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Effects of Amphetamine and Phenylpropanolamine on Latency to Feed and Cumulative Liquid Diet Intake in Rats[1]

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WELLMAN, P. J. AND R. COCKROFT. *Effects of amphetamine and phenylpropanolamine on latency to feed and cumulative liquid diet intake in rats.* PHARMACOL BIOCHEM BEHAV 32(1) 147–150, 1989.—The purpose of the present experiment was to compare the actions of d-amphetamine sulfate (AMP) and phenylpropanolamine hydrochloride (PPA: d,l-norephedrine) on feeding of a liquid diet. Adult rats were deprived of food for 22 hours and treated with 1.0 and 2.0 mg/kg AMP and with 15 and 30 mg/kg PPA 30 minutes prior to a 60-minute feeding test. Latency to begin feeding was recorded at the start of the feeding session with cumulative liquid diet intake recorded every 5 minutes during the test. Amphetamine and PPA significantly increased latency to feed but PPA, at the doses used here, produced a greater reduction in overall feeding during the test sessions than did amphetamine. Eating rate was reduced by both AMP and PPA during the first 20 minutes of the test session, but eating rate after AMP was significantly higher during the latter portion of the test session. Although amphetamine and PPA are often likened as similar in structure and function, these results suggest important qualitative differences between the anorexic activities of these two drugs.

Phenylpropanolamine Amphetamine Anorexia Latency to feed Cumulative food intake Stereotypy

PHENYLPROPANOLAMINE (PPA), a racemic mixture of d- and l-norephedine, induces anorexia and weight loss (6, 7, 9, 10, 16, 18, 24) that is dose- and isomer-dependent (9, 17, 24). The mechanism by which PPA reduces feeding remains unknown but may be related to catecholaminergic mechanisms within the lateral hypothalamus (8) or to alteration of rate of gastric emptying (20). In contrast, the anorexic action of amphetamine has been linked to activation by amphetamine of dopaminergic and/or beta-adrenergic receptors within the perifornical hypothalamus (1, 4, 11–13) and the pathways by which amphetamine reduces feeding have been described from the brainstem to the perifornical region (13). Moreover, a number of studies have documented important differences in anorexic magnitude, isomer potency and mechanism of action between amphetamine and PPA (5, 6, 15, 22, 24). Few studies, however, have sought to compare the qualitative actions of PPA and amphetamine on feeding behavior in rats. Blundell (2), for example, reported that amphetamine significantly increased latency to feed, reduced overall food intake, yet increased eating rate. The purpose of the present experiment was to determine the effects of PPA (15 and 30 mg/kg) on latency to feed as well as the cumulative intake curve during a 60-minute feeding trial and to compare these effects with that of dose levels of amphetamine (1.0 and 2.0 mg/kg) that produce comparable levels of anorexia.

METHOD

Animals

The animals were 16 male Sprague-Dawley albino rats (obtained from Timco, Inc.; Houston, TX) weighing 174–199 grams at the beginning of the study. The rats were housed individually in standard plastic rodent cages in a colony room maintained at 23.0°C under a 12-hr/12-hr illumination schedule (lights *on* at 0800 hr). The rats were provided continuous access to tap water throughout the experiment but limited access to a liquid diet as described in the schedules below.

Diet

The rats were offered a nutritionally complete liquid diet (Bioserv Inc., Control Diet) which provided 1 kcal/ml and consisted of 18% protein, 35% fat and 47% carbohydrate. The liquid diet was prepared fresh daily and was offered to the rats in 100 milliliter calibrated Wahmann drinking tubes.

[1]The procedures of this study were reviewed and approved by the Texas A&M University Laboratory Animal Care Committee.

Drugs

In this experiment, a vehicle solution was prepared using sterile distilled water and 0.9% (w/v) sodium chloride. Amphetamine solutions (1.0 and 2.0 mg/ml) and phenylpropanolamine solutions (15 and 30 mg/ml) were prepared using d-amphetamine sulfate and phenylpropanolamine hydrochloride (Sigma Chemical Company) mixed into the vehicle solution.

Procedure

The rats were maintained in the colony for a week prior to the start of the experiment to acclimate them to daily handling procedures.

Beginning on Day 1 of a nine-day baseline period, the rats were offered a single tube of the liquid diet as well as a water tube in the home cage at 1715 hr. Latency to begin feeding (in seconds) was recorded for each rat at the start of the session. If feeding was not initiated within 60 seconds, the latency measure was terminated and a score of 60 (sec) was recorded for that rat. At 5-minute intervals after the start of the session, the volume remaining in the drinking tube was recorded for each rat during a 60-minute period. After the end of the 60-minute test, the rats were offered the liquid diet for an additional hour (so as to allow the rats sufficient time to consume their daily required calories). The liquid diet was then removed from the home cage with water, but not food, available to each rat in the home cage during the 22-hour deprivation interval that separated each baseline test. On Days 7–9 of the baseline period, each rat received a single injection of vehicle (1.0 mg/kg, IP) 30 minutes prior to the start of the test session so as to accustom them to the injection procedure.

The drug test sequence consisted of 10 trials of which 4 (Days 1, 4, 7 and 10) were preceded by one of the amphetamine or PPA doses, whereas the remainder consisted of blocks of 2 vehicle trials between drug tests as noted below:

TEST INJECTION:
DRUG-VEH-VEH-*DRUG*-VEH-VEH-*DRUG*-VEH-VEH-*DRUG*

TEST DAY:
1 2 3 4 5 6 7 8 9 10

Each rat received either 1.0 or 2.0 mg/kg amphetamine or 15 or 30 mg/kg PPA 30 minutes prior to the ingestive test. Every rat received every drug type and dose once during the 4 test days but in a randomly counterbalanced order. The vehicle tests between successive drug trials served to allow rats to recover baseline levels of feeding after drug injection and to minimize carryover effects.

Statistical Analyses

In the present experiment, two drugs (AMP, PPA) and two dose levels (LOW, HIGH) were used in a factorial design in which every rat received every level of the two factors. Inspection of the latency measures revealed the presence of a variable number of zero scores as well as proportional means and variances; these measures were subjected to a square-root transformation. The latency data were analysed using a 2×2 factorial model with the within-group factors of DRUG (AMP, PPA) and DOSE (LOW, HIGH). The liquid diet intakes during the 5-minute intervals of each 60-minute test session after drug treatment underwent separate analyses using cumulative liquid diet intake and eating rate. The latter measure was calculated as the diet consumed per minute per 5-minute block. The cumulative intake and eating rate data were analysed using a 2×2×12 factorial model with the within-group factors of DRUG, DOSE and TIME (5, 10, 15, 20, 25, 30, 35, 40, 45, 50, 55 and 60 minutes) (3). The criterion for statistical significance was set at 0.05.

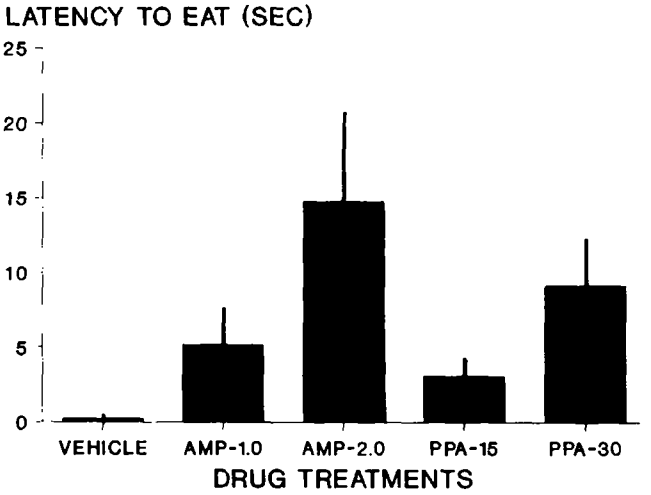

FIG. 1. Mean group latency to initiate feeding (seconds) for rats treated (IP) with vehicle, amphetamine (AMP: 1.0 and 2.0 mg/kg) and phenylpropanolamine (PPA: 15 and 30 mg/kg). The line above each bar represents the mean plus one SEM.

RESULTS

Figure 1 depicts mean group latency to feed after vehicle and after low and high doses of amphetamine and PPA. After vehicle treatment, the rats exhibited very short latencies (0.23 sec) to begin feeding. In contrast, both amphetamine and PPA lengthened the latency to begin feeding when given 30 minutes before the feeding trials. Moreover, increases in drug dose, for both amphetamine and PPA, resulted in increased latency scores. These differences were confirmed by analyses of variance which revealed a significant effect of DOSE, $F(1,15)=6.1$, $p<0.03$, but no significant effect of DRUG ($p<0.33$) or of the interaction between the factors of DRUG and DOSE ($p<0.55$). Thus, although there was a trend for amphetamine to induce longer latency scores than those observed after PPA, this difference was not statistically significant nor was there a significant interaction between drug type and drug dose on latency to eat.

Figure 2 depicts the changes in cumulative milk intake observed after treatment with vehicle, amphetamine and PPA. After vehicle treatment, the rats exhibited a rapid rate of eating as evident in the steep slope of the curve in Fig. 2, with the eating rate slowing during the remaining 40 minutes of the test session. The low doses of amphetamine and PPA exhibited comparable cumulative intake curves, whereas the curves for the high doses of amphetamine and PPA exhibited divergent patterns. The slope of the PPA high dose curve remained essentially constant, whereas the slope of high dose of amphetamine curve was positively accelerated during the last third of the test session. Analyses of variance

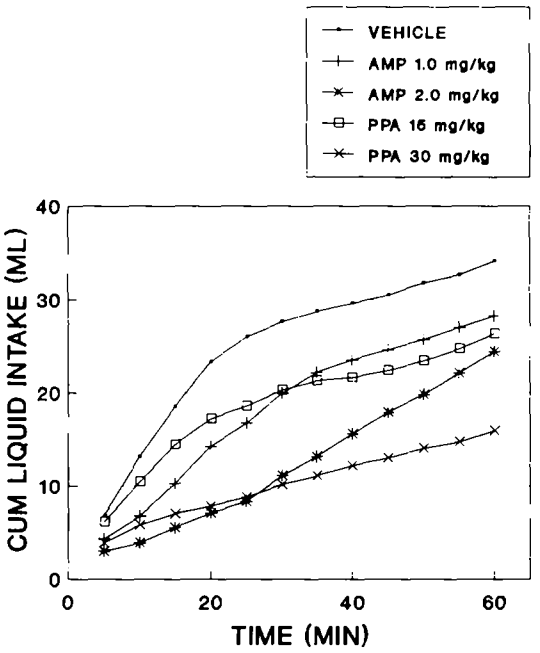

FIG. 2. Mean group cumulative liquid diet intake curves over a 60-minute period for rats treated with vehicle, amphetamine and PPA.

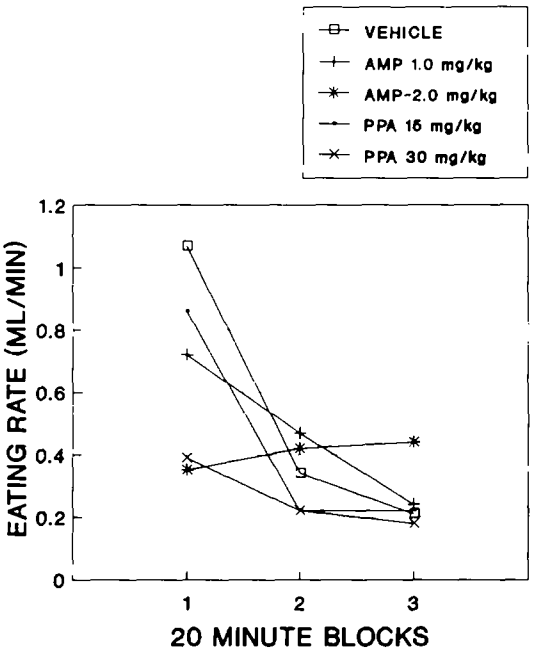

FIG. 3. Mean group eating rate (ml/min) averaged over twenty-minute blocks during 60-minute test trials after treatment with vehicle, amphetamine and PPA.

revealed significant effects of DRUG, $F(1,15)=17.2$, $p<0.0009$, of DOSE, $F(1,15)=25.3$, $p<0.0001$, and of the interaction between the factors of DRUG and DOSE, $F(1,15)=5.4$, $p<0.03$. PPA induced a greater overall suppression of feeding than did amphetamine, the high dose produced a greater overall suppression of feeding than did the low dose and the difference in the anorexic activity of the PPA high dose was greater than that of the high dose of amphetamine.

To further explore the anorexic activity of these two drugs, additional analyses were computed using eating rate (calculated using the volume of liquid diet consumed during a 5-minute interval expressed per minute); these data are presented in Fig. 3 for the three 20-minute intervals of each 60-minute test session. The rapid change in eating rate observed after vehicle treatment in the cumulative intake data are evident when the data are expressed as eating rates. The interesting aspect of Fig. 3 relates to the changes in eating rate observed in the high dose amphetamine and PPA conditions. Although rate of eating after treatment with the high dose of amphetamine was greatly slowed in these rats during the first 20 minutes of the session (0.35 ml/min), their eating rate after 2.0 mg/kg amphetamine was higher (relative to the eating rate after vehicle) during the last 20 minutes of the test session (0.44 ml/min). In contrast, the rates of eating after 30 mg/kg PPA for the three phases of the test period were 0.39, 0.22 and 0.18 ml/min. These differences in eating rate were confirmed by analyses of variance which revealed a significant interaction between the factors of DRUG and TIME, $F(11,165)=3.8$, $p<0.0001$.

DISCUSSION

In the present experiment, latency to feed and eating rate were recorded for rats treated with low and high doses of both amphetamine and phenylpropanolamine. Amphetamine and PPA, at each dose level, produced comparable lengthening of latency to initiate feeding, but PPA produced a greater overall reduction of feeding at the high dose relative to that of amphetamine. Moreover, PPA exerted a maintained inhibitory effect on feeding rate, whereas amphetamine initially reduced feeding rate but then increased eating rate during the latter third of the test session.

Although amphetamine and PPA had similar effects on latency to feed, differences in feeding pattern were noted in the present study. Eating rates after PPA gradually declined during the test session, whereas that observed following the high dose of amphetamine actually increased. Moreover, informal observations of the rats during the feeding trials revealed the presence of stereotypy and arousal after amphetamine, whereas after PPA, the rats exhibited a prone posture, pilomotor erection, salivation and exopthalmos. These differential effects of AMP and PPA on behavior have been noted in the past literature (5, 6, 8, 21) and suggest that these drugs may have qualitatively different effects related to their systemic and central nervous system actions. Amphetamine readily induces stereotypy owing to its potent effect on central dopaminergic synapses, whereas PPA is a relatively weak dopamine agonist (14). Moreover, amphetamine readily enters brain, whereas PPA, because of its low lipid solubility, less easily gains access to brain (19). Amphetamine anorexia is prevented by pretreatment with dopamine antagonists (1, 11, 12, 23), whereas the anorexia induced by PPA is not (23). Further studies are likely to reveal that although amphetamine and PPA induce superficially similar anorexia, these effects are the result of activation of pharmacologically and anatomically distinct mechanisms (9, 11-13, 22, 23).

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