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Effects of Imipramine, Chlorimipramine, and Fluoxetine on Cataplexy in Dogs[1,2]

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BABCOCK, D. A., E. L. NARVER, W. C. DEMENT AND M. M. MITLER. *Effects of imipramine, chlorimipramine, and fluoxetine on cataplexy in dogs.* PHARMAC. BIOCHEM. BEHAV. 5(6) 599–602, 1976. – Four narcoleptic dogs with cataplexy were given trials with the serotonin uptake blockers imipramine and chlorimipramine (known to be effective in treating cataplexy in humans). An even more selective serotonin uptake blocker, fluoxetine, was also tested. Injections of placebo, test compound, and placebo were given respectively on 3 successive days. Anticataplectic effects were measured approximately 30 min, 3 hr, and 6 hr postinjection by recording elapsed time and number of cataplectic episodes during the dogs' attempts to eat ten pieces of a desired food presented in a standard fashion. Imipramine (1 mg/kg) and fluoxetine (1.5 and 3.0 mg/kg) significantly improved performance, while chlorimipramine (0.5–5 mg/kg) had no clear effect. Data were not totally consistent with the notion that serotonin uptake blockers improve cataplexy in dogs, since chlorimipramine was not effective in these animals.

Narcolepsy Cataplexy 5-HT uptake blockade

IN HUMANS the sleep disorder, narcolepsy [13, 14, 15, 17, 18], is an incurable central nervous system disease characterized by excessive sleepiness and one or more of the following symptoms: sleep paralysis, hypnagogic hallucinations, and cataplexy. Recently, a naturally occurring animal model of the disorder has been described in dogs [7, 10–13]. The most apparent clinical sign in such animals is cataplexy (complete or partial flaccid paralysis of postural muscles precipitated by excitement).

Part of our narcolepsy program involves conducting pharmacological trials in narcoleptic dogs with compounds known to influence symptoms in human narcoleptics. This series of trials concerns the role serotonin uptake blockade may play in the control of cataplexy.

Imipramine, an uptake blocker of serotonin and norepinephrine [1, 5, 8], as well as imipramine-like compounds such as desmethyl-imipramine and chlorimipramine (uptake blockers more specific for norepinephrine and serotonin respectively) [2,9] have been shown effective in treating cataplexy in humans [4,6]. We now report studies in four cataplectic dogs. This work involved three drugs thought to be successively more specific serotonin uptake blockers: imipramine, chlorimipramine, and fluoxetine (Lilly 110140 [16]).

METHOD

Animals

The animals used were 4 cataplectic poodles (2 males) from a present colony of 8 narcoleptic dogs at Stanford University School of Medicine.

Procedure

Behavioral testing. In order to quantify fluctuations in cataplexy, we developed the Food Elicited Cataplexy Test (FECT). In the test, ten 1 cm^3 pieces of cheese or other desired food are placed in a row and spaced 30.5 cm apart. Most normal and cataplectic dogs are easily trained to eat the bits of food one after the other while the experimenter sits quietly at the end of the row, recording both how long it takes the dog to eat all ten pieces (the Elapsed Time), and how many cataplectic attacks and partial attacks occur before the last piece of food is swallowed and the dog is again capable of voluntary movement. The experimental criteria for cataplectic attacks are as follows: complete attack – while in the process of eating or walking toward food the dog must drop to the ground with head resting on the floor; partial attack – while in the process of eating or walking toward food the dog must drop to the ground, while keeping head above the floor. Pilot FECTs with normal dogs show that no cataplectic attacks occur and that all food is eaten within 45 sec. FECTs with cataplectic dogs usually require from 2.5–30 min and involve 2–20 complete or partial attacks. Pilot data also indicated that food deprivation or recent feeding in accordance with the normal diet does not affect FECT performance. Therefore, all dogs were fed along with other members of the colony between 0800 and 0900 daily.

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Experimental design. The protocol involved 3 FECTs on each of three days: on Days 1 and 3, 1–2 cc of saline was injected intravenously (or on 4 days in one dog, subcutaneously given near the target vein, because this dog's vessels were too damaged for easy and quick venipuncture. These saline injections served as pre- and postdrug controls. Test compounds in 1–3 cc volumes were always administered intravenously through easily accessible superficial veins in the fore or hind limbs. For imipramine we used a dose level of 1.00 mg/kg. For chlorimipramine we studied doses of 0.50 mg/kg, 1.00 mg/kg, and 5.00 mg/kg. The initial dose levels of imipramine and chlorimipramine were determined from extrapolations of clinically effective levels with humans. For fluoxetine we used 1.50 mg/kg and 3.00 mg/kg dose levels. We began with dose levels of 1.50 and 3.00 mg/kg for fluoxetine since these levels were among the lowest oral dose levels safely used in blood platelet studies on dogs.

All injections were administered at 0930 ± 30 min on each day of the protocol. The first FECT of the day was run 20–30 min after the injection. Data from Guilleminault *et al.* [4] indicate that in humans such a postadministration testing interval was sufficient to show maximum anticataplectic activity. Two more FECTs were given at 3 hr intervals (1230 and 1530). At least 4 days were allowed for the dogs to recuperate between drug trials.

The data were statistically analyzed by Days × Tests repeated measures analyses of variance.

RESULTS

Our three dependent variables – elapsed time, number of complete attacks, and number of partial attacks – are necessarily intercorrelated. Inspection of our data disclosed that elapsed time was the single most descriptive dependent variable for FECT performance. We therefore present results for this measure in detail and discuss relevant aspects of the data for number of complete and partial attacks.

Imipramine (1.0 mg/kg). Table 1 presents elapsed time in terms of mean number of seconds. Performance was significantly better (shorter elapsed time) on Day 2, when the active compound was administered, than on the preceding and following placebo days, $F(2,24) = 4.14$, $p<0.05$. There was no significant Test effect or Day × Test interaction. Imipramine's anticataplectic effect was also reflected by a significant Days effect on number of complete attacks ($p<0.05$). The number of partial attacks showed no statistically significant effects, although the ordering of the means exactly paralleled those for elapsed time and number of complete attacks.

TABLE 1

IMIPRAMINE 1.0 MG/KG ELAPSED TIME OF FECT

		Tests 1	2	3	Row Means
Days	1	416.50	557.25	330.75	434.83
	2	179.50	331.00	383.75	298.08
	3	516.50	456.50	429.50	467.50
Column Means		370.83	448.50	429.50	

F (2,24) = 4.14, $p<0.05$ (days effect).
F = 1 (test effect).
F (4,24) = 1.8 (day x time interaction).

TABLE 2

CHLORIMIPRAMINE 0.50 MG/KG ELAPSED TIME OF FECT

		Tests 1	2	3	Row Means
Days	1	777.25	532.25	493.00	600.83
	2	617.25	704.75	672.00	664.67
	3	503.75	522.25	527.00	517.67
Column Means		632.75	586.42	564.00	

F (2,24) = 1.28 (days effect).
F = 1 (test effect).
F = 1 (day x test interaction).

TABLE 3

CHLORIMIPRAMINE 1.0 MG/KG ELAPSED TIME OF FECT

		Tests 1	2	3	Row Means
Days	1	342.25	436.00	372.50	383.58
	2	295.75	544.00	689.50	509.75
	3	511.50	516.50	386.00	471.33
Column Means		383.17	498.83	482.66	

F (2,24) = 1.18 (days effect).
F (2,24) = 1.11 (test effect).
F (4,24) = 1.67 (day x test interaction).

Chlorimipramine (0.50 mg/kg). Table 2 shows that there was no significant change in elapsed time under this condition, and no evidence of a trend towards a reduced elapsed time on Day 2. Neither was there any significant change in number of attacks. The number of partial attacks significantly increased on Day 2 over the placebo days ($p<0.05$).

Chlorimipramine (1.00 mg/kg). Table 3 shows no significant change in the elapsed time at this dose level. Furthermore, inspection of the means for elapsed time and number of partial attacks failed to disclose trends toward improved scores on Day 2. The only significant change occurred in the number of attacks, which decreased on Day 2 ($p<0.05$).

Chlorimipramine (5.00 mg/kg). Table 4 indicates that there were no significant changes for elapsed time. Data on number of attacks and number of partial attacks were also unremarkable.

Fluoxetine (1.5 mg/kg). Table 5 suggests that elapsed time was significantly shorter on Day 2 than on Days 1 and 3 (see Table 5). Drug effects were also apparent in fewer number of attacks on Day 2 ($p<0.05$). Partial attacks were slightly more frequent on Day 2.

Fluoxetine (3.0 mg/kg). Table 6 shows that there was a significant change in elapsed time for Day 2 (see Table 6). The number of attacks on that day was also reduced, and partial attacks were slightly more frequent on the drug day.

DISCUSSION

A major problem in evaluating drug action in these animals is variability within and between animals. One dog could walk no more than five feet without an attack lasting

TABLE 4

CHLORIMIPRAMINE 5.0 MG/KG ELAPSED TIME OF FECT

		Tests 1	2	3	Row Means
Days	1	484.75	505.50	473.25	487.83
	2	349.50	447.50	480.75	425.92
	3	582.25	553.50	584.75	573.50
Column Means		472.17	502.17	512.92	

F (2,24) = 2.31 (days effect).
F = 1 (test effect).
F = 1 (day x test interaction).

TABLE 5

FLUOXETINE 1.5 MG/KG ELAPSED TIME OF FECT

		Tests 1	2	3	Row Means
Days	1	575.00	671.75	430.25	559.00
	2	248.50	329.50	460.50	346.17
	3	598.75	572.25	558.25	586.42
Column Means		474.08	524.50	495.00	

F (2,24) = 4.55, $p<0.05$ (days effect).
F = 1 (test effect).
F (4,24) = 1.07 (day x test interaction).

TABLE 6

FLUOXETINE 3.0 MG/KG ELAPSED TIME OF FECT

		Tests 1	2	3	Row Means
Days	1	428.50	419.25	605.25	484.33
	2	330.24	235.75	232.25	266.08
	3	293.00	622.50	639.25	518.25
Column Means		350.58	425.83	492.25	

F (2,24) = 3.44, $p<0.05$ (days effect).
F = 1 (test effect).
F (4,24) = 1.13 (day x test interaction).

several minutes. Another may not have an attack for over 15 min. For example, during the fluoxetine 3.0 mg/kg trial, one male went for several hours with no attacks even on placebo days. Inspection of individual data from that run suggests that much greater statistical significance would have been found if this animal's data were excluded. Clinically, we have noted temporary changes in the frequency of cataplexy. We have no indication as to why this variability occurs. Factors to be considered include fluctuations in endocrine function levels, enzyme activity, and metabolism of putative neurotransmitters.

We have noticed also that general arousal leads to some improvement, at least initially, in cataplexy. When the dogs are excited, such as when being bathed or clipped, there are very few complete or partial attacks. Those attacks that do occur are shorter than normal and are more easily reversed by clapping hands or calling the dog's name. Such observation suggests that apparent anticataplectic effects of any compound might stem from general arousal.

On all drug days there was a decrease in one, two, or three of the dependent variables over the pre- and post-placebo days, although changes were not always statistically significant. The runs in which both the elapsed time and the number of attacks are significantly reduced suggest that cataplexy was blocked. The cases in which the elapsed time is significantly decreased while the number of attacks remains the same may indicate that the average duration of the attacks is lessened.

Partial attacks were significantly less frequent for only one trial (chlorimipramine at 0.50 mg/kg). In 4 of the 6 trials, means for partial attacks increased on the drug day. This finding suggests that at some levels of drug activity limited improvement may be characterized by fewer complete attacks and more frequent partial attacks.

In conclusion, the data are not totally consistent with the hypothesis that serotonin uptake blockade improves cataplexy in dogs. Imipramine, the least specific serotonin uptake blocker, appears to be as effective as fluoxetine, the most selective agent. For reasons not yet understood, chlorimipramine at 0.50, 1.00, and 5.00 mg/kg dose levels was ineffective in alleviating the cataplexy. Interestingly, pilot data (not reported) using only two dogs and a 0.25 mg/kg dose of chlorimipramine showed a marked reduction in the elapsed time and number of attacks. At present we can only comment that the apparent discrepancy we have found between the chlorimipramine response in dogs and humans may be due to species differences in the disorder and/or the catabolism of the compound. Future work in humans with fluoxetine may shed light on this issue.

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