Comparative Pharmacodynamic Activity of Single and Divided Doses of Benzphetamine Hydrochloride

CARL A. SCHLAGEL and ELMER C. SANBORN

Abstract □ A battery of pharmacologic methods was employed to determine in man the relative degree and time-course of activity of benzphetamine given as a single, 75-mg. dose, the same total dose of drug given hourly in 15-mg. portions, and placebo. The objectives were to provide guidelines for formulation research and to help ascertain the clinical potential for a sustained-release preparation. The end points for determining central and peripheral activity before treatment and for 12 hr. after medication included the mean integrated voltages of the EEG at various frequencies and several cardiovascular, respiratory, autonomic, and psychometric tests. While the pattern of response-time plots varied with different endpoints, the general trend and statistically significant results showed a more peaked and marked profile of action after the single-drug dose, with the onset of action and profile of the divided-dose regimen being more gradual and intermediate in degree, respectively, between that of the single dose and placebo. The data indicate the utility of the methodology for evaluating dosage forms of the benzphetamine-type drugs and suggest good therapeutic potential for sustained-action preparations.

Keyphrases □ Benzphetamine HCl activity—single, divided dose effect □ Pharmacological response, comparative—time profile □ EEG—CNS effect, benzphetamine HCl □ Psychometric testing—benzphetamine HCl effect □ Bioassay, clinical—benzphetamine HCl activity

A major problem encountered in the design of a sustained-release formulation of benzphetamine hydrochloride,[1] an oral anorexigenic agent, was the availability of adequate *in vivo* procedures for determining which preparations possessed the greatest potential for producing the desired pattern of drug availability and the optimal therapeutic activity. In an initial study using radio-labeled drug to provide data on absorption, metabolism, and excretion, the results provided informative but confounding information because of the questionable activity and potency of a number of metabolites. The present report describes the results obtained using a variety of pharmacologic methods to compare the time-course of activity of benzphetamine hydrochloride[2] when administered as a large single dose and at the same total dose on a divided schedule, the latter to simulate a sustained-release preparation. A placebo was employed as a control.

EXPERIMENTAL

Study Design—The study was conducted as a double-blind, three-way crossover in 24 male volunteers of age 21 to 45 years. They were judged healthy based on physical examinations and clinical laboratory evaluations. The subjects were divided into four groups of six volunteers. Each group received medication and underwent pharmacologic testing 1 day per week over a 3-week period. The three medications given once each to the subjects were: (*a*) a 75-mg. tablet of benzphetamine (SD[3]) given at 0 hr.; (*b*) five 15-mg. tablets of benzphetamine (DD[4]) given as one tablet hourly at 0, 1, 2, 3, and 4 hr.; and (*c*) placebo (P[5]). Extra placebo tablets were given so the three treatments consisted of two tablets at 0, 1, 2, 3, and 4 hr. The three treatments were administered according to eight Latin Squares using two different Latin Squares per group of six subjects as indicated:

Subjects	Week 1	Week 2	Week 3
1	SD	DD	P
2	DD	P	SD
3	P	SD	DD
4	SD	P	DD
5	DD	SD	P
6	P	DD	SD

Test Procedures—A battery of pharmacological tests was performed on each subject prior to the study as an acclimation trial, just prior to medication for control values (0 hr.) and at 1, 2, 3, 4, 6, 8, 10, and 12 hr. after the initial dose of medication. The procedures, listed in the order applied, were: (*a*) a 10-min. electroencephalogram (EEG) from a monopolar left occipital lead using the ears as reference and the left clavicle region as ground; (*b*) systolic and diastolic blood pressure (indirect); (*c*) pulse; (*d*) respiratory rate; (*e*) oral temperature; (*f*) pupil diameter (Polaroid photograph and caliper measurement); (*g*) 1-min. finger-tapping rate (telegraph key with mechanical counter); (*h*) a 5-min. digit-letter coding task as reported by Smith (1); and (*i*) a 48-item check list of the authors' design using six adjectives and six antonyms per category to obtain subjective profiles relative to states of arousal, nervousness, mood, and general feeling.

The subjects were recumbent for the first five tests, with their eyes closed for the EEG.

The EEG's were recorded over 12-min. periods to provide 10 min. of data after editing for artifacts. The signals were recorded graphically on a Type R Dynograph,[6] using a 0.3 time constant, and simultaneously on analog magnetic tape.[7] The taped signals were filtered by low band and high pass filters[8] to separate the EEG into 0–8, 8–13 and 13–30 c.p.s. frequency components. The output from these filters as well as the nonfiltered EEG were integrated electronically,[9] while the digital values representing the integrated voltages cumulated over 20 sec. EEG epochs were recorded with an IBM 026 card punch. Thus, for a 10-min. EEG sample, thirty 20-sec. values were obtained for each of the three frequency ranges and for the total or nonfiltered EEG records.

The basic principles for this EEG integrating approach were similar to those reported by Drohocki (2). The utility of the selection of 20-sec. EEG epochs and 10-min. recordings has been validated by Goldstein (3).

In this study the mean integrated EEG voltages were obtained by calibration of the six integrators to give 75 pulses or counts/20 sec. using a 10-c.p.s. sine wave having an amplitude of 50 μv. peak to peak.

Study Restrictions—The subjects received the initial dose of medication 2 hr. after a light breakfast. They then fasted except for 6 oz. of milk and a candy bar or apple at the 7th, 9th, and 11th hr. of testing. Sanka but no caffeinated beverages were permitted.

Data Analyses—The results from 2 Latin Squares per subject group giving a total of 8 Latin Squares were combined for a Latin Square analysis (4) which accounted for week, subject, and square variability in testing for treatment effects. When statistically significant differences occurred among the three treatments, the results were evaluated by Duncan's Multiple Range test (5) to determine

[1] Didrex, The Upjohn Company, Kalamazoo, Mich.
[2] Referred to herein as benzphetamine.
[3] SD = single dose of benzphetamine.
[4] DD = divided dose of benzphetamine.
[5] P = placebo.
[6] Beckman Instruments, Inc., Spinco Div., Lincolnwood, IL 60649
[7] Honeywell model 8100, Honeywell, Denver, CO 80217
[8] Designed and constructed by C. Alway, The Upjohn Co., Kalamazoo, MI 49001
[9] *Ibid.*

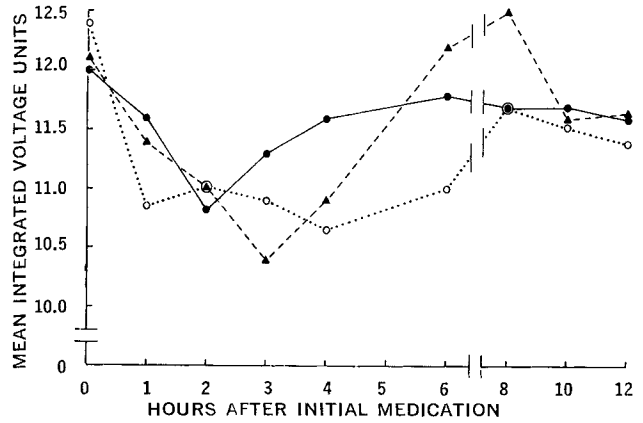

Figure 1—*Response-time curves for 0–8 c.p.s. EEG integrated voltage activity. Key: ●—●, SD; O···O, DD; ▲- - -▲, P. Statistically significant changes: none.*

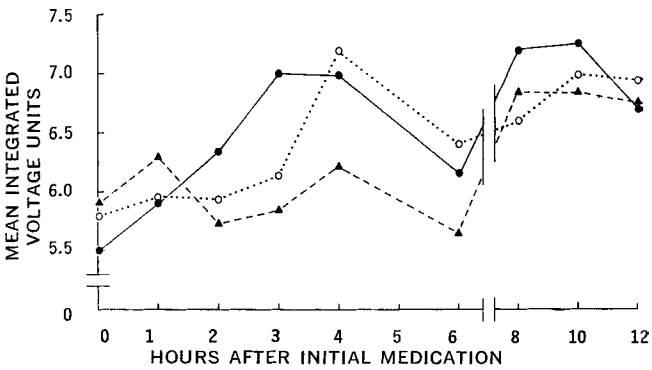

Figure 3—*Response-time curves for 13–30 c.p.s. EEG integrated voltage activity. Key: ●—●, SD; O···O, DD; ▲- - -▲, P. Statistically significant changes: SD > P at 3 and 6 hr.; SD > DD at 3 hr.; DD > P at 6 hr.*

significant differences between treatment means at the 0.05 level or beyond.

The integrated EEG data were analyzed without any form of transformation. The coefficients of variation (CV) of the EEG data were determined using the log of the standard deviation of the log of the original value. This yielded values which could be evaluated statistically and which compared to the 4th decimal place with the CV as calculated in the standard manner.

RESULTS AND DISCUSSION

All of the test methods yielded results showing statistically significant differences among the treatments consisting of a large single dose of benzphetamine (SD), the same total drug dose given in divided portions (DD), and placebo (P). The response-time patterns reflecting these differences revealed an earlier onset and generally more pronounced action after the SD, with the DD regimen resulting in a delayed and gradually increasing response that through 6 hr. after medication was intermediate in slope between that of the SD and the P treatments. Thus, with the EEG and digit-letter coding procedures, the significant changes after the SD occurred from 2–6 hr. after drug, while after the DD the activity reached significance only at 6 hr. With systolic BP, pulse, temperature, and the finger-tapping test, both drug regimens produced significant effects which persisted for 10–12 hr. post-treatment. Only the SD of benzphetamine had significant effects on diastolic BP, respiration, and pupil diameter. These response-time pattern variations with different end points implicate clearly the utility and need for using multiple testing procedures in comparing the activity of different dosage forms of the same drug.

Electroencephalograms—The action of benzphetamine on the EEG was characterized by a lack of significant changes in the mean integrated voltages of the 0–8 c.p.s. component, by the highest and most frequently significant mean integrated voltage increases in the 8–13 c.p.s. band, and by fewer and smaller yet significant mean integrated voltage increases in the 13–30 c.p.s. spectrum. The voltages of the total (0–30 c.p.s.) or nonfiltered EEG were also increased by benzphetamine in a pattern quite similar to that of the 8–13 c.p.s. band. Finally, the coefficients of variation (CV), reflecting variability in the EEG waveforms, were reduced significantly at all frequencies by drug treatment. These results are shown in Figs. 1–4.

An examination of these plots as well as the legends indicating the time periods when significant differences occurred between treatments, revealed that the drug relative to placebo EEG effects were greater for the SD than for the DD, and that the magnitude of the drug-induced changes were greater for the 8–13 c.p.s. or alpha component than for the 13–30 c.p.s. or beta segment of the EEG.

This action of benzphetamine on the EEG is interpreted as a mild CNS stimulant effect as determined by: (*a*) the absence of any significant increases in 0–8 c.p.s. activity indicating that the drug is devoid of primary CNS sedative or depressant properties; (*b*) the appreciable increase in alpha or 8–13 c.p.s. energy content reflecting maintenance of alertness despite acquisition of the EEG with the subjects recumbent, with eyes closed, and in a noise-free environment; and (*c*) the increased beta or 13–30 c.p.s. activity which indicates EEG "desychronization" and which reflects CNS hyperactivity or stimulation (6–8). The drug engendered reduction in the CV of the EEG is also indicative of the activity profile of CNS stimulants (9).

The benzphetamine-induced increase in total or nonfiltered EEG mean energy content is at variance with the decrease in this activity as reported by Goldstein *et al.* (7, 9) for 15 mg. p.o. of the related drug, *d*-amphetamine.

No good explanation is available for this difference using basically similar EEG methodologies. However, the disparity is not dependent on differences in drug action for the authors have observed in two recent studies that 15 mg. p.o. of *d*-amphetamine produces

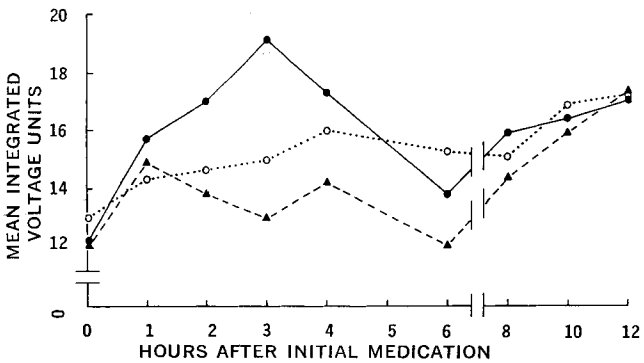

Figure 2—*Response-time curves for 8–13 c.p.s. EEG integrated voltage activity. Key: ●—●, SD; O····O, DD; ▲- - -▲, P. Statistically significant changes: SD > P at 2, 3, 4, and 6 hr.; SD > DD at 2 and 3 hr.; DD > P at 6 hr.*

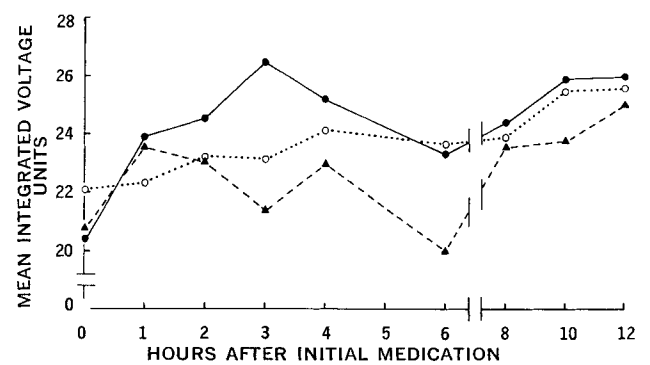

Figure 4—*Response-time curves for total or 0–30 c.p.s. integrated EEG voltage activity. Key: ●—●, SD; O···O, DD; ▲- - -▲, P. Statistically significantly changes: SD > P at 3, 4, and 6 hr.; SD > DD at 3 hr.; DD > P at 6 hr.*

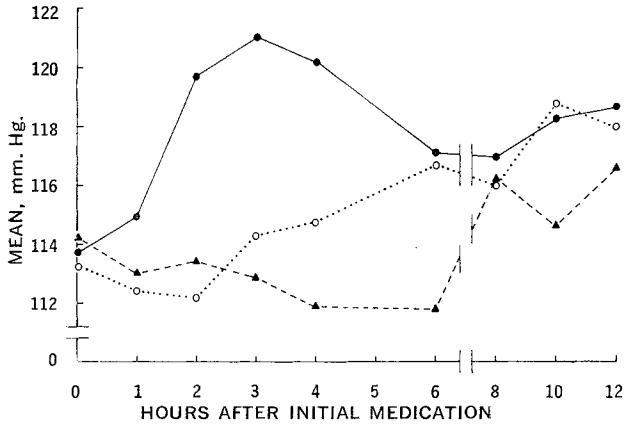

Figure 5—*Response-time plots of systolic blood pressure. Key: ●—●, SD; ○···○, DD; ▲---▲, P. Statistically significant changes: SD > P at 2, 3, 4, 6, and 10 hr.; SD > DD at 2, 3, and 4 hr.; DD > P at 6 and 10 hr.*

an EEG activity profile analogous in all respects to that of the SD of benzphetamine used in this study.

Several theoretical reasons for the disparity are that Goldstein studied subjects individually with the EEG alone while the authors utilized subject groups and a battery of test procedures, and Goldstein used fewer subjects and a very short post-drug observation period.

Systolic Blood Pressure—The major blood pressure elevations after benzphetamine as shown in Fig. 5 followed the same general pattern as the EEG alpha activity inasmuch as the elevations after the SD were significant from 2–6 hr. after drug, while the effect after the DD was delayed, more gradual, and reached a significantly elevated level only by the 6th hr. An exception was that both drug treatments also elevated the pressure significantly at 10 hr. postmedication.

The increased systolic pressure following the SD of drug was more pronounced and peaked, relative to the response to the DD, than were the comparative responses of any of the other endpoints. This provides a logical basis for the use of a divided dose schedule or a properly formulated, sustained-release preparation of benzphetamine to reduce sequelae from abrupt blood pressure changes.

The diastolic blood pressure displayed an upward trend throughout the 12-hour period following all three treatments. The SD of benzphetamine caused a slight peaking effect which at the summit at 4 hr. was significantly higher than that of both the SD and P medication.

Pulse—The data on the pulse depicted in Fig. 6 indicate the reduction in rate for the first 6 hr. following P treatment. This trend was reversed early by the SD of benzphetamine which maintained a significantly higher pulse, relative to P, from the 2nd through the 6th hr. This reversal was delayed following the DD of drug so that significant differences were manifested at the 4th and 6th hr. The action of both drug treatments was evident and significantly different from P after 10 hr.

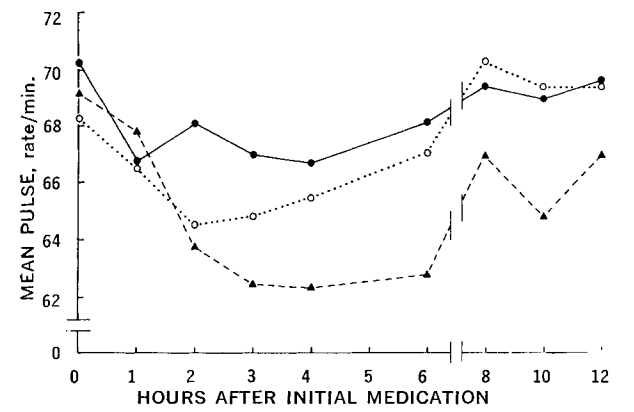

Figure 6—*Pulse change with time. Key: ●—●, SD; ○···○, DD; ▲---▲, P. Statistically significant changes: SD > P at 2, 3, 4, 6, and 10 hr.; SD > DD at 2 hr.; DD > P at 4, 6, and 10 hr.*

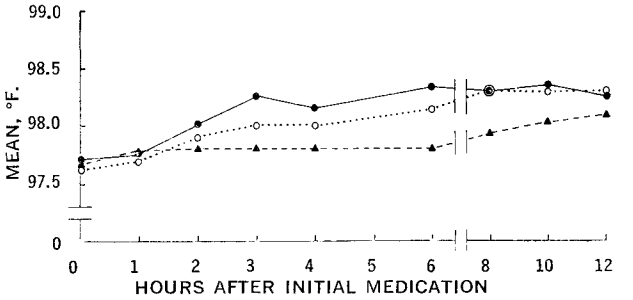

Figure 7—*Response-time changes in oral temperature. Key: ●—●, SD; ○···○, DD; ▲---▲, P. Statistically significant changes: SD > P at 2, 3, 4, 6, 8, and 10 hr.; SD > DD at 3, 4, and 6 hr.; DD > P at 1–12 hr.*

Oral Temperature—The generally smoother character of the response-time curves for oral temperature renders this end point quite valuable for delineating clearly the comparative action profiles for the treatments under study. The early elevation of the temperature by the SD as well as the early but more gradual hyperthermic action of the DD of drug are quite apparent from the plots in Fig. 7. Both drug treatments produced persistent effects with the hyperthemia being significant, relative to P, even at the 12th hour of testing.

Respiratory Rate—The fluctuations in the respiratory rates demonstrated in Fig. 8 contrast dramatically with the stability of the oral temperature measurements shown previously. Although the response of the SD of drug was significantly higher than that of the DD at 2 hr. and than that of the P at 2, 4, and 6 hr., the generally elevated rate produced by the DD was never significantly greater than the P effect.

Pupil Diameter—The rather feeble mydriatic action of oral sympathomimetic benzphetamine, was sufficiently adequate after the SD to stabilize this normally fluctuating endpoint and to provide significant differences relative to the DD of drug at 3 and 4 hr. and to the P at 3, 8, and 10 hr. The responses to the simulated sustained-release regimen of benzphetamine were never significantly different from those of the P treatment. These data are shown in Fig. 9.

Finger-Tapping Rate—The activity of CNS stimulating drugs in this psychomotor procedure is reflected in the production and maintenance of the maximal rate possible for the test subjects. Placebo responses are generally slower and more undulating depending on the relative mood, diligence, and perseverance of the subjects at each testing session. In this study the psychomotor stimulating action of the SD of benzphetamine shown in Fig. 10 was evidenced by the acquisition and upholding of a maximal rate of about 340 taps per min. The higher initial tapping rate observed prior to medication with the SD relates to initial adeptness and subject testing order and is of little consequence when the maximal rate is attained. The gradually increasing rate observed after the DD of drug was typical of the delayed response seen in the other test procedures following this treatment. Both drug schedules displayed prolonged activity with significantly higher tapping rates than P during the 6–12 hr. period of testing.

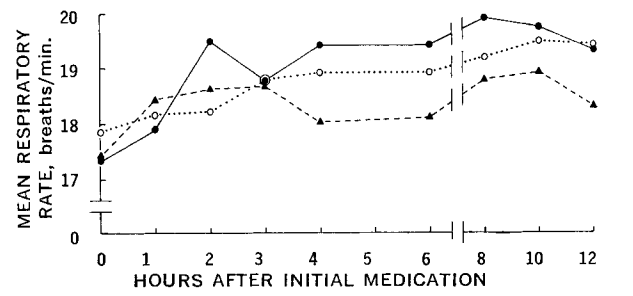

Figure 8—*Respiratory rate changes over time. Key: ●—●, SD; ○···○, DD; ▲---▲, P. Statistically significant changes: SD > P at 2, 4, and 6 hr.*

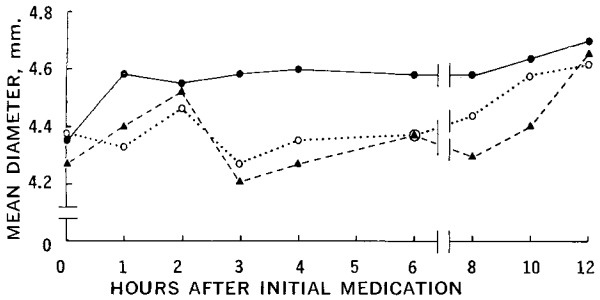

Figure 9—*Pupil diameter changes. Key:* ●—●, *SD;* O···O, *DD;* ▲- - -▲, *P. Statistically significant changes: SD > P at 3, 8, and 10 hr.; SD > DD at 3 and 4 hr.*

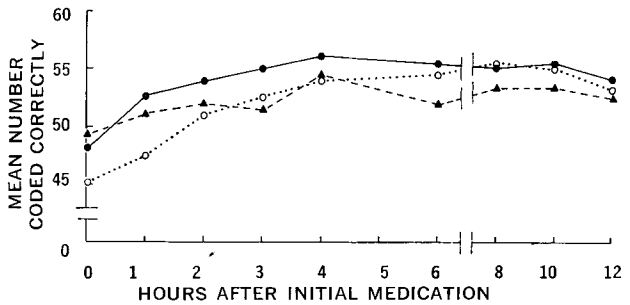

Figure 11—*Digit-letter coding proficiency. Key:* ●—●, *SD;* O···O, *DD;* ▲- - -▲, *P. Statistically significant changes: SD > P at 3 and 6 hr.; SD > DD at 1 hr.; SD > P at 6 hr.; P > DD at 0 and 1 hr.*

Digit-Letter Coding Task—CNS stimulant action as determined in this confusing, monotonous test is also based on maintenance of a high, relatively stable plateau of coding speed. Coding accuracy is not improved under these specific test conditions.

The data in Fig. 11 demonstrate, though not dramatically, the better subject performance when on the SD of drug than when on P. The steady improvement after the DD of benzphetamine depicts again the delayed and slowly increasing effect of the multiple, small-dose regimen of drug. These results are in agreement with those obtained by Smith (1) and Weitzner (10) for *d*-amphetamine.

The increased performances and the response-time patterns obtained in both the tapping and coding tasks are also in accord with the EEG effects indicating drug-induced increases in alertness and mental acuity. These psychomotor task-EEG correlations have been strengthened by the results from a more recent study with benzphetamine and *d*-amphetamine wherein the sensitivity of the tapping and coding tasks has been improved appreciably by increasing the duration of tapping from 60–90 sec. and the coding from 5–10 min. This reduced the impact on the data of the subject's initial speed resulting from the self-mustered vigilance and determination which wanes sooner after placebo than after CNS stimulant drugs.

Adjective Check List—The patterns of the response-time plots comparing the SD of benzphetamine and the P treatments were all in the direction of drug-induced increases in the levels of arousal, mood, calmness, and feeling of well-being. The curves for the DD of drug were again intermediate in position between those of the SD and of the P medication. The action of the SD lasted only for 6 hr., at which time the responses to the two drug regimens were equal. During the 6–12 hr. interval, the response to the DD was maintained relative to those of the SD and P treatments.

These data are not shown because of the number of figures involved and because variations in the control (0 hr.) scores reduced the relevance of many of the significant response differences. The definite trends remained, however, and reflected CNS stimulant-type drug action. It should be noted that the increased calmness is typical of the initial effect of acute doses of the amphetamine-type stimulants. Nervousness is a secondary, rebound phenomenon.

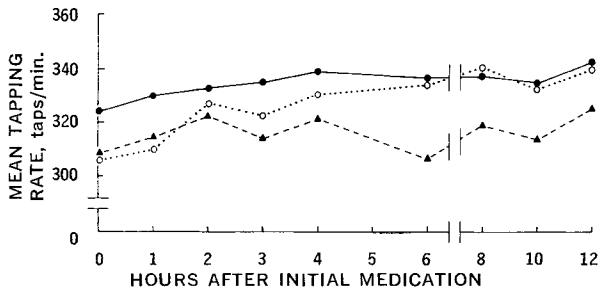

Figure 10—*Finger tapping rate versus time. Key:* ●—●, *SD;* O···O, *DD;* ▲- - -▲, *P. Statistically significant changes: SD > P at 0, 3, 4, 6, 8, 10, and 12 hr.; SD > DD at 0 hr.; DD > P at 6, 8, 10, and 12 hr.*

CONCLUSIONS

The data indicate that the methodology employed is capable of distinguishing comparatively the response-time characteristics of different formulations of acutely administered benzphetamine.

The differences in response-time patterns and durations of activity obtained with different end points reflect the value of multiple methods for delineating more completely drug action profiles.

The simulated sustained-release regimen of benzphetamine generally produced more delayed, more gradual, and less pronounced responses than the large dose of drug. This suggests that a properly formulated, sustained-release formulation may have virtue by producing more blunted and smoother pharmacologic responses. The relative therapeutic (anorexigenic) advantages of different benzphetamine preparations must still be determined under use conditions.

REFERENCES

(1) G. M. Smith, M. Weitzner, S. R. Levenson, and H. K. Beecher, *J. Pharmacol. Exptl. Therap.*, **141,** 100(1963).

(2) Z. Drohocki, *Rev. Neurol.*, **94,** 804(1956).

(3) L. Goldstein and R. A. Beck, *Intern. Rev. Neurobiol.*, **8,** 265(1965).

(4) W. S. Cochran and G. M. Cox, "Experimental Designs," 2nd ed., Wiley, New York, N. Y., 1957, pp. 117–130.

(5) R. G. D. Steel and J. H. Torrie, "Principles and Procedures of Statistics," McGraw-Hill, New York, N. Y., 1960, pp. 107–109.

(6) D. B. Lindsey, "Handbook of Physiology," sect. 1, "Neurophysiology," vol. 3, Am. Physiological Soc., Washington, D. C., 1960, pp. 1575–1577.

(7) D. B. Tyler, J. Goodman, and T. Rothman, *Am. J. Physiol.*, **149,** 185(1947).

(8) J. E. P. Toman and J. P. Davis, *Pharmacol. Rev.*, **1,** 425 (1949).

(9) L. Goldstein, H. B. Murphree, and C. C. Pfeiffer, *Ann. N. Y. Acad. Sci.*, **107,** 1045(1963).

(10) M. Weitzner, *J. Psychol.*, **60,** 71(1965).

ACKNOWLEDGMENTS AND ADDRESSES

Received May 15, 1969 from the *Experimental Medicine Unit, The Upjohn Company, Kalamazoo, MI 49001*

Accepted for publication August 8, 1969.

The authors wish to thank Miss M. L. Henninger of The Upjohn Co. for the statistical analyses. We are also grateful to C. D. Alway and L. Jones of The Upjohn Co. for the design and construction of the instrumentation for EEG integration and frequency analysis. The generous advice given by L. Goldstein of The N. J. Neuropsychiatric Institute, Princeton, N. J., on his analytical EEG approach is also appreciated.