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Intravenous Cocaine Precipitates Panic-Like Flight Responses and Lasting Hyperdefensiveness in Laboratory Rats

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HEBERT, M. A., D. C. BLANCHARD AND R. J. BLANCHARD. *Intravenous cocaine precipitates panic-like flight responses and lasting hyperdefensiveness in laboratory rats.* PHARMACOL BIOCHEM BEHAV **63**(3) 349–360, 1999.— There is an emerging body of clinical evidence that cocaine use in humans can result in serious fear or panic-related emotional disturbances. The present study evaluated the effects of intravenous cocaine administration upon defensive responses of rats to a threatening conspecific in a test situation, an oval runaway, permitting the display of the full range of the rat defensive repertoire. A battery of tests was employed to evaluate avoidance/escape, flight, freezing, defensive upright and defensive attack behaviors. In the first experiment male Long–Evans rats implanted with a chronic indwelling jugular catheter were placed in the runway and tested immediately after administration of either 0, 1, or 4 mg/kg of cocaine hydrochloride. The 4-mg/kg dose produced a dramatic flight response, the direction of which depended upon the direction of the approaching threat source. The same dose produced increased defensive upright postures during forced contact with the stimulus animal. Experiment 2 examined the time course for cocaine-induced hyperdefensiveness. Rats were administered either saline or 4 mg/kg cocaine intravenously and were tested following a delay of either 0, 5, 15, or 30 min following infusion. Cocaine-treated rats again displayed high levels of flight, which declined with increased time between infusion and testing. However, increased defensiveness persisted even at the 30 min delay for several defensive measures including avoidance, freezing, and defensive upright posture. Thus, following an initial period of rapid flight with intravenous cocaine administration, there was a lasting hyperdefensiveness in cocaine-treated rats. The present results suggest that cocaine may exert its panic-producing effects by acting upon neurobehavioral systems subserving defensive behavior, and that understanding of these systems is critical for understanding the neurobiology of panic disorder. © 1999 Elsevier Science Inc.

Fear Anxiety Panic Defense Drugs of abuse Intravenous administration

THERE is accumulating evidence that cocaine can produce emotional effects such as anxiety or panic in human users (9), and, that these "anxiogenic" effects are manifest during use of the drug, in addition to periods of withdrawal. Panic anxiety/panic attacks appear to be particularly closely associated with cocaine use (9,21): case reports (12,13), correlational evidence (5,30), and experiments involving cocaine abusers (23) all suggest that cocaine can, at least in some individuals, elicit or potentiate panic. Although the risk of panic appears to increase with long-term or high-magnitude cocaine use, panic attacks may also appear after occasional use of small amounts of cocaine (13).

Although a number of the studies using animals models to investigate the anxiogenic effects of cocaine have focused on cocaine withdrawal effects (7,18,24,28,33), there is considerable evidence that, in animals as in humans, cocaine administration itself can produce anxiety-like behaviors. Acute cocaine increases thigmotaxis, a defensive behavior, in mice (34), and increases the aversive response to the white area of a black–white test box (8). Both acute and chronic cocaine increase defensive withdrawal in rats (35), while acute cocaine increases anxiety-like behavior in mice (35) or, at smaller doses, for some subgroups of mice only (29), in the elevated plus maze. All the above studies have involved rodents, but cocaine has also been reported to produce hypervigilance and "panic-like" flight behaviors in monkeys (10).

There have been few attempts to examine cocaine effects in contexts designed to allow for the full expression of the de-

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fensive behavior repertoire including defensive immobility, flight/escape, and defensive threat and attack. Studies in mice using the Mouse Defense Test Battery (MDTB) showed that of the range of defensive behaviors, flight is selectively responsive to panicogenic compounds such as yohimbine (5), as well as to chronic administration of panicolytic compounds such as alprazolam (17), imipramine, and fluoxetine (16).

In contrast to laboratory mice and wild rats, which exhibit high baseline levels of escape, flight, and biting behavior, laboratory rats display these behaviors only under conditions of extreme threat, notably those involving pain (4). This difference of laboratory rats and wild rats appears to reflect generations of selective breeding, in which docile animals (i.e., those not biting, struggling, or fleeing to human contact) were used as breeders. The result is that laboratory rats show sharply reduced levels of both flight and defensive threat/attack compared to wild *R. norvegicus* (1). Thus, it was of interest to determine if cocaine administration could reinstate defensive behavior in laboratory rats under conditions that would not typically produce such responses.

In the present study, laboratory rats were administered cocaine intravenously and their defensive responses to a threatening conspecific were evaluated. A battery of tests similar to that of the MDTB was employed to examine effects of cocaine on a full range of defensive behaviors. Experiment 1 examined defensive responses under a range of doses. In the second experiment, the time course for the cocaine effects was characterized.

EXPERIMENT 1

The present experiment examined the relationship between cocaine dose and defensive behavior. All subjects were tested immediately following intravenous administration of three doses of cocaine, one dose per test day. Behavioral tests were conducted in an oval runway permitting the display of flight and other defensive behaviors (2). The first phase of the test, discriminative/avoidance, assessed avoidance and escape behavior when the animals was approached by a conspecific from a distance. This was followed by a chase test in which the subject was pursued around the runway by the conspecific. To evaluate whether flight in this test was directed as opposed to random or protean in nature, following three laps in the initial chase direction the subject was approached from the opposite direction and chased for two additional laps. The speed of flight as well as directionality was assessed in this phase of the test. Following the chase test, the runway was converted into a straight alley by partitioning the runway at one end and the stimulus conspecific was presented at fixed, but increasingly closer, distances from the subject for evaluation of defensive immobility. Finally, the stimulus animal was forced into contact with the subject to evaluate defensive threat and attack behavior. Cocaine was predicted to increase defensive behavior in a dose-dependent fashion.

Method

Animals. Subjects were male Long–Evans rats from University of Hawaii Laboratory breeding colony. They were singly housed and experimentally naive prior to this study and were 110–150 days of age at the time of testing. Food and water were available at all times except during testing. Animals were singly housed under a 12:12 LD, lights on at 0600 h.

Experimental Design. Each subject underwent behavioral testing (described below) on 3 consecutive days following administration of one of the three doses (0, 1.0, or 4.0 mg/kg, 1 ml/kg volume) of cocaine hydrochloride (RBI) suspended in sterile 0.9% physiological saline. Subjects received a different dose on each test day, and the order in which doses were administered was completely counterbalanced.

Apparatus. Behavioral tests were conducted in an oval runway, 0.40 m wide, 0.30 m high, and 4.8 m in total length, consisting of two 2-m straight segments joined by two 0.4-m curved segments and separated by a median wall ($2.0 \times 0.30 \times 0.06$ m). The apparatus was elevated to a height of 0.80 m from the floor to enable the experimenter to place and maneuver the stimulus rat while minimizing the subject's visual contact with the experimenter. All parts of the apparatus were made of black Plexiglas. The floor of the runway was marked every 20 cm to facilitate distance measurement. Activity was recorded with video cameras mounted above the apparatus.

Procedure

Surgery. A chronic, indwelling venous catheter was implanted in each subject. Animals were surgically anesthetized with a combination of sodium pentobarbital (25 mg/kg, IP) and ketamine hydrochloride (40 mg/kg, IP). A 50-cm portion of Micro-Renethane MRE-040 (Braintree Scientific) tubing (0.040 in. o.d. $\times$ .025 i.d.) containing a sterile 10% heparin/saline solution was inserted into the right jugular vein and moved to within 5 mm of the right atrium. Three loops of 3-0 surgical silk around the vein and the catheter anchored the catheter in place. A 4×2-cm patch of Velcro (4 cm long $\times$ 2.5 cm wide) was fixed to the back of each rat in the midscapular region using Vetbond (3M) and nonabsorbable 3-0 nylon suture. The catheter was then passed subcutaneously over the shoulder region, externalized through a small hole in the middle of the back, and coiled within the Velcro patch. This configuration allowed rats to move about untethered, while providing protection for the tubing. Each day following surgery, the catheter was removed from the Velcro pouch and flushed with a 10% heparin/saline solution to help maintain patency. Subjects were given 48 h to recover from surgery before behavioral testing.

Drug infusion. During each test, the rat was placed in a 60 cm-long closed compartment at one end of the alley. The catheter was uncoiled from the Velcro patch and flushed with a 0.5 ml 10% heparin/saline solution. The drug (or saline for the 0 dose) was then infused through the catheter followed by the infusion of an additional -.5 ml heparanized saline to ensure complete delivery of the drug. The entire drug infusion process from the time the rat was placed into the alley until the drug was fully delivered was less than 2 min. Immediately following drug infusion, the partitions were removed from the alley and behavioral test began.

Behavioral testing. On each testing day, subjects were administered a battery of tests in the order presented below:

1. Discriminated avoidance: Starting from the opposite end of the alley, the subject was approached by a hand-held terminally anesthetized (CO_2 inhalation) male stimulus rat at the approximate speed of 1.0 m/s. For this and the other tests the stimulus rat was held such that its head faced the subject. The experimenter stopped the approach when the subject began to flee. Avoidance distance, the distance between the subject and stimulus when the subject began to avoid and escape distance, and the distance that the subject moved, was recorded. If the subject failed to exhibit avoidance behavior, a zero distance was recorded for both measures. The stimulus rat was removed from the runway

TABLE 1
MEAN AVOIDANCE AND ESCAPE DISTANCES (CM) IN EXPERIMENT 1

		Avoidance			Escape		
		Dose (mg/kg)					
		0	1	4	0	1	4
Trial 1	Mean	5.45	6.36	0.00	0.46	2.27	0.91
	SEM	5.45	3.88	0.00	0.46	1.24	0.91
	n	1/11	3/11	0/11	1/11	3/11	1/11
Trial 2	Mean	16.4	9.09	0.00	2.73	0.91	0.00
	SEM	11.1	6.25	0.00	1.41	0.61	0.00
	n	3/11	2/11	0/11	3/11	2/11	0/11

n indicates proportion of animals exhibiting behavior.

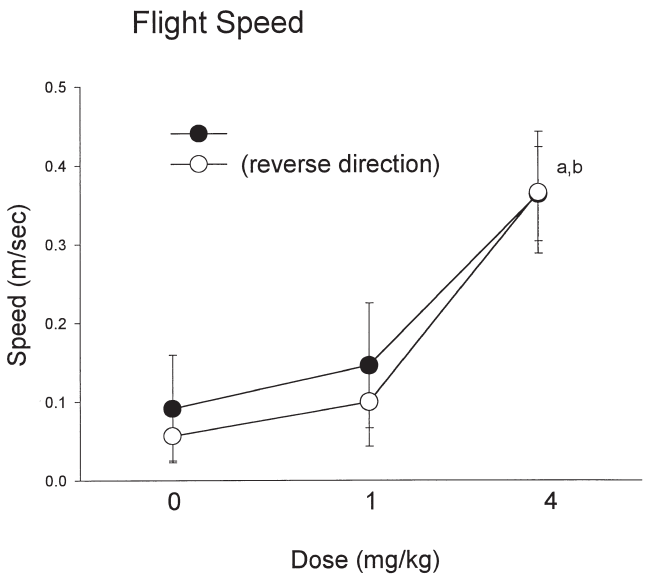

FIG. 1. Mean (and SEM) flight speed during chase test as a function of cocaine dose for both chase directions in Experiment 1. [a]Indicates significant difference from 0 mg/kg dose ($p < 0.01$). [b]Indicates significant difference from 1 mg/kg group ($p < 0.05$).

for 10 s between the two trials. Measures were taken at the time of testing. This test took approximately 2 min in total.

2. Flight test: The subject was approached by the (same) hand-held stimulus rat at approximately 2 m/s. If the subject did not flee, the experimenter continued to make approaches with the stimulus rat until flight was initiated or 2 min had elapsed from the initial approach. If the subject fled, the experimenter "chased" the subject with the stimulus rat maintaining a distance of approximately 20 cm between the head of the stimulus animal to the tail of the subject. Chase continued until the subject completed three laps or 2 min had elapsed from the first approach. The number of jump escapes, stops, orientations (rat stops and turns toward the stimulus rat), and the time required to complete the three laps were recorded. If the trial ended before three laps were completed, the total distance fled was recorded. Immediately following the first trial, subjects were given a second trial in which the subject was approached from the opposite direction (to assess whether fight was directed). The second trial was identical to the first, except the trial was terminated after two laps or 2 min. Flight time and distance was recorded at the time of testing. Other behaviors were scored from the video recordings. This test was administered approximately 2 min following infusion and lasted 2–8 min, depending upon how quickly the animal completed the laps.
3. Fixed distance approach: A door at one end of the alley was closed to prevent escape and the subject was placed in the (first) square nearest the door. The stimulus animal was then held by the experimenter for 15 s at each of three distances (1.2, 0.8, and 0.4 m) from the subject, beginning at the farthest distance, with no time between each distance. For each distance, time out of the first square (i.e., time when the subject moved toward the threat stimulus), time in contact with the stimulus, and immobility time were recorded. The closest distance between the stimulus and the subject was also measured. These measures were scored from video tapes.
4. Forced contact: The door of the runway remained in the closed position and the subject was placed in the first square. The experimenter quickly moved the nose of the stimulus rat into contact with the head (mouth) of the subject and recorded whether the subject exhibited any of the following behaviors: bite, upright defense, jump attack, vocalization, flight, and jump escape. Behaviors were scored at the time of testing. Three trials were given, each 5 s in duration with 5 s between trials. The last two procedures were run about 10-min post-injection.

Statistics. Equipment failure on one of the test days resulted in loss of data for some behaviors that were not scored at the time of testing. For measures in which no data points were missing, a one-way repeated-measures ANOVA was performed in combination with the Fisher's Least Significant Difference Test. For measures affected by the equipment fail-

TABLE 2
MEAN FREQUENCY OF BEHAVIORS DURING CHASE TEST IN EXPERIMENT 1

		Chase 1			Chase 2		
		Dose (mg/kg)					
		0	1	4	0	1	4
Reversals	Mean	0	0	0.22	0.50	0.13	0.11
	SEM	0	0	0.15	0.27	0.13	0.11
	n	0/10	0/8	2/9	3/10	1/8	1/9
Orientations	Mean	1.80	1.38	4.44	4.40	1.75	3.33
	SEM	0.85	0.94	2.44	1.06	0.82	1.67
	n	4/10	2/8	4/9	10/10	5/8	4/9
Stops	Mean	3.30	2.00	6.78	4.40	3.38	5.67
	SEM	2.17	1.59	2.71	1.39	1.28	2.22
	n	3/10	3/8	7/9	7/10	6/8	8/9
Def. uprights	Mean	0	0.50	0.56	0.20	0.13	0.11
	SEM	0	0.27	0.18	0.13	0.13	0.11
	n	0/10	3/8	5/9	2/10	1/8	1/9

n indicates proportion of animals exhibiting behavior.

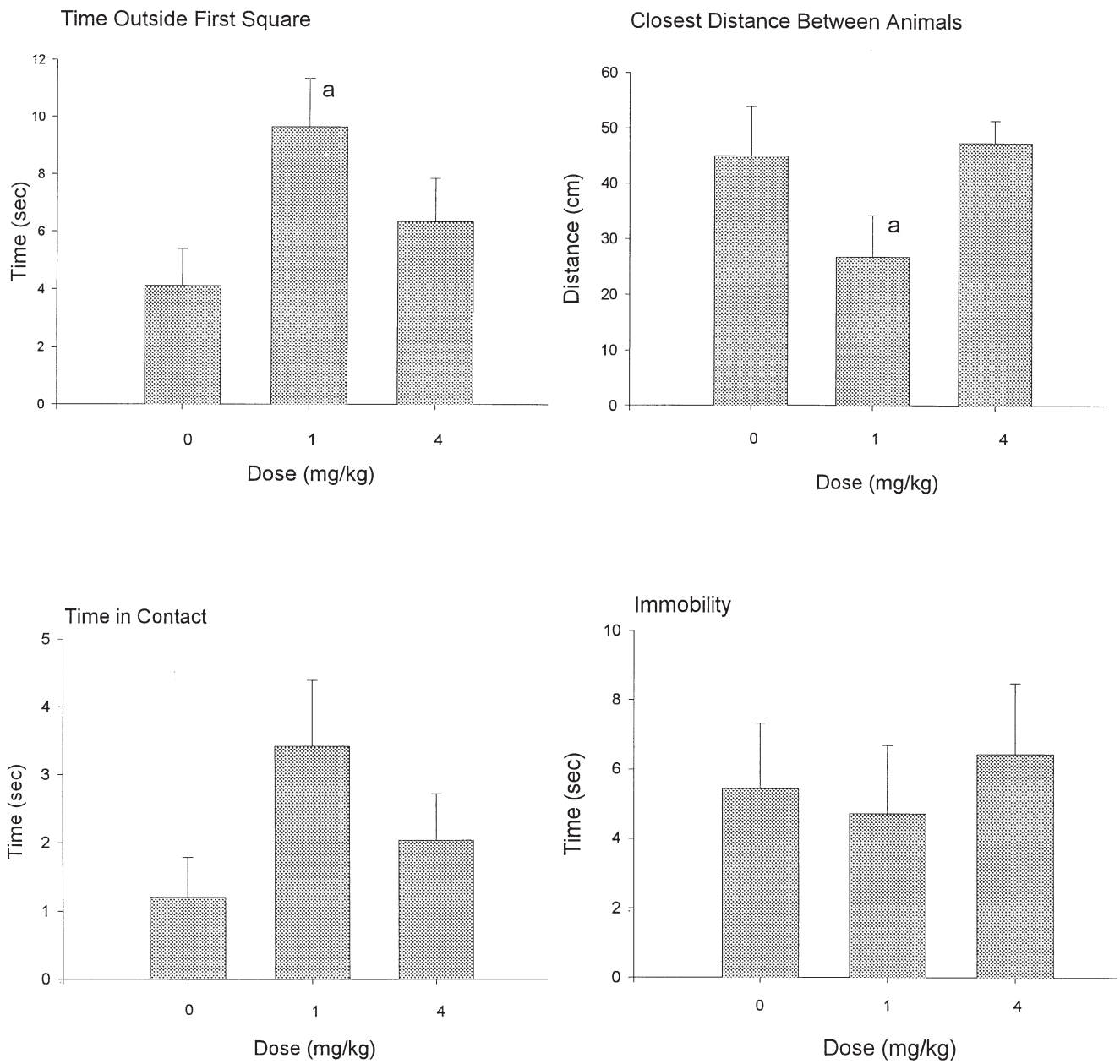

FIG. 2. Behavioral measures from Fixed Distance Approach Test in Experiment 1. Means and SEMs shown for each drug dose. [a]Indicates significant difference from 0 mg/kg dose ($p < 0.05$).

ure, dependent *t*-tests were performed to make pairwise group comparisons. For measures with low frequencies or skewed distributions, the nonparametric Wilcoxon Signed Ranks test was performed.

Results

Discriminative/avoidance. The proportion of animals exhibiting avoidance and escape on the two discriminative/avoidance trials are given in Table 1 along with the mean avoidance and escape distances on each trial. Five animals showed avoidance of the approaching stimulus or escape behavior. However, one-way ANOVAs indicted no differences among the drug doses for avoidance distance on trial 1, $F(2, 20) = 0.95$, $p = 0.40$, or trial 2, $F(2, 20) = 1.82$, $p = 0.19$, nor differences in escape distances on these trials, $F(2, 20) = 1.51$, $p = 0.24$, and $F(2, 20) = 2.69$, $p = 0.09$, respectively.

Chase test. Flights speeds during the two chase trials are shown in Fig. 1 for each of the three doses. There was a sharp increase in flight speed with the high cocaine dose for both directions of approach, indicating there was well-oriented flight response with this drug dose. On both trials, there was a main effect of drug dose on flight speed, $F(2, 20) = 6.2$, $p < 0.01$, and $F(2, 20) = 10.21$, $p < 0.001$. Fisher's post hoc analyses re-

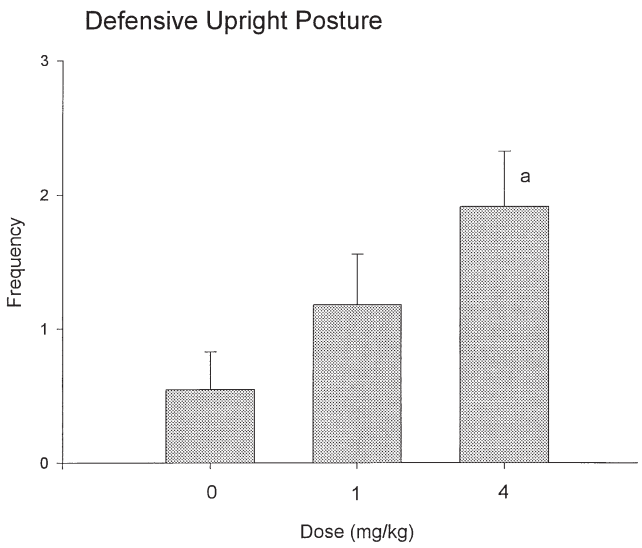

FIG. 3. Mean (and SEM) number of defensive upright postures during forced contact test in Experiment 1. [a]Indicates significant difference from 0 mg/kg dose ($p < 0.05$).

vealed a significant difference between flight speed in the saline and 4 mg/kg conditions, $p < 0.01$. The same analysis indicated a significant difference between the 1 and 4 mg/kg conditions for both trials, $p < 0.05$. The saline and 1 mg/kg conditions did not differ, $p > 0.05$. Results for other measures taken during the chase test are given in Table 2. Multiple Wilcoxon tests for matched pairs indicated no significant differences among any of the drug conditions for any of these behavioral measures, $p > 0.05$.

Fixed distance approach. Behavioral measures for this test are presented in Fig. 2. There was an overall tendency for rats at the 1 mg/kg dose to exhibit increased activity, but no apparent increase in immobility in high dose animals. Animals under the 1 mg/kg dose spent significantly more time out of the first square than animals in the saline, group, $t(7) = 3.26$, $p < 0.01$. The 4-mg/kg condition approached but just failed to reach significance compared to either the saline condition, $t(8) = 2.3$, $p = 0.055$, or the 1-mg/kg condition, $t(7) = 2.09$, $p = 0.08$, on this measure. Rats given 1 mg/kg exhibited a significant reduction in the closest distance between animals measure compared to either the saline, $t(7) = 2.79$, $p < 0.03$, or the 4-mg/kg condition, $t(7) = 3.66$, $p < 0.01$. The saline and 4-mg/kg conditions did not differ significantly on this measure, $t(8) = 0.33$, $p = 0.75$. Animals under the 1-mg/kg dose spent the most time in contact with the stimulus animal, a differences that approached but failed to reach significance in comparison with the saline condition, $t(7) = 2.37$, $p = 0.06$. The saline and 4-mg/kg conditions did not differ, $t(8) = 1.81$, $p = 0.11$, nor did the 1- and 4-mg/kg conditions, $t(7) = 1.67$, $p = 0.15$. There were no differences among the three drug conditions in immobility time, $0.57 < p < 0.86$.

Forced contact. Figure 3 shows the mean number of defensive postures exhibited by subjects under the three doses of the forced contact test. There was a dose-dependent increase in the number of upright defensive postures indicating residual hyperdefensiveness following the initial flight effect. The Wilcoxon test indicated significantly more upright postures exhibited in the 4 mg/kg than the saline condition, $t(11) = 2.37$, $p < 0.02$. Although there were more upright postures in the 1-mg/kg condition than in the saline condition, the difference was not significant, $t(11) = 1.36$, $p = 0.017$. The 1-mg/kg condition was not significantly different than the 4-mg/kg condition, $t(11) = 1.52$, $p = 0.13$. There were no occurrences of jump attack, vocalization, flight, or jump escape for any dose. No animals in the saline condition, and only one animal in the 1- and 4-mg/kg groups' exhibited biting.

Discussion

Intravenous administration of cocaine at the 4-mg/kg dose produced explosive flight behavior in laboratory rats, in tests initiated approximately 2 min following IV administration. As cocaine-treated animals typically reached the five-lap criteria in the chase test within 1–2 min, the observed effect occurred between 2–4 min from the time of infusion. The same rats showed absolutely no flight in the same situation when tested without cocaine. The flight response was directed in nature. That is, rats fled away from the approaching threat source initially, then reversed direction when the threat source was shifted such as to approach from the opposite direction. Thus, cocaine administration enhanced flight but without compromising sensorimotor defensive systems involved in spatial localization of the threat stimulus and in orientation of the appropriate directional response.

Cocaine administration did not increase avoidance behavior, evaluated immediately after drug administration. However, the high cocaine dose did produce lasting hyperdefensiveness following the initial flight response: In the forced contact test, the last test administered, there was an increase in upright defense behavior in the high dose condition. Saline and low dose cocaine animals rarely exhibited this behavior, whereas high dose cocaine animals did so quite readily.

The low cocaine dose appeared to affect some measures of defensiveness, notably risk assessment behavior. Animals given 1 mg/kg exhibited cautious investigation of the stimulus animal in the fixed distance approach test; they spent the most time out of the first square and exhibited the closest approach distance. Clearly, however, there was a dose-dependent increase in the intensity of defensive behavior.

EXPERIMENT 2

Experiment 1 indicated strong enhancing effects of intravenous cocaine on defensive behavior, particularly flight at the 4-mg/kg dose. The present experiment was conducted to determine the time course for this effect. Thus, rats were administered 4 mg/kg cocaine (or saline) intravenously and then tested at various intervals following infusion.

Method

Animals. Subjects were 16 male Long–Evans rats from University of Hawaii breeding colony. They were singly housed and experimentally naive prior to this study and were 100–140 days of age at the time of testing. Food and water were available at all times except during testing. Animals were singly housed under a 12 :12 LD cycle, lights on at 0600 h.

Experimental design. Subjects were randomly assigned to either the saline or cocaine condition ($n = 8$ per group) and tested on 4 consecutive days (one test per day) beginning 48 h following surgery. Each subject was tested following a delay of either 0, 5, 15, or 30 min from the time of infusion. The order in which the delays were administered was randomized and each animal was tested once at each delay.

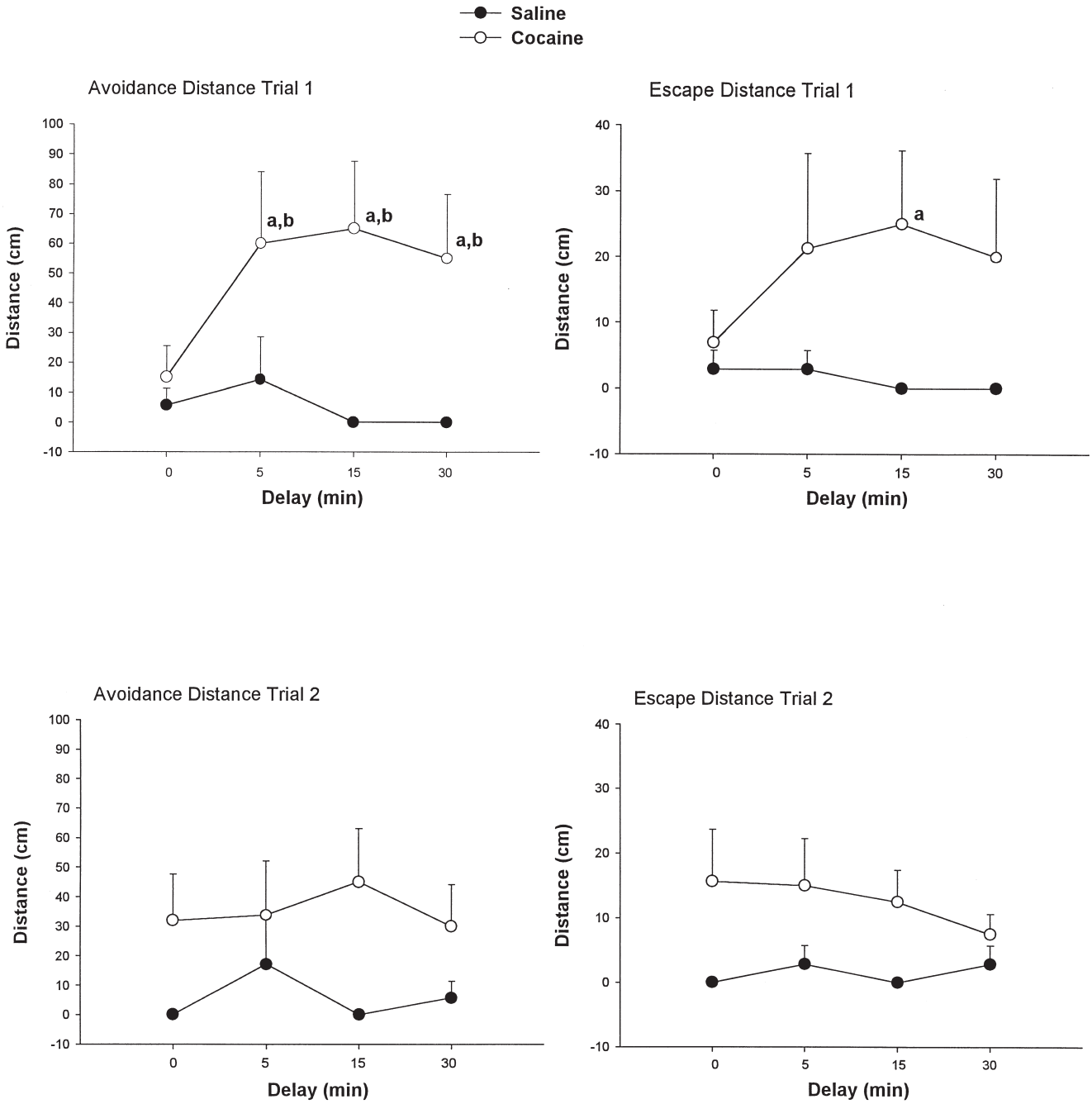

FIG. 4. Mean (and SEM) avoidance and escape distances on both discriminative/avoidance trials of Experiment 2. [a]Indicates significant difference from saline group ($p < 0.01$). [b]indicates significant difference from 0 min delay group ($p < 0.05$).

Apparatus. The runway was the same as that described in Experiment 1.

Procedure

Surgery. Venous catheters were implanted as described in Experiment 1.

Behavioral testing. On each test day, subjects were placed in a closed compartment of the alley and, depending on group infused with either saline or cocaine (4 mg/kg, 1 m/kg) as described in Experiment 1. Subjects remained in the closed compartment of the alley during the delay period. A sheet of clear Plexiglas was placed over the compartment to prevent escape while permitting videotaping of the animals from cameras mounted above the alley. The same behavioral measures were taken as in Experiment 1.

Statistics. Where possible, a two factor mixed ANOVA was performed on each behavioral measure, with drug as the

TABLE 3
MEAN (SEM) FREQUENCIES OF BEHAVIORS DURING CHASE TEST IN EXPERIMENT 2

		Trial 1				Trial 2			
		Delay (min)							
		0	5	15	30	0	5	15	30
Reversal	Saline	1.14 (0.63)	0.86 (0.55)	0.86 (0.40)	1.29 (0.64)	1.14 (0.77)	0.86 (0.55)	1.28 (0.71)	0.57 (0.43)
	n/7	3	2	4	3	2	2	3	2
	Cocaine	0.25 (0.25)	0.00 (0.00)	0.25 (0.16)	0.88 (0.52)	0.13 (0.13)	0.00 (0.00)	0.50 (0.33)	0.00 (0.00)
	n/8	1	0	2	3	1	0	2	0
Orientation	Saline	1.43 (0.75)	1.28 (0.52)	1.00 (0.53)	2.14 (0.91)	1.86 (0.63)	1.86 (0.63)	1.14 (0.46)	0.86 (0.26)
	n/7	3	5	4	5	6	6	4	5
	Cocaine	0.75 (0.49)	0.25 (0.25)	0.75 (0.49)	1.25 (0.77)	0.50 (0.27)	0.00 (0.00)	0.50 (0.50)	0.63 (0.38)
	n/8	3	1	3	3	3	0	1	3
Jump attack	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0	0	0	0	0
	Cocaine	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/8	0	0	0	0	0	0	0	0
Jump escape	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0	0	0	0	0
	Cocaine	0.50 (0.33)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.13 (0.13)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/8	2	0	0	0	1	0	0	0
Wall climb	Saline	0.00 (0.00)	0.29 (0.29)	0.00 (0.00)	0.14 (0.14)	0.00 (0.00)	0.00 (0.00)	0.14 (0.14)	0.14 (0.14)
	n/7	0	1	0	1	0	0	1	1
	Cocaine	0.13 (0.13)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/8	1	0	0	0	0	0	0	0
Stops	Saline	2.91 (0.92)	1.57 (0.48)	3.00 (1.13)	2.71 (0.84)	2.71 (0.52)	3.14 (0.96)	3.00 (0.95)	2.43 (0.95)
	n/7	5	6	5	5	6	6	5	5
	Cocaine	2.25 (1.13)	2.38 (1.15)	1.50 (0.87)	2.50 (1.21)	1.38 (0.65)	1.38 (1.24)	1.50 (0.82)	1.25 (0.56)
	n/8	5	5	4	6	4	2	3	4

n indicates number of animals exhibiting behavior.

between-group factor and delay as the repeated measure. Pairwise group comparisons were made using the Fishers Least Significant Difference test. Low-frequency measures and measures for which data were severely skewed were subjected to nonparametric Kruskal–Wallis tests. Alpha was set at 0.05 for all tests.

Results

Subject mortality. One subject in the saline condition had a dislodged catheter and was therefore sacrificed, leaving seven subjects for which data were available in this condition.

Discriminative/avoidance. Avoidance and flight distances for the two trials are shown in Fig. 4. There was an overall increase in avoidance behavior in cocaine-treated animals. There was a significant main effect of drug on avoidance distance for trials 1, $F(1, 13) = 10.755$, $p < 0.006$, and 2, $F(1, 13) = 6.53$, $p < 0.02$. Increased avoidance in cocaine animals persisted despite increased delays between infusion and testing. There was no main effect of delay on avoidance distance in either trial, $F(3, 39) = 1.15$, $p = 0.34$, and $F(3, 39) = 3.39$, $p = 0.88$, respectively, nor were there significant drug × delay interactions on either trial, $F(3, 39) = 1.24$, $p = 0.31$, and $F(3, 39) = 0.43$, $p = 0.73$. Escape behavior was also enhanced by cocaine administration. There was a significant main effect of drug on escape distance in both trial 1, $F(1, 13) = 6.80$, $p < 0.02$, and trial 2, $F(1, 13) = 13.42$, $p < 0.003$. There was no significant effect of delay on flight distance in either trial, $F(3, 39) = 0.57$, $p = 0.64$, and $F(3, 39) = 0.43$, $p = 0.74$, respectively. There was no significant drug × delay interaction on either trial as well, $F(3, 39) = 0.346$, $p = 0.63$, and $F(3, 39) = 0.43$, $p = 0.74$, respectively.

Chase test. Cocaine-treated animals exhibited high levels of flight during the chase test, with speed decreasing as the time between drug infusion and testing was increased. Chase speeds for both chase directions are shown in Fig. 5. There was a significant main effect of drug on flight speed for both trials, $F(1, 13) = 7.75$, $p < 0.01$, and $F(1, 13) = 7.88$, $p < 0.02$, respectively. The was a significant main effect of delay on flight speed for both trials as well, $F(3, 39) = 3.25$, $p < 0.03$, and $F(3, 39) = 3.26$, $p < 0.03$, and a drug × delay interaction on both trials, $F(3, 39) = 3.61$, $p < 0.02$, and $F(3, 39) = 3.27$, $p < 0.03$. Flight speeds were significantly higher for cocaine-treated animals compared to controls at all four delays in trial 1 and at all but the 30 min delay in trial 2 (ps < 0.01). On both trials there was a delay-dependent decrease in flight speed in the cocaine animals. Flight speeds at the 0 and 5 min delay were not significantly different on either trial, but there was a significant decrease in flight speeds at the 15 and 30 min delays compared to the shorter delays ($p < 0.01$). Data for other behavioral measures taken during the chase test are presented in Table 3.

All animals, regardless of drug and delay, showed equally low levels of these behaviors during this phase of the test.

Fixed distance approach. Behavioral measures taken during this phase of the test are presented in Table 4.

Cocaine-treated rats tended to freeze more than saline controls when confronted by the stimulus animal at fixed dis-

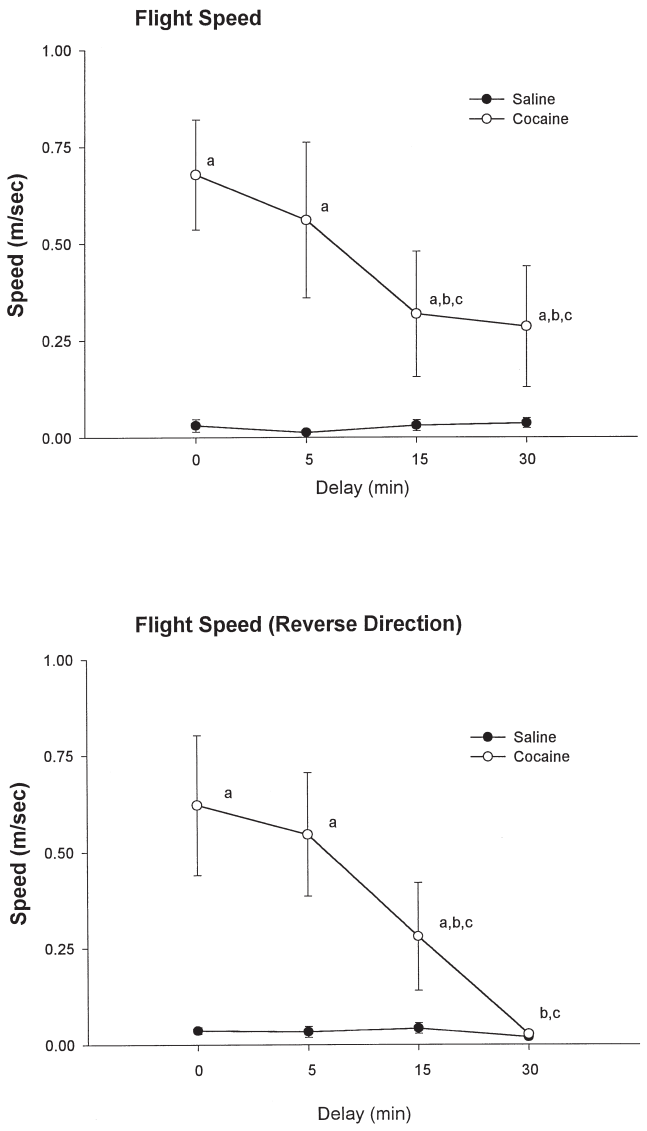

FIG. 5. Mean (and SEM) flight speed during chase test as a function of cocaine dose for both chase directions in Experiment 1. [a]Indicates significant difference from saline group ($p < 0.01$). [b]Indicates significant difference from 0 min delay group ($p < 0.01$). [c]Indicates significant difference from 5-min delay group ($p < 0.05$).

tances. There was a nearly significant drug × delay interaction for immobility time, $F(3, 39) = 2.50$, $p < 0.07$, but no significant main effect of ether drug, $F(1, 13) = 2.25$, $p = 0.16$, or delay, $F(3, 39) = 0.35$, $p = 0.79$. At every delay except 5 min, cocaine-treated rats spent significantly more time immobile than saline treated animals, according to the Fisher's post hoc test (ps < 0.01). Cocaine-treated rats at the 5-min delay spent less time immobile than at the 0-min delay ($p < 0.05$).

There was no significant main effect of either drug, $F(1, 13) = 1.88$, $p = 0.193$, or delay, $F(3, 39) = 3.39$, $p = 0.22$, on the closest distance between animals. However, the drug × delay interaction for this measure was significant, $F(3, 39) = 5.01$, $p < 0.01$. Fisher's post hoc analyses indicated a significant difference between the 0- and 15-min delay saline-treated animals ($p < 0.01$). At the 0- and 15-min delays, cocaine produced significantly greater distances between the stimulus and the subjects than saline ($p < 0.05$ and 0.01, respectively). There was no main effect of drug, $F(1, 13) = 0.35$, $p = 0.56$, or delay, $F(3, 39) = 1.46$, $p = 0.24$, on time out of first square, nor was the drug × delay interaction significant for this measure, $F(3, 39) = 1.76$, $p = 0.17$.

There was no main effect of drug on contact time, $F(1, 13) = 1.52$, $p = 0.24$, but there was a significant main effect of delay, $F(3, 39) = 3.37$, $p < 0.03$. The drug × delay interaction was not significant, $F(3, 39) = 1.52$, $p = 0.23$.

Forced contact. As Fig. 6 shows, cocaine increased upright defense and biting during the three forced-contact trials. The Kruskal–Wallis ANOVA indicated a significantly higher number of upright defense postures in cocaine-treated rats compared to saline controls at the 0 delay, $H(1, 15) = 10.37$, $p < 0.001$, 15-min delay, $H(1, 15) = 4.2$, $p < 0.04$, and the 30-min delay, $H(1, 15) = 4.45$, $p < 0.03$. The 5-min delay approached, but did not achieve statistical significance, $H(1, 15) = 2.81$, $p = 0.09$. Cocaine-treated rats exhibited biting, whereas none of the saline animals did so. At the 5-min delay, but not the other delays, the number of bites by cocaine treated subjects was significantly higher than that of saline controls, $H(1, 15) = 4.45$, $p < 0.03$. Biting levels at the 0 delay approached significance, $H(1, 15) = 3.06$, $p = 0.08$. Data for other measures taken during the forced-contact test are summarized in Table 5. Saline animals exhibited no jump attacks, jump escapes, flight, or vocalization, but cocaine-treated rats exhibited these behaviors occasionally.

Discussion

As in the first experiment, rats given 4 mg/kg cocaine intravenously in Experiment 2 displayed a well-oriented flight response. The intensity of the flight response declined as a function of time between infusion and testing but remained significantly higher for cocaine-dosed animals than for saline controls for 30 min. Cocaine increased avoidance and escape behavior during the discriminative/avoidance trials, and the magnitude of this effect did not decline overall with increased delay between drug infusion and testing. However, avoidance and escape behavior was not increased during the first of the two discriminative avoidance trials for animals in the 0 delay condition. Consistent with the results of Experiment 1, these results suggest a small window of the order of 1–2 min following intravenous administration before which the drug affects defensive behavior.

In addition to the time-dependent decrease in flight, there was evidence of residual hyperdefensiveness that persists at least 30 min, the longest delay employed in this study. Rats in the second experiment displayed increased avoidance and escape, and, in immobility and upright defense even at the longest delays; the enhancement of upright defense was consistent with results obtained in the first experiment. There was also a tendency for increased biting across all delays during the forced contact test, but only the 5-min delay was significant. Thus, intravenous administration of 4 mg/kg cocaine resulted in an initial flight response that appeared to peak approximately 5 min following infusion and declined significantly by 15 min postinfusion. Heightened defensiveness, as measured by avoidance, escape, immobility, and upright defense, persisted for at least 30 min, suggesting prolonged, residual effects of cocaine on a variety of defensive behaviors.

The magnitude of the cocaine effect on defense was impressive in that the drug produced defensive behaviors not typically observed in laboratory rats. In fact, the pattern of de-

TABLE 4
MEAN (SEM) RESPONSES DURING FIXED DISTANCE TEST IN EXPERIMENT 2

		Delay (min)			
		0	5	15	30
Closest distance between animals	Saline	51.43 (8.96)	55.71 (9.42)	34.29 (12.32)†	54.29 (9.59)
	Cocaine	63.54 (4.23)*	57.08 (2.22)	65.00 (4.67)*	59.08 (3.33)
Time out of first square (s)	Saline	3.10 (1.24)	2.05 (1.84)	6.76 (2.58)	2.43 (1.66)
	Cocaine	2.50 (1.34)	3.42 (1.82)	2.38 (1.56)*	1.58 (1.58)
Immobile time (s)	Saline	11.10 (2.30)	12.14 (2.10)	10.29 (2.22)	11.76 (2.13)
	Cocaine	14.88 (0.09)*	12.62 (0.48)†	14.67 (0.29)*	14.21 (0.50)*
Contact time (s)	Saline	1.19 (0.70)	0.66 (0.54)	3.19 (1.60)†	1.14 (0.77)
	Cocaine	0.00 (0.00)	0.80 (0.44)	1.04 (0.70)*	0.42 (0.42)

*Indicates significant difference between cocaine and saline group ($p < 0.05$).
†Indicates significant difference from 0 min delay condition.

fensive behaviors of rats receiving a high dose of intravenous cocaine resembled that of wild rats tested in a similar context (3). Thus, the current findings suggest that cocaine can exert powerful effects on normally quiescent laboratory rat defense systems.

The effective dose in the present study (4 mg/kg), while higher than a single bolus amount typically given in self-administration procedures (6,11), was within the range of doses others have used to investigate aversive and rewarding aspects of cocaine in other experimental paradigms involving IV administration. A study by O'Dell and colleagues (26) examined conditioned place preference (CPP) or avoidance (CPA) in rats given daily infusions of 4.2 and 5.6 mg/kg, and another by Nomikos and Spyraki (25) studied the same phenomena using 2.5, 5, and 10 mg/kg. The two studies produced opposite results for doses around 5 mg/kg: the former reported CPP at both the 4.2 and 5.6 mg/kg dose, whereas the latter showed CPP at 2.5 but CPA avoidance at the 5 and 10 mg/kg dose. One possible explanation for the discrepancy between these results was that more conditioning trials were used in the study that produced CPP (26). In another paradigm used to evaluate reward and aversive properties of cocaine, rats were

TABLE 5
MEAN (SEM) FREQUENCY OF BEHAVIORS IN FORCED CONTACT TEST IN EXPERIMENT 2

		Delay (min)			
		0	5	15	30
Bites	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0
	Cocaine	1.13 (0.55)	1.50 (0.57)	0.63 (0.42)	0.75 (0.49)
	n/8	3	4	2	2
Upright	Saline	0.29 (0.29)	0.14 (0.14)	0.14 (0.14)	0.00 (0.00)
	n/7	1	1	1	0
	Cocaine	2.50 (0.33)	1.50 (0.57)	1.63 (0.53)	1.50 (0.57)
	n/8	8	4	5	4
Jump attack	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0
	Cocaine	0.25 (0.16)	0.63 (0.32)	0.25 (0.16)	0.25 (0.25)
	n/8	2	3	2	1
Vocalization	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0
	Cocaine	0.13 (0.13)	0.38 (0.38)	0.13 (0.13)	0.50 (0.33)
	n/8	1	1	1	2
Flight	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0
	Cocaine	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.25 (0.25)
	n/8	0	0	0	1
Jump escape	Saline	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)	0.00 (0.00)
	n/7	0	0	0	0
	Cocaine	0.75 (0.49)	0.13 (0.13)	0.00 (0.00)	0.13 (0.13)
	n/8	2	1	0	1

n indicates number of animals exhibiting behavior.

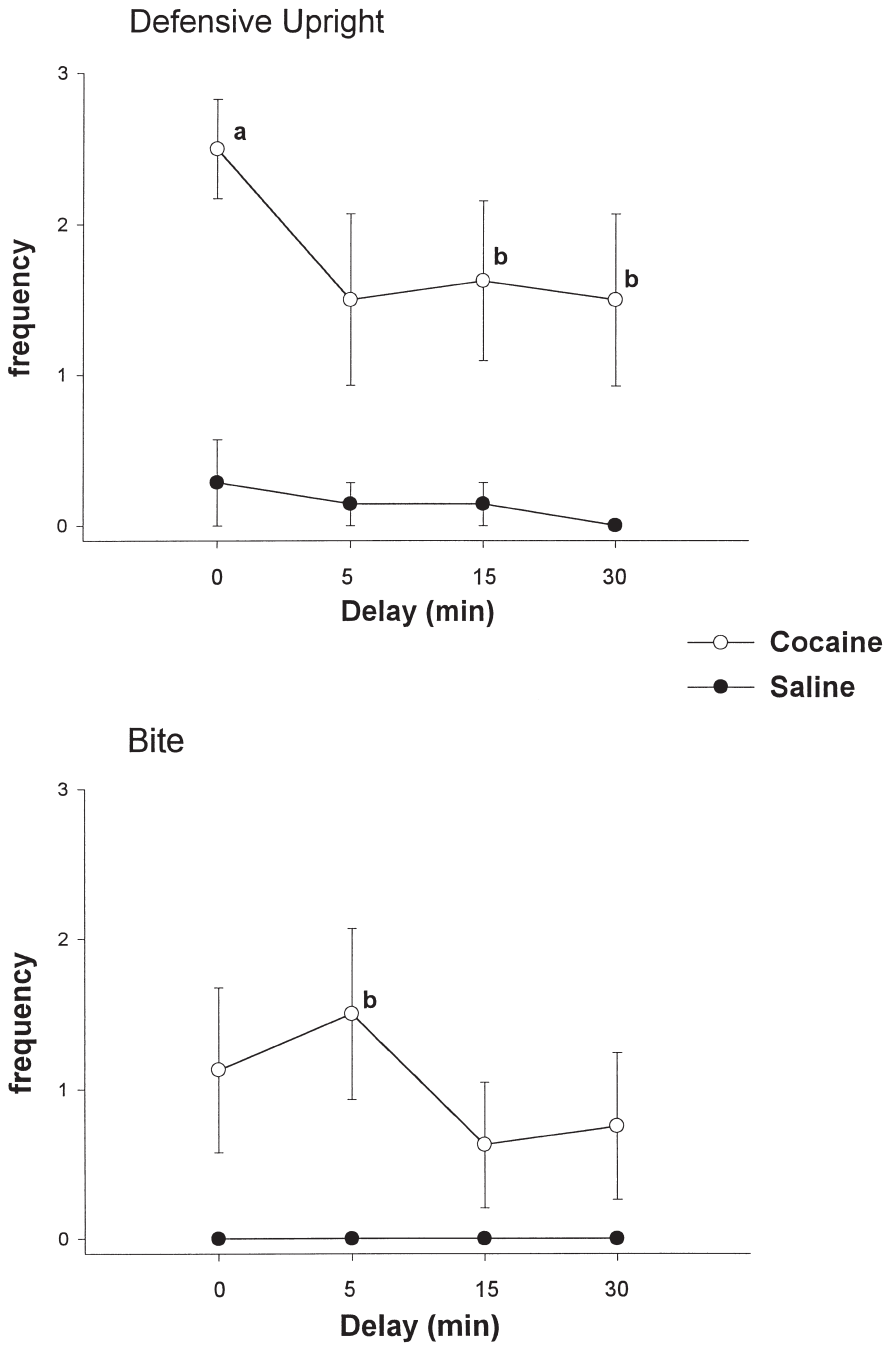

FIG. 6. Mean (and SEM) number of defensive upright and bites during forced contact test in Experiment 2. [a]Indicates significant difference from saline group ($p < 0.01$). [b]Indicates significant difference from saline group ($p < 0.05$).

trained to traverse a straight alley for five discrete (5 s each) 0.75 mg/kg IV injections of cocaine totaling 3.75 mg/kg within a single, 2-min injection period (11,19). In the present study, nearly the same amount of cocaine (4 mg/kg) was infused (albeit continuously) within the same amount of time (2 min). Thus, our selection of 4 mg/kg IV cocaine was consistent with doses used in other studies of related phenomena.

The major focus of the present study was to provide a ethologically relevant situation in which to examine rat defensive behavior following IV administration of cocaine, aimed at the creation of models relevant to analysis of the negative emotional effects of cocaine in humans. However, in view of the many studies indicating reward effects of IV cocaine at these (11,19) and lower (14,15) IV cocaine doses, the present

findings raise the question of the relationship between the defensive and the rewarding effects of cocaine. One possibility is that doses of cocaine that are initially aversive can become rewarding (26), perhaps through habituation of defensive effects and sensitization of reward. (The present results, showing no interaction of cocaine effects with repeated testing, do not indicate rapid habituation of defense effects, but this was over only four trials.) Alternatively, both defense and reward effects may be dose dependent such that defense becomes a factor only at higher doses. Although the present results do not provide any specific view of the relationship of these effects, they do indicate a defense component to cocaine effects that must be considered in conjunction with analysis of its reward characteristics; and they suggest that an adequate conceptualization of cocaine effects must take into account both processes and their potential interactions.

We can only speculate on the underlying mechanisms for cocaine effects on defense. There is increasing evidence that activation of the hypothalamic–pituitary–adrenal (HPA) axis is a necessary condition for cocaine self-administration (14,15,22,27) and the development of cocaine-induced stereotypy (22,32) in rats. Thus, it is possible that circulating corticosteroids may have played an important permissive role for the effects observed in the present study.

The present findings in laboratory rats are consistent with clinical reports of panic and anxiety associated with use of cocaine in humans (10,13,31). Results of preclinical pharmacological studies suggest that flight and escape responses may be useful indicators of panic. Panicogenic compounds such as yohimbine have been shown to selectively potentiate these behaviors in mice in the MDTB (5), while panicolytic drugs, alprazolam (17), fluoxetine, and imipramine (16) reduce flight, but only (again consonant with clinical findings) when given on a chronic basis. In the present study the flight effect had a sudden onset and, although intense, was short lasting. Thus, temporal characteristics of the flight component sharply parallel those of panic or anxiety attacks in humans. Longer lasting increases in other (less intense) defensive behaviors suggest a period of residual hyperdefensivenss following the peak ("panic") episode, which, again, is typical of residual anxiety following "attacks" of panic in humans. Taken together, the current results indicate that acute intravenous administration of cocaine has potent, time-dependent effects on defensive behaviors in laboratory rats, and that these effects may be particularly related to cocaine-associated panic or anxiety-related disorders in humans.

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