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Sensitivity to the dopaminergic regulation of prepulse inhibition in rats: Evidence for genetic, but not environmental determinants

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Abstract

Prepulse inhibition (PPI), a measure of sensorimotor gating, is reduced in schizophrenia patients and in rats treated with dopamine (DA) agonists. Reported strain and supplier-based differences in sensitivity to PPI-disruptive effects of DA agonists presumably reflect the differential impact of genetics and/or environment on DAergic substrates regulating PPI. In 2000, Harlan Laboratories established a Texas Sprague–Dawley line (SDHt; facility 211) using breeders from Indianapolis (SDHi; facility 202A). SDHi rats had been used, approximately 11 years earlier, to establish a colony in San Diego (SDHsd; facility 235). SDHt and SDHi rats are thus genetically similar, but raised in distinct environments; approximately 11 years of genetic "drift" separates SDHsd rats from both SDHi and SDHt rats. Harlan Long–Evans hooded rats (LEH; Madison, WI; facility 207) are genetically distinct from albino SDH. All except SDHsd rats were shipped to our facility by air freight. We used SDHt, SDHi, SDHsd, and LEH rats to assess genetic and environmental contributions to the DAergic regulation of PPI. Acoustic startle/PPI were assessed in rats treated with the D1/D2 agonist apomorphine (APO), the D2 agonist quinpirole, or the D1 agonist SKF 82958. The relative sensitivites to the PPI-disruptive effects were: *APO*: SDHt = SDHsd = SDHi >> LEH; *SKF 82958*: SDHt = SDHsd = SDHi (LEH not sensitive); *quinpirole*: SDHt = SDHsd = SDHi; SDHi > LEH. Strain/supplier differences in sensitivity to drug effects on startle magnitude did not correspond to patterns of PPI sensitivity. In these rats, strain differences in the DAergic regulation of PPI are most easily explained by genetic, rather than environmental influences that differentially impact both D1 and D2 substrates. This finding is consistent with published reports in other strains. Pharmacogenetic studies of PPI in rats may identify a genetic basis for a model of deficient sensorimotor gating in schizophrenia. © 2001 Elsevier Science Inc. All rights reserved.

Keywords: Apomorphine; Dopamine; Genetics; Prepulse inhibition; Schizophrenia; Startle; Strain

1. Introduction

Both genetic and nongenetic factors might contribute to strain differences in behavior in laboratory animals. By clarifying the role of these factors, we can identify behaviors which may be best understood in terms of their genetic underpinnings, and thereby contribute to our understanding of the role of specific genes in regulating brain and behavioral functions.

The startle reflex is inhibited when the startling stimulus is preceded 30–500 ms by a weak prepulse. Prepulse inhibition (PPI) is thought to reflect the activation of brain mechanisms designed to briefly "protect" the information contained in the prepulse; arrival of the startling stimulus during this "protected" period results in a relative diminution ("gating") of the startle response (cf. Swerdlow et al., 2000a). PPI is reduced in specific neuropsychiatric disorders, including schizophrenia (Braff et al., 1978, 1992, 1999, in press; Grillon et al., 1992; Kumari et al., 1999; Weike et al., 2000), and is also impaired in unaffected relatives of schizophrenia probands (Cadenhead et al., 2000), and in medication-free, nonpsychotic individuals with schizotypal personality disorder (Cadenhead et al., 1993, 2000) who are believed to carry a significant genetic "loading" for schizophrenia. Thus, diminished PPI may be a measure of impaired sensorimotor gating that is genetically determined in some individuals, and which conveys a risk for the development of specific forms of psychopathology.

PPI is measured in laboratory animals and humans using similar stimuli to elicit comparable response characteristics (Swerdlow et al., 1992, 1994). In both inbred and outbred laboratory mice and rats, there are significant strain and

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substrain differences in both "basal" levels of PPI (e.g., Palmer and Printz, 1999; Paylor and Crawley, 1997), and in the sensitivity to the PPI-disruptive effects of pharmacologic agents, including dopamine (DA) agonists (Dulawa et al., 2000; Hitchcock et al., 1999; Rigdon, 1990; Rothchild et al., 1999; Swerdlow et al., 2000b). Our laboratory reported greater sensitivity to the PPI-disruptive effects of the mixed D1/D2 DA agonist apomorphine (APO) in Sprague–Dawley rats from Harlan Laboratories ("SDH") than in Wistar rats from Harlan Laboratories ("WH") (Swerdlow et al., 1997), or SD and Wistar rats from Bantin–Kingman Laboratories (Swerdlow et al., 2000b). Others have reported greater PPI-disruptive effects of APO, among several other compounds, in Sprague–Dawley rats from Harlan vs. Charles River suppliers (Hitchcock et al., 1999). If strain or substrain differences in the sensitivity to the PPI-disruptive effects of DA agonists, or other pharmacological manipulations, reflect genetic differences among these animals, then these measures might be valuable tools for understanding the genetic regulation of a phenotype (diminished PPI) that in humans appears to characterize an inherited marker for increased risk of specific neuropsychiatric disorders.

Environmental factors might contribute to some of the observed strain differences in PPI drug sensitivity, and these factors must be assessed before we can convincingly ascribe these phenotypes to underlying genetic differences. For example, early developmental conditions clearly impact adult levels of PPI, as well as PPI drug sensitivity in rodents (e.g., Geyer et al., 1993; Overstreet et al., 2000; Rothchild et al., 1999; Vaillancourt and Boksa, 2000). Differential drug sensitivity in rats from different breeding facilities might thus reflect subtle differences in the rearing environment. Some startle measures, and behavioral sensitivity to DA agonists, are also sensitive to stressors, and thus it is conceivable that differential PPI drug sensitivity in rats from different facilities might reflect stress or hearing damage/noise exposure associated with different forms of transportation from breeding facility to testing location.

An opportunity to assess the potential impact of "nongenetic" factors on PPI drug sensitivity recently arose in January, 2000, when Harlan Laboratories established a new line of SDH rats in Houston, TX (SDHt; facility 211) from a parental stock in Indianapolis, IN (SDHi; facility 202A; established 1983 and never repopulated). Thus, in the immediate aftermath of this process, rats from the SDHi and SDHt lines should be genetically identical, but reared in different environments. Travel for both of these rats to our San Diego laboratory is via air freight, which should yield comparable levels of stress and barotrauma across these facilities. In the present study, we characterized the PPI-disruptive effects of several DA agonists in albino SDHi and SDHt rats, and compared them to the PPI-disruptive effects of these drugs in a genetically distinct group of hooded rats (Long–Evans, Harlan Laboratories, Madison, WI, facility 207; LEH). Sensitivity was assessed using the mixed D1/D2 agonist APO, as well as the relatively selective D2-family agonist quinpirole, and full D1 agonist SKF 82958. Finally, these results were compared with findings from similar tests of APO effects in SDH rats from a local supplier facility (Harlan Laboratories, San Diego, facility 235; "SDHsd"), which had been derived from the SDHi parental stock in 1989 (and never repopulated), and which had been tested without the potential impact of air freight transportation. Breeding females average five litters per 9-month period in these Harlan facilities; given staggered breeding schedules, over the 11 years during which SDHsd rats were genetically isolated from their SDHi founders, it is likely that SDH lines advanced several hundred generations at each of these facilities.

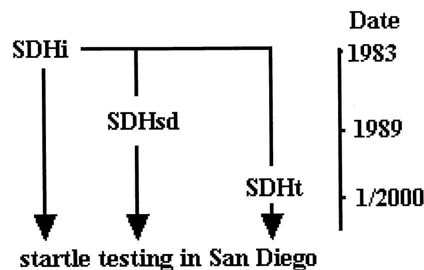

Fig. 1. Pedigree of SDH rats in the present study. The SDHi colonies were populated by breeders in 1983. Offspring were then used to populate the SDHsd facility in 1989, and the SDHt facility in 2000.

A schematic representation of the geneology of these different rat facilities/strains is seen in Fig. 1. If there is a strong genetic basis for the PPI drug-sensitivity phenotype, we would predict similar patterns of sensitivity to the PPI-disruptive effects of DA agonists in SDHi and SDHt rats, due to their likely genetic homology, despite their environmental differences. In this case, findings with LEH rats might be expected to be quite distinct from those of SDHi and SDHt rats, due to the genetic differences between these strains. If the rearing environment, or the consequences of freight travel are major determinants of this drug-sensitivity phenotype, we might expect a number of other possible outcomes. For example, we might predict: (1) substantial differences in SDHi, SDHt, SDHsd, and LEH drug sensitivity, if rearing environment is a critical factor; or (2) substantial differences between SDHsd rats vs. SDHi, SDHt, and LEH rats, if type of freight travel has a major impact on startle measures.

2. Methods and materials

2.1. Experimental animals

A total of 34 adult male SDHi rats, 33 adult male SDHt rats, 17 adult male SDHsd rats, and 34 adult male LEH rats were used in these experiments. Most rats were obtained within the same 3-month period, beginning approximately 3 months after the opening of the new Houston breeding

facility in January 2000. Studies in adults were limited to male rats, based on findings of estrous cyclicity of the PPI-disruptive effects of APO in adult female rats (Koch, 1998). Adult male rats were housed in same-sex rooms, in groups of two or three. Methods for housing and all behavioral testing were consistent with the substantial literature of startle measures in rodents (cf. Geyer and Swerdlow, 1998). For example, a reversed 12-h light/dark cycle was used (lights on at 1900 h, off at 0700 h) for at least 1 week prior to testing. After shipment arrival, rats were maintained in the housing facility for at least 1 week prior to behavioral testing. All testing and drug administration occurred between 1000 and 1700 h. Weiss et al. (1999) recently reported that circadian time does not modify either PPI or its disruption by APO. Rats were handled regularly prior to any procedures to minimize stress during behavioral testing, and were given ad libitum access to food and water except during behavioral testing. Throughout these studies, all efforts were made to minimize animal suffering and to reduce the number of animals used. All experiments conform to guidelines of the National Institute of Health for the use of animals in biomedical research and were approved by the Animal Subjects Committee at the University of California, San Diego (protocol #0224908).

2.2. Drugs

APO (0.1% ascorbate/saline vehicle, 0.1, 0.25, or 0.5 mg/kg), quinpirole (saline vehicle, 0.1, 0.2, and 0.5 mg/kg), and SKF 82958 (saline vehicle, 0.1, 1.0, and 5.0 mg/kg) were administered subcutaneously to rats immediately prior to testing (APO) or 10 min prior to testing (quinpirole, SKF 82958), in a volume of 1 ml/kg.

2.3. Apparatus

Startle experiments used four startle chambers (SR-LAB; San Diego Instruments, San Diego, CA) housed in a sound-attenuated room with a 60-dB ambient noise level. Each startle chamber consisted of a Plexiglas cylinder (8.7 cm internal diameter) resting on a 12.5 × 25.5-cm Plexiglas stand. Acoustic stimuli and background noise were presented via a Radioshack Supertweeter mounted 24 cm above the Plexiglas cylinder. Startle magnitude was detected and recorded as transduced cylinder movement via a piezoelectric device mounted below the Plexiglas stand. Response sensitivities were calibrated (SR-LAB Startle Calibration System) to be nearly identical in each of the four startle chambers (maximum variability < 1% of stimulus range and < 5% of response ranges). Chambers were also balanced across all experimental groups. Sound levels were measured and calibrated with a sound-level meter (Quest electronics: Oconomowoc, WI), A scale (relative to 20 μN/M2) with a microphone placed inside the Plexiglas cylinder. Methodological details can be found in published material (Geyer and Swerdlow, 1998).

2.4. Startle testing procedures

Approximately 7 days after shipment arrival, rats were exposed to a brief "matching" startle session, as described previously (Geyer and Swerdlow, 1998). Rats were placed in a startle chamber, and exposed to 5 min of 70 dB background noise followed by 17 pulse trials of 40-ms, 120-dB noise bursts ("pulse") and 3 prepulse + pulse trials consisting of a 20-ms, 82-dB (12 dB above background) prepulse followed by a 100-ms, 120-dB pulse (onset to onset). Data from this session were used to assign rats to balanced dose groups according to their average pulse startle magnitude.

Behavioral testing continued 2–4 days after the "matching" session. Rats were brought to the laboratory in individual cages, approximately 1 h before testing. For the initial test, APO (0, 0.1, 0.25, or 0.5 mg/kg sc) was administered, and rats were placed immediately into the startle test chambers. Test sessions were approximately 16 min long and consisted of 5 min of 70 dB background followed by five trial types: pulse noise bursts, prepulse trials (20 ms noise bursts 5, 10, or 15 dB above background followed 100 ms by a pulse), and NOSTIM trials (stabilimeter recordings obtained when no stimulus was presented). The session consisted of initial and final blocks of 4 pulse trials, separated by two blocks that included 8 PULSE trials and 15 prepulse trials (the latter divided equally among 5, 10, and 15 dB prepulse intensities); NOSTIM trials were interspersed between all trials. For these "NOSTIM" trials, stabilimeter readings were recorded during periods where no stimulus was presented; these trials were used to assess gross motor activity during the test session, but were not included in the calculation of intertrial intervals, which were variable and averaged 15 s. Reflex "habituation" was determined based on the change in startle magnitude from the initial to the final block of PULSE trials. Using this design, PPI is measured during a portion of the session in which startle magnitude is relatively constant.

After 7–10 days, startle and PPI were assessed in these same rats, 10 min after administration of either quinpirole (saline vehicle, 0.1, 0.2, and 0.5 mg/kg) or SKF 82958 (saline vehicle, 0.1, 1.0, and 5.0 mg/kg). For these quinpirole and SKF 82958 experiments, rats were tested twice, at a 1-week interval. During each week, half of the rats from each facility/strain were tested with one of four doses of either SKF 82958 or quinpirole, with drug order (SKF 82958 vs. quinpirole) balanced across test weeks, and dose groups randomized across weeks.

2.5. Data analysis

PPI was calculated as a percentage of reduction in startle magnitude on prepulse trials compared to pulse trials. Any drug effects on percent PPI (%PPI) prompted separate analyses to assess the relationship of these effects to drug-induced changes in startle magnitude on pulse and prepulse

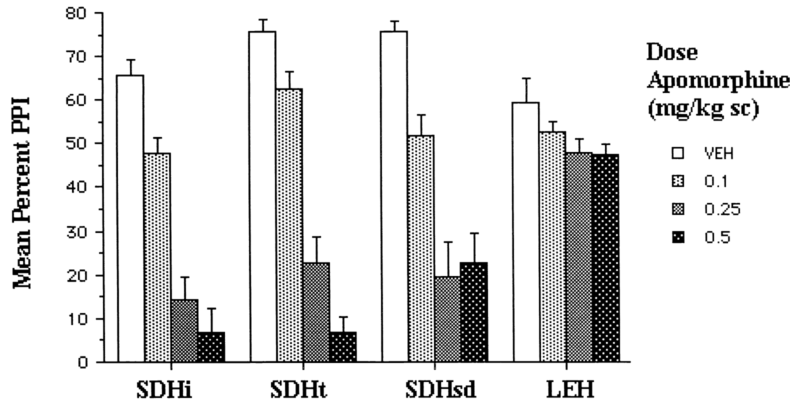

Fig. 2. Mean %PPI in SDHi, SDHt, SDHsd, and LEH rats in response to APO (0–0.5 mg/kg sc). APO sensitivity was comparable across the three SDH substrains, wherein each was significantly more sensitive than LEH rats.

trials. Drug-induced changes in startle magnitude on pulse trials — even in the absence of changes in startle magnitude on prepulse trials — can change the amount of calculated %PPI (cf. Swerdlow et al., 2000a). The most unequivocal *changes in sensorimotor gating* occur when the reflex-inhibiting effects of prepulses are modified significantly — demonstrated by a significant change in startle magnitude on prepulse trials — without obligatory significant changes in startle magnitude on pulse trials. It is clearly possible for changes in sensorimotor gating to occur together with significant increases or decreases in startle magnitude, but the interpretation of a change in %PPI under such conditions is complex (discussed in Swerdlow et al., 2000a). Thus, for each facility and strain, data were assessed to determine whether drug-induced changes in the calculated amount of %PPI reflected actual changes in sensorimotor gating per se.

All startle data were analyzed using an analysis of variance (ANOVA) with drug treatment and facility/strain as between-subject factors and trial block and trial type as within-subject repeated measures. Post hoc comparisons of significant interaction effects and relevant main factor effects were conducted using the Tukey–Kramer and one-factor ANOVA tests. Alpha was set at .05. One rat was excluded from one analysis due to a very low startle response magnitude (mean startle on pulse-alone trials = 1.0; SDHt quinpirole study), that precluded a meaningful calculation of PPI. For ease of presentation, unless otherwise stated, several normal parametric effects can be assumed to be statistically significant in all startle analyses: effects of trial block on startle magnitude, and effect of prepulse intensity on PPI. Also, unless otherwise stated, reported values of mean %PPI can be assumed to be collapsed across all prepulse intensities and trial blocks. For most instances, only statistically significant effects, or those relevant to the critical comparisons, will be reported in detail.

3. Results

The effects of APO on PPI in SDHi, SDHt, SDHsd, and LEH rats are seen in Fig. 2. ANOVA revealed significant effects of facility/strain (F = 4.47, *df* 3,102, P < .01), APO dose (F = 35.41, *df* 3,102, P < .0001), and a significant Facility/Strain × Dose interaction (F = 3.15, *df* 9,102, P < .005).

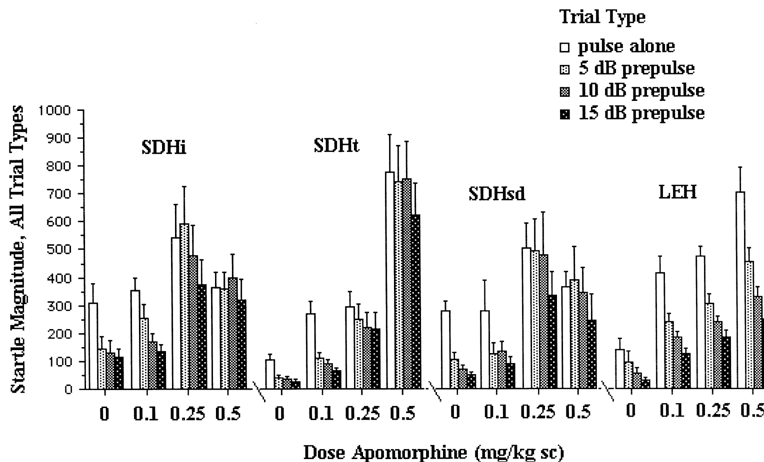

Fig. 3. Startle magnitude on pulse-alone and prepulse trial types, in SDHi, SDHt, SDHsd, and LEH rats. A clear APO disruption of sensorimotor gating was evident in all SDH rats (e.g., compare doses: SDHi, 0 vs. 0.5 mg/kg; SDHt, 0 vs. 0.25 mg/kg; SDHsd, 0 vs. 0.5 mg/kg), but not in LEH rats.

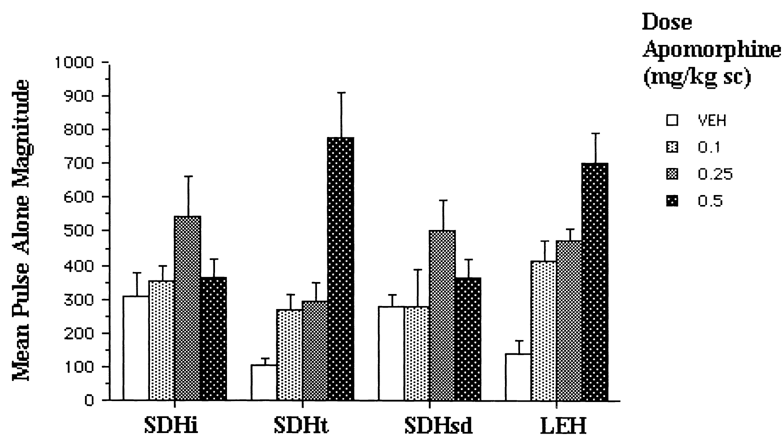

Fig. 4. Effects of APO on startle magnitude on pulse-alone trials. APO increased startle magnitude with a monotonic dose relationship in SDHt and LEH rats, while in SDHi and SDHsd rats, an "inverted U" function was noted. Unlike APO effects on PPI, its effects on startle magnitude were not strain-specific.

Comparison of PPI levels among rats treated with APO vehicle revealed no significant effect of facility/strain (F = 1.28, df 3,28, ns). There was a significant interaction of facility/strain and prepulse intensity, reflecting the reduced "baseline" PPI levels for 5-dB prepulses in LEH rats [mean (S.E.M.) %PPI = 36.49 (12.09)], vs. all other strains [62.63 (6.19), 65.46 (4.52), and 67.50 (4.48) for SDHi, SDHt, and SDHsd, respectively]. For active APO doses (0.1, 0.25, and 0.5 mg/kg), ANOVA revealed a significant effect of facility/strain (F = 4.47, df 3,102, P < .01) and post hoc Tukey comparison revealed lower (P < .05) PPI levels in SDHi, SDHsd, and SDHt rats, compared to LEH rats, but no differences in PPI between SDHi, SDHsd, and SDHt rats. Within each facility/strain, ANOVA revealed significant effects of APO on PPI in SDHi rats (F = 13.835, df 3,30, P < .0001), SDHsd rats (F = 4.63, df 3,13, P < .025), and SDHt rats (F = 26.03, df 3,29, P < .0001), but not in LEH rats (F < 1).

Inspection of startle magnitude on pulse and prepulse trials revealed that the loss of PPI in SDHi, SDHsd, and SDHt rats reflected a clear loss of sensorimotor gating, i.e., a reduction in the ability of the prepulse to reduce startle magnitude (Fig. 3). Specifically, in SDHi rats, startle magnitude on pulse trials was not significantly increased by 0.5 mg/kg APO, while startle magnitude on the prepulse trials increased significantly after 0.5 mg/kg APO; the identical pattern is seen in SDHsd rats. In SDHt rats, startle magnitude on pulse trials was increased by APO (see below), but a comparison between the 0.1- and 0.25-mg/kg doses of APO revealed no change in startle magnitude on pulse trials, but a significant increase in startle magnitude on prepulse trials. These patterns contrast markedly from that exhibited by LEH rats. The impact of facility/strain on sensorimotor gating was supported by the critical interaction of Facility/Strain × Trial Type × APO Dose (F = 3.51, df 27,306, P < .0001). These data also revealed strain/facility differences in the effects of APO on startle magnitude on pulse-alone trials (Fig. 4). When this measure was examined carefully, ANOVA revealed a significant effect of APO (F = 9.48, df 3,102,

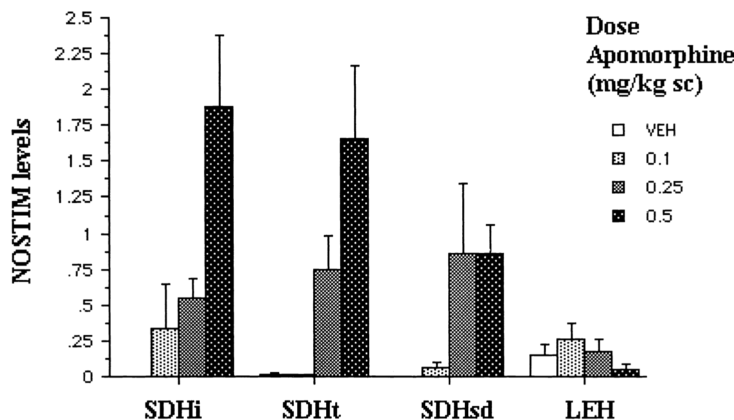

Fig. 5. Effects of APO on activity readings between startle trials ("NOSTIM" levels). Note that APO increased NOSTIM activity in all SDH rats, but not in LEH rats. While there was some rough correspondence between these sensitivities to APO effects on NOSTIM activity and PPI in SDH rats, this relationship — as previously reported (Swerdlow et al., 2000b) — was not particularly robust [e.g., compare effects of 0.1 vs. 0.25 mg/kg APO in SDHi rats on PPI (effects of 0.25 >> 0.1 mg/kg) and on NOSTIM activity (effects of 0.25 ≈ 0.1 mg/kg).

$P < .0001$), no significant effect of facility/strain ($F < 1$), and a significant Facility/Strain × APO interaction ($F = 2.60$, *df* 9,102, $P < .001$). Post hoc comparisons revealed significant effects of APO on pulse-alone magnitude in SDHt and LEH rats, but not in SDHsd or SDHi rats (F's = 9.82, 9.42, 1.21, and 1.00, respectively, P's < .001, .002, ns, and ns, respectively). Tukey comparisons revealed significant effects of APO on pulse startle magnitude for the highest dose of APO (0.5 mg/kg) in SDHi rats, and for all doses of APO (0.1, 0.25, and 0.5 mg/kg) in LEH rats.

An assessment of startle magnitude in the initial vs. final blocks of pulse-alone trials revealed robust habituation that did not differ across facilities/strains (not shown). Assessment of NOSTIM activity revealed APO-induced increases in NOSTIM levels in SDHt, SDHi, and SDHsd rats, but not in LEH rats (Fig. 5). This was supported by a significant interaction of Facility/Supplier × APO Dose ($F = 2.35$, *df* 9,102, $P < .02$), and post hoc analyses revealing significantly increased NOSTIM levels after the highest dose of APO in SDHt, SDHsd, and SDHi rats (P's < .02, .003, and .004, respectively), but not in LEH rats (ns).

The effects of quinpirole and SKF 82958 on PPI and startle measures in SDHi, SDHt, SDHsd, and LEH rats are seen in Tables 1 and 2, respectively. In general, the findings paralleled those seen with APO, except that the magnitude of the SKF 82958 effect on PPI was relatively smaller in the SDHi, SDHt, and SDHsd rats, consistent with our previous findings (Swerdlow et al., 2000b; Vaillancourt and Boksa, 2000). In other words, SDHi, SDHt, and SDHsd rats exhibited comparable sensitivities to the PPI-disruptive effects of quinpirole and SKF 82958, while LEH rats were relatively less sensitive (quinpirole) or insensitive (SKF 82958) to these effects. We previously reported that SDH rats can exhibit extreme behavioral excitation and even generalized seizures at the highest dose of SKF 82958 (5.0 mg/kg; Swerdlow et al., 2000b); in the present studies, these seizures were evident in almost all of the Sprague–Dawley rats treated with the 5.0-mg/kg dose (but none of the LEH rats) within 10 min of completion of behavioral testing, and thus behavioral data from this one dose were not included in the statistical analyses. Inspection of startle magnitude on pulse-alone trials (Table 1), as well as prepulse + pulse trials, revealed three general trends: (1) both quinpirole and SKF 82958 tended to *reduce* startle magnitude on pulse-alone trials in *SDHi*, *SDHt*, and *SDHsd* rats, and to *increase* startle magnitude on pulse-alone trials in *LEH* rats; (2) due to changes in startle magnitude on pulse-alone trials, clear effects of quinpirole and SKF 82958 on sensorimotor gating per se (ability of the prepulse to inhibit startle magnitude) were evident only at specific doses (e.g., 0.2 mg/kg quinpirole and 1.0 mg/kg SKF 82958 for SDHt rats); and (3) LEH rats were sensitive to modest PPI-disruptive (and "gating"-disruptive) effects of quinpirole, but actually exhibited modestly increased PPI (and "gating") in response to SKF 82958.

Table 1
Effects of quinpirole on startle variables

Facility/strain	Dose (mg/kg)	Mean % PPI (S.E.M.)	Pulse-alone magnitude [mean (S.E.M.)]	NOSTIM magnitude [mean (S.E.M.)]
SDHi	0	81.09 (1.85)	314.76 (53.35)	0.02 (0.01)
	0.1	60.45 (2.83)	243.49 (35.66)	0.40 (0.34)
	0.2	51.11 (4.74)	261.47 (46.82)	0.30 (0.17)
	0.5	42.24 (4.29)	212.02 (53.48)	1.44 (0.69)
SDHt	0	72.75 (2.30)	414.04 (96.68)	0.09 (0.08)
	0.1	58.22 (3.88)	327.31 (36.70)	0.24 (0.13)
	0.2	19.45 (7.00)	233.55 (37.13)	0.65 (0.22)
	0.5	20.44 (12.34)	183.40 (20.53)	1.36 (0.45)
SDHsd	0	78.34 (3.45)	367.59 (51.33)	0.10 (0.10)
	0.1	53.47 (6.55)	137.95 (22.35)	0.06 (0.04)
	0.2	56.17 (4.78)	220.52 (22.13)	0.11 (0.09)
	0.5	33.25 (6.11)	173.69 (53.79)	2.56 (1.91)
LEH	0	76.62 (2.79)	258.89 (68.12)	0.13 (0.04)
	0.1	52.69 (3.00)	481.02 (38.30)	0.76 (0.36)
	0.2	55.94 (3.60)	337.20 (52.65)	1.81 (0.75)
	0.5	51.35 (3.46)	295.11 (36.06)	0.62 (0.25)

Table 2
Effects of SKF 82958 on startle variables

Facility/strain	Dose (mg/kg)	Mean % PPI (S.E.M.)	Pulse-alone magnitude [mean (S.E.M.)]	NOSTIM magnitude [mean (S.E.M.)]
SDHi	0	75.94 (2.67)	350.29 (41.72)	0.05 (0.04)
	0.1	71.75 (2.40)	320.13 (62.90)	0.06 (0.04)
	1.0	59.98 (4.19)	212.18 (40.24)	0.34 (0.20)
SDHt	0	84.35 (1.85)	266.18 (35.15)	0.40 (0.30)
	0.1	74.41 (2.55)	301.87 (60.94)	0.03 (0.02)
	1.0	66.15 (2.87)	194.93 (26.11)	0.18 (0.08)
SDHsd	0	74.76 (2.28)	315.11 (29.38)	0.02 (0.01)
	0.1	75.56 (3.47)	475.70 (125.55)	0.00 (0.00)
	1.0	24.91 (7.53)	205.73 (38.75)	2.26 (0.69)
LEH	0	67.29 (3.07)	261.81 (64.96)	0.11 (0.04)
	0.1	74.33 (2.84)	517.89 (92.62)	0.41 (0.39)
	1.0	69.87 (2.40)	875.23 (119.05)	0.00 (0.00)
	5.0	64.36 (2.09)	637.03 (62.02)	0.05 (0.03)

4. Discussion

The present study tested the hypothesis that genetic background plays a major role among the many factors that might contribute to strain- and substrain-specific patterns of sensitivity to the PPI-disruptive effects of DA agonists in rats. Outbred Sprague–Dawley rats from nearly identical gene pools, raised in three different facilities (Indianapolis, Texas, and San Diego) exhibited very similar sensitivities to the PPI-disruptive effects of APO, quinpirole, and SKF 82958. These patterns were also quite similar to those exhibited by another line of Sprague–Dawley rats (SDHsd), which arrived at our testing facility without the stresses of air freight travel, and which had been genetically isolated from the Indianapolis and Houston breeding lines (and thus subject to "drift") for approximately 11 years. Clearly,

either "drift" or environmental factors might contribute to PPI differences *within strain, across suppliers*, that have been reported previously in the literature (e.g., Hitchcock et al., 1999; Swerdlow et al., 2000b). Equally striking in the present study was the fact that these patterns of DA agonist sensitivity in SDH rats differed significantly from those exhibited by outbred Long–Evans rats. PPI sensitivity to APO clearly *did not* differ significantly among genetically similar rats reared in different facilities (SDHi vs. SDHt); it *did not* differ significantly among genetically similar rats transported to the testing facility via different methods with different associated stressors (SDHi/SDHt vs. SDHsd); it *did* differ significantly among genetically disparate rats transported to testing via similar methods with similar associated stressors (SDHi/SDHt vs. LEH). Thus, these studies confirm that even among outbred rats, genetic background plays a major role in determining this DA agonist PPI "phenotype." Succinctly, with regards to determinants of this specific phenotype, "it's who you are, not where you're from, or how you got here."

Strain differences have also been reported between Sprague–Dawley and Long–Evans rats in the regulation of PPI by nicotine (Faraday et al., 1999). It is not clear whether this pattern also reflects strain differences within DAergic systems, although some evidence would argue against such an interpretation (Bejar et al., 1997).

Outbred rat strains, including Sprague–Dawley and Long–Evans rats, differ in their expression of specific genetic polymorphisms for D2-family receptors (Luedtke et al., 1992; Scott et al., 1995). It is possible that some of these genetic differences might ultimately contribute to the observed phenotypes of PPI sensitivity to DA agonists. Differences in D1 agonist sensitivity (seen with SKF 82958) might also suggest genetic variations of the D1 receptor among these strains. However, because D1 and D2 receptors appear to interact in the regulation of PPI (Peng et al., 1990; Wan et al., 1996), it is equally possible that sensitivity to the PPI-disruptive effects of SKF 82958 actually reflect genetic differences within loci coding for the D2 receptor. For example, we reported that blockade of D2 receptors prevents the PPI-disruptive effects of SKF 82958 (Wan et al., 1996). Thus, the lack of sensitivity to SKF 82958 among LEH rats in the present studies might conceivably reflect reduced sensitivity among D2 receptors (seen with blunted quinpirole sensitivity), that normally contribute—perhaps via some "permissive" role—to the D1 regulation of PPI.

The present study also identified distinct patterns of drug effects on pulse-alone startle magnitude, and NOSTIM activity levels, among SDHi, SDHt, SDHsd, and LEH. Some patterns may have reflected strain differences. For example, quinpirole and SKF 82958 tended to reduce pulse-alone startle magnitude in SDHi, SDHsd, and SDHt rats, while both drugs produced inverted U-shaped dose effects on pulse-alone startle magnitude in LEH rats (i.e., maximally increased startle magnitude at intermediate doses). Meloni and Davis have reported that these doses of SKF 82958 increase pulse-alone startle magnitude in Sprague–Dawley rats from Charles River Laboratory facilities in Massachusetts (Meloni and Davis, 2000) and North Carolina (Meloni and Davis, 1999). The effects of APO on startle magnitude did not follow strain-specific patterns, but instead, were most similar in SDHt and LEH rats (substantial, monotonic dose-dependent increase), compared to SDHi and SDHsd rats (no significant increases in startle magnitude). NOSTIM activity levels also followed no clear strain-related pattern: they were increased by APO in all SDH (but not LEH) rats, by SKF 82958 in SDHi and SDHsd (but not SDHt or LEH) rats, and by quinpirole in all rats (with LEH rats exhibiting an inverted U dose sensitivity).

The observed patterns of sensitivity to the PPI-disruptive effects of APO, quinpirole, or SKF 82958 did not correspond in any simple way with those for the drug effects on pulse-alone startle magnitude or NOSTIM activity. This is generally consistent with the relative independence of drug effects on PPI with those on other startle characteristics (e.g., Swerdlow et al., 1986, 1994, 2000b). Also consistent with our previous findings with APO, quinpirole, and SKF 82958, in most instances in the present studies, drug effects on PPI were accompanied by clear reductions in sensorimotor gating, i.e., the startle-reducing effects of prepulses (e.g., Swerdlow et al., 2000b).

In summary, the present findings suggest that sensitivity to the PPI-disruptive effects of DA agonists are very sensitive to genetic differences among two outbred rat strains—albino Sprague–Dawley and hooded Long–Evans rats—but are relatively insensitive to differences in rearing environments or transportation stressors. These PPI "phenotypes" are largely conserved among three lines of SDH rats that originated from a common founder population, but were then genetically isolated for 11 years. Patterns of other startle and drug-sensitivity "phenotypes" did not segregate neatly according to rat strain, rearing environment, or transportation stress. Sensitivity to the PPI-disruptive effects of DA agonists in at least two outbred rat strains appears to have strong genetic determinants that are conserved across hundreds of generations, and which are relatively insensitive to a number of environmental variables. This phenotype may thus be a useful tool for understanding the genetics of diminished PPI in humans, which in certain populations may be associated with a genetic predisposition for schizophrenia.

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