped dose-effect curve for nicotine's effect on startle (3), reduced metabolism could result in greater nicotine effects. Although nicotine appears to have a lesser effect in younger rats, nicotine's dose-effect curve for ASR amplitude and PPI is such that low nicotine doses produce increases in amplitude and PPI, whereas higher doses produce decreases (3). A greater nicotine effect could result in less increase in ASR amplitude and PPI because of the inverted U-shaped curve. Therefore, less increase in ASR and PPI for younger rats may indicate a greater behavioral effect of nicotine resulting from reduced metabolism. Alternatively, younger rats may be less sensitive to the effects of nicotine, but further studies, including more complete dose-effect curves for different age groups, would be required to resolve this issue.

Effects of nicotine in rats of 70 days of age (350 g) were generally similar to those previously reported in rats of this or greater age (1-3). Nicotine administration increased ASR amplitude and PPI in older animals, in contrast to the younger Sprague-Dawley rats in this experiment. Although only young Wistar and Long-Evans rats were tested in Experiment 1, it is likely that the lack of nicotine effects in these animals was related to their age at the time of study. Age and strain are clearly important variables that should be considered in future experiments of nicotine effects in rats.

ACKNOWLEDGEMENTS

Thanks to Dr. Jeffrey Witkin for helpful comments on the manuscript, and to Stephanie Nespor for assistance in conducting the experiments.

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