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Effect of β-Phenylethylamine and d-Amphetamine on Electrical Self-Stimulation of Brain[1]

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(Received 28 September 1976)

HOWARD, J. L., G. T. POLLARD, K. W. ROHRBACH, N. E. HARTO. *Effect of β-phenylethylamine and d-amphetamine on electrical self-stimulation of brain.* PHARMAC. BIOCHEM. BEHAV. 5(6) 661–664, 1976. – β-phenylethylamine (PEA) has been viewed as amphetamine-like in its effects on behavior. Support for this putative similarity of action has been derived primarily from observations that both of these structually related compounds increase locomotor activity in a dose-related manner and at higher doses evoke stereotypies. Since d-amphetamine (d-A) produces a dose-related increase in the rate of bar pressing for electrical stimulation of the medial forebrain bundle, the effect of PEA on this behavioral paradigm was examined. Male Long-Evans rats implanted with bipolar electrodes self-administered 250 msec 60 Hz constant current sine wave trains over a 30–70 μA range of intensities in daily 20-min tests. Over a range of 1–40 mg/kg IP of PEA, a dose-related decrease in self-stimulation rate was observed; pretreatment with para-chlorophenylalanine or alpha-methyl-para-tyrosine did not alter the response to 2.5 or 30 mg/kg IP of PEA. Since within the dose range of PEA used in this study a dose-related increase in locomotor activity was observed, and since d-A increases self-stimulation rate at doses that increase locomotor activity, it would seem that there are qualitative differences in the actions of d-A and PEA on behavior.

β-phenylethylamine d-Amphetamine Self-stimulation

β-PHENYLETHYLAMINE (PEA), a close structural analogue of amphetamine, has been shown to be a stimulant with amphetamine-like effects on locomotor activity [4, 8, 11]. PEA also resembles amphetamine in its effects on other behaviors, its electrophysiological actions and its electrotonic properties [15]; it blocks reuptake of catecholamines [3] and releases catecholamines [5,10] as does amphetamine.

PEA appears to be an endogenous substance, having been found in brains of a number of species [16]. In light of this, and because of its similarity of action to amphetamine, PEA has been characterized as an endogenous amphetamine.

There are, however, differences in action between amphetamine and PEA. PEA does not induce the full stereotyped behavior pattern that can be evoked by amphetamine [12] and does not by itself produce an amphetamine-like cue in drug discrimination work [7; C. Jones and J. Howard, unpublished observations]. PEA alone also did not produce an amphetamine-like increase in intracranial self-stimulation (ICSS) [18]. The narrow dose range and limited amount of quantitative reporting of results in the work of Stein [18] prompted the more complete evaluation of the effect of PEA on ICSS in the present study. The effect of PEA over a wide dose range is compared to the effect of d-amphetamine (d-A) on electrical self-stimulation of brain and on locomotor activity.

METHOD

Animals

Male Long-Evans rats obtained from Blue Spruce Farms were used in all studies. Animals were maintained on a 12-hr light/12-hr dark reversed cycle and had ad lib access to food and water.

Apparatus

Self-stimulation determinations were carried out in two identical operant chambers and enclosures (Coulbourn Instrument Co.), each equipped with a single manipulandum, six cue lights, a houselight and a mercury commutator through which brain stimulation was delivered. Stimulation consisted of 250 msec 60 Hz sine waves of different intensities produced from two channels of a Microtronics constant current stimulator. Intensity of stimulation for each channel was determined by a resistor network. Experimental control and data acquisition were

[1] Portions of this paper were presented at the Fall pharmacology meeting in New Orleans, La., August 16–19, 1976.

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provided via an Interact interface (Lehigh Valley Electronics) and a Nova 2/10 computer (Data General Corp.).

Locomotor activity data was gathered in 12 identical circular photocell cages (Woodard), and data acquisition and timing were provided by the Interact System.

Procedure

Animals to be used for self-stimulation experiments were implanted at 350–400 g with a bipolar electrode (Plastic Products) in the left medial forebrain bundle according to coordinates derived from Pellegrino and Cushman [13] – 0.5 mm posterior to Bregma, 1.6 mm lateral to the sagittal suture, 9.1 mm from skull surface – using conventional stereotaxic techniques under 50 mg/kg IP sodium pentobarbital anesthesia. Following at least one week of recovery from surgery, animals were allowed to shape themselves for self-stimulation during a 12-hr session [14] and then to stabilize on daily 20-min sessions (6 days per week) identical to the sessions later employed for testing. Five different ICSS intensities were used in every session: 30, 40, 50, 60, and 70 μA. Four min after the animal was placed in the operant chamber for testing, the houselight came on, 10 priming stimuli at the middle intensity were given, and the first 10-min self-stimulation period occurred; then followed a 2-min time-out and a second 10-min self-stimulation period. Within each 10-min period, each of the 5 intensities was presented for a block of 2 min; during the first period the order of presentation was 50, 70, 40, 60, and 30 μA, and during the second period it was 30, 70, 40, 60, and 50 μA. Each intensity was signalled by a unique combination of cue lights, and a priming stimulus at that intensity started each block. After response rates stabilized within 20% from day to day, the effect of drugs was determined. In general, drugs were administered on Tuesday and Friday, and the other 4 days of the week were used to determine baseline response rates. Following the study, animals were sacrificed and electrode placement was confirmed by histological evaluation.

Animals used in locomotor activity data experiments were acclimated to the laboratory for 2 to 3 weeks and weighed 180–210 g when run. They were placed in the activity cages for a 1-hour habituation period before being injected and immediately replaced in the cages. Following injection, activity counts were accumulated into 2 min time bins.

Drugs used. All drugs were injected intraperitoneally and administered in approximately equal volumes. Doses are specified in terms of the salt. β-phenylethylamine (PEA) was crystalized as a HCl salt from the free base (Eastman). d-Amphetamine SO_4 (d-A) was purchased from Elkins-Sinn. Para-chlorophenylalanine (PCPA) was a gift of Pfizer and alpha-methyl-para-tyrosine (αMT) of Merck, Sharp and Dhome.

Statistics. Differences between treatments within groups were analyzed with the Wilcoxon matched pairs signed ranks test [17], and differences between groups were assessed by the Mann-Whitney Test [17]. A $p<0.05$ was considered significant.

RESULTS

Figure 1 shows the results of injection of a wide range of doses of PEA on electrical self-stimulation of brain. No dose-dependent increase in self-stimulation rate was found at any intensity during either the first or second 10-min block of time. During the first 10 min, 30 mg/kg decreased self-stimulation rates at all intensities, and doses of 10 and 20 mg/kg decreased self-stimulation rates at the 40 and 50 μA intensities. During the second 10 min, self-stimulation rates were significantly decreased at the 40 μA intensity by 10 and 20 mg/kg, at the 50 μA intensity by 10 mg/kg, and at the highest intensity by the 20 and 30 mg/kg doses.

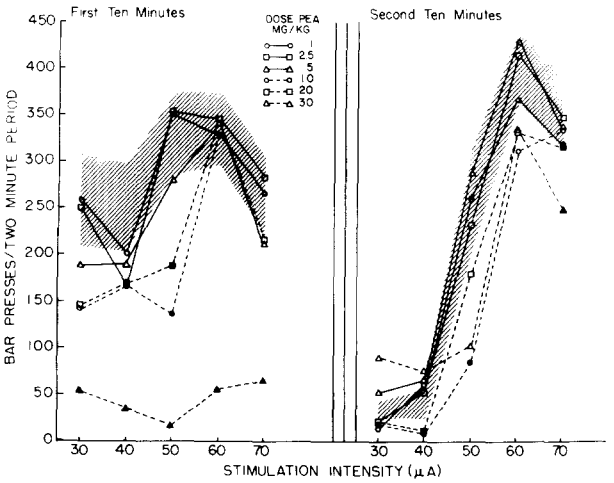

FIG. 1. Self-stimulation rate following various doses of β-phenylethylamine (○——○ 1, □——□ 2.5, △——△ 5, ○- - -○ 10, □- - -□ 20, △- - - -△ 30 mg/kg IP) over a range of current intensities during the periods 4–14 min (left panel) and 16–26 min (right panel) after injection. Each point is the mean of six animals, and the shaded portion represents the area of plus and minus one standard error of the mean following saline injection. Filled symbols denote points significantly different from control by a Wilcoxon Test.

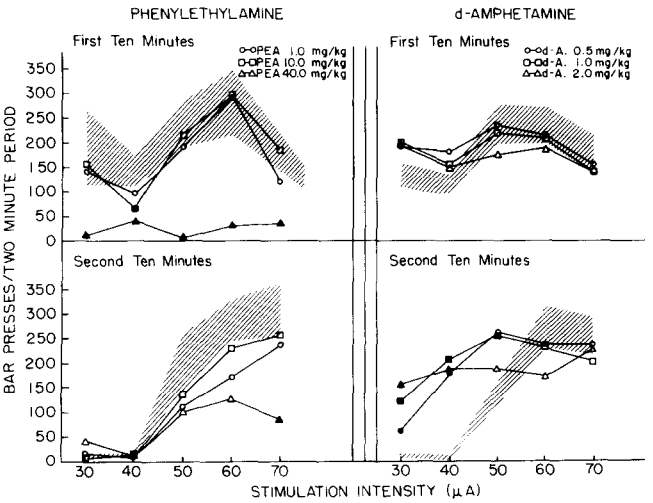

FIG. 2. Self-stimulation rates following β-phenylethylamine (○——○ 1, □——□ 10, △——△ 40 mg/kg IP left panels) or d-amphetamine (○——○ 0.5, □——□ 1, △——△ 2 mg/kg IP right panels) over a range of current intensities during the periods 4–14 min (top panels) and 16–26 min (bottom panels) after injection. Other information in Fig. 1.

Figure 2 shows a direct comparison of three dose levels of PEA and d-A in another group of rats. During the first 10 min of self-stimulation, PEA at 40 mg/kg showed significant rate-decreasing effects at all intensities, and 10 mg/kg decreased the rate at the 40 μA intensity; during the same time period, d-A was without effect. The only

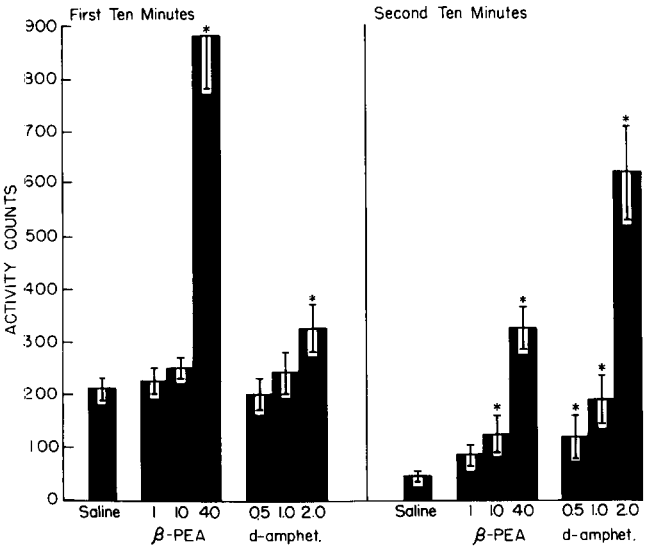

FIG. 3. Locomotor activity following three doses of β-phenylethylamine (Ns = 12), three doses of d-amphetamine (Ns = 12), or saline (N = 36) during the periods 4–14 min (left panel) or 16–26 min (right panel) following injection. Animals were habituated for one hour to the activity cages before injection. Insert bars represent one standard error of the mean. Asterisks over the bars indicate a significant difference from saline control by the Mann-Whitney Test. Doses are in mg/kg IP.

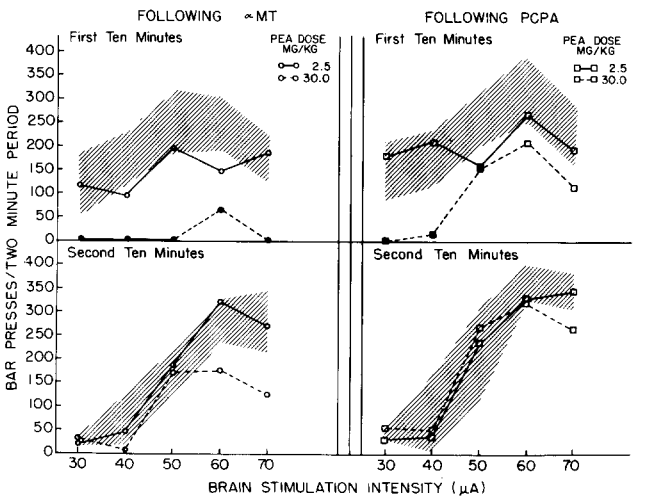

FIG. 4. Self-stimulation rates following saline (shaded portion represents plus and minus one standard error of the mean), 2.5 ○——○ or 30 ○- -○ mg/kg of β-phenylethylamine in animals (N = 6) pretreated with alphamethylparatyrosine (left panels) or parachlorophenylalanine (right panels) over a range of current intensities during the periods 4–14 min (top panels) and 16–26 min (bottom panels) after injection. Alphamethylparatyrosine treatment was 100 mg/kg IP 3 hr prior to a session. Parachlorophenylalanine was administered immediately following the session for four consecutive days: Day 1, 100 mg/kg; Days 2–4, 50 mg/kg. The data presented was collected on Days 3–5 with treatment in counterbalanced order. Filled points represent points significantly different from control by a Wilcoxon Test.

significant change in the second 10 min following PEA injections was after 40 mg/kg and was a rate decrease at the highest intensity. Over the same time period, d-A at 0.5, 1.0 and 2.0 mg/kg reliably increased self-stimulation rates at 30 and 40 μA intensities, and 0.5 and 1.0 mg/kg also increased rates at the 50 μA intensity.

Figure 3 shows the effects of the same three dose levels for each drug on locomotor activity during the same period of time after injection as the data in Fig. 2 on self-stimulation. During the first 10 min, PEA at 40 mg/kg and d-A at 2.0 mg/kg stimulated locomotor activity. During the second 10 min, dose-related increases in locomotor activity were observed following both PEA and d-A. That is, PEA and d-A produced very similar effects upon locomotor activity, whereas they produced very dissimilar effects in the self-stimulation paradigm.

The effect in the self-stimulation paradigm of blocking catecholamine and serotonin synthesis before administration of PEA was also tested (Fig. 4). Following either αMT or PCPA, PEA at 2.5 mg/kg still did not alter response rates. The rate decreasing effect of 30 mg/kg PEA appeared to be slightly greater following αMT and appeared to be attenuated at higher intensities following PCPA. However, neither trend was significantly different from the effect produced by 30 mg/kg PEA without pretreatment.

DISCUSSION

PEA did not exert an amphetamine-like effect on ICSS over the dose range in which it did exert an amphetamine-like effect on locomotor activity. Whereas d-A reliably increased self-stimulation rates at low current intensities, the only significant effects of PEA at any dose or any current intensity were rate-decreasing ones. These results replicate and extend observations reported previously [18].

In spite of the lack of effect of PEA alone on ICSS, Stein [18] concluded that PEA was amphetamine-like in its effect on ICSS because, following monoamine oxidase inhibition, PEA did increase self-stimulation rates. A similar conversion of the effects of PEA to amphetamine-like ones following monoamine oxidase inhibition was found in the drug discrimination paradigm [7]. In both of the above studies, the rationale for PEA becoming like amphetamine following monoamine oxidase inhibition was that the prevention of destruction of PEA by monoamine oxidase, the route by which 90% of PEA is metabolized [2], allowed PEA to remain in the brain long enough to exert its action. A number of problems exist with this interpretation. The first is that PEA is able to increase locomotor activity without pretreatment with a monoamine oxidase inhibitor. The second is that treatment with a monoamine oxidase inhibitor does more than prevent the breakdown of PEA; monoamine oxidase inhibition increases the levels of all amines in brain and alters the metabolism of all amines, including PEA.

The alteration of metabolism of PEA may be more important in converting its effects to amphetamine-like ones than the increased availability of the other amines. In the present study it was shown that PEA still exerted a rate-decreasing effect on self-stimulation following inhibition of synthesis of catecholamines and serotonin. In addition, the deaminated metabolites of PEA, the formation of which would be prevented by monoamine oxidase inhibition, have been shown to be behavioral depressants [6,9].

Thus the role of endogenous PEA in behavior remains to be clarified. Nevertheless, the qualitative difference be-

tween the actions of amphetamine and PEA on self-stimulation limit somewhat the conclusion that endogenous PEA is the important mediator of amphetamine's actions [1].

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