ELSEVIER

"Early" and "Late" Effects of Sustained Haloperidol on Apomorphine- and Phencyclidine-induced Sensorimotor Gating Deficits

Zoë A. Martinez, M.A., Jeff Oostwegel, Mark A. Geyer, Ph.D., Gaylord D. Ellison, Ph.D., and Neal R. Swerdlow, M.D., Ph.D.

Both dopamine (DA) agonists and NMDA antagonists produce prepulse inhibition (PPI) deficits in rats that model PPI deficits in schizophrenia patients. While DA agonist effects on PPI are reversed by acute treatment with either "typical" high-potency D2 DA antagonists or "atypical" antipsychotics, PPI deficits produced by phencyclidine (PCP) are preferentially reversed by acute treatment with "atypical" antipsychotics. Acute effects of antipsychotics may not accurately model the more clinically relevant effects of these drugs that emerge after several weeks of continuous treatment. In the present study, sustained treatment with haloperidol via subcutaneous minipumps blocked the PPI-disruptive effects of apomorphine and attenuated the PCP-induced disruption of PPI. Restoration of PPI in apomorphine-treated rats was evident within the first week of sustained haloperidol administration. A partial reversal of PCP effects on PPI did not develop until the second week of sustained haloperidol treatment, followed a fluctuating course, but remained significant into the seventh week of sustained haloperidol administration. The delayed emergence of anti-PCP effects of haloperidol suggests that the brain substrates responsible for the DAergic and NMDA regulation of PPI are differentially sensitive to acute and chronic effects of antipsychotics.
[Neuropsychopharmacology 23:517–527, 2000]

KEY WORDS: *Apomorphine; Haloperidol; Phencyclidine; Prepulse inhibition; Schizophrenia; Sensorimotor; Startle*

Prepulse inhibition (PPI), the reduction in startle magnitude when a weak prestimulus is presented 30–500 ms prior to a startling stimulus, is an operational measure of sensorimotor gating that is deficient in schizophrenia spectrum patients (Braff et al. 1978, 1992, 1999; Grillon et al. 1992; Cadenhead et al. 1993; Bolino et al. 1994; Perry and Braff 1994). In rats, PPI is disrupted by acute treatment with DA agonists (Swerdlow et al. 1986, 1994; Mansbach et al. 1988; Martinez et al. 1999) or NMDA antagonists (Mansbach and Geyer 1989, 1991). These different drug effects appear to be mediated in part by distinct substrates that are sensitive to different pharmacological manipulations. Specifically, while acute treatment with either "typical" high-potency D2 DA antagonists or "atypical" antipsychotics can reverse DA agonist-induced PPI deficits, clinically "atypical" antipsychotics preferentially reverse the PPI deficits produced by the NMDA antagonist phencyclidine (PCP) (Keith et al. 1991;

From the Department of Psychology (ZAM, GDE), University of California, Los Angeles, CA; Faculty of Pharmacy (JO), University of Utrecht, Utrecht, The Netherlands; and Department of Psychiatry (MAG, NRS), UCSD School of Medicine, La Jolla, CA.

Address correspondence to: Dr. N.R. Swerdlow, UCSD School of Medicine, Department of Psychiatry, La Jolla, CA 92093-0804. Tel.: 619-543-6270; Fax: 619-543-2493; E-mail: nswerdlow@ucsd.edu

Received February 2, 2000; revised April 25, 2000; accepted May 4, 2000.

Published by Elsevier Science Inc.
655 Avenue of the Americas, New York, NY 10010

0893-133X/00/$–see front matter
PII S0893-133X(00)00147-0

Swerdlow and Geyer 1993; Hoffman et al. 1993; Swerdlow et al. 1994; Bakshi et al. 1994; Swerdlow and Geyer 1998). These findings support the suggestion that the behavioral effects of DA agonists may model some aspects of "positive" symptoms of schizophrenia, which respond to treatment with clinically typical antipsychotics, while the behavioral effects of NMDA antagonists may model some aspects of "negative" symptoms of schizophrenia, which have been reported to respond preferentially to treatment with clinically atypical antipsychotics (Kane et al. 1988).

Previous reports have suggested that acute behavioral effects of antipsychotics may not be fully informative in models of the clinical properties of these drugs, since their full clinical effectiveness requires repeated or sustained administration (Pietraszek and Ossowska 1998; Feifel and Priebe 1999). The present study was designed to assess the effects of sustained exposure to the high-potency D2 DA antagonist haloperidol on the PPI-disruptive effects of apomorphine and phencyclidine. Rats were treated for 28 or 49 days with haloperidol via osmotic minipumps, at a dose that reversed the apomorphine-induced disruption of PPI at the earliest time point tested. During administration of haloperidol, rats were tested weekly, to determine the effects of haloperidol on both PCP- and apomorphine-induced PPI deficits. Although haloperidol initially had no effect on PCP-induced PPI deficits, the PPI-disruptive effects of PCP were significantly reversed by haloperidol at two different time points: after 2–3 weeks of sustained haloperidol administration ("early") and after 7 weeks of sustained haloperidol administration ("late").

METHODS

Rats

A total of 89 male Sprague-Dawley rats, weighing 250–500 g (Harlan Laboratories, San Diego, CA), were used in these experiments. Rats were housed in groups of two or three, until equipped with minipumps, at which time they were housed singly, to prevent excessive perturbation of the pumps. A reversed 12-h light/dark cycle was used (lights on at 1900, off at 0700); surgery and all testing occurred between 1000 and 1700. Rats were handled prior to any procedures to minimize stress during behavioral testing, and were given ad libitum access to food and water except during surgery and behavioral testing.

Drugs

For sustained administration, osmotic minipumps (Alza Corporation, Palo Alto, CA) filled to deliver approximately 1 mg/kg/d haloperidol were used. Control rats received a "sham pump" (either silastic polymer pellets of approximately the same size and shape as the minipumps (Lipton et al. 1991), or an actual minipump filled with saline). In studies examining acute drug effects on PPI, saline vehicle, 1.25 mg/kg or 1.5 mg/kg phencyclidine (PCP HCl) or 0.1% ascorbate/saline vehicle or 0.5 mg/kg apomorphine were administered subcutaneously (sc). These doses of apomorphine and PCP have been shown to produce consistent and robust disruptions of PPI in rats (Mansbach et al. 1988; Mansbach and Geyer 1989).

Surgery

Implantation of subcutaneous minipumps was performed under halothane anesthesia. Rats were anesthetized initially by exposing them to a bell jar containing vaporized halothane. After this, a small area on the back, directly behind the head, was shaved and the rat was placed into a stereotaxic apparatus with a nose cone that delivered a mixture of halothane and air. The area to be incised was cleaned with an alcohol swab, while tail pinch and visual observation of respiratory movements were used to assess sufficient levels of anesthesia. Once the animal was anesthetized, a small incision was made with a scalpel, a small subcutaneous pocket was created using blunt dissection, a pump or "sham pump" was inserted and the incision was closed with wound clips. Explantation involved a similar procedure, with an incision made below the previous incision through which the pump or sham pump was removed.

Apparatus

All experiments utilized 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 chamber consisted of a Plexiglas cylinder 8.2 cm in diameter resting on a 12.5 $\times$ 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 by a microcomputer and interface assembly, which transduced cylinder movement via a piezoelectric device mounted below the Plexiglas stand. Startle magnitude was defined as the average of 100 readings collected every 1 ms beginning at the onset of the acoustic noise burst. Acoustic stimulus intensities and response sensitivities were calibrated (using an 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 microphone placed inside the Plexiglas cylinder.

Methodological details can be found in published material (Geyer and Swerdlow 1998).

Testing Procedures

Eight to 14 days after arrival (2–4 days prior to surgery), rats were exposed to a brief "matching" startle session. Rats were placed in a startle chamber (closed Plexiglas cylinder) and exposed to 5 min of 70-dB background noise followed by 17 PULSE trials of 40 ms 120-dB noise bursts ("PULSE ALONE") and 3 PREPULSE + PULSE trials consisting of a 20 ms 82 dB (12 dB above background) prepulse followed 100 ms by a 120-dB pulse. Data from this session were used to assign rats to balanced groups (vehicle or 1 mg/kg/d haloperidol) according to their average PULSE ALONE startle magnitude.

Beginning on post-operative day 8 (7 days after implantation of minipump or pellet), rats were brought to the laboratory for 30 min and then treated with either vehicle or drug immediately (apomorphine) or 10 min before (PCP) a test session. Each session was approximately 19 min long and consisted of 5 min of 70-dB background followed by four trial types: PULSE ALONE noise bursts; and PREPULSE trials which consisted of 20-ms noise bursts 3, 6, or 12 dB above 70-dB background noise followed 100 ms by a PULSE. The session consisted of four "blocks,": blocks 1 and 4 included four PULSE ALONE trials, and blocks 2 and 3 included both PULSE ALONE (8 trials per block) and 3, 6 and 12 dB PREPULSE+PULSE trials (5 trials each per block), presented in pseudorandom order with a variable intertrial interval (average of 15 sec). In addition, between each stimulus trial, 100 ms of response was recorded during periods where no stimulus was presented. These trials were called "NOSTIM" trials and were used to assess gross motor activity during the test session but were not included in the calculation of intertrial intervals.

Treatment and Test Schedule

Group 1. Rats were equipped with 28-day minipumps delivering haloperidol ($n = 36$) or control pellets ($n = 32$). Animals were tested 7, 14, and 21 days post-implantation after treatment with saline vehicle or PCP (1.25 mg/kg or 1.5 mg/kg) and 10, 17, and 24 days post-implantation after treatment with 0.1% ascorbate/saline vehicle or 0.5 mg/kg apomorphine. Dose group (vehicle vs. PCP or apomorphine) was assigned randomly prior to each test.

Group II. A subset of Group I rats was equipped with a second 28-day minipump delivering haloperidol ($n = 17$) or with control pellets ($n = 17$) on post-operative day 25 (24 days after implantation of the first minipump). Haloperidol vs. control pellet/pump was kept constant for all rats. At this time, the original minipump or pellets were removed. These animals were also tested with an acute treatment of vehicle or PCP (1.25 mg/kg or 1.5 mg/kg) 28, 35, 42, and 49 days post-implantation of the initial minipump/pellets and with an acute treatment of vehicle or 0.5 mg/kg apomorphine 31, 38, 45, and 52 days post-implantation of the initial minipump/pellets.

Data Analysis

Data were analyzed using an analysis of variance (ANOVA) with drug treatments as between-subject factors and block and trial type as within-subject repeated measures. Post hoc comparisons were conducted using a one-factor ANOVA. Alpha was set at 0.05. Because treatment (vehicle vs. PCP or vehicle vs. apomorphine) was randomized for each test, duration of haloperidol exposure could not be used as a within-subject variable for assessment of treatment × haloperidol interactions. Instead, separate ANOVAs were calculated for each test date.

RESULTS

Group I

During the first 10 days of sustained haloperidol treatment, haloperidol reversed the PPI-disruptive effects of apomorphine, but not PCP. At several time points between 14–24 days of sustained haloperidol treatment, haloperidol reversed the PPI-disruptive effects of both apomorphine and PCP.

Startle magnitude was potentiated by the interaction of apomorphine and haloperidol during the first 3 weeks of testing. Data are presented in Table 1. At 10, 17 and 24 days post-implantation, post-hoc analyses revealed elevated startle magnitude in the "apomorphine + haloperidol" group compared to either "apomorphine + vehicle", or "vehicle + haloperidol" groups ($p < 0.05$, all comparisons).

Apomorphine produced a decrease in PPI that was reversed by haloperidol at all of the time points tested (Figure 1A–C). Ten and 17 days post-implantation, ANOVAs revealed significant effects of haloperidol on PPI ($F(1,84) > 16.24$, $p < .001$), significant effects of apomorphine ($F(1,84) > 103.32$, $p < .0001$) and significant haloperidol × apomorphine interactions ($F(1,84) > 26.51$, $p < .001$). Twenty-four days post-implantation, ANOVA revealed no significant effect of haloperidol on PPI ($F(1,84) = 2.38$, ns), a significant effect of apomorphine ($F(1,84) = 114.85$, $p < .0001$) and a significant haloperidol × apomorphine interaction ($F(1,84) = 6.42$, $p < .015$). Post hoc comparison revealed significant haloperidol-

Table 1. Mean (SEM) Startle Magnitude

		Treatment						Treatment			
	Day	Sham/Sal	Sham/PCP 1.25 mg/kg	Sham/PCP 1.5 mg/kg	Hal/Sal	Hal/PCP 1.25 mg/kg	Hal/PCP 1.5 mg/kg	Day	Sham/Veh	Sham/Apo 0.5 mg/kg	Hal/Veh	Hal/Apo 0.5 mg/kg
Week 1	7[a]	388.32 (24.3)	474.84 (39.3)	508.61 (63.2)	394.98 (40.6)	597.03 (47.9)	621.81 (77.04)	10[b]	320.40 (25.2)	437.16 (39.6)	341.11 (29.2)	651.28 (53.2)
Week 2	14[c]	313.4 (35.2)	517.47 (32.5)	517.38 (74.1)	367.79 (26.1)	589.70 (53.9)	502.52 (48.4)	17[d]	356.82 (35.3)	364.07 (30.1)	329.19 (26.5)	525.05 (46.5)
Week 3	21[e]	381.39 (31.5)	559.35 (42.8)	276.04 (30.9)	388.66 (34.9)	543.41 (47.3)	429.40 (62.5)	24[f]	330.30 (21.3)	354.90 (24.5)	332.26 (28.5)	549.13 (50.9)
Week 4	28[g]	335.11 (32.4)	476.81 (46.6)	523.61 (29.9)	330.08 (17.9)	418.78 (29.0)	588.78 (68.4)	31[h]	306.97 (28.0)	406.13 (49.2)	341.24 (41.2)	551.30 (61.1)
Week 5	35	389.45 (62.0)	367.70 (41.5)	369.06 (26.1)	407.18 (41.4)	352.17 (57.3)	529.52 (127.8)	38	307.48 (35.3)	297.81 (44.0)	318.81 (38.1)	484.92 (69.4)
Week 6	42	228.05 (32.9)	528.52 (96.7)	468.83 (50.2)	375.79 (71.5)	477.39 (59.9)	416.43 (71.8)	45[i]	333.52 (44.8)	236.35 (33.7)	322.03 (55.6)	561.23 (47.2)
Week 7	49[j]	383.00 (46.38)	419.08 (72.5)	523.78 (40.2)	240.93 (25.6)	597.09 (106.8)	719.07 (94.5)	52	349.48 (60.9)	360.86 (50.8)	340.92 (29.8)	502.41 (49.9)

[a] Effect of PCP $F(2,83) = 6.03$, $p < .004$.
[b] Effect of haloperidol $F(1,85) = 4.98$, $p < 0.03$; effect of apomorphine $F(1,85) = 16.45$, $p < .0001$.
[c] Effect of PCP $F(2,83) = 12.55$, $p < .0001$.
[d] Effect of apomorphine $F(1,84) = 4.55$, $p < .04$; Apo × Hal interaction $F(1,84) = 3.92$, $p < .051$.
[e] Effect of PCP $F(2,81) = 6.77$, $p < .002$.
[f] Effect of haloperidol $F(1,83) = 4.083$, $p < .05$; effect of apomorphine $F(1,83) = 6.18$, $p < .015$; Apo × Hal interaction $F(1,83) = 3.92$, $p < .051$.
[g] Effect of PCP $F(2,33) = 5.29$, $p < .015$.
[h] Effect of apomorphine $F(1,33) = 6.33$, $p < .02$.
[i] Effect of haloperidol $F(1,33) = 75.38$ $p < .03$; Apo × Hal interaction $F(1,33) = 6.19$, $p < .02$.
[j] Effect of PCP $F(2,31) = 9.57$, $p < .0006$; PCP × Hal interaction $F(2,31) = 3.62$, $p < .04$.

induced increases in PPI in apomorphine treated rats at 10, 17 and 24 days post-implantation.

PCP produced a significant increase in startle magnitude that was not reversed or facilitated by haloperidol at any of the time points tested (Table 1). Seven, 14 and 21 days post-implantation, ANOVAs revealed no significant effects of haloperidol on startle magnitude ($F(1,83) < 2.41$, ns), significant effects of PCP ($F(2,83) > 6.03$, $p < .004$) and no significant haloperidol × PCP interactions ($F(2,83) < 1$).

PCP disrupted PPI, and this effect was opposed by haloperidol 14 and 21 days post-implantation, but not 7 days post-implantation (Figure 2A–C). Seven days post-implantation, ANOVA revealed no significant effects of haloperidol ($F(1,83) < 1$), an effect of PCP ($F(2,83) = 98.39$, $p < .0001$) and no significant haloperidol × PCP interaction ($F(1,83) < 1$). Fourteen days post-implantation, ANOVA revealed an effect of haloperidol ($F(1, 83) = 4.42$, $p < .04$), an effect of PCP ($F(2,83) = 52.65$, $p < .0001$) and a haloperidol × PCP interaction ($F(2,83) = 3.30$, $p < .045$). Twenty-one days post-implantation, ANOVA revealed no significant effects of haloperidol ($F(1,83) = 2.66$, ns), an effect of PCP ($F(1,83) = 72.84$, $p < .0001$) and a haloperidol × PCP interaction ($F(1,83) = 6.07$, $p < .0035$). Post hoc comparisons revealed significant haloperidol-induced increases in PPI in rats treated with 1.5 mg/kg PCP at days 14 and 21.

Group II

A subset of rats from Group I were then tested during 28–52 days of exposure to haloperidol or placebo pellets or pumps. In these Group II animals, results were similar to those obtained in animals during the first week of testing.

Apomorphine increased startle magnitude on 31 days post-implantation but had no significant effect on startle magnitude during tests on days 38 and 45 post-implantation (Table 1). Apomorphine significantly reduced PPI 31, 38, and 45 days post-implantation (Figure 3A–C). This effect was reversed by haloperidol 38 and 45 days after implantation of the first minipump. Thirty-one days post-implantation, ANOVA revealed no significant effect of haloperidol ($F(1,33) = 2.23$, ns), a significant effect of apomorphine ($F(1,33) = 69.35$, $p < .0001$) and no significant haloperidol × apomorphine interaction ($F(1,33) = 3.86$, $p < .06$). At 38 days post-implantation, ANOVA revealed a significant effect of haloperidol ($F(1,33) = 11.83$, $p < .002$, a significant effect of apomorphine ($F(1,33) = 223.41$, $p < .0001$) and a significant haloperidol × apomorphine interaction ($F(1,33) = 7.08$, $p < .02$). At 45 days post-implantation, ANOVA revealed no significant effect of haloperidol ($F(1,33) = 3.93$, $p < .06$), a significant effect of apomorphine ($F(1,33) = 128.30$, $p < .0001$) and a significant haloperidol × apomorphine interaction ($F(1,33) = 5.34$, $p < .03$). Although ANOVA did not reveal a significant haloperi-

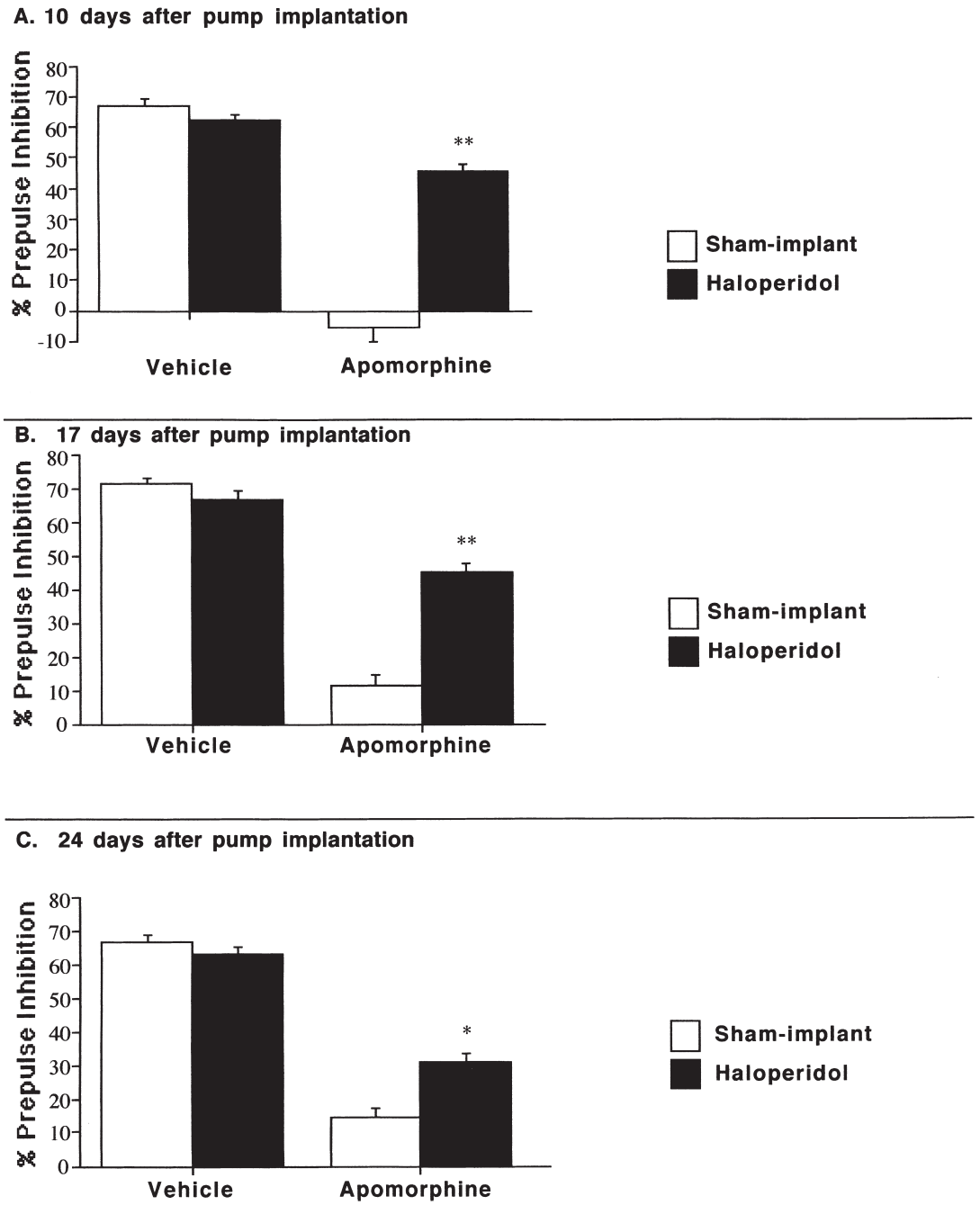

Figure 1. Effects of sustained haloperidol and apomorphine on PPI 10 days **(A)**, 17 days **(B)** and 24 days **(C)** post-implantation of minipumps. *$p < .05$ in animals treated with apomorphine by ANOVA following significant haloperidol × apomorphine interaction. **$p < .0001$ in animals treated with apomorphine by ANOVA following significant haloperidol × apomorphine interaction. Error bars represent S.E.M.

dol × apomorphine interaction at 31 days post-implantation, there was a nonsignificant tendency for haloperidol to reverse apomorphine-induced PPI deficits at this time also [mean PPI (S.E.M.) in apomorphine-treated rats: vehicle group = 6.4 (3.5); haloperidol group = 26.9 (4.8)]. The haloperidol effect size on post-implantation day 31 (0.66) was actually larger than it was on post-implantation days 38 (0.62) or 45 (0.58) (Figure 3A).

PCP significantly increased startle magnitude 28 days post-implantation and significantly reduced PPI 28, 35, and 42 days post-implantation. Haloperidol had no significant effects on startle magnitude and neither reversed nor facilitated the effect of PCP on startle magnitude or PPI 28, 35, or 42 days post-implantation of the first minipump. Startle data are presented in Table 1, PPI data are presented in Figure 4.

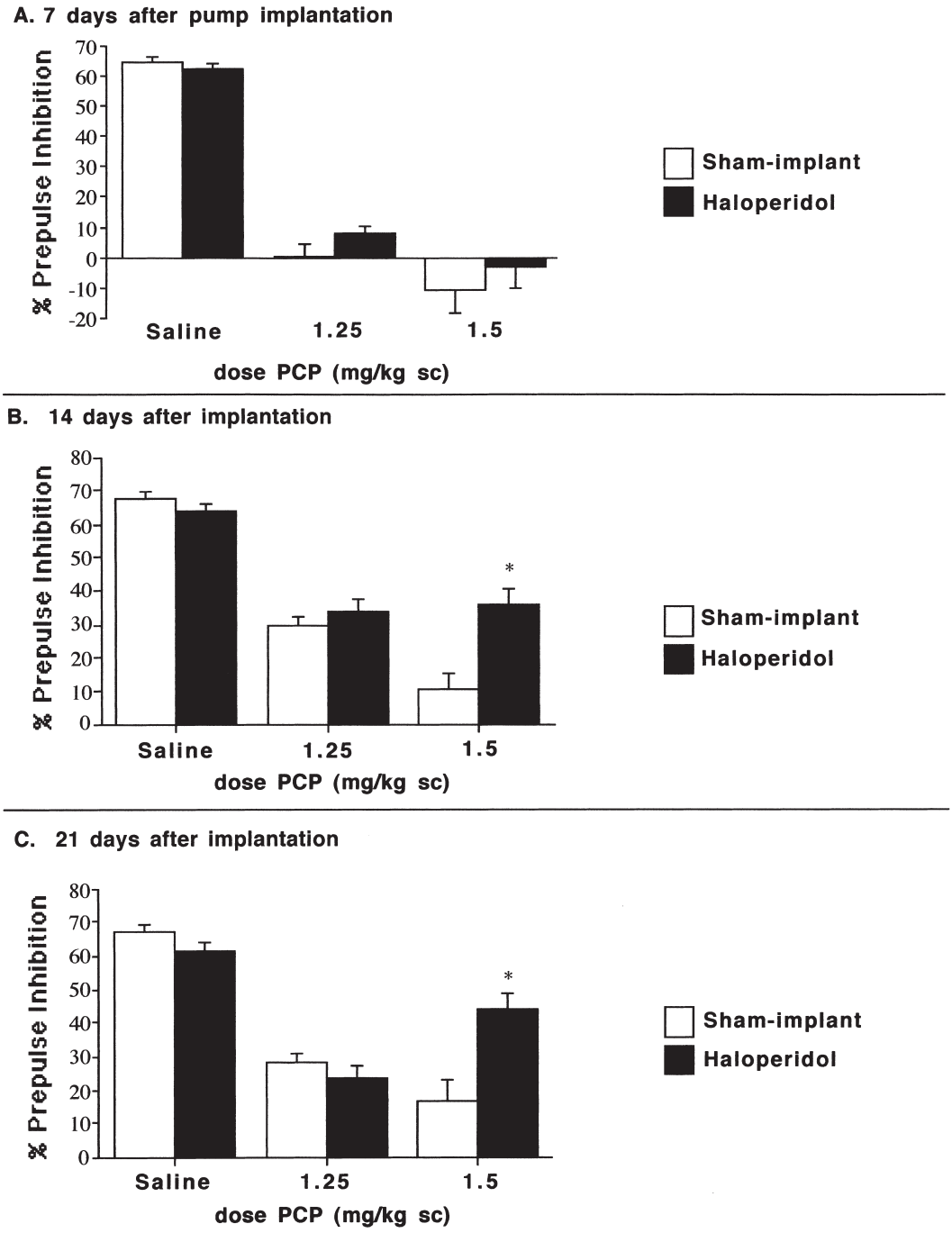

Figure 2. Effects of sustained haloperidol and PCP on PPI 7 days **(A)**, 14 days **(B)** and 21 days **(C)** post- implantation of minipumps. *$p < .05$ in animals treated with 1.5 mg/kg PCP by ANOVA following significant haloperidol × PCP interaction. Error bars represent S.E.M.

Forty-nine days after implantation of the first minipump (approximately 4 weeks after implantation of the second minipump), PCP significantly increased startle magnitude (Table 1). In addition, there was a significant haloperidol × PCP interaction (Table 1). This interaction reflected the reduction in startle magnitude in rats treated with haloperidol + vehicle, compared to the nonsignificant increase in startle in rats treated with haloperidol and either dose of PCP. Haloperidol attenuated the PCP-induced PPI deficit (Figure 4). ANOVA revealed significant effects of haloperidol ($F(2,31) = 7.66$, $p < .009$), and PCP ($F(2,31) = 46.73$, $p < .001$), as well as

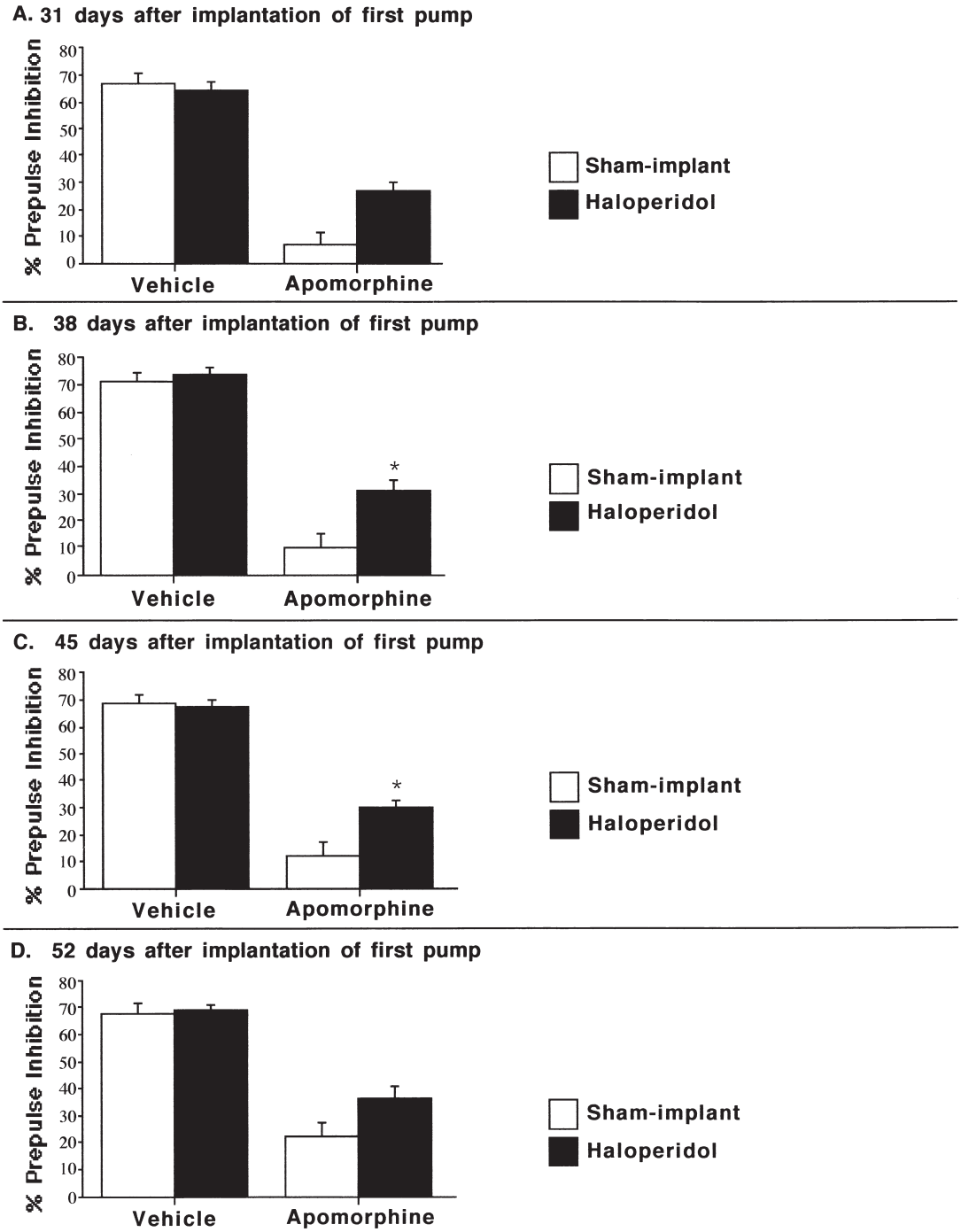

Figure 3. Effects of sustained haloperidol and apomorphine on PPI 31 days **(A)**, 38 days **(B)**, 45 days **(C)**, and 52 days **(D)** post-implantation of minipumps. *$p < .05$ in animals treated with 0.5 mg/kg apomorphine by ANOVA following significant haloperidol × apomorphine interaction. Error bars represent S.E.M.

a significant haloperidol × PCP interaction ($F(2,31) = 8.05$, $p < .0015$). Across both doses of PCP (1.25 and 1.5 mg/kg), PPI was increased significantly in haloperidol vs. vehicle minipump rats ($F(1,15) = 10.09$, $p < .007$).

After the second set of pellets had been implanted for 28 days, some of the effects of haloperidol on apomorphine-disrupted PPI appeared to wane. Thus, 52 days after implantation of the first minipump (4 weeks after implantation of the second minipump), the interactions of apomorphine and haloperidol in measures of both startle magnitude (Table 1) and PPI (Figure 3) exhibited only nonsignificant trends in the expected directions. ANOVA

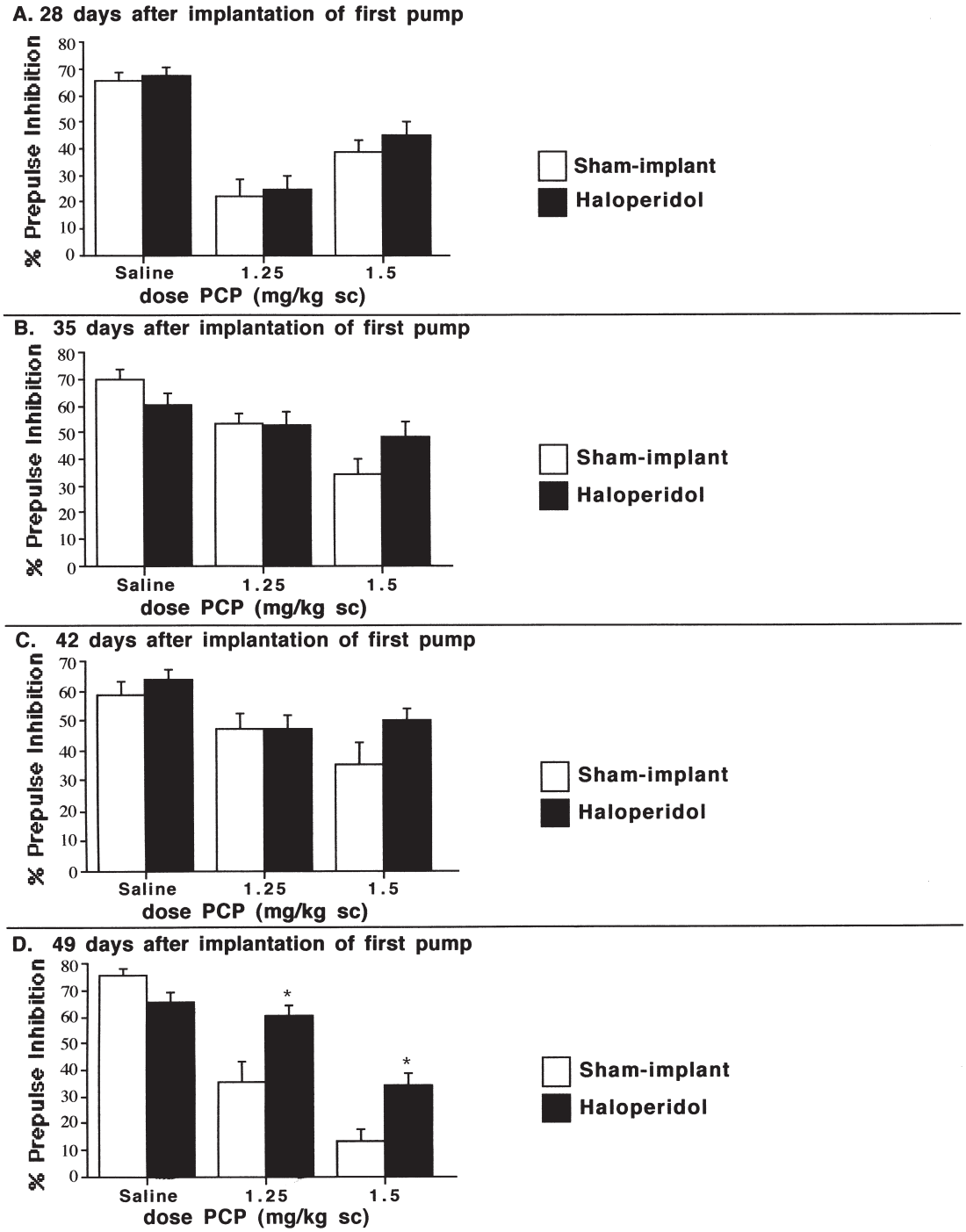

Figure 4. Effects of sustained haloperidol and PCP on PPI 28 days **(A)**, 35 days **(B)**, 42 days **(C)** and 49 days **(D)** post-implantation of minipumps. *$p < .05$ in animals treated with 1.25 and 1.5 mg/kg PCP by ANOVA following significant haloperidol × PCP interaction. Error bars represent S.E.M.

revealed no significant effects of haloperidol or apomorphine on startle magnitude, and no significant haloperidol × apomorphine interaction (Table 1). ANOVA of PPI revealed no significant effect of haloperidol ($F(1,33) = 2.09$, ns), a significant effect of apomorphine ($F(1,33) = 57.42$, $p < .0001$), and no significant haloperidol × apomorphine interaction ($F(1,33) = 1.68$, ns). Among rats treated with apomorphine, more PPI was still exhibited by those who had received haloperidol minipumps [mean (S.E.M.) %PPI = 36.39 (4.29)] vs. control pumps [mean (SEM) %PPI = 22.28 (4.79)], but this difference did not reach statistical significance ($F(1,16) = 2.44$, ns).

DISCUSSION

Reports from several groups have demonstrated consistently that acute treatments with high-potency D2 antagonists, such as haloperidol, do not reverse PPI deficits produced by NMDA antagonists such as PCP (Geyer et al. 1990; Keith et al. 1991; Hoffman et al. 1993). Recent evidence, however, indicates that sustained or subchronic exposure to high-potency D2 antagonists via drinking water or repeated daily injections does attenuate or reverse PCP- or dizocilpine (MK-801)-induced PPI deficits (Pietraszek and Ossowska 1998; Feifel and Priebe 1999). The present study assessed the effects of sustained subcutaneous infusion of haloperidol on apomorphine- and PCP-induced PPI deficits.

As predicted based on its effects after acute injection, sustained administration of haloperidol significantly reversed the PPI-disruptive effects of apomorphine over 10 to 45 days. This reversal of the apomorphine effect provided evidence for bioactivity of the sustained haloperidol treatments, because the effects of haloperidol as a DA antagonist should block the effects of apomorphine as long as adequate concentrations of haloperidol are present in the brain. Although initially haloperidol had no significant effect on PCP-induced PPI deficits, 14 and 21 days after minipump implantation, haloperidol significantly reversed PCP-induced PPI deficits. During the first 21 days of exposure to the second haloperidol minipumps (study days 28–42) haloperidol failed to reverse PCP-induced PPI deficits, but haloperidol significantly reversed these PCP effects 24 days after the second minipumps were implanted (study day 49). Although the magnitude of this haloperidol effect was modest, it was consistent with the magnitude of chronic haloperidol effects in one other report (Pietraszek and Ossowska 1998), and with the magnitude of atypical antipsychotic effects on the PCP-induced disruption of PPI (Bakshi et al. 1994). In addition, although these studies differ in the routes of antipsychotic administration (minipumps vs. drinking water or injections) and the exact time at which D2 antagonists were found to be effective at reversing PCP- or MK-801-induced PPI deficits, the fundamental findings are the same: acute administration of a clinically "typical" antipsychotic is ineffective in reversing NMDA antagonist-induced PPI deficits, while sustained or subchronic antipsychotic administration reverses these effects after a delay of many days to weeks.

These results indicate that sustained exposure (>1 week) to the D2 antagonist haloperidol results in time-dependent changes that are capable of opposing specific behavioral effects of the NMDA antagonist PCP. In this study, these effects are variable over time (i.e., between 28–45 days haloperidol did not reverse the PPI-disruptive effect of PCP). This fluctuation of the ability of haloperidol to reverse specific PCP-induced behavioral effects may reflect changes in haloperidol blood levels, caused by the limitations of the minipumps used in these studies. Throughout this period of variability, haloperidol significantly diminished the PPI-disruptive effects of apomorphine, suggestive of functional D2 receptor blockade. Nevertheless, it is conceivable that during this period, haloperidol levels dropped below a threshold required to sustain anti-PCP effects in this model, despite remaining above a threshold required to reverse the PPI-disruptive effects of apomorphine. This explanation, however, could not easily account for the reinstatement of the haloperidol-blockade of PCP effects 49 days after implantation of the first minipumps, and approxiamtely 24 days after implantation of the second minipumps.

There are several possible explanations for the ability of haloperidol to reverse PCP-induced PPI deficits. First, this effect may result from haloperidol-induced changes in DA receptors, that are not evident after less than 1 week of sustained haloperidol exposure. Such delayed effects might conceivably involve cellular substrates or receptor populations that are not involved in the behavioral impact of acute DA receptor blockade. The present studies did not assess the possibility that the emergence of haloperidol effects after 7 weeks of sustained administration might simply have reflected the passage of time after an initial acute exposure to haloperidol, rather than the cumulative effects of 7 weeks of sustained exposure. Such a delayed physiological response to a single or acute drug treatment has been implicated previously in the delayed clinical response to antidepressants (Antelman and Gershon 1998). At the least, the present results confirm that sustained blockade of D2 receptors for up to 1 week is not a sufficient condition for the restoration of PPI in PCP-treated rats.

A second possible explanation for the delayed emergence of anti-PCP effects of haloperidol in this paradigm is that these effects may result from a direct augmentation of NMDA receptor processes (Banerjee et al. 1995), or via changes in specific interactions between glutamate and DA receptors in frontal cortex (Cepeda et al. 1992), striatum (Amalric et al. 1994; Smith et al. 1994) or nucleus accumbens (Svensson et al. 1994) — all regions implicated in the regulation of PPI (Swerdlow et al. 1992). Other reports suggest that subchronic perturbations of glutamatergic substrates via repeated injections of PCP are capable of modifying behavioral and neurochemical properties of brain DA systems (Jentsch et al. 1997, 1998).

In a recent study, the ability of antipsychotics to restore PPI in PCP-treated rats correlated significantly with their affinity for 5HT2A receptors (Yamada et al. 1999), and it is thus conceivable that the time-dependent effects of haloperidol in this model reflect changes in brain serotonergic activity. One caveat in extrapolating these findings across studies is that Wistar rats were used by Yamada et al. (1999), while the present study

utilized Sprague-Dawley rats. Our group (Swerdlow et al. 1997, 2000) and others (Rigdon 1990; Kinney et al. 1999) have reported substantial differences in drug sensitivity that presumably reflect differences in the underlying neural substrates across rat strains and suppliers. However, Varty et al. (1999) reported that M100907 is equally effective in Sprague-Dawley and Wistar rats in reversing dizocilpine-induced PPI deficits, indicating that for this aspect of possible 5HT-NMDA interactions, drug effects on PPI may be comparable across strains.

The present results confirm that the distinction between "typical" and "atypical" antipsychotic effects in this model is not absolute. While the ability to restore PPI in PCP-treated rats is evident after a single acute injection of clozapine (Bakshi et al. 1994), olanzapine (Bakshi and Geyer 1995) or quetiapine (Swerdlow et al. 1996) and other atypical antipsychotics (Varty et al. 1999), haloperidol "acquires" this ability after 2–4 weeks of sustained administration. It is not known whether the common ability of atypical antipsychotics and chronic haloperidol in this model reflect changes in a common underlying neural substrate.

PPI deficits in schizophrenia patients may be "normalized" by antipsychotics, although this has not been reported in within-subject longitudinal studies, nor is it clear whether this effect is greater in atypical vs. typical antipsychotics (Weike et al. 2000; Kumari et al. 1999). The present studies suggest that, compared to atypical antipsychotics, the PPI restorative effects of typical antipsychotics might be later to emerge in schizophrenia patients; this might account for the relative ineffectiveness of typical antipsychotics to restore PPI in schizophrenia patients in acute settings (Kumari et al. 1999) compared to patients studied after chronic medication (Weike et al. 2000; Young et al. 1995). Negative symptoms of schizophrenia are relatively insensitive to typical antipsychotics. Thus, while the reversal of PCP-induced PPI deficits after acute drug administration may predict clinical utility against negative symptoms, the same cannot be said for the reversal of PCP-induced PPI deficits after chronic drug administration. Perhaps most importantly, this study adds to an emerging literature, suggesting that time-dependent clinical effects of antipsychotics can be effectively studied using the PPI model, to help clarify the effect of chronic medication exposure on sensorimotor gating deficits in schizophrenia.

ACKNOWLEDGMENTS

This study was supported by MH01436 and MH53484 (NRS), DA02925 (MAG) and VA grant (MAG, ZAM), Department of Veterans Affairs VISN22 Mental Illness Research, Education and Clinical Center (MIRECC) The authors gratefully acknowledge the expert technical assistance of Mr. Navid Taaid and Ms. Pamela Auerbach.

REFERENCES

Amalric M, Ouagazzal A, Baunez C, Nieoullon A (1994): Functional interactions between glutamate and dopamine in the rat striatum. Neurochem Intl 25:123–131

Antelman SM, Gershon S (1998): Clinical application of time-dependent sensitization to antidepressant therapy. Prog Neuropsychopharm Biol Psychiat 22:65–78.

Bakshi VP, Geyer MA (1995): Antagonism of phencyclidine-induced deficits in prepulse inhibition by the putative atypical antipsychotic olanzapine. Psychopharmacology 122:199–201

Bakshi VP, Swerdlow NR, Geyer MA (1994): Clozapine antagonizes phencyclidine-induced deficits in sensorimotor gating of the startle response. J Pharmacol Exp Ther 271:787–794

Banerjee SP, Zuck LG, Yablonsky-Alter E, Lidsky TI (1995): Glutamate agonist activity: implications for antipsychotic drug action and schizophrenia. NeuroReport 6:2500–2504

Braff D, Grillon C, Geyer M (1992): Gating and habituation of the startle reflex in schizophrenic patients. Arch Gen Psychiatry 49:206–215

Braff D, Stone C, Callaway E, Geyer MA, Glick ID, Bali L (1978): Prestimulus effects on human startle reflex in normals and schizophrenics. Psychophysiology 15:339–343

Braff DL, Swerdlow NR, Geyer MA (1999): Symptom correlates of prepulse inhibition deficits in male schizophrenic patients. Am J Psychiatry 156:596–602

Bolino F, Di Michele V, Di Cicco L, Manna V, Danelluzzo E, Casacchia M (1994): Sensorimotor gating and habituation evoked by electro-cutaneous stimulation in schizophrenia. Biol Psychiatry 36:670–679

Cadenhead KS, Geyer MA, Braff DL (1993): Impaired startle prepulse inhibition and habituation in patients with schizotypal personality disorder. Am J Psychiatry 150:1862–1867

Cepeda C, Radisavljevic Z, Peacock W, Levine MS, Buchwald NA (1992): Differential modulation by dopamine of responses evoked by excitatory amino acids in human cortex. Synapse 11:330–341

Feifel D, Priebe K (1999): The effects of subchronic haloperidol on intact and dizocilpine-disrupted sensorimotor gating. Psychopharmacology 146:175–179

Geyer MA, Swerdlow NR (1998): Measurement of startle response, prepulse inhibition, and habituation. In Crawley JN, Skolnick P (eds), Current Protocols in Neuroscience. New York, John Wiley & Sons, unit 8.7, pp 1–15

Geyer MA, Swerdlow NR, Mansbach RS, Braff DL (1990): Startle response models of sensorimotor gating and habituation deficits in schizophrenia. Brain Res Bull 25:485–498

Grillon C, Ameli R, Charney DS, Krystal J, Braff D (1992): Startle gating deficits occur across prepulse intensities in schizophrenic patients. Biol Psychiatry 32:939–943

Hoffman DC, Donovan H, Cassella JV (1993): The effects of haloperidol and clozapine on the disruption of sensorimotor gating induced by the noncompetitive glutamate antagonist MK-801. Psychopharmacology 111:339–344

Jentsch JD, Tran A, Le D, Youngren KD, Roth RH (1997): Subchronic phencyclidine administration reduces mesoprefrontal dopamine utilization and impairs prefrontal cortical-dependent cognition in the rat. Neuropsychopharmacology 17:92–99

Jentsch JD, Taylor JR, Roth RH (1998): Subchronic phencyclidine administration increases mesolimbic dopaminergic system responsivity and augments stress- and psychostimulant-induced hyperlocomotion. Neuropsychopharmacology 19:105–113

Kane J, Honigfeld G, Singer J, Meltzer H (1988): Clozapine for the treatment-resistant schizophrenic: a double-blind comparison with chlorpromazine. Arch Gen Psychiatry 45:789–796

Keith VA, Mansbach RS, Geyer MA (1991): Failure of haloperidol to block the effects of phencyclidine and dizocilpine on prepulse inhibition of startle. Biol Psychiatry 30:557–566

Kinney GG, Wilkinson LO, Saywell KL, Tricklebank MD (1999): Rat strain differences in the ability to disrupt sensorimotor gating are limited to the dopaminergic system, specific to prepulse inhibition and unrelated to changes in startle amplitude or nucleus accumbens dopamine receptor sensitivity. J Neurosci 19:5644–5653

Kumari V, Soni W, Sharma T (1999): Normalization of information processing deficits in schizophrenia with clozapine. Am J Psychiatry 156:1046–1051

Lipton J, Zeigler S, Wilkins J, Ellison G (1991): A silicone pellet for continuous cocaine: comparison with continuous amphetamine. Pharmacol Biochem Behav 38:927–930

Mansbach RS, Geyer MA (1989): Effects of phencyclidine and phencyclidine biologs on sensorimotor gating in the rat. Neuropsychopharmacology 2:299–308

Mansbach RS, Geyer MA (1991): Parametric determinants in pre-stimulus modification of acoustic startle: interaction with ketamine. Psychopharmacology 105:162–168

Mansbach RS, Geyer MA, Braff, DL (1988): Dopaminergic stimulation disrupts sensorimotor gating in the rat. Psychopharmacology 94:507–514

Martinez ZA, Ellison GD, Geyer MA, Swerdlow NR (1999): Effects of sustained cocaine exposure on sensorimotor gating of startle in rats. Psychopharmacol 142:253–260

Perry W, Braff D (1994): Information-processing deficits and thought disorder in schizophrenia. Am J Psychiatry 151:363–367

Pietraszek M, Ossowska K (1998): Chronic treatment with haloperidol diminshes the phencyclidine-induced sensorimotor gating deficits in rats. Arch Pharmacol 357:466–471

Rigdon GC (1990): Differential effects of apomorphine on prepulse inhibition of acoustic startle reflex in two rat strains. Psychopharmacol 102:419–421

Smith Y, Bennett BD, Bolam JP, Parent A, Sadikot AF (1994): Synaptic relationships between dopaminergic afferents and cortical or thalamic input in the sensorimotor territory of the striatum in monkey. J Comp Neurol 344:1–19.

Svensson A, Carlsson ML, Carlsson A (1994): Glutamatergic neurons projecting to the nucleus accumbens can affect motor function in opposite directions depending on the dopaminergic tone. Prog NeuroPsychopharmacol Biol Psych 18:1203–1218.

Swerdlow NR, Bakshi V, Geyer MA (1996): Seroquel restores sensorimotor gating in phencyclidine-treated rats. J Pharmacol Exp Ther 279:1290–1299

Swerdlow NR, Braff DL, Geyer MA, Koob GF (1986): Central dopamine hyperactivity in rats mimics abnormal acoustic startle response in schizophrenics. Biol Psychiatry 21:23–33

Swerdlow NR, Braff DL, Taaid N, Geyer MA (1994): Assessing the validity of an animal model of deficient sensorimotor gating in schizophrenic patients. Arch Gen Psychiatry 51:139–154

Swerdlow NR, Caine SB, Braff DL, Geyer MA (1992): The neural substrates of sensorimotor gating of the startle reflex: a review of recent findings and their implications. J Psychopharmacol 6:176–190

Swerdlow NR, Geyer MA (1993): Clozapine and haloperidol in an animal model of sensorimotor gating deficits in schizophrenia. Pharmacol Biochem Behav 44:741–744

Swerdlow NR, Geyer MA (1998): Using an animal model of deficient sensorimotor gating to study the pathophysiology and new treatments of schizophrenia. Schizophrenia Bull 24:285–302

Swerdlow NR, Martinez ZA, Hanlon FM, Platten A, Farid M, Auerbach P, Braff DL, Geyer MA (2000): Toward understanding the biology of a complex phenotype: Rat strain and substrain differences in the sensorimotor gating-disruptive effects of dopamine agonists. J. Neurosci 20:4325–4336

Swerdlow NR, Varty GB, Geyer MA (1997): Discrepant findings of clozapine effects on prepulse inhibition of startle: Is it the route or the rat? Neuropsychopharmacol 18:50–56

Weike AI, Bauer U, Hamm AO (2000): Effective neuroleptic medication removes prepulse inhibition deficits in schizophrenia patients. Biol Psychiat 47:61–70

Varty GB, Bakshi VP, Geyer MA (1999): M100907, a serotonin 5-HT2A receptor antagonist and putative antipsychotic, blocks dizocilpine-induced prepulse inhibition deficits in Sprague-Dawley and Wistar rats. Neuropsychopharmacol 20:311–321

Yamada S, Harano M, Annoh N, Nakamura K, Tanaka M (1999): Involvement of serotonin 2A receptors in phencyclidine-induced disruption of prepulse inhibition of acoustic startle in rats. Biological Psychiatry 46:832–838

Young KA, Brady MD, Poche C, Hicks PB (1995): Changes in startle, evoked potentials, SPEM, sustained attention and negative symptoms frame a "therapeutic window" for neuroleptics. Abstr Soc Neurosci 21:170S