Induction by antipsychotics of "win-shift" in the drug discrimination paradigm

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benzoyl-N-(((2S)-7-chloro-2,3-dihydro-1,4-benzodioxin-1-yl)methyl)-8-azabicyclo(3.2.1)octane-3-methanamine; SLV313, 1-(2,3-dihydrobenzo[1,4]dioxin-5-yl)-4-[5-(4-fluorophenyl)-pyridin-3-ylmethyl]-piperazine; 8-OH-DPAT, (+/-)-8-hydroxy-2-(di-n-propylamino)tetralin; DA, dopamine; DD, drug discrimination; DL, drug-appropriate lever; SL, saline-appropriate lever; FR, fixed ratio; FRF, sum of the responses made on either lever before the first reinforcement; D_{50}, dose that produced an effect in 50% of the animals; ip, intraperitoneal; sc, subcutaneous

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Abstract

In a two-lever, food-rewarded, drug-discrimination paradigm, behavior appears to be governed by a win-stay/lose-shift rule; rats continue to press the lever that yields food and, when not rewarded, shift responding to the alternative lever. We here report on the effects which antipsychotics and further neuropharmacological agents exert in those conditions. At higher doses, antipsychotics disrupt most or all behavioral parameters in this paradigm. However, at lower doses, rats may select the appropriate lever with normal latency and accuracy, obtain a first food pellet (i.e., “win”) and then, remarkably, shift responding to the alternative lever (“win-shift”). This suggests that antipsychotics can block the effects of reward selectively, i.e. at doses where the initial, secondarily-reinforced behavior including the initiation of lever pressing, remains intact. Indeed, saline-treated rats that are given no reward (i.e., “lose”) after having selected a lever, also press the alternative lever (“lose-shift”). The induction of selective win-shift is specific to antipsychotics but varies greatly among them. Perhaps relating to its alleged “incisive” action on delirium and hallucinations, and surprisingly in view of its extrapyramidal actions, acutely administered haloperidol (0.04-0.08 mg/kg) demonstrates win-shift to an exceptional extent, shared only with the newly proposed agent, F15063 (0.31-0.63 mg/kg); the more sedative antipsychotic, chlorpromazine demonstrated little selectivity. The paradigm offers a novel tool to characterize antipsychotics with regard to presumably pathogenic motivational processes; mixed D2-antagonist/5-HT1A-agonist agents such as F15063 may
conceivably share haloperidol’s powerful antipsychotic action while avoiding the sensitization that develops to haloperidol’s extrapyramidal effects and consequent negative symptoms.
Introduction

In a typical drug discrimination (DD) experiment, subjects are trained to discriminate a given training drug from saline. For example, rats can be trained to press one of two levers (drug-appropriate lever; DL) for food in sessions preceded by the injection of the opioid, fentanyl, and to press the alternative lever (saline-appropriate lever; SL) in sessions preceded by saline injection (e.g., Colpaert and Janssen, 1984).

Rats that have been trained to discriminate fentanyl from saline can demonstrate an exquisite discrimination; the lever selection that is to be made early in the session is correct in nearly 100% of the sessions and occurs with great speed (short latency) and accuracy (the animal pressing the inappropriate lever little if at all before completing the first FR10 schedule on the appropriate lever). After having selected the appropriate lever, the rats continue to lever press, totaling some 1,500 responses/session which typically are all made on the appropriate, reward-associated lever (Colpaert, 1978, 1987; Colpaert and Janssen, 1984). This performance emerges after a training in the course of which all of the parameters progressively improve (Colpaert et al., 1980a, 1980b, 2001) and is presumably governed by an acquired “win-stay/lose-shift” rule (Harlow, 1949; Restle, 1958). In particular, the improvement of appropriate-lever pressing after lever selection has occurred and a first reward has been delivered, reflects that the animal increasingly learns to continue to press the reward-delivering lever and to shift to the other lever when not rewarded. This win-
stay/lose-shift and other rules (such as its opposite: win-shift/lose-stay) offer a powerful description of much of the behavior (e.g., foraging) that occurs in species ranging from insects to man (e.g., Dietrich, 2003; Dietrich et al., 2001; Ferguson et al., 2001; Tremblay et al., 1998; Matsen and Nowak, 2004); with humans, the rules are considered as an attribute of consciousness (Harlow, 1949; Dietrich, 2003).

In rats that have been trained to discriminate fentanyl from saline, the injection of the antipsychotic, haloperidol, not surprisingly, causes SL selection. Remarkably, however, after having selected the SL and having obtained a first food reward, the animals press the other lever, may again shift levers in the further course of the session, and eventually discontinue responding (Colpaert et al., 1977). Thus, while leaving the ability of conditioned stimuli to control (secondarily reinforced) behavior unaffected, haloperidol blocks the response control that is otherwise exerted by the primary reward; similar findings were obtained in saline-treated but non-rewarded animals (Colpaert et al., 1977; Colpaert, 1987). The haloperidol effect also occurred in rats discriminating the psychostimulant, dl-cathinone from saline, indicating that the effect is not specific to the particular stimuli that are being discriminated (Goudie et al., 1986). Similarly, pimozide blocked the food-rewarded pressing of a single lever without affecting motor performance (Wise et al., 1978). These findings originated the anhedonia hypothesis of the role of dopamine in motivational, and also cognitive processes (Wise, 2004; Berridge, 2006). In DD, the “shift” to the alternative lever after a reward has been obtained (“win”), constitutes a dramatic departure from the
governing win-stay/lose-shift rule; where the effect occurs in a selective manner, we here will refer to it as “win-shift” induction.

Under the win-stay/lose-shift rule, a shift occurs if and when the organism loses. In one of the experiments reported here, DD-trained rats were treated with saline, allowed to select either lever, but were given no reward; the experiment determined to what extent lose-shift effectively occurs under the conditions of the present study. Other experiments examined and compared the ability of antipsychotics and further CNS agents to induce win-shift while care was taken to ensure the effect’s behavioral specificity.
Methods

Animals

Male Sprague Dawley rats (Ico: OFA SD (I.O.P.S. Caw) Iffa Credo, Lyon, France), weighing between 240 to 260 g at the beginning of the studies were used. Animals were housed in individual cages (Iffa Credo, Lyon, France; 28 cm x 21 cm x 18 cm) with metal grid floors in air-conditioned rooms (21±1 °C; relative humidity 55 ± 5%) under a 12-hr light-dark cycle (lights on at 0700). Filtered (0.22 µ) water was freely available, but access to standard laboratory food (SAFE, Epinay sur Orge, France) was limited to 10 g per day, except during weekends when food was freely available between 1700 hours Friday and 1400 hours Sunday. Experiments were conducted between 0900 and 1700 hours, Monday through Friday. Animals were cared for in accordance with guidelines set by the U.S. Department of Health and Human Services for humane treatment of animals (Guide for the Care and Use of Laboratory Animals, U.S. DHHS, PHS, National Institutes of Health publication No. 85-23, revised 1985) and the experimental protocol approved (No. 009) by the institutional Ethical Committee.

Apparatus

Experiments were conducted in standard operant conditioning chambers (model E10-10, Coulbourn Instruments, Lehigh Valley, PA, USA) housed in light-
and sound-attenuating enclosures that were ventilated by a fan, which also produced a masking noise. Each chamber contained a house-light that was mounted above a food pellet receptacle located between two levers, which were situated 2.5 cm above the grid floor. Food pellets (45 mg dustless pellets, Bioserv, Frenchtown, NJ, USA) were delivered by a pellet dispenser (model E14-12, Coulbourn Instruments, Lehigh Valley, PA, USA). Scheduling of reinforcement contingencies, reinforcement delivery and data recording were controlled by a MED Associates interface and WMPC® software, (Med Associates Inc., VT, USA) implemented on a desktop computer.

Drug discrimination

The procedures have been described in detail elsewhere (e.g., Colpaert and Janssen, 1984). After rats had initially been trained to complete a fixed-ratio:10 (FR10) schedule for food reward on either of the two levers, drug discrimination training was instituted. Thirty min before every daily (5 days/week) 15-min “training” session, rats received a subcutaneous (s.c.) injection of either 0.04 mg/kg fentanyl or saline, and could obtain food pellets by pressing (FR10) the drug-appropriate lever (DL) or the saline-appropriate lever (SL), respectively. Responses on the inappropriate lever had no programmed consequences. The sum of the responses made on either lever before the first reinforcement (FRF) occurred, was recorded. Every week, each rat was trained once a day, Monday through Friday. Daily drug (D) or saline (S) injections were given according to
two, monthly alternating sequences, i.e., 1) DSSDS, SDDSD, SDSDD, DSDSD and 2) SDDSS, DSDSD, DSSDD, SDSDS. The discrimination was considered acquired when during 10 consecutive sessions the rat made at most two responses on the injection-inappropriate lever before the first reinforcement (and, thus, before having completed the first FR10 schedule on the injection-appropriate lever; FRF ≤ 12).

“Test” sessions were run on Fridays only, whereas training sessions continued to be conducted on other days. During test sessions, the lever on which 10 responses accumulated first was defined as the selected lever and the FRF and the latency (in sec) with which lever selection occurred, were recorded. Unless specified otherwise, after lever selection, the rat received a first food pellet and subsequent reinforcement was made contingent upon pressing the selected lever. The number of responses made on both levers after lever selection and throughout the 15 min session, was also recorded. To ensure the stability of performance and avoid carry-over effects of test conditions, testing was postponed to the next scheduled test day if (1) on either of the two most recent training days, the FRF value exceeded 15, (2) on either of the two most recent training days, the total response rate was less than 80% of that observed during the preceding training session of the same type (i.e., drug or saline), or (3) during the most recently preceding saline training session, the total number of responses was less than 500. Also, test data were discarded and the test condition later retested if the test session was followed by a training session of which the FRF value exceeded 15.
After the animals had reached the DD acquisition criterion and to familiarize them with the test procedure, a fentanyl dose-response was conducted, involving tests with randomized 0.0025, 0.005, 0.01, 0.02 and 0.04 mg/kg doses of the training drug being injected s.c., 30 min before test sessions (data not shown). The experiments described below involved 89 rats that had reached this stage. The experiments called for various test conditions to which the animals were assigned randomly.

Though a priori any discriminandum and any discrimination procedure might be utilized for the purposes set out above, the experiments were conducted in fentanyl-saline discriminating rats. This is because, in our experience with this procedure, it has proven difficult to obtain with other training drugs or, for that matter with external discriminanda, the degree of accuracy and reliability that can be achieved with fentanyl-saline discrimination. Also, more so than most alternatives, the particular DD procedure used here usefully affords the assessment of different parameters in addition to stimulus discrimination.

Experiments
The experiments determined the dose-dependent effects of haloperidol and of other clinically established antipsychotics (i.e., chlorpromazine, thioridazine, raclopride, aripiprazole, nemonapride, ziprasidone, olanzapine, clozapine, and risperidone) as well as of further agents currently being developed for the treatment of schizophrenia [i.e., F 15063 (Depoortère et al., 2007), bifeprunox,
SLV313 McCreary et al., 2007), SSR181507 (Claustre et al., 2003). Also tested were compounds that may interfere with DD performance by different molecular and/or neurobiological mechanisms (see: Discussion; i.e., scopolamine, dizocilpine) or that constitute useful tools in examining the possible involvement of some major neurotransmitter systems (e.g., ritanserin, naltrexone, prazosin).

Drugs were injected (n=7/dose) either s.c. or intraperitoneally (i.p.), as specified below, 60 min before the test session; 30 min before the session, and in order to conduct the tests in otherwise familiar training conditions (where either saline or 0.04 mg/kg fentanyl were injected s.c. at that time), saline was injected s.c.

To control for the drug tests described above, saline injections were given, 60 min before the test session, either s.c. (n=12) or i.p. (n=10), followed by another (s.c.) saline injection 30 min before the session; here, as with drug tests, reward was delivered upon lever selection, and these animals are collectively referred to as “saline, reward” controls. Two further conditions served to control for two non-pharmacological manipulations. One examined the effects of withdrawing the primary reinforcer; rats were given saline either s.c. or i.p. (n=7 in both cases) 60 min as well as 30 min (s.c.) before the session, and were administered a test session as described above except that no food pellets were delivered upon selecting or pressing the selected or, for that matter, the alternative lever (the empty food pellet dispenser nonetheless being activated and generating the same sound as that associated with actual food delivery, this condition is referred to as “saline, no-reward, sound”). In the DD paradigm used here, several
stimulus conditions (e.g., injection procedure, introduction into the apparatus) predict or are associated with the eventual delivery of the food pellet/primary reinforcer and are potentially amenable to conditioning; one of those stimuli is the seemingly stark and salient sound produced by the pellet dispenser. Thus, another manipulation sought to explore the effects of dispenser-generated sound; rats were given a s.c. or i.p. (n=7 in both cases) injection 60 min, and a s.c. saline injection 30 min before, and were then administered a test session as described above except that the pellet dispenser was de-activated, delivering neither the pellet nor the sound (“saline, no-reward, no-sound”; collectively, the two latter conditions are referred to as “saline, no-reward”).

Haloperidol and F 15063 excelled the other antipsychotics tested: they induced “win-shift” (see below) to the same (86%) extent to which maximal “lose-shift” was observed in saline, no-reward (control) animals; both compounds also maintained that effect at two consecutive doses (see Results). To examine the reproducibility of these finding, haloperidol and F 15063 were examined also in two different groups (n=7/group) of newly trained rats that had not been tested before with any antipsychotic. In these further experiments, haloperidol and F 15063 were tested at incremental doses in an additional effort to minimize the possible carry-over effects that might have occurred in the first series of experiments.

Data analysis
Test sessions generated data on five variables: 1) the selected lever, i.e., SL or DL, 2) the latency to lever selection, i.e., the time between the start of the session and the occurrence of lever selection, 3) the FRF value, i.e., the total number of responses that were made on both levers before either lever was selected, 4) the percentage of responses on the selected lever after lever selection, and 5) the rate of responding after lever selection, i.e., the total number of responses made on both levers after lever selection divided by the number of seconds between lever selection and the end of the session. Effects on selection latency, FRF value, and percentage- and rate of responses on the selected lever were examined for significance by comparing the mean value obtained during a test session with the mean control value of five saline training sessions most recently preceding the test session by means of the Wilcoxon matched-pairs signed-rank test (NCSS; Kaysville, UT, USA). In addition, the significance of effects in individual animals was evaluated by determining whether or not responding was within ±2 S.D. of the control values calculated from the five saline training sessions most recently preceding the test sessions. If a drug had significant effects in 50% or more of the animals, the dose that produced an effect in 50% of the animals (D50) was estimated by linear interpolation.

“Win-shift” was considered to have occurred when all of the following five conditions were met. (1) The animal selected the saline lever. Relative to the values calculated from the five saline training sessions that most recently preceded the test session, (2) the latency to lever selection and (3) the FRF were less than
the mean + 2 standard deviations (S.D.), and (4) the percentage of responses made on the non-selected lever after lever-selection was more than the mean + 2 S.D. Finally, and avoiding that data be based on excessively small samples, (5) at least 5/7 (or 71%) of the animals tested made a lever selection. These criteria were used to calculate for each treatment condition the percentage of animals showing “win-shift”.

Note that this composite operational definition not only requires that a “shift” occurred, but also ensures as much as possible that the “win” occurred in a behaviorally selective manner. Indeed, antipsychotics can depress different behaviors, including total responding in the DD paradigm (e.g., Colpaert et al., 1977; Goudie et al., 1986) and all the paradigm’s parameters can be fully disrupted by different further agents and neurobiological mechanisms. A striking example is the deterioration by scopolamine of each of the parameters that are assessed before and after lever selection occurs and such that the behavior eventually resembles that seen at the very outset of discrimination training (Colpaert et al., 2001). With scopolamine, the shift thus is non-specific; it and the compound’s other effects in the DD paradigm result from scopolamine causing state-dependent retrieval and a dose-dependent, regressive, temporally graded retrograde amnesia. Scopolamine eventually induces a failure to retrieve all that was learned in the paradigm, including the win-stay rule and the “win” itself (i.e., the instrumental relationship between appropriate lever pressing and the obtainment of food reward; Colpaert et al., 2001; see also: Koek et al., 1995).
For each drug, the maximum percentage animals showing “win-shift” was determined. The relationship between this maximum and the therapeutic classification of each drug as antipsychotic or non-antipsychotic was quantified by means of the point biserial correlation coefficient (NCSS).

The results obtained during the different saline tests were analyzed by means of the Mann-Whitney U-test (NCSS).

Drugs

Fentanyl citrate, haloperidol, S(-) raclopride tartrate, chlorpromazine HCl, thioridazine HCl, clozapine, risperidone, 8-OH-DPAT HBr [(±)-8-hydroxy-2-(dipropylamino)tetratin hydrobromide], phentolamine mesylate, chlorpheniramine maleate, prazosin HCl, propranolol HCl, naltrexone HCl, diazepam, (+) dizocilpine maleate, and ritanserin were purchased from Sigma/RBI (Natick, MA), scopolamine HBr was purchased from Acros (Geel, Belgium), and cocaine HCl was purchased from Coopération Pharматeutique Française (Melun, France). F15063 (3-cyclopent-1-enyl-benzyl)-[2-(2,2-dimethyl-2,3-dihydro-benzofuran-7-yloxy)-ethyl]-amine fumarate), ziprasidone HCl, aripiprazole, nemonapride, olanzapine, bifeprunox (DU127090), and SSR181507 HCl ((3-exo)-8-benzoyl-N-[(2S)-7-chloro-2,3-dihydro-1,4-benzodioxin-1-yl]methyl]-8-azabicyclo-[3.2.1]octane-3-methanamine hydrochloride) were synthesized in-house. SLV313 (piperazine, 1-(2,3-dihydro-1,4-benzodioxin-5-yl)-4-[[5-(4-fluorophenyl)-3-pyridinyl]methyl] was kindly donated by Solvay Pharmaceuticals (Weesp, The
Netherlands). Haloperidol, nemonapride, olanzapine, thioridazine, clozapine, risperidone, prazosin, ritanserin, and SLV313 were dissolved in distilled water with a drop of lactic acid, after which the pH was adjusted to 5 to 7 with 1 N sodium hydroxide. F 15063, aripiprazole, ziprasidone, diazepam, and bifeprunox were suspended in distilled water by adding Tween 80 (2 drops/10 ml). All other compounds were dissolved and administered in distilled water. Drugs and saline were injected in a volume of 1 ml/100 g; doses refer to the weight of free base.
Results

In addition to a s.c. saline injection 30 min before the session, saline tests involved another s.c. or i.p. injection of saline 60 min before the session; there were no differences associated with the route of this injection, and the data were pooled.

In (saline, reward) tests that controlled for drug administrations, SL selection occurred in 95% of the 22 animals, with a mean (SEM) selection latency of 24 (3.3) s, a mean FRF value of 10.4 (0.26), a mean percentage of responses on the selected lever of 100 (0.01), and a mean rate of responding after lever selection of 2.04 (0.10) responses/s (Fig. 1, open circles labeled “saline, reward”). Also, none of the animals failed to select a lever, and the percentage of animals with increased selection latency, increased FRF values, and decreased response rate after lever selection was 4.5, 9, 4.5, and 0%, respectively (Fig. 1, closed circles labeled “saline, reward”). Most importantly, only one rat (4.5%) demonstrated a decreased percentage of responses on the selected lever (i.e., “shifted” to the alternative lever), and “win-shift” thus occurred in 4.5% of the animals.

In tests that controlled for the omission of reward (saline, no-reward, sound) and of the sound of a stimulus (dispenser sound) that was otherwise associated with reward (saline, no-reward, no-sound), the results obtained with saline in the absence of reward were similar to the aforementioned saline test results in terms of
lever selection, latency and FRF. However, in the saline, no-reward, sound condition (n=14), the absence of reward significantly decreased the mean percentage of responses on the selected lever to 84 (4.1) (P<0.0001; such that 100% of the animals demonstrated decreased responding on that lever) as well as the mean response rate after lever selection to 0.70 (0.11) responses/s (P<0.0001; such that 100% of the animals demonstrated a decreased response rate).

When saline was tested in the absence of both reward and feeder sound (saline, no-reward, no-sound; n=14), the response rate after lever selection was even more decreased, to 0.33 (0.05) responses/s (P<0.01), but none of the other measures were changed further. The combined results of all saline tests in the absence of reward (; n=28) are represented in Fig. 1 by the symbols labeled “saline, no-reward”. Most importantly, under these conditions, all 28 animals demonstrated a decrease in the percentage of responding that was made on the selected lever; 4 of them performed poorly on other measures (i.e., lever selection, FRF or latency), so that a composite “lose-shift” criterion (identical to the “win-shift” criterion, but no food was actually delivered) was satisfied in 86, not 100% of the animals. In summary, after saline treatment, the composite criterion in rewarded and non-rewarded conditions was satisfied in 4.5% (“false positives”) and 86% (“true positives”) of the observations, respectively.

Detailed accounts of data obtained with different pharmacological agents are provided in Table 1 and Figs. 1-3 and 5; a summary of the win-shift results is provided in Figs. 4 and 6.
Haloperidol exerted no effect on the SL selection, the lowest percentage being 86% (Fig. 1, upper left panel). As the dose of haloperidol was increased, the percentage of animals failing to select a lever increased, as did the lever selection latency; FRF was not significantly affected, and the percentage of responses on the selected lever as well as the rate of responding after lever selection were significantly decreased (Fig 1, left panels). Note that the results obtained at intermediate doses of haloperidol were similar to the results obtained when saline was administered but no food pellets were delivered (Fig.1, “saline, no-reward”) : in both cases, the behavior before lever selection occurred (i.e., SL selection, selection latency, FRF) was normal (i.e., similar to the behavior during saline training sessions), but the percentage of responses on the selected lever and rate of responding after lever selection were decreased.

All other antipsychotics had similar effects in that they tended to decrease the percentage of selected lever responding (i.e., to produce a “shift”) and the rate of responding after lever selection (see next-to-lowest and lowest panels, respectively, in Figs. 1-3). However, the doses at which these latter (post-lever-selection) effects occurred relative to those at which effects occurred on the other, antecedent parameters appeared to differ among the antipsychotics. Note that the further antipsychotics in Figs. 1-3 are shown in an order based on the number of doses that significantly decreased the percentage of responses on the selected lever. This number of doses ranged from 5 with aripiprazole to 0 with clozapine and risperidone, in spite of the fact that clozapine and risperidone, like the other
compounds, were tested up to doses that significantly increased lever selection latency as well as the percentage of animals that failed to select a lever.

Based on the results shown in Figs 1-3, the percentage of animals showing win-shift (i.e., normal saline lever selection parameters, followed by increased responding on the non-selected lever) was determined (Fig. 4). The antipsychotics differed markedly in their ability to induce win-shift; the maximum percentage of animals showing win-shift ranged from 86% for haloperidol to 29% with clozapine and risperidone (Fig. 4, Table 1). Non-antipsychotics were either unable to produce win-shift, or produced it in at most 29% of the animals (Fig. 4 insert, Table 1). Note that all non-antipsychotics, except phentolamine, chlorpheniramine, naltrexone and ritanserin, were tested up to and including doses that significantly increased lever selection latency. The maximum percentage win-shift obtained with the different compounds correlated significantly with their therapeutic classification as “antipsychotic” or “other” (Fig. 4 insert; point biserial correlation $r = 0.80$, $P<0.0001$). In this correlation, haloperidol appeared to be a significant outlier (externally studentized residual $= 2.47$), being more efficacious to induce win-shift than expected from the results obtained with all other compounds. Similar results were obtained with haloperidol when incremental doses were tested in a separate, newly trained group of rats that had not been tested before with any antipsychotic (Fig. 4, open triangles).
The effects of the potential antipsychotics F 15063, bifeprunox, SLV313, and SSR181507 were similar to those obtained here with established antipsychotics: all four compounds decreased the percentage of selected lever responding and the rate of responding after lever selection at doses lower than those affecting lever selection (Fig. 5, Table 1) and produced “win-shift” in 71-100% of the animals tested (Fig. 6, Table 1). The results obtained with F 15063 when incremental doses were tested in a separate group of newly trained rats that had not been tested before with any antipsychotic (Fig. 6, open triangles) were similar to those observed with random-ordered doses of F 15063 in rats that had been tested with other antipsychotics (Fig. 6, open circles).
Discussion

The present data confirm that, in rats discriminating 0.04 mg/kg of fentanyl from saline in a two-lever discrimination paradigm, behavior is largely governed by a win-stay/lose-shift rule that is acquired during discrimination training. Saline-treated rats, when reward was delivered upon their selecting any lever in test sessions (saline, reward), consistently continued to press the selected lever (“win-stay”; 95.5%). When no reward was delivered, the (saline, no-reward) animals consistently demonstrated significant alternative-lever responding (“lose-shift”; 100%); in 86% of the tests, this shift occurred in the absence of any significantly aberrant behavior prior to lever selection.

The results also confirm that when administered haloperidol, and in spite of obtaining food reward upon lever selection (“win”), the rats significantly “shift” responding to the alternative, non-reinforced lever (Colpaert et al., 1977; Goudie et al., 1986). The data extend this finding to other established antipsychotics albeit that the effect did not reach statistical significance with clozapine and risperidone. With several antipsychotics, the shift significantly occurred (Figs. 1-3; next-to-lowest panel) at doses that also began to impair lever selection and any shift that clozapine and risperidone might possibly produce, may have been masked by such an impairment.

Compounds in the present paradigm may produce a shift and, in fact, act to deteriorate some or all of the further four parameters that characterize behavior in
this paradigm (see: Methods); we therefore analyzed the data (Figs. 4,6) to determine the extent to which antipsychotics may cause the shift in a behaviorally specific manner, i.e., in the absence of any significant effect on the parameters that characterize behavior before any shift can occur (that is, prior to lever selection).

This analysis shows that all established antipsychotics tested produced behaviorally specific “win-shift”; the extent to which they did so varied from 29 to 86% and starkly differentiated (P< 0.0001) antipsychotics from a host of other CNS agents. Some currently developed, putative anti-psychotics also produced win-shift, and this to an extent that compared favorably with established antipsychotics. Thus, the ability to induce win-shift appears to constitute a distinguishing feature of antipsychotic agents.

The largest extent to which win-shift occurred with established antipsychotics was by 86% and this occurred with two consecutive haloperidol doses (i.e., 0.04 and 0.08 mg/kg). That is, the animals, appropriately, selected the saline lever and did so with normal speed and accuracy; thereafter, the rats shifted to the alternative lever and little further responding on any lever occurred during the remainder of the session. The extent to which this occurred with haloperidol was identical to that (i.e., 86%) to which the omission of reward did so in saline-treated rats. This indicates that haloperidol can counteract the effects on behavior of (positive) primary reinforcement while leaving intact the (antecedent) behavior that is driven
Haloperidol demonstrated this capability in a complete fashion over a two-fold dose range; with higher doses, selectivity with haloperidol and other antipsychotics was compromised, causing the win-shift dose-response curve to reach asymptote or assume an inverted-U shape (Fig. 4,6). This suggests that all antipsychotics may perhaps be capable of inducing a shift, but that the extent to which a selective win-shift is observed is obscured, to a varying degree, by other, detrimental effects.

The motor, extrapyramidal actions and sedation that DA antagonists may produce, are unlikely to account for the present findings; behavior prior to the advent of reward was adequately initiated and performed normally, and the win-shift actually required the rats to “productively” initiate a behavior (i.e., alternative lever responding) that did not occur in (rewarded) saline-control conditions. There also is no apparent involvement of antipsychotic drug effects on memory; in fact, haloperidol-treated rats did not merely stop responding on the selected lever, and their alternative-lever responding suggests that they actually continue to implement the acquired, lose-shift part of the rule. Though random responding and response bias can occur in the present paradigm (Colpaert,1978), haloperidol at 0.04 and 0.08 mg/kg led all rats to select the saline lever and only then consistently press the alternative lever.

While chlorpromazine produced (selective) win-shift in no more than 33% of rats, haloperidol here was particularly effective; it induced win-shift to an extent (i.e.,
that is identical to the maximal extent to which lose-shift occurred in non-rewarded saline-control animals, and maintained that effect at two consecutive doses (see the surface-under-the-curve in Fig. 4). Haloperidol has been described clinically as a particularly “incisive” antipsychotic (Bobon et al., 1972; Flach, 1975; Gerard et al., 1984), referring to the selective ability to reduce such productive features as delirium and hallucinations as opposed to inducing the prominent sedation that is characteristic of chlorpromazine (Simon et al., 1984; Colonna et al., 1989). Haloperidol blocks dopamine receptors selectively, contrasting with chlorpromazine’s potent noradrenaline and histamine-antagonist properties (Niemegeers and Janssen, 1979; Leysen et al., 1993; Blin, 1999); thus, the differential extent to which haloperidol and chlorpromazine induced win-shift is consistent with a proposed incisive- versus -sedative discriminant in the analysis of clinical antipsychotic drug action (Simon et al., 1984). Interestingly, chlorpromazine and haloperidol are two of the only three antipsychotics which the WHO considers as essential medicines. Having been introduced in 1958, haloperidol continues to be widely used, often as a first treatment option, both with newly diagnosed, acutely agitated patients and as a chronic therapy (Hamann et al., 2003); it also continues to serve as the comparator when new antipsychotics are introduced (Andrezina et al., 2006). Haloperidol’s exceptional selectivity, both in inducing win-shift and in blocking clinical features of psychosis, are surprising inasmuch as the compound is known to potently induce extrapyramidal effects. However, in rats, D₂-antagonist-induced catalepsy becomes prominent by a
neuroadaptive, sensitization mechanism that develops after repeated injection (Schmidt et al., 1999); in schizophrenics, relative improvement declines with prolonged treatment (Kapur et al., 2005). Further work should determine to what extent haloperidol maintains its selectivity after repeated administration in the present conditions. The potential antipsychotics characterized here also induced win-shift to a relatively large extent (Fig. 6); like all established antipsychotics, these compounds act at D$_2$ receptors (Leysen et al., 1993; Blin, 1999; Newman-Tancredi et al., 2006) but, in addition, activate 5-HT$_{1A}$ receptors (Depoortère et al., 2007). 5-HT$_{1A}$ receptor activation counteracts DA receptor-blockade-induced catalepsy (Broekkamp et al., 1988). Conceivably therefore, these compounds may share haloperidol’s clinical antipsychotic features while avoiding sensitization to extrapyramidal effects.

That antipsychotics can selectively block the response control exerted by primary food reward has been demonstrated in the DD (Colpaert et al., 1977; Goudie et al., 1986) as well as in non-discriminative paradigms (Wise et al., 1978; Ettenberg et al., 1986), and these agents reportedly block the reinforcing action of diverse rewards (Salamone et al., 2003; Wise, 2004). It is unclear whether antipsychotics can block the control of negative primary reinforcers in a similarly selective fashion; in stark contrast with the present findings, in the conditioned (shock) avoidance paradigm, antipsychotics demonstrate an opposite selectivity, reportedly requiring doses to inhibit the conditioned response that are lower than
or equal to, but never higher than those inhibiting escape (Wadenberg and Hicks, 1999).

That antipsychotics block the response control by rewards suggests that their therapeutic action may in part consist of blocking the control that rewards, in schizophrenics, exert in a pathogenic, possibly excessive manner. One current theory suggests that excessive incentive learning is instrumental in causing such pathological features as delirium (Schmidt and Beninger, 2006; Kapur et al., 2005), but the role of dopamine and the effects of DA receptor blockade in reward-related processes remains complex and a matter of debate (Wise, 2004; Berridge, 2006).

In conclusion, in a two-lever, food-reinforced, DD paradigm that is governed by a win-stay/lose-shift rule, antipsychotics induce win-shift; in spite of receiving food reward upon the selection of any lever, rats shift responding to the alternative, non-rewarded lever. The ability to induce behaviorally selective win-shift (i.e., in the absence of detrimental effects on secondarily reinforced behaviors) constitutes a feature that is specific to and perhaps a prerequisite for antipsychotics. Possibly relating to its alleged “incisive” action on delirium and hallucinations haloperidol demonstrates win-shift to an exceptional extent. It would be of interest for further studies to investigate the receptor mechanisms of win-shift and to similarly analyze antipsychotic drug actions while implementing negative reinforcement.
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References:


McCreary AC, Glennon JC, Ashby CR Jr, Meltzer HY, Li Z, Reinders JH,


Legends for Figures:

Fig. 1. Effects of haloperidol, aripiprazole and ziprasidone on responding before and after lever selection occurred in rats (n=7 per dose) trained to discriminate 0.04 mg/kg fentanyl from saline using a two-lever, fixed-ratio 10, food-reinforced procedure. Also shown are data obtained with two different saline control conditions, one involving the normal delivery of food pellets (“saline, reward”; n=22) and one during which food pellets were not delivered (“saline, no-reward”; n=28). During test sessions, the lever on which 10 responses accumulated first was defined as the selected lever, and subsequent reinforcement could be obtained only by pressing the selected lever. The upper three panels show the percentage of animals selecting the saline lever, the mean lever selection latency, and the mean sum of the responses made on either lever before the first reinforcement (FRF) (open circles, left ordinates), and the percentage of animals failing to select a lever, showing increased selection latency, and showing increased FRF values (closed circles, right ordinates). Measures derived from responding after lever selection occurred are shown in the lower two panels, i.e., the mean percentage of responses on the selected lever and the mean response rate (open circles, left ordinates), and the percentage of animals showing decreased responding on the selected lever (shift behavior), and showing decreased response rate (closed circles, right ordinates). Mean lever selection latency, percentage animals with
increased lever selection latency, and percentage animals failing to select a lever were calculated based on all 7 rats tested; other means and percentages are presented only when lever selection took place in at least 5 animals. In individual animals, selection latencies and FRF values were considered increased, and percentage and rate of responding on the selected lever were considered decreased, when they differed by more than two standard deviations from the mean of the five saline control sessions most recently preceding the test. Asterisks indicate a statistically significant difference (P<0.05; one-tailed Wilcoxon matched-pairs signed-ranks test) between a mean value obtained during a test session and the mean control value of the five saline sessions most recently preceding the test. Vertical bars represent 1 S.E.M.

Fig. 2. Effects of raclopride, chlorpromazine and thioridazine on responding before and after lever selection occurred in rats (n=7 per dose) trained to discriminate 0.04 mg/kg fentanyl from saline using a two-lever, fixed-ratio 10, food-reinforced procedure. See legend to Fig. 1.

Fig. 3. Effects of nemonapride, olanzapine, clozapine and risperidone on responding before and after lever selection occurred in rats (n=7 per dose) trained to discriminate 0.04 mg/kg fentanyl from saline using a two-lever, fixed-ratio 10, food-reinforced procedure. See legend to Fig. 1.
Fig. 4. Induction by antipsychotics of win-shift behavior as determined from responding before and after lever selection occurred in rats (n=7 per dose) trained to discriminate 0.04 mg/kg fentanyl from saline using an operant, two-lever, fixed-ratio 10, food-reinforced procedure. During test sessions, the lever on which 10 responses accumulated first was defined as the selected lever, and subsequent reinforcement could be obtained only by pressing the selected lever. Win-shift behavior involves a decreased percentage responding (“shift”) on the selected lever, i.e., the lever that had been pressed to obtain the first reinforcement (“win”). In individual animals, the percentage responses on the selected lever was considered decreased if it was more than two standard deviations lower than the mean of the five saline control sessions most recently preceding the test. Here, win-shift behavior is defined as a significantly decreased percentage of responses on the selected lever after otherwise normal lever selection, i.e., after the selection of either lever in the absence of a significant increase of lever selection latency and FRF. The percentage animals showing win-shift was calculated and is shown only when lever selection and responding after lever selection occurred in at least five of the seven animals tested. The insert shows the maximum percentage of win-shift that was produced by any dose of the antipsychotics and other compounds listed in Table 1. Haloperidol was also tested at incremental doses in a separate group of rats not tested before with antipsychotics (open triangles).
Fig. 5. Effects of putative antipsychotics during on responding before and after lever selection occurred in rats (n=7 per dose) trained to discriminate 0.04 mg/kg fentanyl from saline using a two-lever, fixed-ratio 10, food-reinforced procedure. See also legend to Fig. 1.

Fig. 6. Induction by putative antipsychotics of win-shift behavior, as determined from responding before and after lever selection occurred in rats (n=7 per dose) trained to discriminate 0.04 mg/kg fentanyl and saline using a two-lever, fixed-ratio 10, food-reinforced procedure. The insert shows the maximum percentage of win-shift that was produced by any dose of the potential antipsychotics, of the antipsychotics and of the other compounds listed in Table 1. F 15063 was also tested at incremental doses in a separate group of rats not tested before with antipsychotics (open triangles). See legend to Fig. 4.
Table 1. Effects of antipsychotics, potential antipsychotics, and other compounds on win-shift behavior, as determined from responding before and after lever selection occurred, in rats trained to discriminate 0.04 mg/kg fentanyl from saline.

<table>
<thead>
<tr>
<th>Win-shift</th>
<th>Saline lever selection</th>
<th>No lever selection</th>
<th>Lever selection latency</th>
<th>FRF</th>
<th>Percentage responses on selected lever</th>
<th>Rate of responding after lever selection</th>
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<tr>
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<td>dose</td>
<td>min</td>
<td>%</td>
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<td>100</td>
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"min" and "max" indicate minimum and maximum effects; "r/s" indicates responses per second; "med" indicates minimum effective dose. Means and percentages were calculated only when based on at least five animals showing lever selection, except mean lever selection latency, % animals with increased lever selection latency, and % animals failing to select a lever, which were calculated based on all 7 animals that were tested at each dose. D50 values were calculated by linear interpolation; when more than one value could be calculated, the highest value, marked with an asterisk, is reported. Minimum effective doses are the lowest doses with effects significantly higher (lever selection latency, FRF) or lower (percentage responses on the selected lever, rate of responding after lever selection) than control (one-tailed Wilcoxon matched-pairs signed-ranks test). "-" indicates that a value could not be calculated. Dose, D50, and med values are in mg/kg. All compounds were injected sc, except aripiprazole, ziprasidone, olanzapine, clozapine, bifeprunox, F 15063, and diazepam, which were injected ip.
Fig. 1
Fig. 2
Fig. 4

- **Haloperidol**
- **Raclopride**
- **Aripiprazole**
- **Olanzapine**
- **Memonapride**

% Animals showing win-shift vs. dose (mg/kg)

- **Ziprasidone**
- **Thioridazine**

% Animals showing win-shift vs. dose (mg/kg)

- **Chlorpromazine**
- **Clozapine**
- **Risperidone**

% Animals showing win-shift vs. dose (mg/kg)
Fig. 6