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Effects of Combinations of Phencyclidine and Pentobarbital on Schedule-Controlled Behavior in the Squirrel Monkey

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CHAIT, L. D. AND ROBERT L. BALSTER. *Effects of combinations of phencyclidine and pentobarbital on schedule-controlled behavior in the squirrel monkey.* PHARMAC. BIOCHEM. BEHAV. 9(2) 201–205, 1978.—Three squirrel monkeys trained on a variable interval schedule of food presentation were used to examine the interaction between phencyclidine (PCP) and pentobarbital (PB). First, dose-response curves for each drug given alone were obtained. PCP caused small response rate increases at low doses, and a dose-dependent decrease in responding at higher doses. PB caused only dose-dependent decreases in responding. The PB dose-response curve was then redetermined in the presence of four doses of PCP. Little support was found for the hypothesis that PCP enhances the depressant properties of PB. In fact, most dose combinations caused less disruption of responding than expected from simple addition of the effects of each drug given alone. These results are discussed in terms of species differences, measurement of different dependent variables and rate-dependency.

Phencyclidine Pentobarbital Schedule-controlled behavior Squirrel monkey Drug interactions

PHENCYCLIDINE (l-(1-phenylcyclohexyl) piperidine; PCP) is a drug with an unusually wide spectrum of central nervous system activity [8]. Originally developed as a general anesthetic, it has emerged as a major drug of abuse in recent years [1]. Although it is often classified as a hallucinogen, it does not characteristically produce visual hallucinations as do LSD and mescaline. Instead, it produces a dissociated state often with dramatic disturbances of body image, proprioception and affect [4, 5, 13].

Numerous deaths have been attributed recently to PCP use [2]. Although most of these fatalities are incidental to the behavioral toxicity caused by the drug, others are thought to represent true cases of pharmacological overdose. In these cases, the cause of death appears to be respiratory depression [2].

In view of the frequency of polydrug abuse in recent years [15], one possible explanation for some of these deaths could be an interaction of the effects of PCP with those of another drug. Likely candidates for such an interaction are ethanol and barbiturates. These agents are widely abused, and the dangerous interactions between CNS depressants are well-known. Observations in our own laboratory, where PCP is used routinely to aid in preparing rhesus monkeys for surgery, also suggest that PCP may enhance the effects of CNS depressants. Invariably, PCP pretreatment greatly reduces the amount of pentobarbital (PB) needed to induce and maintain a level of surgical anesthesia. This apparent synergistic effect has been noted by others [12].

Thus, it was decided to explore the possible interactions of PCP and pentobarbital in a systematic manner. Squirrel monkeys were chosen as subjects because of previous experience in our laboratory with PCP in this species [3]. An operant schedule of food presentation was used as the behavioral task because the behavior is easily quantifiable and because of the lack of published experiments systematically evaluating the effects of drug combinations on behavior maintained by schedules of food presentation. Specifically, a variable interval (VI) schedule [9] was chosen for this study because it is a simple schedule which typically generates stable, linear, intermediate rates of responding. An intermediate response rate was desirable in order to be able to show both increases and decreases in rate of responding as a result of drug treatments.

METHOD

Animals

Three male adult squirrel monkeys *(Saimiri sciureus)* were used initially in this experiment. They were experimentally- and drug-naive at the start of the study. Initial free-feeding weights ranged from 1038 to 1093 g. Animals were maintained at approximately 85% of free-feeding weight throughout the study. Diet consisted of Purina Monkey Chow and supplementary vitamin C. Animals were housed in individual cages in an isolated room with a 12 hr light-dark cycle. Water was continuously available in their home cages, but not during experimental sessions.

Apparatus

During experimental sessions animals were restrained about the waist in a Plexiglas primate chair, described in detail in a previous publication [3]. The chair was equipped with stimulus lights, a response lever and an automatic feeder which delivered 97 mg Noyes banana flavored food pellets into a brass food trough. For experimental sessions the chair was placed in a light- and sound-attenuating isolation cubicle. A fan provided both adequate ventilation and masking noise. Solid state programming equipment, counters and a cumulative recorder were located in an adjacent room.

Procedure

Training. Animals were initially trained to lever press on a schedule of continuous food presentation (Fixed Ratio 1). The schedule was then changed to a variable interval (VI) 15 sec. Over the next month the average interval was gradually lengthened to a VI 100 sec. The monkeys were maintained under this schedule for an additional month in order to stabilize baseline rates and to allow habituation to saline injections.

The duration of intervals was determined by a probability gate (BRS/LVE PP-201). Because the output of this gate varied from day to day, the average interval during a particular session varied from a minimum of 71 to a maximum of 129 sec, with a mean of approximately 100 sec. An upper limit of 4 min was imposed to prevent cessation of responding during unusually long intervals.

Experimental sessions. In order to determine onset of drug action, sessions began immediately after placing the chaired animal into the isolation cubicle or immediately after the injection on days when drug or saline was given. A white stimulus light was illuminated for the duration of the session, which was normally 2 hr. It was anticipated that drug duration of action could be measured in a 2 hr session.

Sessions were run in a 4-day cycle. The first day was a warm-up day on which data were collected but not analyzed. Data from the second day comprised the non-drug baseline data. On the third day drug or saline was given before the session. No session was held on the fourth day of the cycle. This sequence continued for the duration of the study (about 22 weeks). Monkeys were run in the same order, and at approximately the same time of day, each day a session was held. They received from 56 to 101 food pellets during an experimental session (unless their responding was disrupted by drug). The balance of their daily food intake (and vitamin supplement) was given in their home cages one hour after the end of the session.

Treatment regimens. After the training period, a dose-response curve was obtained from each animal for PCP. Doses were given in a mixed order, and each animal received each dose one time. Next, a dose-response curve for PB was obtained in the same manner. Finally, a series of combinations of the two drugs was given. Because of the possibility of fatal interactions, this series of combinations was generally given in an ascending order of PCP dose. Each animal received each combination one time. In addition, doses of PCP were given in combination with saline before being given in combination with PB to determine whether any change in sensitivity to PCP had occurred since obtaining the original dose-response curve.

Drugs

Phencyclidine hydrochloride (Sernylan, Bio-Ceutic Laboratories, Inc., St. Joseph, MO) was diluted with physiological saline to the appropriate concentrations to give an injection volume of 0.25 ml/kg. Sodium pentobarbital was diluted with 90% saline–10% propylene glycol to give the same injection volume. Pentobarbital solutions were made up fresh each day as needed. All doses are expressed as the appropriate salt. Injections were given IM into the thigh muscle immediately before start of the session. During the interaction part of the study one drug was injected into one leg, and the other drug (or saline) into the other leg.

Data Analysis

Because rates of responding tended to decrease toward the end of the 2 hr session, as well as become more variable, only data from the first hour of the session were used for analysis. Response rates (as responses per min) were determined from counters, while onset and duration of drug effect were measured from cumulative records. Response rates are expressed as percent of baseline; that is, (response rate during the first hour of a drug session/ response rate during the first hour of the preceding day's session) $\times$ 100. This was done to control for gradually shifting baseline rates.

Response rates after drug combinations were compared to those that would be expected if the effects of each drug when given alone (or in combination with saline) were simply additive. This is a model that has been used to characterize drug interactions [11].

RESULTS

Baseline Performance

Figure 1 shows typical baseline session cumulative records for each animal. Rates of responding were linear throughout the session, but usually decreased towards the end of the session. The reason for this rate decline was never established, but it did not seem to be due to either satiation or thirst. Overall rates of responding during the first hour of the session were fairly constant from day to day.

Table 1 shows mean baseline rates of responding for each animal during each of the three treatment regimens. Baseline rates of responding for animals J. G. and K. G. were stable both within and between treatment regimens. Animal M. H., however, showed considerable variability in his baseline rates of responding, especially within the PCP+PB treatment regimen, when his response rate dropped from a high of 50 resp/min to 3 resp/min. Due to this extreme instability, this animal was dropped from the study until his baseline rates of responding stabilized. Unfortunately, this monkey died before any useful data could be obtained. Therefore, only data from the remaining two animals are presented.

Effects of PCP and PB, Given Alone

Figure 2 shows the dose-response curve for PCP alone in each animal. Low doses (0.02–0.08 mg/kg) produced small increases in response rates. Higher doses produced dose-dependent decreases in rate of responding. Animal K. G. was more sensitive to doses of PCP greater than 0.23 mg/kg than was animal J. G. The onset of disruption of responding at the two highest doses (0.32 and 0.64 mg/kg) was abrupt and rapid, always occurring within the first 10 min, and usu-

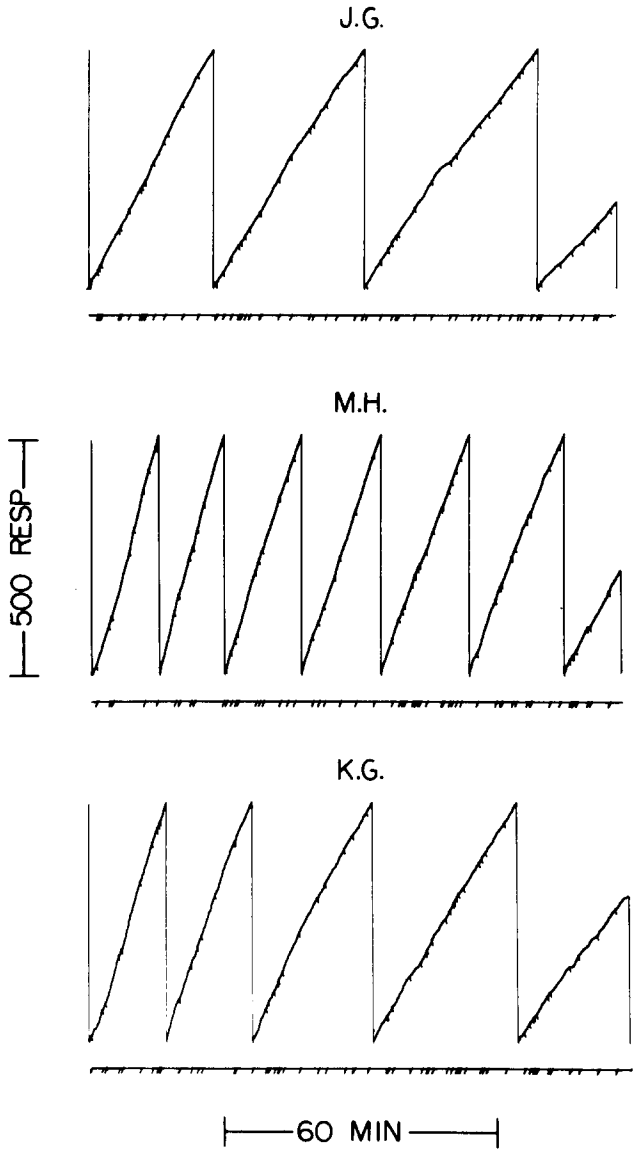

FIG. 1.Representative baseline session cumulative records for each animal. Diagonal slashes along the bottom pen tracing indicate that a food pellet would be delivered upon the next lever press. Diagonal slashes along the upper (response) pen tracing indicate delivery of a food pellet. The response pen reset after every 500 responses.

TABLE 1

MEAN BASELINE RESPONSE RATES IN INDIVIDUAL ANIMALS

Treatment	Subject J.G.	M.H.	K.G.
		resp/min ± SD	
PCP	15.9 ± 3.1	26.9 ± 3.1	25.1 ± 4.1
PB	21.7 ± 3.3	43.0 ± 4.7	23.3 ± 2.3
PCP + PB	20.8 ± 6.5	19.7 ± 13.5	30.2 ± 6.9

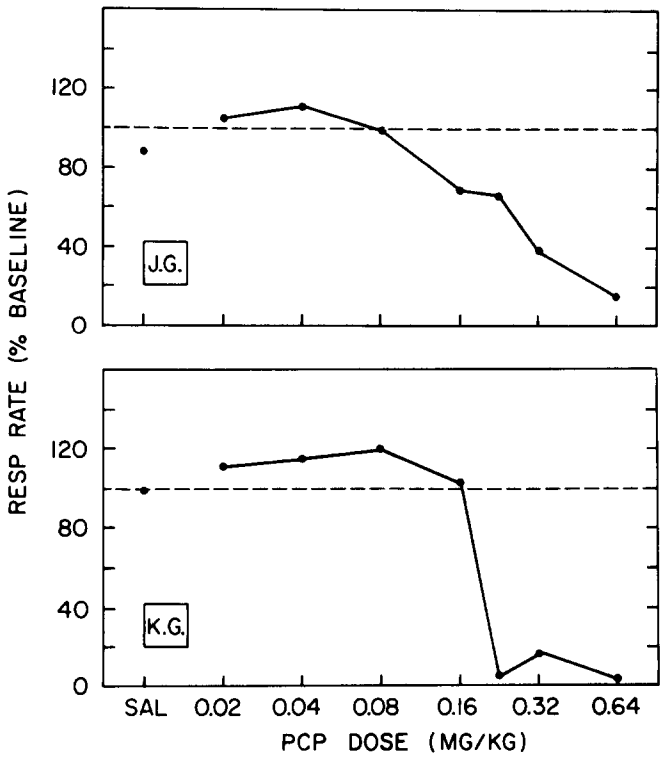

FIG. 2. Individual dose-response curves for PCP. Each point represents one determination. SAL refers to saline injection.

ally the first 5 min, of the session. Recovery was gradual and only partial by the end of the two hr session.

Individual dose-response curves for PB alone are displayed in Fig. 3. PB produced a dose-related decrease in rate of responding; low doses did not increase response rates. At 8.0 and 16.0 mg/kg PB, the mean onset of response disruption was 13.5 and 4.7 min, respectively. Unlike the case with PCP, recovery was almost immediate with PB. At 8.0 mg/kg the mean recovery time was 67.3 min. Recovery did not occur within the two hr session at 16.0 mg/kg.

The period of disruption under PB was characterized by almost complete suppression of responding. Observation of the animals at this time showed them to be collapsed motionless in a corner of the chair, but they could be aroused by stimulation. The sudden recovery of responding which occurred at 8.0 mg/kg no doubt was due to recovery of consciousness. This contrasts with the effects of the maximally disruptive doses of PCP (0.32 and 0.64 mg/kg). These doses did not produce complete suppression of responding, and observation of the monkeys showed them to be alert, flailing around and engaging in repetitive behavior, occasionally accidentally hitting the response lever, but not eating the food pellet delivered.

Effects of PCP and PB, Given in Combination

The results of the PCP-PB combination regimen in each animal are shown in Fig. 4. The left-hand sets of bars indicate that the effect of PCP when combined with saline was not consistently different from the effect of PCP when given alone during the initial dose-response determination (Fig. 2).

Response rates for combinations of PCP and PB are

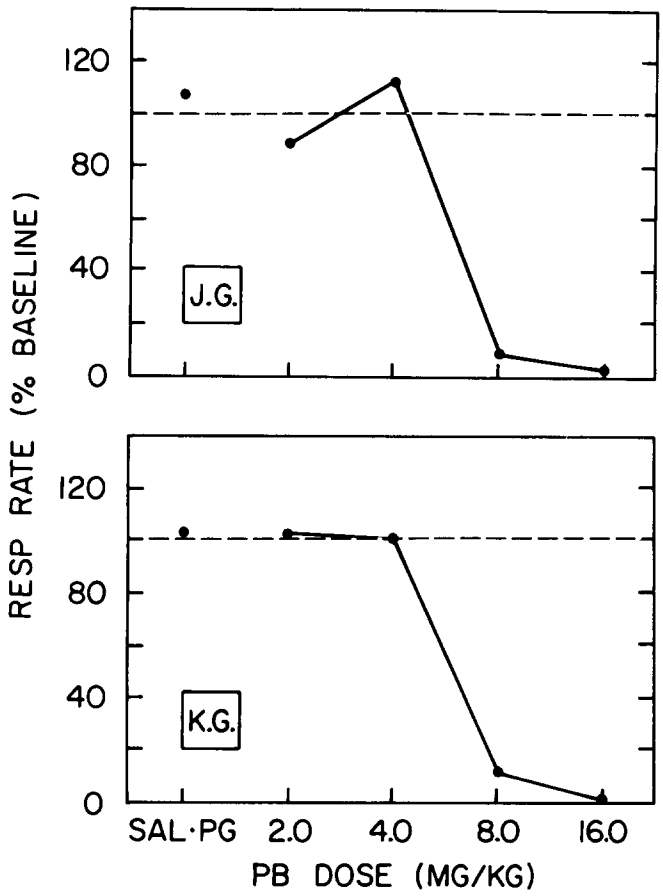

FIG. 3. Individual dose-response curves for PB. Each point represents one determination. SAL-PG refers to vehicle injection, 90% saline—10% propylene glycol.

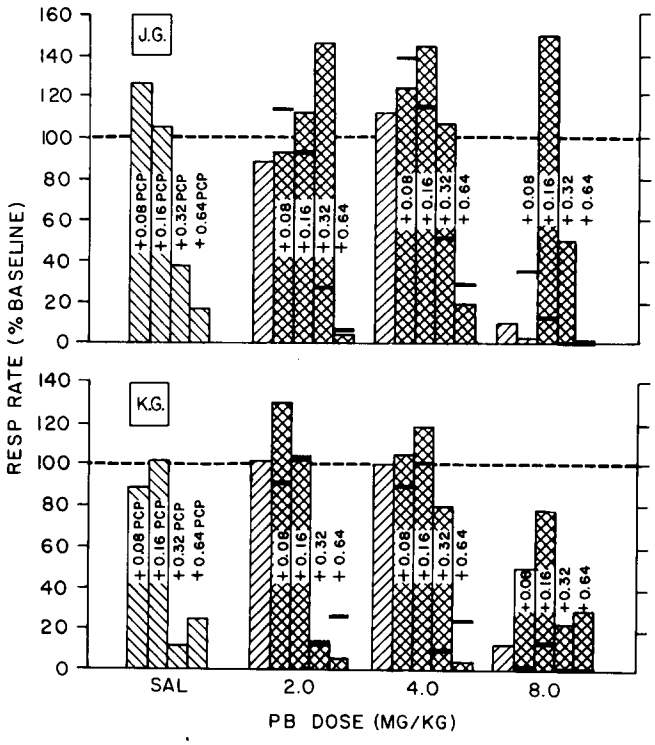

FIG. 4. Effects of PCP on the PB dose-response curve in individual animals. Each vertical bar represents one determination of that dose or dose combination. The left-hand set of bars shows the effects of PCP given in combination with saline. Cross-hatched bars show the effects of PCP-PB dose combinatins. Solid horizontal lines at each combination represent the rates of responding expected if the effects of PB alone and PCP + SAL were additive. Unlabelled bars represent the effect of PB alone during the initial dose-response determination.

shown by cross-hatched bars in Fig. 4. In addition, the effects of PB alone (unlabelled bars) from the initial dose-response determination are shown for comparison. In animal J. G., 0.16 mg/kg and 0.32 mg/kg PCP consistently produced response rates greater than additive (solid horizontal lines) when combined with PB. 0.08 mg/kg PCP, a no-effect or rate-increasing dose in this animal, consistently produced response rates less than additive when combined with PB. The highest dose of PCP tested (0.64 mg/kg), when combined with PB, resulted in little deviation from a purely additive effect.

In the other animal, all doses of PCP except 0.64 mg/kg yielded rates of responding equal to or greater than additive when combined with PB. The results with 0.64 mg/kg were inconsistent in this animal.

DISCUSSION

The dose-response curve obtained for PCP is very similar to that obtained in squirrel monkeys responding on a chain fixed-interval fixed-ratio schedule of food presentation [3]. Small increases in response rate were observed at low doses (0.02–0.08 mg/kg), but these increases were not seen consistently in the same animal when a dose was repeated.

Low doses of barbiturates have been demonstrated to increase response rates under many different schedules of reinforcement [14], including VI schedules. No dose of PB increased rates of responding in the present study. The reason for this is unknown, but species differences do not appear to be a factor, since Hanson *et al.* [10] obtained increases in rates of responding after low doses of PB in squirrel monkeys responding for food on a VI component of a multiple schedule of reinforcement.

The results of the combinations of PCP and PB were unexpected. In view of our observations in rhesus monkeys that low doses of PCP greatly enhance the depressant effect of PB, it was anticipated that low doses of PCP would enhance the disruption of responding produced by PB in the present study. In fact, except for one dose of PCP (0.08 mg/kg) in animal J. G., the results obtained suggest that just the opposite is true. Out of the 12 combinations of PCP and PB, seven combinations in one animal and 10 combinations in the other yielded rates of responding greater than expected based on additive effects. Some of these response rate increases over additivity were large (over 100% baseline), whereas in no case were negative deviations from additivity greater than 33% baseline response rate.

One possible explanation for this infraadditive effect of PCP-PB combinations is discussed by Dews [7]. He suggests that many cases of apparent drug antagonism on operant

baselines could be explained on the basis of the rate-dependent effect of drugs, and do not represent examples of true physiological antagonism. It has been shown for many CNS active drugs, including PCP [3,16] and barbiturates [6] that the same dose can lower response rates when they are high or raise them when they are low; that is, the effect of the drug is dependent on the baseline rate of responding. For example, if a dose of drug A which by itself lowers response rates is given along with a dose of drug B which also lowers response rates, the combination might cause less of a decrease in rate of responding than expected because each drug is acting upon a baseline rate that has been lowered by the other drug. This effect would result in what appears to be drug antagonism. The role that rate-dependency may have played in the present study cannot be determined because the VI schedule of food presentation generated a range of response rates too narrow for rate-dependency analysis to be performed. Ideally, any study examining the effects of drug combinations on rates of responding should be designed to allow evaluation of this possible component of drug antagonism.

Besides rate-dependency, there are two other possible explanations for the results of the present study. One is that the effect of PCP-PB combinations is species-dependent. Evidence in support of this hypothesis has been obtained recently in our laboratory (submitted for publication). An observational rating scale of CNS depression was employed to compare the effects of a PCP-PB combination in rhesus and squirrel monkeys. In three rhesus monkeys, a no-effect dose of PCP (0.20 mg/kg IM) combined with a moderately depressant dose of PB (25.0 mg/kg IM) produced surgical anesthesia of over 1 hr duration in all animals. In contrast, 0.16 mg/kg PCP (IM) failed to enhance the depressant effect of 12.5 mg/kg PB (IM) in four squirrel monkeys, despite the fact that these two doses alone had a greater effect on the rating scale than the doses used in the rhesus monkeys.

This demonstration that the squirrel monkey is less sensitive to the enhancing effect of PCP on PB-induced CNS depression may explain, in part or in full, the results of the present study. However, the remaining possibility is that the behavioral measure employed in the present study (rate of lever pressing for food presentation) was not a suitable measure for detecting a drug interaction which manifests itself in other forms of behavior. This possibility could be effectively evaluated by performing in rhesus monkeys an experiment similar to the present one.

It is reasonable to assume that the nature of interaction which a specific drug combination exhibits would be dependent on the specific dependent variable being measured. For this reason, and because of the species difference shown in the study cited above, the results of the present study obviously cannot rule out the possibility of dangerous interactions between PCP and CNS depressants in humans.

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