Pharmacology Biochemistry and Behavior, Vol. 44, pp. 181–189, 1993
Printed in the U.S.A.

0091-3057/93 $6.00 + .00

Differentiation of Haloperidol and Clozapine Using a Complex Operant Schedule in the Dog

J. BRUHWYLER,[1] E. CHLEIDE, G. HOUBEAU, N. WAEGENEER AND M. MERCIER

Department of Experimental Psychology, University of Namur, 61, Rue de Bruxelles, 5000 Namur, Belgium

Received 23 April 1992

BRUHWYLER, J., E. CHLEIDE, G. HOUBEAU, N. WAEGENEER AND M. MERCIER. *Differentiation of haloperidol and clozapine using a complex operant schedule in the dog.* PHARMACOL BIOCHEM BEHAV 44(1) 181–189, 1993.—This study aimed to differentiate chronically administered typical (haloperidol) and atypical (clozapine) neuroleptics in the dog using a complex temporal regulation schedule combining operant, voluntary, and involuntary motor parameters. Although clozapine and haloperidol showed some characteristics of neuroleptics, justifying their adherence to the same class of compounds, differences have also been highlighted and compared to the clinical observations. Haloperidol induced catalepsy, tremor, dystony, hyperkinesia, and stereotypy. Subjects produced anticipated responses before any stimulus. Incomplete and delayed responses were also produced. An interpretation in terms of akathisia and anhedonia has been suggested. Clozapine induced tremor, exploration, dystony, and hypersalivation. Subjects produced disinhibitory responses to the negative stimulus and incomplete responses but these latter were submitted to tolerance. The simultaneous presence of tranquilizing and disinhibitory effects has been reported on the clinical potential of clozapine both in cases of positive and negative schizophrenic symptomatologies.

Clozapine Haloperidol Atypical neuroleptic Neuroleptic Dog DRRD Extrapyramidal symptoms Chronic administration Akathisia Anhedonia Disinhibition

SINCE it was synthesized in 1960 by Sandoz, Wander Ltd. (37,52), much has been written about clozapine (CLZ) (35). Recently, three theoretical reviews have been devoted to this substance (6,13,28).

While antipsychotic and extrapyramidal actions are often considered the main properties of classic neuroleptic drugs like haloperidol (HAL) and chlorpromazine, CLZ has been described as efficiently inhibiting psychotic reactions in man without producing any clearcut extrapyramidal side effects (4,21,38). In animals, a classic neuroleptic has been defined according to its cataleptic properties, its ability to antagonize apomorphine and amphetamine stereotypies and suppress the conditioned avoidance response (47). With CLZ, most of these properties are no longer strictly in force (23,26,50). So, it calls in question the validity of the preclinical tests carried out to detect the antipsychotic potential of compounds. Moreover, it arouses considerable interest and poses questions not only in connection with the dopaminergic hypothesis of schizophrenia but also about the pharmacological approaches being utilized in searching for new and improved antipsychotic drugs (6,13).

In previous studies, a complex operant conditioning schedule has been used as a screening test in the dog to compare the behavioral, neurophysiological, and motor effects of a large series of acutely administered barbiturates, benzodiazepines, and neuroleptics. Differences have been highlighted not only between these main classes of psychotropics but also within classes, between 1,4-benzodiazepines and 1,5-benzodiazepines, between hypnotic and nonhypnotic benzodiazepines, and between classic and atypical neuroleptics (10,14,15). The aim of this study is to apply the same test to specifically differentiate the effects of HAL and CLZ when chronically administered.

METHOD

Subjects

Ten naive, male dogs (2–4 years old) of Beagle breed (Velaz, Czechoslovakia) weighing from 11–15 kg were used in these experiments. They were housed in separate cages and fed at the end of the day with Cervo Expan diet (250 g).

Test Room

The test room (Fig. 1) was 5.6 × 3.5 m. At the entrance, in the right-hand corner there was a board (60 × 50 × 2 cm) fastened to the ground. In the opposite corner, at the end of the room, the food dispenser (50 × 76 × 52 cm) was situ-

[1] Requests for reprints should be addressed to Dr. Jacques Bruhwyler, Quai au Foin, 15 Bte 14, 1000 Brussels, Belgium.

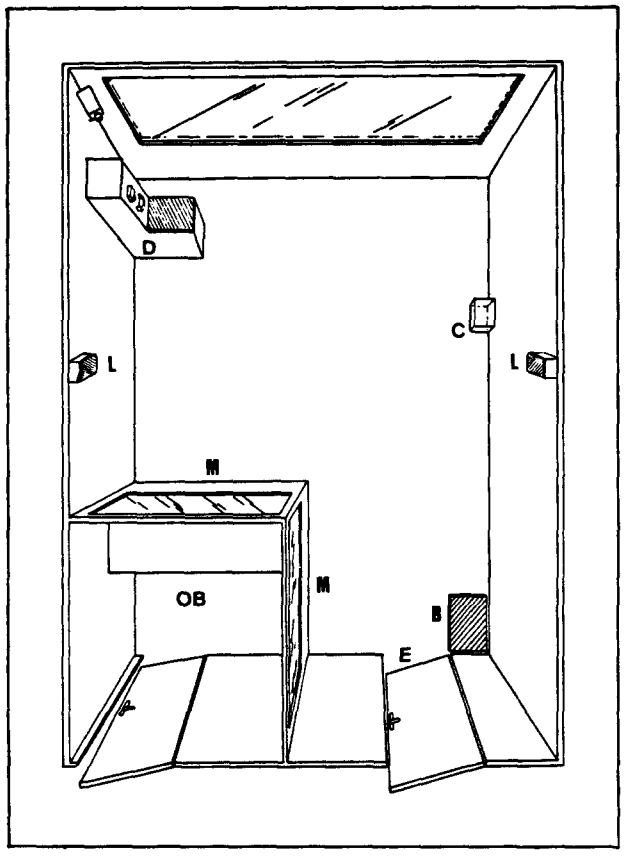

FIG. 1. Test room. (B). Board. (C). Water container. (D). Food dispenser. (E). Entrance. (L) Loud speakers. (M). Mirrors. (OB). Observation booth.

ated. The auditory signals for the test were emitted from two loudspeakers incorporated in the ceiling. Water was available throughout the session. The experimenter stood in an observation booth fitted with two-way mirrors. The booth contained all the controls of the external stimuli and the distribution of reinforcements, as well as materials for observing and recording the sessions. The observation and recording materials consisted of two cameras, one giving an overall view of the room and the other filming the dog on the board. These pictures were recorded on videotape and analyzed.

Procedure

The shaping procedure has largely been described in previous studies (11,46). The final procedure was a schedule of differential reinforcement of response duration (DRRD) with limited hold (LH) and positive and negative external cues (10,14). It consisted of the random alternation of two kinds of trials. Each trial started when the dog spontaneously took place on the board. In the first type of trial, a maintenance response lasting 9 s on the board was required for obtaining reinforcement. At the end of this time delay, an auditory stimulus of 1.5 s was given to the animal. Every time it left the board between 9 and 10.5 s and then jumped on the food dispenser, it received a piece of meat (5 g). The second type of trial differed from the first by the addition of the same auditory stimulus, randomly presented between the third and sixth seconds of the time delay. Both auditory stimuli presented between 3 and 6 s and at 9 s were physically identical and had the same duration (1.5 s); the animal could only discriminate between them according to their location in time. Both kinds of trials were presented in an equal number and distributed randomly during the session. Thus, the added stimulus was double random, first, because it was not given on each trial and, second, because it was given at random between the third and sixth seconds. In every case, the only reinforced response was the response to the stimulus at 9 s; any anticipated (<9 s) or delayed (>10.5 s) response was not reinforced. The intertrial interval and maximum amount of time a trial lasted were not limited. Experimental sessions (one per day) were limited by the subject obtaining eight reinforcements and/or by a maximum time (net time and time elapsed between trials) of 900 s. Performance was considered stabilized when 70% of responses relating to all trials were correct (after 30 sessions or 240 trials).

Drug Administration

After stabilization of the learning performance, two groups of five subjects were constituted. They received either HAL (Haldol®, 0.3 mg/kg) or CLZ (Sandoz Ltd., Basel, Switzerland, 7 mg/kg) orally in capsule form each day (between 0900–1100 h) during 13 days. The experimental sessions took place on days 1, 2, 3, 5, 7, 9, 11, 12, and 13 4 h following drug administration. The withdrawal effects of both drugs were also measured during 4 days. An adequate estimate of the baseline performance was determined by the average on the last 5 drug-free conditioning days. On these days, subjects received a placebo and took part in a session 4 h later. Chronic doses were chosen to be approximately in the human therapeutic maintenance dose range (i.e., 0.08–0.25 mg/kg p.o. HAL and 5–8.3 mg/kg p.o. CLZ) (5,20,23,31,38) but also to be significantly active in this procedure. Our previous acute studies in the dog showed that 7 mg/kg CLZ was the lowest effective dose and 0.1 mg/kg HAL was not significantly effective (10,14,15). So, we decided to use 7 mg/kg CLZ and 0.3 mg/kg HAL in this study.

Parameters

The following measures were taken always by the same rater working in simple blind check:

1. *Operant behavior.* Total and correct response rates (responses/min), temporal distribution of response durations, and number of subjects producing incomplete responses, the latter defined as a correct response duration followed by the dog moved off the board but not followed in the 10 consecutive s by the jump on the food dispenser.
2. *Motor effects.* Effects on involuntary movements included the frequency (number/min) of ataxic movements, defined as a lack of coordination when standing or performing during test trials. Unsteady gait and/or fall were considered evidence of ataxia (23); the frequency of cataleptic positions, diagnosed when the rigid aspect of the posture lasted more than 60 s (23); the frequency of akinetic movements included all the problems occurring during locomotion, such as the cog wheel effect, incoherent zig-zag walking, and hopping features of the animal's gait; the frequency of trembling positions, evaluated when the animal was immobile; the frequency of dystonic symptoms diagnosed if muscular contractions or grimaces affected the peribuccal area.

3. *Voluntary movements.* Voluntary movements included the frequency (number/min) of exploration defined as the sniffing of a specific object with a clear orientation of the head, the frequency of barking, and the frequency of stereotypy and hyperkinesia, diagnosed when the subject repeated a given movement such as scratching, licking, rotating, or self-grooming for more than 30 s or when it walked for more than 60 s without pausing.

The number of subjects showing sialorrhea was also noted.

Statistical Analysis

Statistical analysis was obtained from analysis of variance (ANOVA) for repeated measures, with the factors treatment and day as classification criteria, followed by posthoc Dunnett's *t*-tests. Two separate analyses were undertaken for the chronic administration period compared to the baseline performance and for the withdrawal period compared to the 13th day of administration. The Kolmogorov-Smirnov test was used to compare the temporal distributions of response durations. The Cochran's *Q*-statistic for dichotomous data was used to analyze the effects of drugs on hypersalivation and incomplete responses (58).

RESULTS

Chronic Administration

The effect of the factors treatment and day on total response rate [$F(1, 80) = 51.8$, $F(9, 80) = 5.9$, respectively] and correct response rate [$F(1, 80) = 144$, $F(9, 80) = 13.5$, respectively] were significant ($p < 0.01$). These two parameters were significantly ($p < 0.05$) decreased by HAL and CLZ during the whole period of administration (Fig. 2). The interactions between factors were not significant ($p > 0.05$).

Figure 3 shows the evolution of the temporal distribution of response durations during the pharmacological treatment compared to the baseline performance. For the placebo, it was typically bimodal with the principal mode (75%) centered on 9 s, corresponding to the correct response durations, and with the secondary mode (15%) situated between 3 and 6 s, being the moment at which the negative stimulus was presented. The Kolmogorov-Smirnov test revealed that the distribution was significantly ($p < 0.01$) disturbed by the two drugs but with a significantly ($p < 0.01$) greater intensity for HAL. The two drugs induced an extension of the temporal distribution toward shorter and longer response durations. For CLZ, the principal mode was slightly reduced ($p < 0.05$) to the advantage of the secondary mode between 3 and 6 s. For HAL, the principal mode was shifted ($p < 0.01$) from 9 to 1-3 s, a period where no stimulus was presented to the subject.

Table 1 shows the evolution of the number of subjects producing incomplete responses. Both CLZ ($Q = 22.6$, $p < 0.01$) and HAL ($Q = 18$, $p < 0.05$) significantly increased this parameter. However, with CLZ incomplete responses completely disappeared from day 11.

The effect of treatment on catalepsy, $F(1, 80) = 13.9$, $p < 0.01$, and dystony, $F(1, 80) = 6.6$, $p < 0.05$, was significant. HAL induced a significant ($p < 0.05$) catalepsy the first 2 days of administration. CLZ never induced such an effect. HAL induced a significant dystony ($p < 0.05$) from the seventh day of administration while CLZ had the same effect during the whole period (Table 2). The effect of treatment on ataxia, akinesia, and tremor was nonsignificant ($p > 0.05$). The effect of the factor day on ataxia, $F(9, 80) = 3.7$, $p < 0.01$, and tremor, $F(9, 80) = 2.0$, $p < 0.05$, was significant. However, for akinesia it was nonsignificant ($p > 0.05$). The two drugs induced a significant ataxia and tremor ($p < 0.05$) the first day of administration. The interactions between factors were not significant ($p > 0.05$) except for catalepsy ($p < 0.01$).

The effect of treatment on exploration, $F(1, 80) = 8.1$, $p < 0.01$, and stereotypy/hyperkinesia, $F(1, 80) = 8.6$, $p < 0.01$, was significant. CLZ induced a higher frequency of exploration than HAL. However, the inverse relation was true for stereotypy/hyperkinesia (Table 3). The effect of treatment on vocalization was nonsignificant ($p > 0.05$) but the effect of day was significant, $F(9, 80) = 12$, $p < 0.01$. The two drugs significantly ($p < 0.05$) decreased barking during the whole period of administration. The interactions between factors were not significant ($p > 0.05$). CLZ induced a significant hypersalivation in three to four dogs ($Q = 22.8$, $p < 0.01$) during the 13 days of treatment. This effect was never observed for HAL.

Withdrawal

When comparing the last day of administration with the 4 days of withdrawal, the effect of treatment on total response rate, $F(1, 40) = 22.8$, $p < 0.01$, and correct response rate, $F(1, 40) = 86.5$, $p < 0.01$, was significant (Fig. 2). The differences between the two drugs were maintained. No recovery was measured for CLZ and the apparent recovery for HAL was nonsignificant ($p > 0.05$). The interactions between factors were not significant ($p > 0.05$).

The Kolmogorov-Smirnov test revealed a significant ($p < 0.01$) difference between HAL and CLZ in the temporal distributions of response durations during withdrawal. For HAL, response durations of 1-3 s and >13 s still subsisted ($p < 0.01$). For CLZ, no difference could be measured in comparison to the baseline performance (Fig. 3).

No more incomplete responses were measured for CLZ. However, for HAL one to three dogs still produced incomplete responses and no significant recovery was recorded ($Q = 5.6$, $p > 0.05$) (Table 1). The effect of treatment on akinesia, $F(1, 40) = 9.4$, $p < 0.01$, and dystony, $F(1, 40) = 4.4$, $p < 0.05$, was significant. While HAL still induced akinesia and dystony, a recovery was observed with CLZ (Table 2). No more catalepsy and tremor were noted. Ataxia remained present with the two drugs but at a nonsignificant level ($p > 0.05$).

The effect of treatment on exploration, $F(1, 40) = 4.4$, $p < 0.05$, was significant. This parameter remained higher for CLZ compared to HAL (Table 3). The effects of the factors treatment and day on vocalization and stereotypy/hyperkinesia were nonsignificant ($p > 0.05$). No significant recovery was measured for the two drugs. The interactions between factors were not significant ($p > 0.05$). Hypersalivation significantly decreased ($Q = 10$, $p < 0.05$) and completely disappeared the second day of withdrawal of CLZ.

DISCUSSION

These results largely corroborated our previous acute study (10) showing that HAL and CLZ tended to decrease the total and correct response rates, disturb the temporal distributions of response durations with a shift toward both delayed and anticipated responses, and induce an increase in incomplete responses. They also reinforced the observations made by others that a rapid tolerance (within 2 days) developed in experi-

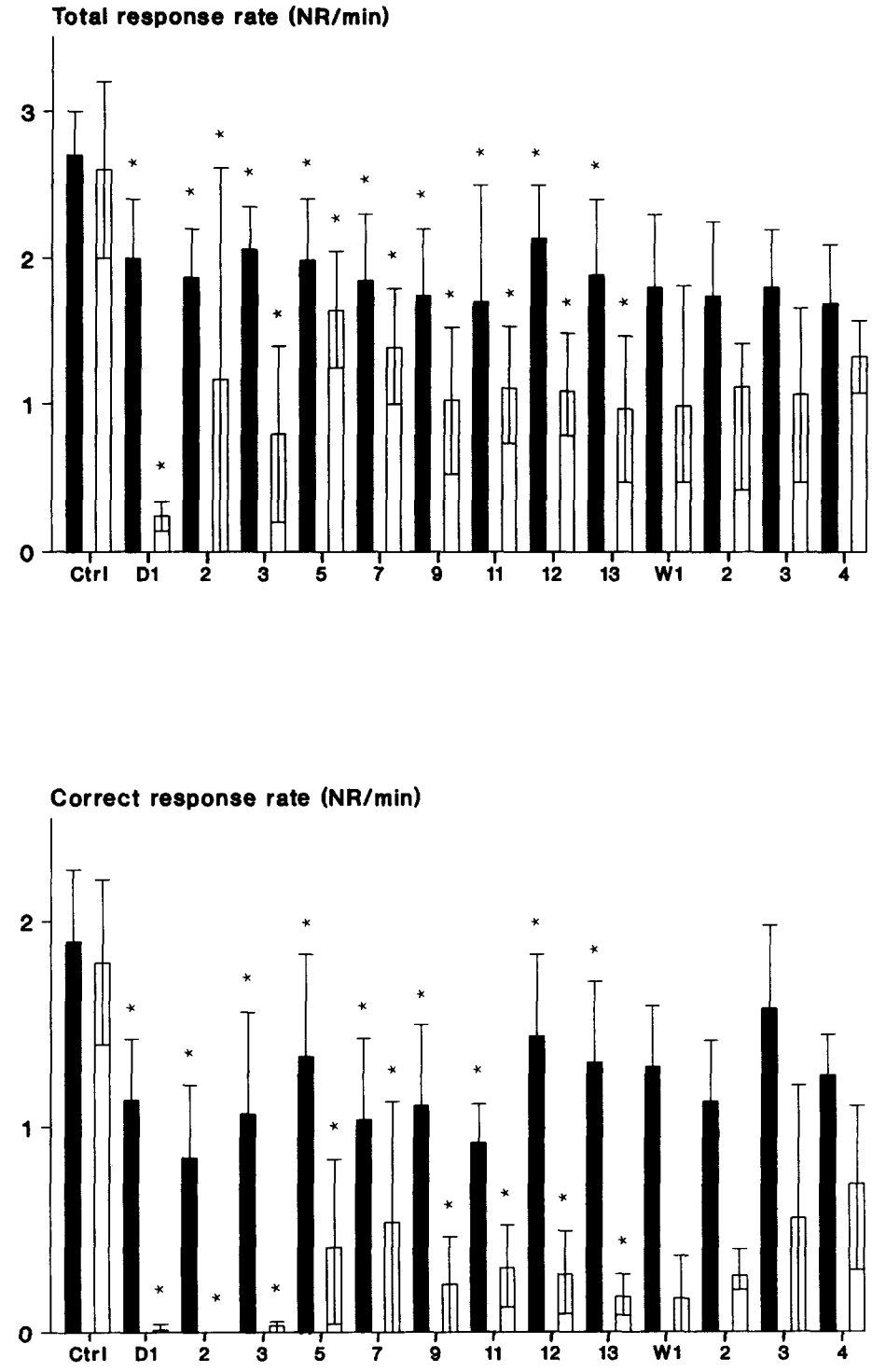

FIG. 2. Evolution of the total response rate (TRR) and the correct response rate (CRR) during chronic administration (D) of clozapine (solid bars) and haloperidol (open bars) and during withdrawal (W) compared to the baseline performance (Ctrl). NR/min, number of responses per minute; *$p < 0.05$ (Dunnett's t-tests).

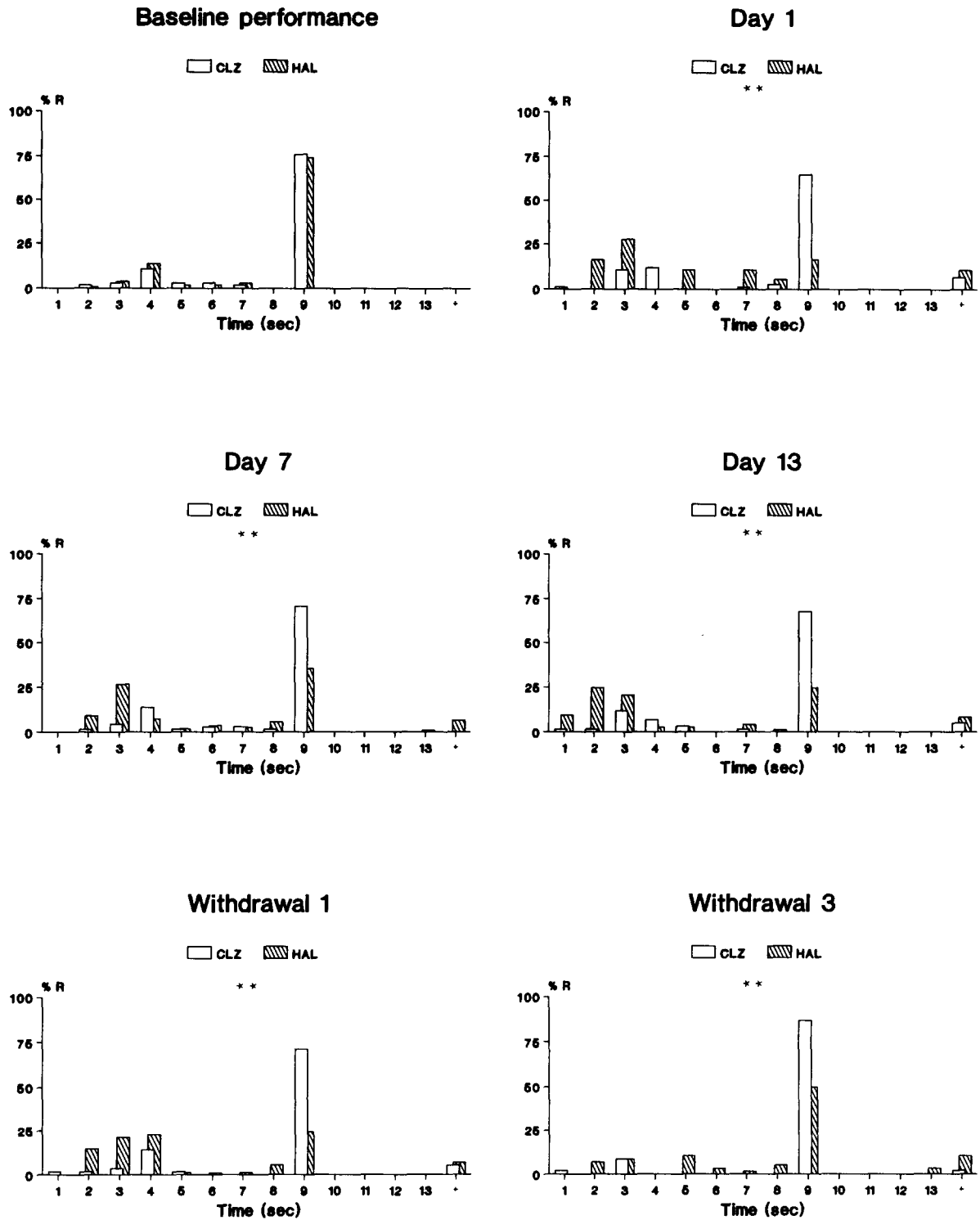

FIG. 3. Evolution of the temporal distribution of response durations during chronic administration of clozapine (CLZ) and haloperidol (HAL) and during withdrawal compared to the baseline performance. +, response durations superior to 13 s; % R, percentage of responses; $^{**}p < 0.01$ (Kolmogorov-Smirnov test).

TABLE 1

INCOMPLETE RESPONSES DURING CHRONIC ADMINISTRATION OF CLOZAPINE AND HALOPERIDOL AND DURING WITHDRAWAL

	Clozapine (n = 5)		Haloperidol (n = 5)	
Baseline	0		0	
Administration				
Day 1	3		3	
2	3		2	
3	2		2	
5	3	*	3	†
7	0		2	
9	1		3	
11	0		3	
12	0		3	
13	0		3	
Withdrawal				
Day 1	0		1	
2	0		2	
3	0		3	
4	0		2	

Data are given as number of subjects producing incomplete responses.

*$p < 0.01$; †$p < 0.05$; Cochran's Q-statistic comparison between days and baseline.

mental animals to the extrapyramidal side effects of chronically administered neuroleptics (3,8,16,29). It was the case for catalepsy and tremor in this study. However, akinesia (nonsignificant) and dystony (significant) remained present until the end of the administration period. Further, no significant withdrawal effect was measured, indicating that neuroleptics did not induce physical dependence (2,5).

Although some of the involuntary motor effects were rapidly submitted to tolerance, the operant performance remained significantly affected and incomplete responses increased during the whole period with HAL and during 8 days with CLZ. An intrasession increase in the rate of incomplete responses accompanied by an intrasession decrease in the total response rate were always noted (data not shown). Moreover, the difficulty to jump on the food dispenser might not be invoked to explain such incomplete responses because the ability to jump persisted when anticipatory and late errors or rare correct responses were made. These results were arguments supporting the hypothesis of neuroleptic-induced anhedonia (7,34,36,42) against the hypothesis of neuroleptic-induced motor deficit (30,51) and corroborated the interpretation of Wise (59), who argued that "subjects treated with a neuroleptic retain the motor ability to produce the operant response but do not do so."

Quantitative differences between the two drugs might also be highlighted. All operant parameters taken into account were much more affected by HAL than by CLZ. The doses of HAL (0.3 mg/kg) and CLZ (7 mg/kg) used in this study were approximately in the range of clinical doses (23,31,33). However, the dog could be more sensitive to HAL compared to CLZ because the ED_{50} measured by Cohen (23) for suppression of conditioned avoidance was 0.08 mg/kg p.o. for HAL and 32 mg/kg p.o. for CLZ. Available pharmacokinetic data were sufficient to suggest that HAL had a longer duration of action (24,48) than did CLZ (19,20,57) and its action could result in a cumulative process. However, the fact that CLZ remained active even during withdrawal seriously limited this single interpretation. Nevertheless, a more complete study of the chronic effects of the two drugs on operant responding, using several doses, is needed before dose-effect relationships in the dog could be compared to the clinical results.

Qualitatively, whatever the dose the effects of HAL and CLZ were distinguishable. As already mentioned in a large number of studies, CLZ did not induce catalepsy while HAL significantly produced this side effect (4,21,23,38). In our study, catalepsy was rapidly submitted to tolerance and fully disappeared after 3 days.

Dystony remained at a significant level during the whole period of administration of CLZ. With HAL, dystony was significant from the seventh day of treatment. These dystonic symptoms were mainly observed when the subject had to eat the reinforcement and in particular during swallowing. The absence of dystony, at the beginning of the treatment with HAL, could simply be explained by the fact that subjects did not obtain reinforcements during the first days. The relationship between dystony and reinforcement consumption agrees with Sovner and DiMascio (54) according to which the involvement of the pharyngeal musculature in dystony could produce dysphagia or respiratory distress.

CLZ and HAL also produced a significant increase in tremor on the first day of treatment. For HAL, tremor was always associated with catalepsy and concerned the whole body. For CLZ, tremor was never associated with catalepsy and concerned only the paws. Sovner and DiMascio (54) noted that akinesia, tremor, and rigidity individually or in combination were the most frequent manifestations of drug-induced parkinsonism.

Sialorrhea had already been mentioned as a potent side effect of CLZ both in animals (23,40) and in man (21,38), important to detect at the preclinical level because its clinical incidence could be 23% (6,32,38,49). It was interesting to note that while most of the side effects of CLZ were submitted to tolerance hypersalivation remained present in three of the five dogs during the whole chronic period. As it was difficult to account for such a symptom with a potent anticholinergic drug, sialorrhea could represent a particular extrapyramidal side effect of CLZ. Like dystony, this effect could be linked to pseudo-parkinsonism or a deglutition difficulty (27,54), although it was contested by Copp et al. (25), showing that amitriptyline could reduce drooling substantially. "Whether or not the benefit from amitriptyline was related to its antimuscarinic effect remains to be seen" (25).

HAL induced a higher frequency of stereotypy/hyperkinesia than CLZ. Such a significant difference could explain a part of the discrimination between HAL and CLZ at the operant performance level. HAL increased the percentage of response duration before any stimulus (1-3 s) but these responses were rarely followed by a jump on the food dispenser. Moreover, the frequency of hyperkinesia and stereotypy was also increased (not significantly). According to Adler et al. (1), the syndrome of akathisia was composed of both subjective feelings like sense of inner restlessness, sensation of needing to move, and anxiety, but also of objective motor signs. In the moderate and severe forms of this side effect (tasikinesia), affected patients were only comfortable when in motion (54). Although there remained a nonmeasurable subjective dimension in akathisia, the hypothesis could be suggested that to avoid catalepsy the dog tended to move excessively. When

TABLE 2

INVOLUNTARY MOTOR EFFECTS OF CLOZAPINE AND HALOPERIDOL DURING CHRONIC ADMINISTRATION AND DURING WITHDRAWAL

	Clozapine					Haloperidol				
	Ataxia	Catalepsy	Akinesia	Tremor	Dystony	Ataxia	Catalepsy	Akinesia	Tremor	Dystony
Baseline	0.04(0.09)	0.00(0.00)	0.00(0.00)	0.00(0.00)	0.00(0.00)	0.06(0.13)	0.00(0.00)	0.00(0.00)	0.00(0.00)	0.00(0.00)
Administration										
Day 1	0.58(0.34)*	0.00(0.00)	1.38(1.04)	0.81(0.88)*	0.70(0.23)*	0.33(0.27)*	0.20(0.12)*	0.31(0.24)	0.29(0.51)*	0.20(0.40)
2	0.08(0.05)	0.00(0.00)	0.50(0.50)	0.07(0.07)	0.59(0.19)*	0.21(0.17)	0.12(0.17)*	0.31(0.33)	0.09(0.21)	0.00(0.00)
3	0.00(0.00)	0.00(0.00)	0.35(0.49)	0.03(0.07)	0.60(0.32)*	0.11(0.11)	0.01(0.03)	0.51(0.92)	0.65(1.46)	0.20(0.40)
5	0.10(0.13)	0.00(0.00)	0.35(0.33)	0.00(0.00)	0.35(0.27)*	0.11(0.07)	0.00(0.00)	0.30(0.52)	0.00(0.00)	0.24(0.34)
7	0.20(0.35)	0.00(0.00)	0.32(0.25)	0.00(0.00)	0.50(0.40)*	0.14(0.05)	0.00(0.00)	0.38(0.72)	0.01(0.03)	0.50(0.44)*
9	0.29(0.41)	0.00(0.00)	0.22(0.25)	0.00(0.00)	0.55(0.26)*	0.04(0.09)	0.00(0.00)	0.48(0.56)	0.24(0.54)	0.53(0.50)*
11	0.09(0.08)	0.00(0.00)	0.21(0.22)	0.00(0.00)	0.72(0.31)*	0.11(0.17)	0.00(0.00)	0.63(0.45)	0.00(0.00)	0.42(0.48)*
12	0.03(0.06)	0.00(0.00)	0.05(0.07)	0.00(0.00)	0.38(0.34)*	0.31(0.23)	0.01(0.03)	0.58(0.76)	0.01(0.03)	0.52(0.48)*
13	0.08(0.13)	0.00(0.00)	0.29(0.22)	0.00(0.00)	0.53(0.45)*	0.25(0.17)	0.00(0.00)	0.83(1.01)	0.00(0.00)	0.48(0.50)*
Withdrawal										
Day 1	0.08(0.11)	0.00(0.00)	0.13(0.30)	0.00(0.00)	0.23(0.10)†	0.06(0.09)	0.00(0.00)	0.50(0.52)	0.00(0.00)	0.43(0.52)
2	0.03(0.07)	0.00(0.00)	0.03(0.07)†	0.00(0.00)	0.10(0.10)†	0.30(0.30)	0.00(0.00)	0.51(0.59)	0.00(0.00)	0.66(0.36)
3	0.08(0.12)	0.00(0.00)	0.06(0.13)†	0.00(0.00)	0.25(0.15)†	0.03(0.04)	0.00(0.00)	0.31(0.37)	0.00(0.00)	0.31(0.31)
4	0.17(0.16)	0.00(0.00)	0.00(0.00)†	0.00(0.00)	0.08(0.07)†	0.21(0.20)	0.00(0.00)	0.40(0.50)	0.00(0.00)	0.26(0.16)

Results are given as frequency (number/min) means (SD).
*$p < 0.05$, Dunnett's t-tests comparison between days and baseline.
†$p < 0.05$, Dunnett's t-tests comparison between withdrawal and day 13.

TABLE 3

VOLUNTARY MOTOR EFFECTS AND SALIVATION DURING CHRONIC ADMINISTRATION OF CLOZAPINE AND DURING WITHDRAWAL

	Clozapine				Haloperidol			
	Exploration	Vocalization	Hyperkinesia/ Stereotypy	Hypersalivation	Exploration	Vocalization	Hyperkinesia/ Stereotypy	Hypersalivation
Baseline	0.90(0.35)	0.26(0.22)	0.03(0.04)	0	0.82(0.29)	0.28(0.27)	0.02(0.02)	0
Administration								
Day 1	1.31(0.36)	0.00(0.00)*	0.10(0.11)	4†	0.61(0.11)	0.00(0.00)*	0.36(0.31)	0
2	1.40(0.24)*	0.00(0.00)*	0.11(0.24)	4†	1.11(0.43)	0.00(0.00)*	0.38(0.21)	0
3	1.16(0.22)	0.00(0.00)*	0.09(0.14)	3†	1.01(0.41)	0.00(0.00)*	0.31(0.17)	0
5	1.30(0.23)	0.00(0.00)*	0.04(0.09)	3†	0.99(0.32)	0.00(0.00)*	0.22(0.21)	0
7	1.04(0.42)	0.00(0.00)*	0.07(0.16)	3†	1.29(0.27)	0.00(0.00)*	0.29(0.27)	0
9	1.04(0.17)	0.00(0.00)*	0.17(0.26)	3†	0.92(0.26)	0.00(0.00)*	0.18(0.22)	0
11	1.15(0.41)	0.00(0.00)*	0.18(0.33)	3†	0.86(0.21)	0.00(0.00)*	0.19(0.14)	0
12	1.09(0.15)	0.00(0.00)*	0.10(0.23)	3†	0.95(0.22)	0.00(0.00)*	0.20(0.14)	0
13	1.01(0.43)	0.00(0.00)*	0.12(0.27)	3†	0.98(0.36)	0.01(0.03)*	0.18(0.30)	0
Withdrawal								
Day 1	1.00(0.13)	0.00(0.00)	0.02(0.05)	2	0.81(0.21)	0.01(0.03)	0.10(0.12)	0
2	1.17(0.44)	0.34(0.75)	0.10(0.15)	0‡	0.88(0.32)	0.04(0.09)	0.10(0.14)	0
3	0.99(0.38)	0.10(0.23)	0.21(0.34)	0‡	0.69(0.12)	0.00(0.00)	0.15(0.17)	0
4	1.30(0.65)	0.24(0.53)	0.22(0.33)	0‡	1.04(0.27)	0.03(0.06)	0.11(0.21)	0

Results are given as frequency (number/min) means (SD) except for salivation, for which the number of subjects is calculated.
*$p < 0.05$, Dunnett's t-tests.
†$p < 0.01$, Cochran's Q-test comparison between days and baseline.
‡$p < 0.05$, Cochran's Q-test comparison between withdrawal and day 13.

catalepsy was submitted to tolerance (after the third day), a slight decrease was also measured in stereotypy/hyperkinesia.

The situation was different for CLZ. It increased the percentage of response duration between 3 and 6 s, the moment at which the negative stimulus was produced. Our previous works (11,14,46) demonstrated that this moment was in particular important and required a strong inhibition of behavior. It could thus be said that CLZ led to a disinhibition of behavior in the sense that it produced the reappearance of a suppressed conditioned response due to the presentation of an external stimulus (9). From this point of view, CLZ presented similarity with anxiolytics, which also produced disinhibition in this schedule (10). Different comparisons of CLZ and benzodiazepines had shown that these drugs could have qualitatively similar effects under several test conditions (FI + food, FI + shock termination, DRL, openfield) (12,17,18,55).

Moreover, on one hand CLZ induced an increase in incomplete responses but only during the 6 first days and on the other hand response rate and barking remained at an abnormally low level. It could be suggested that the simultaneous presence of tranquilizing and disinhibitory effects could be at the basis of the therapeutic potential of CLZ both in cases of negative or positive schizophrenic symptomatology (22,38, 39,43,53). According to Weinberger (56), the cortical dopaminergic defect led to negative symptoms, while the enhanced subcortical dopaminergic tone would contribute to positive symptoms. The ability of CLZ to reduce both positive and negative symptoms might be associated with its ability to enhance prefrontal cortex dopaminergic function while decreasing dopaminergic transmission in subcortical sites (44). It had also been suggested that CLZ might act via its ability to block serotonin [5-hydroxytryptamine$_2$ (5-HT$_2$)] receptors in the frontal cortex relative to its dopamine (D_2) receptor binding (45). The frontal cortex is rich in 5-HT$_2$ receptors and CLZ had been reported to bind extensively to the human frontal cortex (41).

In conclusion, this study using a complex operant schedule in a superior species allowed to differentiate the classic neuroleptic HAL from the atypical neuroleptic CLZ. The replication of the same experiments with a larger range of doses should allow to precise some correlations with the clinical effects observed with HAL and CLZ. Nevertheless, these results also allowed to clearly dissociate motor from motivational and operant effects. This could be relevant at the clinical level to better understand drug response of schizophrenic patients to CLZ or HAL as a function of their symptomatology.

ACKNOWLEDGEMENTS

The authors are grateful to Sandoz Ltd. for its generous gift of clozapine, to J. P. Peters and V. Mineur for computer processing of their data, and to M. Fourneau for statistical assistance.

REFERENCES

1. Adler, L. A.; Angrist, B.; Reiter, S.; Rotrosen, J. Neuroleptic-induced akathisia: A review. Psychopharmacology (Berl.) 97:1-11; 1989.
2. Allain, H.; Merot, J. L.; Decombe, R.; Bentue-Ferrer, D. Le sevrage brutal des médicaments à visée neuroleptique. Concours Méd. 113:119-123; 1991.
3. Asper, H.; Baggiolini, M.; Bürki, H. R.; Lauener, H.; Ruch, W.; Stille, G. Tolerance phenomena with neuroleptics. Catalepsy, apomorphine stereotypies and striatal dopamine metabolism in the rat after single and repeated administration of loxapine and haloperidol. Eur. J. Pharmacol. 22:287-294; 1973.
4. Bailey, P.; Barnes, P.; Rosengren-Keith, B.; Loew, D. M.; Moeglen, J. M.; Wormser, P. Leponex/Clozaril products claims and evidence. Ware, UK: B. Ward; 1990:1-72.
5. Baldessarini, R. J. Drugs and the treatment of psychiatric disorders. In: Gilman, A. G.; Goodman, L. S.; Rall, T. W.; Murad, F., eds. Goodman and Gilman's the pharmacological basis of therapeutics. 8th ed. New York: Pergamon Press; 1990:383-435.
6. Baldessarini, R. J.; Frankenburg, F. R. Clozapine. A novel antipsychotic agent. N. Engl. J. Med. 324:746-754; 1991.
7. Beninger, R. J.; Cheng, M.; Hahn, B. L. Effects of extinction, pimozide, SCH 23390 and metoclopramide on food-rewarded operant responding of rats. Psychopharmacology (Berl.) 92:343-349; 1987.
8. Biazzi, A.; Fregnan, G. B. Behavioral anti-dopaminergic and prohypnotic effects of neuroleptics during and after prolonged treatment. Adv. Biochem. Psychopharmacol. 24:351-357; 1980.
9. Brimer, C. J. Disinhibition of an operant response. In: Boakes, R. A.; Halliday, M. S., eds. Inhibition and learning. New York: Academic Press; 1972:205-227.
10. Bruhwyler, J.; Chleide, E. Comparative study of the behavioral, neurophysiological, and motor effects of psychotropic drugs in the dog. Biol. Psychiatry 27:1264-1278; 1990.
11. Bruhwyler, J.; Chleide, E.; Gueur, H.; Mercier, M. Transformation of a waiting schedule into a temporal regulation schedule (DRRD) by addition of external stimuli in the dog. Behav. Proc. 17:117-129; 1988.
12. Bruhwyler, J.; Chleide, E.; Liégeois, J. F.; Delarge, J.; Mercier, M. Anxiolytic potential of sulpiride, clozapine and derivatives in the open-field test. Pharmacol. Biochem. Behav. 36:57-61; 1990.
13. Bruhwyler, J.; Chleide, E.; Mercier, M. Clozapine: An atypical neuroleptic. Neurosci. Biobehav. Rev. 14:357-363; 1990.
14. Bruhwyler, J.; Chleide, E.; Mercier, M. Effects of low doses of neuroleptics on temporal regulation in a differential reinforcement of response duration (DRRD) schedule in the dog. Pharmacol. Biochem. Behav. 37:607-611; 1990.
15. Bruhwyler, J.; Chleide, E.; Mercier, M. Mise en évidence des effets des psychotropes sur la sphère noétique à partir d'un programme de conditionnement complexe chez le chien. Psychotropes 6:77-83; 1990.
16. Campbell, A.; Baldessarini, R. J. Tolerance to behavioral effects of haloperidol. Life Sci. 29:1341-1346; 1981.
17. Canon, J. G. A comparison of clozapine, chlorpromazine and thioridazine upon DRL performances in the squirrel monkey. Psychopharmacology (Berl.) 64:55-60; 1979.
18. Canon, J. G.; Lippa, A. S. Use of DRL in differentiating anxiolytic and neuroleptic properties of CNS drugs. Pharmacol. Biochem. Behav. 6:591-593; 1977.
19. Cheng, Y. F.; Lundberg, T.; Bondesson, U.; Lindström, L.; Gabrielsson, J. Clinical pharmacokinetics of clozapine in chronic schizophrenic patients. Eur. J. Clin. Pharmacol. 34:445-449; 1988.
20. Choc, M. G.; Hsuan, F.; Honigfeld, G.; Robinson, W. T.; Ereshefsky, L.; Crismon, M. L.; Saklad, S. R. Hirschowitz, J.; Wagner, R. Single versus multiple-dose pharmacokinetics of clozapine in psychiatric patients. Pharmacol. Res. 7:347-351; 1990.
21. Chouinard, G.; Annable, L. Clozapine in the treatment of newly admitted schizophrenic patients. J. Clin. Pharmacol. 16:289-297; 1976.
22. Claghorn, J.; Honigfeld, G.; Abuzzahab, F. S.; Wang, R.; Steinbook, R.; Tuason, V.; Klerman, G. The risks and benefits of clozapine versus chlorpromazine. J. Clin. Psychopharmacol. 7: 377-384; 1987.
23. Cohen, M. Effects of orally administered psychotropic drugs on dog conditioned avoidance responses. Arch. Int. Pharmacodyn. 253:11-21; 1981.
24. Cohen, B. M.; Babb, S.; Campbell, A.; Baldessarini, R. J. Persis-

tence of haloperidol in the brain. Arch. Gen. Psychiatry 45:879–880; 1988.
25. Copp, P. J.; Lament, R.; Tennent, T. G. Amitriptyline in clozapine-induced sialorrhoea. Br. J. Psychiatry 159:166; 1991.
26. Costall, B.; Marsden, C. D.; Naylor, R. J.; Pycock, C. J. Stereotyped behaviour patterns and hyperactivity induced by amphetamine and apomorphine after discrete 6-hydroxydopamine lesions of extrapyramidal and mesolimbic nuclei. Brain Res. 123: 89–111; 1977.
27. Costentin, J.; Petit, M.; Dollfus, S. Les neuroleptiques, collection les grands médicaments. Paris: Ed Ellipses Marketing; 1987: 243–268.
28. Deutch, A. Y.; Moghaddam, B.; Innis, R. B.; Krystal, J. H.; Aghajanian, G. K.; Bunney, B. S.; Charney, D. S. Mechanisms of action of atypical antipsychotic drugs. Implications for novel therapeutic strategies for schizophrenia. Schizophr. Res. 4:121–156; 1991.
29. Ezrin-Waters, C.; Seeman, P. Tolerance to haloperidol catalepsy. Eur. J. Pharmacol. 41:321–327; 1987.
30. Faustman, W. O.; Fowler, S. C. An examination of methodological refinements, clozapine and fluphenazine in the anhedonia paradigm. Pharmacol. Biochem. Behav. 17:987–993; 1982.
31. Gerlach, J.; Koppelhus, P.; Helweg, E.; Monrad, A. Clozapine and haloperidol in a single-blind cross-over trial: Therapeutic and biochemical aspects in the treatment of schizophrenia. Acta Psychiatr. Scand. 50:410–424; 1974.
32. Gudelsky, G. A.; Nash, J. F.; Berry, S. A.; Meltzer, H. Y. Basic biology of clozapine: Electrophysiological and neuroendocrinological studies. Psychopharmacology (Berl.) 99:S13–S17; 1989.
33. Guirguis, E.; Voineskos, G.; Gray, J.; Schlieman, E. Clozapine (Leponex) vs chlorpromazine (Largactil) in acute schizophrenia (a double-blind controlled study). Curr. Ther. Res. 21:707–719; 1977.
34. Heath, G. F.; Rech, R. H. Chronic haloperidol treatment: Effects on operant responding and inter-response time. Prog. Neuropsychopharmacol. Biol. Psychiatry 9:697–701; 1985.
35. Hippius, H. The history of clozapine. Psychopharmacology (Berl.) 99:S3–S5; 1989.
36. Hoffman, D. C.; Beninger, R. J. Preferential stimulation of D1 or D2 receptors disrupts food-rewarded operant responding in rats. Pharmacol. Biochem. Behav. 34:923–925; 1989.
37. Hunziker, F.; Fischer, E.; Schmutz, J. 11-Amino-5*H*-dibenzo(*b*,*e*)-1,4-diazepine-10. Mitteilung über siebengliedrige heterocyclen. Helv. Chim. Acta 50:1588–1599; 1967.
38. Kane, J.; Honigfeld, G.; Singer, J.; Meltzer, H. Clozapine for the treatment-resistant schizophrenic. Arch. Gen. Psychiatry 45: 789–796; 1988.
39. Lieberman, J. A.; Kane, J.; Johns, C.; Vital-Herne, J. Clozapine: Clinical evidence of novel effects. Clin. Neuropharmacol. 9: 140–141; 1986.
40. Lindt, S.; Lauener, H.; Eichenberger, E. The toxicology of 8-chloro-11-(4-methyl-1-piperazinyl)-5*H*-dibenzo(*b*,*e*)-(1,4)diazepine (clozapine). Il Farmaco 26:585–602; 1971.
41. Lundberg, T.; Lindstrom, L. H.; Hartvig, P. Striatal and frontal cortex binding of ^{11}C-labelled clozapine visualized by positron emission tomography (PET) in drug-free schizophrenics and healthy volunteers. Psychopharmacology (Berl.) 99:8–12; 1989.
42. Lynch, M. R.; Carey, R. J. Chronic low-dose haloperidol effects on self-stimulation rate-intensity functions. Psychopharmacology (Berl.) 102:122–129; 1990.
43. Marder, S. R.; Van Putten, T. Who should receive clozapine. Arch. Gen. Psychiatry 45:865–867; 1988.
44. Meltzer, H. Y. Clinical studies on the mechanism of action of clozapine: The dopamine–serotonin hypothesis of schizophrenia. Psychopharmacology (Berl.) 99:S18–S27; 1989.
45. Meltzer, H. Y.; Bastani, B.; Young Kwon, K.; Ramirez, L. F.; Burnett, S.; Sharpe, J. A prospective study of clozapine in treatment-resistant schizophrenic patients. I: Preliminary report. Psychopharmacology (Berl.) 99:S68–S72; 1989.
46. Mercier, M.; Chleide, E.; Bruhwyler, J. Analysis of the mediatory role of collateral behaviour into a differential reinforcement of response duration (DRRD) schedule with external cues in the dog. Psychol. Belgica 28:123–137; 1988.
47. Niemegeers, C. J. E.; Janssen, P. A. J. A systematic review of the pharmacological activities of dopamine antagonists. Life Sci. 24:2201–2216; 1979.
48. Ohman, R.; Larsson, M.; Nilsson, I. M.; Engel, J.; Carlsson, A. Neurometabolic and behavioral effects of haloperidol in relation to drug levels in serum and brain. Naunyn Schmiedeberg's Arch. Pharmacol. 299:105; 1977.
49. Povlsen, U. J.; Noring, U.; Fog, R.; Gerlach, J. Tolerability and therapeutic effect of clozapine. Acta Psychiatr. Scand. 71:176–185; 1985.
50. Robertson, A.; MacDonald, C. The effects of some atypical neuroleptics on apomorphine-induced behaviors as a measure of their relative potencies in blocking presynaptic versus postsynaptic dopamine receptors. Pharmacol. Biochem. Behav. 24:1639–1643; 1986.
51. Salamone, J. D. Different effects of haloperidol and extinction on instrumental behavior. Psychopharmacology (Berl.) 88:18–23; 1986.
52. Schmutz, J.; Hunziker, F.; Stille, G.; Lauener, H. Constitution chimique et action pharmacologique d'un nouveau groupe de neuroleptiques tricycliques. Bull. Chim. Thér. 6:424–429; 1967.
53. Shopsin, B.; Klein, H.; Aaronson, M.; Collora, M. Clozapine, chlorpromazine and placebo in newly-hospitalized, acute schizophrenic patients: A controlled, double-blind comparison. Arch. Gen. Psychiatry 36:657–664; 1979.
54. Sovner, R.; DiMascio, A. Extrapyramidal syndromes and other neurological side effects of psychotropic drugs. In: Lipton, M. A.; DiMascio, A.; Killam, K. F., eds. Psychopharmacology: A generation of progress. New York: Raven Press; 1978:1021–1032.
55. Spealman, R. D.; Kelleher, R. T.; Goldberg, S. R.; DeWeese, J.; Goldberg, D. M. Behavioural effects of clozapine: Comparison with thioridazine, chlorpromazine, haloperidol and chlordiazepoxide in squirrel monkeys. J. Pharmacol. Exp. Ther. 224:127–134; 1983.
56. Weinberger, D. R. Implications of normal brain development for the pathogenesis of schizophrenia. Arch. Gen. Psychiatry 44:660–669; 1987.
57. Wilk, S.; Watson, E.; Stanley, M. Differential sensitivity of two dopaminergic structures in rat brain to haloperidol and to clozapine. J. Pharmacol. Exp. Ther. 195:265–270; 1975.
58. Winer, B. J. Statistical principles in experimental design. London: McGraw-Hill Kogakusha, Ltd.; 1971:303–305.
59. Wise, R. A. Neuroleptics and operant behavior: The anhedonia hypothesis. Behav. Brain Sci. 5:39–87; 1982.