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Chronic Haloperidol Effects on Oral Movements and Radial-Arm Maze Performance in Rats

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LEVIN, E. D., D. M. GALEN AND G. D. ELLISON. *Chronic haloperidol effects on oral movements and radial-arm maze performance in rats.* PHARMACOL BIOCHEM BEHAV **26**(1) 1–6, 1987.—Rats were examined for the development of adverse motor and cognitive effects during and after 24 weeks of chronic haloperidol (HAL) administration using an 8-arm maze and a computerized apparatus for measuring spontaneous oral movements. In the maze, HAL caused a significant decline in choice accuracy only during the first week of administration, whereas it caused a significant decline in locomotor speed throughout drug administration. There were no effects of HAL on maze behavior after withdrawal. Haloperidol reduced the number of mouth movements during drug administration, but after withdrawal there was a significant increase. This replicated a previous finding from our lab. The oral movements which did occur in the HAL-treated rats were slower than normal. The timing of the HAL-induced cognitive dysfunction was similar to the Parkinson-like disorder shown by patients given chronic neuroleptics, whereas the timing of the increase in oral movements after the withdrawal of HAL was more related to the appearance of tardive dyskinesia. There was evidence in both tests of a persisting sedation during chronic neuroleptic administration.

Haloperidol Oral movements Radial-arm maze Rats Memory Tardive dyskinesia

SINCE their introduction thirty years ago, neuroleptic drugs have proven to be quite useful for suppressing psychotic symptoms in schizophrenics. However, they have also been found to cause a number of adverse side effects. The most obvious and widely studied of these, tardive dyskinesia and a Parkinson-like disorder, affect motor systems, but there are indications that adverse cognitive effects may occur as well [7, 10, 13–15, 25]. Cognitive effects may not be as obvious as motor effects because they can be masked either by concurrent motor impairment or the pre-existing cognitive dysfunction often seen in schizophrenics [1, 2, 18, 23]. The present study examined both the motor and cognitive effects of long-term neuroleptic administration in rats.

Cognitive dysfunction has been found to be related to tardive dyskinesia by some investigators [11, 30, 31, 33, 34], in that it is especially apparent in patients who also show signs of tardive dyskinesia. On the other hand, neuroleptic-induced cognitive dysfunction may be more akin to Parkinsonism. As in Parkinsonism [17,19], the neuroleptic-induced disorder [26] seems to involve both bradykinesia (slowness of movement) and bradyphrenia (slowness of thought). The combination of these effects, usually labeled sedation, is probably the most common adverse side effect of neuroleptic drug administration [26,32]. Since the Parkinson-like disorder is usually seen soon after the onset of neuroleptic administration and disappears within weeks and tardive dyskinesia does not become manifest for months or years, the timing of any cognitive effects seen in the present study will provide an indication as to their relationship to the motor effects.

The primary neuropharmacological effect of haloperidol is to block dopamine (DA) receptors. In animal models consequences of decreasing DA activity include sensory inattention [20,21], decreased exploratory behavior [4–6, 12] and impaired memory function [3, 22, 29]. Memory function was examined in the present study because unlike sensory attention it is impaired by relatively mild disruptions of the DA system and unlike exploration it does not loose its validity with repeated testing.

The memory test used in the present study was the radial-arm maze. Beatty and Rush [3] found that acute doses of haloperidol impaired spatial working memory in the radial-arm maze as long as the rats were under the influence of the drug during the recall portion of the test. The sedation induced by HAL did not appear to in itself be responsible for the deficits in choice accuracy. Doses of barbiturates which caused a similar degree of sedation have not been found to impair choice accuracy in the radial-arm maze [3,8].

To complement our test of cognitive function, we measured spontaneous oral movements using a computerized video analysis system we have developed [9]. This sytem has the advantages of being objective and having the ability to quantify the characteristics of each movement. In a previous experiment using the same type of HAL administration [9], we found with this scoring system that, except for very tiny movements, oral activity was not increased during chronic HAL administration. However, after withdrawal there was a significant increase in all sizes of movements.

The goal of the present study was to define the relation-

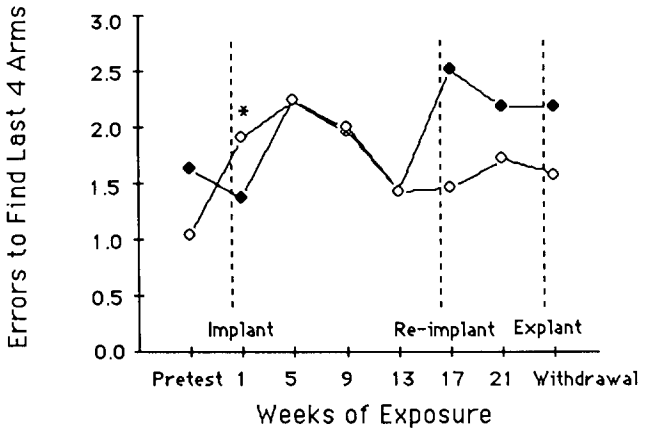

FIG. 1. The number of errors to enter the last four arms. ◆ Control, ◇ haloperidol. *$p<0.05$.

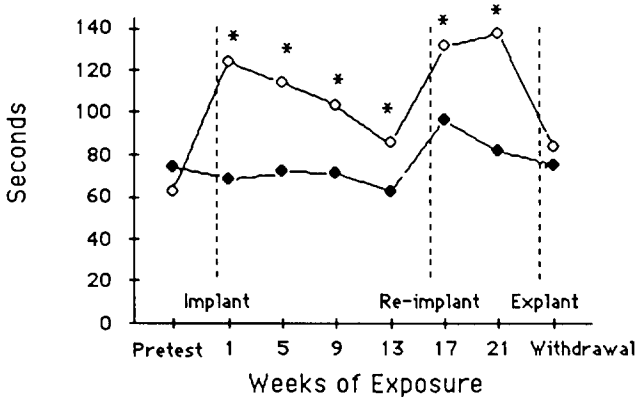

FIG. 2. Latency in seconds to enter the last four arms in the radial arm maze. ◆ Control, ◇ haloperidol. *$p<0.05$.

ship of cognitive and motor effects of long-term HAL administration in terms of severity, time of onset and persistence. This will help in the characterization of possible adverse cognitive effects as being related more to the Parkinson-like disorder or to tardive dyskinesia.

METHOD

Subjects and Exposure

Twenty adult female albino rats (Simonsen, Gilroy, CA) were used in this study. One rat which did not run in the maze was eliminated from the study and one rat died, leaving nine rats in each group. Half of the rats received subcutaneous implants of silastic envelopes developed in our lab for haloperidol (HAL) administration [9]. The implantation was performed under lidocaine local anesthesia. The rats in the HAL group were implanted with envelopes filled with 100 mg of HAL. A release rate of approximately 0.45 mg/kg/day for these envelopes was determined in another set of rats using HPLC analysis of the amount of drug remaining in the explanted envelopes after 6, 12 and 24 weeks of implantation. The average HAL release rate for a 100 mg envelope was 22.9 mg in 24 weeks, 12.3 mg for 12 weeks and 5.2 mg for 6 weeks. The controls were implanted with silastic strips containing no HAL. After 16 weeks of exposure the old silastic envelopes were removed and new ones were implanted. After 24 weeks these implants were removed and withdrawal was studied.

8-Arm Maze

The rats were tested in a black wooden radial-arm maze patterned after the one developed by Olton and his coworkers [24]. It had a central platform 35 cm across and eight arms (10×80 cm) radiating outward at equal angles. The maze was elevated 30 cm from the floor in a room with abundant extramaze visual cues. Food wells were located 2 cm from the end of each arm.

At the beginning of each session the maze was wiped off with water and 1/3 to 1/2 of a Kellogg's Froot Loop was placed in each food well. To start the session the rat was placed inside a Plexiglas ring located on the central platform. After 10 seconds the ring was lifted and the rat was allowed to enter arms (four paws passed the threshold) until either 4

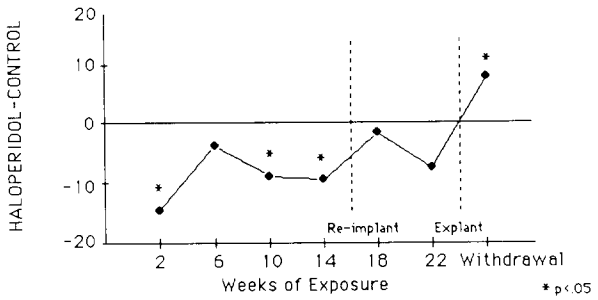

FIG. 3. Movelets/minute: Average of all amplitude categories, the haloperidol group relative to controls. *$p<0.05$.

different arms had been entered or 300 seconds had elapsed. Then the rat was lifted off of the maze and placed back in its home cage either for 0, 1, 10 or 100 minutes. After the delay, the rat was again placed into the central ring for 10 seconds and then allowed to retrieve the last four rewards. The second half of the session lasted until either all of the rewards had been entered or 300 seconds had elapsed. The dependent measures were the number of errors in finding the last four arms and latency in finding the last four arms.

Prior to drug administration the rats underwent 49 sessions of training. Delays were introduced two weeks before the first drug administration. The rats were tested with one delay per day with the order of delays being given in a counter-balanced design. Based on their choice performance at the longest delay the rats were sorted into matched groups and a final week of pretests were given. Starting the first week after implant and every four weeks thereafter, the rats were tested on the maze at each of the four delays. For two days before and during each week of testing on the 8-arm maze the rats were put on a 22 hour food deprivation schedule and were fed each day following testing. The rats were fed ad lib between the weeks of 8-arm testing. At the beginning of each week the rats were tested without delays to reacquaint them with the maze. After the four sessions with delays make-up sessions were conducted for the rats who did not complete one of their sessions.

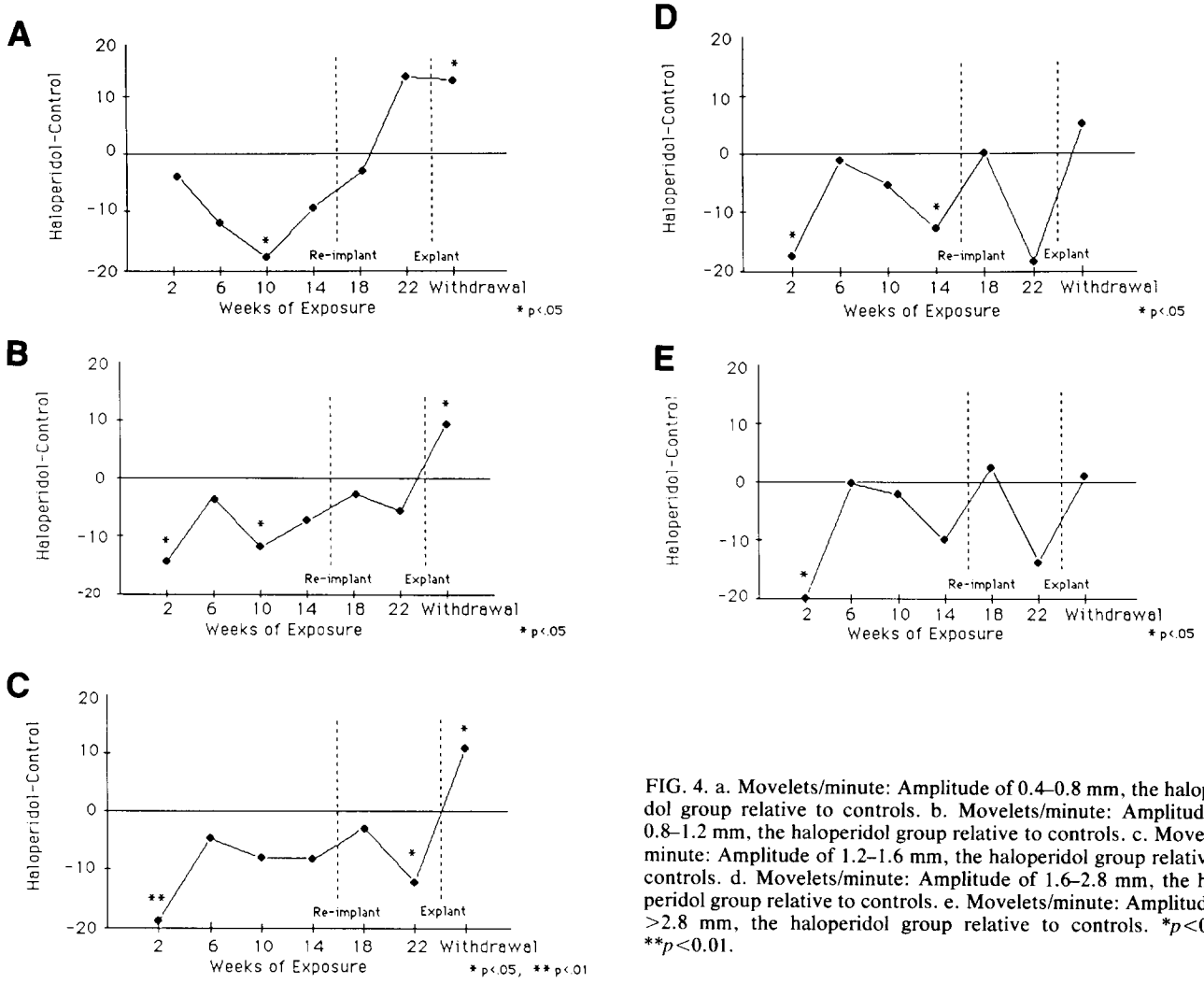

FIG. 4. a. Movelets/minute: Amplitude of 0.4–0.8 mm, the haloperidol group relative to controls. b. Movelets/minute: Amplitude of 0.8–1.2 mm, the haloperidol group relative to controls. c. Movelets/minute: Amplitude of 1.2–1.6 mm, the haloperidol group relative to controls. d. Movelets/minute: Amplitude of 1.6–2.8 mm, the haloperidol group relative to controls. e. Movelets/minute: Amplitude of >2.8 mm, the haloperidol group relative to controls. $*p<0.05$, $**p<0.01$.

Oral Movements

On the weeks after 8-arm maze testing, the rats were tested for the frequency and types of oral movements. Before testing, ultra violet-sensitive fluorescent dots ("Black-Ray Swimming-pool readmission ink" from UVP, Inc., P.O. Box 1501, San Gabriel, CA 91778) were painted on the rats' upper and lower jaws. They were put in a Plexiglas tube with their head poking out of one end toward a video camera. For a six-minute session their oral movements were recorded by a computer program which measured the distance between the upper and lower dots at a rate of 60 times per second. A closed circuit TV camera with a close-up lens was positioned 22 cm in front of the rat. This camera had a UV filter in front of the lens eliminating the background UV light so as to detect only the two fluorescent dots painted on the rat's mouth. The output from this camera was monitored on a TV screen, and the output of the camera was adjusted by potentiameter so that only the two fluorescing spots were visible. The resulting digital signal was fed to a computer with a movement detection circuit (the "MM" board from Biotronic Designs, Tarzana, CA). This circuit calculated the number of TV rasters from the bottom of the top spot to the top of the bottom spot. This distance was stored in computer memory 60 times each second at each vertical synch pulse from the TV camera. This system was designed to be maximally sensitive to mouth movements, such that horizontal movements of the head would be completely undetected and vertical movements of the entire head would be minimized relative to movements of the mouth. These data were then summarized by a Pascal program which classified the "movelets" (single openings or closings) by amplitude. Five different categories were used: 0.4–0.8 mm, 0.8–1.2 mm, 1.2–1.6 mm, 1.6–2.8 mm and >2.8 mm. Finer-grained categories were used for smaller movelets because of the relatively greater number of movelets in this range. The threshold for video noise was one raster (0.3 mm). The smallest amplitude category was just above this threshold. Artifactual movelets were practically all eliminated by only counting movelets that started within $^3/_{60}$ths of a second of a movelet in the opposite direction. The number of movelets in each amplitude category was calculated for each mintute the rat's mouth was in range of the camera for more than 20% of the session were rerun. In addition, the slopes of individual movelets (amplitude/duration) were calculated and averaged for each amplitude category.

In our previous study [9] there was simultaneous computer scoring and visual observation. It was found that

movelets larger than 1.6 mm corresponded very well to observer-scored chewing movements. The smaller-sized movelets corresponded to tremor and movements not seen by the human observer.

Statistics

The data were assessed by the analysis of variance. For the measures from the 8-arm maze test comparisons were made in terms of change from the sessions run during the week before the start of HAL administration. Significant interactions were followed up by post-hoc *t*-tests of simple effects. One-tailed tests were used with the movelets/min measure because a previous experiment from our lab [9] showed that there was a decreased number of movelets during HAL administration and an increased number of movelets after HAL withdrawal, except for the smallest amplitude category, which showed an increase in the number of movelets after six months of drug administration.

RESULTS

8-Arm Maze

Choice accuracy was significantly impaired by HAL administration only during the first week after the start of drug administration, $F(1,16)=8.43$, $p<0.025$. With all of the other test periods during and after HAL administration there were no significant effects (Fig. 1). After the replacement envelope was implanted during the 16th week, the controls appeared to decline in performance, however this effect was not significant. At the end of the study the difference between the HAL and control groups was about the same as it was during the pretest. The HAL× delay interaction was not significant, suggesting that performance at the different delays was not differentially affected by HAL.

The latency measure showed a much more robust and long-lasting HAL effect (Fig. 2). The HAL-treated rats took significantly longer than the controls at each of test weeks during drug administration. The F-ratios ranged from 13.24 to 22.33 and the *p*-values were all less than 0.005. Before implantation of the replacement envelope the latencies in the HAL group were declining although they remained significantly increased. After the replacement envelope was implanted the original magnitude of HAL-control difference in latency was restored. One week after HAL withdrawal, the latencies in the HAL declined toward control levels and were no longer significantly different.

Oral Movements

HAL caused significant decreases in the overall number of movelets/min during drug administration (weeks 2, 10 and 14) and a significant increase after withdrawal (Fig. 3). Given the significant three-way interaction of HAL × session × amplitude category, separate analyses of the simple, simple main effects were conducted for each amplitude category during each test session. As can be seen in Fig. 4a–e, in general there was a decrease in the number of movelets during drug administration and an increase after withdrawal. The decrease in the number of movelets was significant in more amplitude categories during the first run after the start of HAL administration. During the later sessions, more significant decreases were seen in the small to mid-sized movelets than in the large ones. After withdrawal, movelets of all sizes increased in number in the HAL group, but only the small to mid-sized categories showed significant increases over control levels.

Of interest is the apparent rise in the number of smallest sized movelets in the last test session during HAL administration. Although this rise did not reach a level significantly above the controls, it did resemble a similar rise seen in our previous study [9]. In that study the increase in small movelets during HAL administration also predicted a significant increase in oral movement after HAL withdrawal. However, in the present study a conclusion regarding the relative rise in very small movelets is tenuous not only because of the lack of statistical significance but also because this relative rise was due more a decrease in the number of very small movelets by the control group during this test session rather than a rise by the HAL group.

Analysis of the slopes of the movelets showed that the HAL group had significantly lower slopes, $F(1,16)=4.41$, $p<0.05$. Planned comparisons between the groups during drug administration and after withdrawal showed that during the period of drug administration there was a significant HAL-related decrease in slopes, $F(1,16)=6.07$, $p<0.025$, and that after withdrawal there was no significant HAL-related effect. The HAL × sessions interaction in the analysis of data gathered during drug administration was not significant, indicating that the same effect persisted throughout the 24 weeks of exposure.

DISCUSSION

These data show a clear separation in the time course of three different behavioral effects of chronic HAL administration. In the 8-arm maze, HAL only caused a deficit in choice behavior for the first week, but it significantly increased response latency for the entire period of drug administration. The effects on the oral movements showed different pattern, for throughout HAL administration there was a decrease in the number of movelets and in the slopes of movelets, and after withdrawal there were increased numbers of small and mid-sized movelets and a return of slopes to control levels.

The transient effect of HAL on choice behavior suggests that this cognitive deficit was more related to a short-term, neuroleptic-induced (i.e., Parkinson-like) disorder than to tardive dyskinesia. However, the persisting HAL-induced increase in response latency in the maze was unlike the transient bradykinesia seen in this Parkinson-like disorder in humans. The fact that the latency effect in the 8-arm maze persisted aids in the interpretation of the recovery from the cognitive deficit, for this long-term effect on locomotion in the maze demonstrates a continuing effect of the HAL administration. That is, the rats did not seem to develop a general tolerance to the effects of HAL.

One possible explanation for the selective recovery of the choice behavior is that there was selective tolerance by one component of the DA system. The most probable site for this tolerance would be the nigro-striatal DA system because the mesolimbocortical system does not seem to become tolerant to chronic administration of neuroleptic drugs [27,28]. Another explanation may be that the rats learned to solve the maze while in the drugged state. This dissociation of the haloperidol effects on locomotor and choice behavior in the maze suggested that the increased latency was probably not the cause of the impaired choice behavior, a result consistent with the findings of Beatty and Rush [3] and Eckerman *et al.* [8] that sedation in itself does not impair choice behavior in the radial-arm maze.

The biphasic effect of haloperidol administration and withdrawal on the number of movelets replicates our previous finding [9]. That study also found that during HAL administration, there was no increase in movelets except for possibly the very tiny ones. In both this previous study and the present one, in contrast to the findings of Gunne and Haggstom [16], there was a persisting HAL-induced reduction in large amplitude movements. The timing of the increase in smaller oral movements in both studies mirrors the timing in tardive dyskinesia.

The increase in small to mid-sized movelets but not the large movelets following HAL withdrawal indicates that the increase was not due solely to a general hyperactivity but had some specificity to certain types of oral movements. This lack of a general hyperactivity after HAL withdrawal in the oral movement test was consistent with the lack of hyperactivity in the 8-arm maze.

The HAL-induced decrease in slope during drug administration adds to the picture of persisting sedation induced by HAL. In addition to there being fewer movelets, those movelets which did occur had a slower, or sluggish waveform. After withdrawal, the slopes of the movelets returned to control levels, but did not exceed them. This finding is further evidence that the withdrawal-induced increase in oral movements was specific and was not due merely to increased levels of general activity.

This study showed effects during all phases of chronic haloperidol administration. The cognitive deficit was only apparent during the first week of administration; the locomotor sedation was evident throughout drug administration (as was the decrease in large-amplitude oral activity), and the increase in oral activity only became apparent after withdrawal from HAL. These results highlight the complex nature of chronic haloperidol effects and how the nature of the effects can change in character over time.

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REFERENCES

1. Asarnow, R. F. and D. J. MacCrimmon. Residual performance deficit in clinically remitted schizophrenics: A marker for schizophrenia? *J Abnorm Psychol* **87:** 597–608, 1978.
2. Asarnow, R. F. and D. J. MacCrimmon. Span of apprehension deficits during the postpsychotic stages of schizophrenia: A replication and extension. *Arch Gen Psychiatry* **38:** 1006–1011, 1981.
3. Beatty, W. W. and J. R. Rush. Spatial working memory: Effects of monoaminergic antagonists. *Pharmacol Biochem Behav* **18:** 7–12, 1983.
4. Buchel, L., J. Levy and O. Tanguy. The importance of antagonism and synergism in the pharmacological study of neuroleptics. *J Physiol (Paris)* **54:** 771–786, 1962.
5. Cheal, M. Differential effects of haloperidol and clozapine on attention. *Psychopharmacology (Berlin)* **84:** 268–273, 1984.
6. Cunha, J. M. and J. Masur. Evaluation of psychotropic drugs with a modified open field test. *Pharmacology* **16:** 259–267, 1978.
7. Davis, K. L. and G. S. Rosenberg. Is there a limbic system equivalent of tardive dyskinesia? *Biol Psychiatry* **14:** 699–703, 1979.
8. Eckerman, D. A., W. A. Gordon, J. D. Edwards, R. C. McPhail and M. I. Gage. Effects of scopolamine, pentobarbitol and amphetamine on radial arm maze performance in the rat. *Physiol Behav* **12:** 595–602, 1980.
9. Ellison, G. D., R. E. See, E. D. Levin and J. Kinney. Tremorous oral movements caused by chronic haloperidol intoxication in rats. *Psychopharmacology (Berlin)*, in press, 1987.
10. Erickson, W. D., A. M. Yellin, J. H. Hopwood, G. M. Realmuto and L. M. Greenberg. The effects of neuroleptics on attention in adolescent schizophrenics. *Biol Psychiatry* **19:** 745–753, 1984.
11. Famuyiwa, O. O., D. Eccleston and A. A. Donaldson. Tardive dyskinesia and dementia. *Br J Psychiatry* **135:** 500–504, 1979.
12. Fink, J. S. and G. P. Smith. Mesolimbocortical dopamine terminal fields are necessary for normal locomotion and investigatory exploration in rats. *Brain Res* **199:** 359–384, 1980.
13. Goldberg, E. Akinesia, tardive dysmentia, and frontal lobe disorder in schizophrenia. *Schizophr Bull* **11:** 255–263, 1985.
14. Goldstone, S. and W. T. Lhamon. The effects of haloperidol upon temporal information processing by patients with Tourette's syndrome. *Psychopharmacology (Berlin)* **50:** 7–10, 1976.
15. Goldstone, S., H. G. Nurnberg and W. T. Lhamon. Effects of trifluoperazine, chlorpromazine and haloperidol upon temporal information processing by schizophrenic patients. *Psychopharmacology (Berlin)* **65;** 119–124, 1979.
16. Gunne, L. M. and J. E. Haggstrom. Reduction of nigral glutamic acid decarboxylase in rats with neuroleptic-induced oral dyskinesia. *Psychopharmacology (Berlin)* **81:** 191–194, 1983.
17. Lees, A. J. and E. Smith. Cognitive deficits in the early stages of Parkinson's disease. *Brain* **106:** 257–270, 1983.
18. Lehmann, H. E. Negative effects of psychotherapeutic drug treatment. *Prog Neuropsychopharmacol* **3:** 223–229, 1979.
19. Lieberman, A., M. Dziatolowski and M. Kupersmith. Dementia in Parkinson disease. *Ann Neurol* **6:** 355–359, 1979.
20. Marshall, J. F., N. Berrios and S. Sawyer. Neostriatal dopamine and sensory inattention. *J Comp Physiol Psychol* **94:** 833–846, 1980.
21. Marshall, J. F., J. S. Richardson and P. Teitelbaum. Nigrostriatal bundle damage and the lateral hypothalamic syndrome. *J Comp Physiol Psychol* **87:** 808–830, 1974.
22. Oades, R. D. Impairments of search behaviour in rats after haloperidol treatment, hippocampal or neocortical damage suggest a mesocortical limbic role in cognition. *Biol Psychol* **12:** 77–85, 1981.
23. Oades, R. D. *Attention and Schizophrenia: Neurological Bases*. Boston: Pitman Publishing Inc, 1982.
24. Olton, D. S. and R. J. Samuelson. Rememberance of places passed: Spatial memory in rats. *J Exp Psychol: Anim Behav Proc* **2:** 97–116, 1976.
25. Platt, J. E., M. Campbell, D. M. Grega and W. H. Green. Cognitive effects of haloperidol and lithium in aggressive conduct-disorder children. *Psychopharmacol Bull* **20:** 93–97, 1984.
26. Rifkin, A., F. Quitkin and D. Klein. Akinesia: A poorly recognized drug-induced extrapyramidal disorder. *Arch Gen Psychiatry* **32:** 672–674, 1975.
27. Scatton, B., C. Garnett and L. Jolou. Acute and subacute effects of neuroleptics on dopamine synthesis and release in the rat striatum. *Naunyn Schmiedebergs Arch Pharmacol* **289:** 419–434, 1975.
28. Scatton, B., C. Garnett and L. Jolou. Dopamine metabolism in the mesolimbic and mesocortical dopaminergic systems after single or repeated administrations of neuroleptics. *Brain Res* **109:** 184–189, 1976.
29. Simon, H., S. Scatton and M. LeMoal. Dopaminergic A10 neurons are involved in cognitive functions. *Nature* **286:** 150–151, 1980.

30. Spohn, H. E., L. Coyne, R. Lacoursiere, D. Mazur and K. Hayes. Relation of neuroleptic dose and tardive dyskinesia to attention, information-processing, and psychophysiology in medicated schizophrenics. *Arch Gen Psychiatry* **42:** 849–859, 1985.
31. Struve, F. A. and A. E. Willner. Cognitive dysfunction and tardive dyskinesia. *Br J Psychiatry* **143:** 597–600, 1983.
32. Van Putten, T. and P. R. May. Akinetic depression in schizophrenia. *Arch Gen Psychiatry* **35:** 1101–1107, 1978.
33. Wegner, J. T., J. M. Kane, P. Weinhold, M. Woerner, B. Kinon and J. Lieberman. Cognitive impairment in tardive dyskinesia. *Psychiatry Res* **16:** 331–337, 1985.
34. Wolfe, M. E., J. J. Ryan and A. D. Mosnaim. Cognitive function in tardive dyskinesia. *Psychol Med* **13:** 671–674, 1983.