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Amphetamine-Induced Taste Aversion Demonstrated With Operant Behaviour

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STOLERMAN, I. P. AND G. D. D'MELLO. *Amphetamine-induced taste aversion demonstrated with operant behaviour.* PHARMAC. BIOCHEM. BEHAV. 8(2) 107–111, 1978. – Amphetamine can be used to condition strong taste aversions, but little is known about the possible effects of flavour-amphetamine pairings on operant behaviour. Rats were trained to press bars for water reinforcers delivered after every 40 responses (FR 40). Flavoured reinforcers were then substituted for the water and post-session injections of amphetamine (1 mg/kg) were given. Even a single flavour-amphetamine pairing produced some decrement in responding for that flavour, whereas three flavour-amphetamine pairings almost completely suppressed responding. In the same rats, a flavour which was paired with saline injections did not suppress responding. Flavour-amphetamine pairings can therefore have a powerful influence on operant behaviour and the different outcomes of flavour-conditioning and self-administration procedures cannot be attributed simply to the type of response required from the rat.

Amphetamine Conditioned taste aversion Operant behaviour Fixed-ratio schedules

RATS refused to drink distinctively flavoured solutions when their consumption on previous occasions was followed by injections of amphetamine [2, 3, 6, 10, 19]. This aversive property of amphetamine contrasts with its positively reinforcing effect in studies of drug self-administration in rats [8,22], and a similar situation has arisen with several other psychoactive drugs [5,28]. These different effects have been explained by reference to the widely accepted generalisation that drugs can indeed have different effects in different circumstances [5], but if this account is to have predictive power, it is necessary to identify the factor or factors which determine the nature of the drug effect.

One of the many variables which may be relevant is the type of response required from the rat; conditioned taste aversion (CTA) involves drinking but self-administration typically requires bar-pressing. Seligman and Hager are among those who have argued that different classes of response are conditioned most rapidly by different classes of reinforcing or aversive consequences; it was suggested [23] that flavour-drug pairings would exert less effect on an arbitrary operant such as bar-pressing than on a naturalistic consummatory response such as drinking. After pairing saccharin solutions with injections of lithium, Holman [15] concluded that an association between a flavour and sickness did not influence bar-pressing in extinction. Other workers have reported that CTA induced with lithium [20] or apomorphine [1] can influence bar-pressing for flavoured reinforcers. There seems to be little further evidence available to suggest that flavours are inherently unable to influence operant behaviour in ways resembling the more frequently studied auditory or visual stimuli. For example, discriminative reactions to flavours can be shown when sweetened food pellets serve as discriminative stimuli in multiple fixed-ratio fixed-interval schedules [7]. However, despite the intensive study of CTA in recent years, very little is known about the effects of flavour-drug pairings on operant behaviour.

In the present experiments, the effects of flavour-amphetamine pairings were examined on bar-pressing for liquid reinforcement delivered on a fixed-ratio schedule. Many aspects of the procedure (e.g. flavours, drug dose, session length) were similar to those in our earlier investigations of CTA induced with amphetamine [2,10] but in this case, the drug was administered after sessions of operant responding rather than after periods of drinking. Different effects of flavour-amphetamine pairings on bar-pressing and on drinking would support distinctions between the classes of responses of which these are representative [23], and would indicate a factor which might be responsible in whole or part for the contrasting effects of amphetamine in different circumstances.

METHOD

Animals

Four female, hooded rats weighing 150–200 g and bred in the Department of Psychology, University of Birmingham, served as the subjects of the experiment. The rats were housed individually in a room maintained at about 22°C and a regular light-dark cycle was imposed by electric lighting (light from 08.00–20.00 hr). Tap water was made available in the home cages for 1 hr each day throughout the experiment.

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Drugs

(+)-Amphetamine sulphate (Smith, Kline and French) was dissolved in isotonic saline and was injected intraperitoneally at a dose of 1 mg/kg in a volume of 1 ml/kg.

Solutions with synthetic chicken and lemon flavours were adapted from Lovett and Booth [18]. Chicken flavour consisted of monosodium glutamate (12.5 mM) and sodium chloride (128 mM) dissolved in distilled water, whereas lemon flavour consisted of citric acid (1 mM) and sodium saccharin (2 mM) dissolved in distilled water.

Procedure

The rats were first trained to press a bar for water reinforcement in a standard test chamber within a sound-insulated, ventilated enclosure (Campden Instruments). The number of responses required for reinforcement was raised progressively from one to 40 bar-presses. Reinforcement consisted of access to distilled water in a cup (nominally 0.08 ml) presented by a conventional dipper mechanism adjacent to the response bar. The dipper arm was lowered briefly to refill the cup when reinforcement was appropriate, and was otherwise held in the raised position. The rats were maintained on the fixed-ratio (FR 40) schedule for five days per week for several weeks to allow performance to stabilise prior to flavour presentations. Session length was 15 min, to match earlier studies [2,10]. Extraneous sounds were masked by white noise (78 dB above 0.0002 dynes/cm^2) which was present at all times.

For certain sessions in the next stage of the experiment, flavoured solutions were presented in the dipper cup. At the end of each such session, the rats were injected with either amphetamine (1 mg/kg) or isotonic saline, and were then returned to their home cages. This dose of amphetamine was previously shown to induce strong flavour aversions [2,4]. Each rat was presented with both chicken and lemon flavours on different days in an alternating sequence, with distilled water as the reinforcer on intervening days. Two rats were injected with amphetamine after every session of responding for chicken-flavoured water and with saline after every session of responding for lemon-flavoured water. The flavour-injection pairings were reversed for the remaining two rats in order to balance out possible effects of the unconditioned palatabilities of the flavours. Flavoured solutions were presented on Tuesdays and Fridays of every week until all the rats had been presented with each flavour on four occasions. Each flavour was then presented once more, on the Fridays of two successive weeks.

The numbers of bar-presses in each session were recorded by print-out counters and the amounts of liquid reinforcers consumed were estimated by weighing the reservoir before and after each session. The dipper cup and the adjacent areas of the chamber were washed and dried before each session so that the rats were not able to detect whether a flavour was to be presented until the first reinforcer was obtained (i.e. until 40 bar-presses had been emitted).

RESULTS

The pattern of responding maintained by the fixed-ratio schedule was similar to that reported previously [11]. Figure 1 shows cumulative records of responding for Rat G15, in which lemon-flavoured water was repeatedly paired with saline and chicken-flavoured water with amphetamine. The usual, rapid rate of FR40 responding can be seen with marked post-reinforcement pauses. Responding for lemon flavour remained reasonably consistent during the course of the study. However, even a single previous pairing of chicken flavour with amphetamine was sufficient to disrupt responding on the next occasion that the chicken flavour was presented. After six reinforcers were obtained, responding was suppressed for several minutes. An even more marked disruption of responding was seen after further flavour-amphetamine pairings, culminating in response suppression after a single reinforcement on the fifth presentation of chicken flavour. Responding at the very beginning

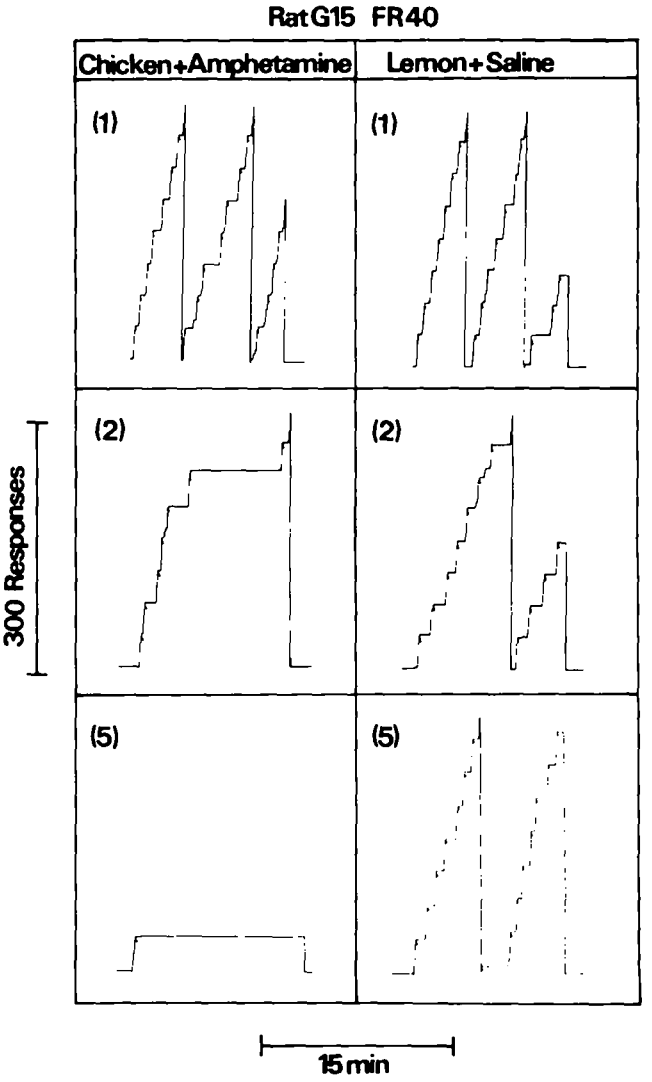

FIG. 1. Performance of Rat G15 under the FR40 schedule. Abscissae, time; ordinates, cumulative number of bar-pressing responses. Short diagonal strokes on the records indicate presentations of flavoured liquid reinforcers. The recorder was reset after approximately every 300 responses and at the end of the 15 min sessions. The records show responding for chicken and lemon flavours on their first (1), second (2) and fifth (5) presentations; responding for chicken flavour was suppressed by pairings with amphetamine (1 mg/kg).

of the session was essentially normal, presumably because the rat was not exposed to the flavour stimulus until it pressed the bar at least 40 times.

The results just described for Rat G15 could merely have indicated an unconditioned effect of chicken-flavour, although the trend over trials makes this unlikely. However, Fig. 2 presents results for Rat G13, in which the flavour-injection pairings were the reverse of those for Rat G15; it can be seen that in this case, suppression of responding developed to lemon flavour whereas responding for chicken flavour remained reasonably constant over its successive presentations.

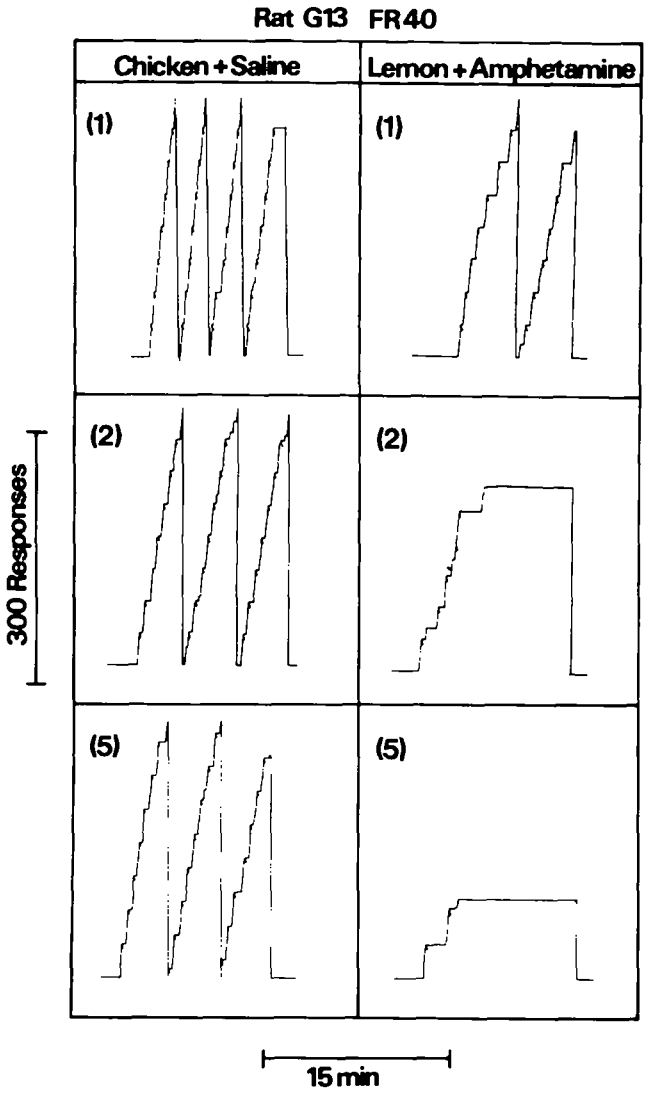

FIG. 2. Performance of Rat G13 under the FR40 schedule of flavoured liquid reinforcement. The records show suppression of responding for lemon flavour after pairings with amphetamine (1 mg/kg). For details, see legend to Fig. 1.

The mean data for all four rats are shown in Fig. 3A. The mean numbers of bar-presses declined rapidly for amphetamine-paired flavours, whereas responding for saline-paired flavours remained relatively constant. A tendency for responding for the saline-paired flavours to decline

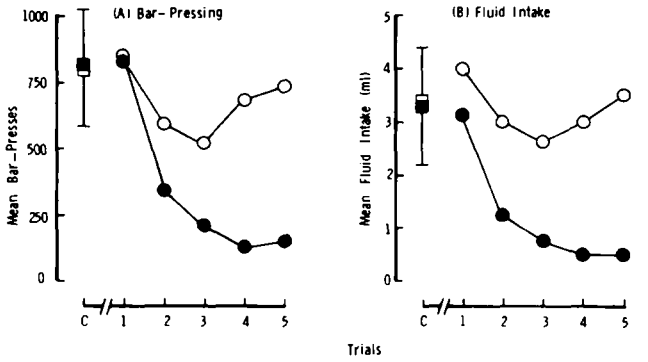

FIG. 3. Flavour-amphetamine pairings suppressed bar-pressing for, and consumption of, flavoured liquid reinforcers. Each point represents the mean number of bar-presses (A) or mean fluid intake (B) for four rats over 15 min sessions. (• amphetamine-paired flavours; ○ saline-paired flavours). Mean scores (± standard deviations) are also shown for distilled water reinforcement on the days immediately preceding presentations of the amphetamine-paired flavours (■) and saline-paired flavours (□).

(Trials 2 and 3) was transient and mainly attributable to one rat (G18). Statistical support for the reliability of the results was sought by means of a two-factor analysis of variance with repeated measures on both factors [27]. The overall difference between responses for the amphetamine- and saline-paired flavours was not significant, $F(1,3) = 4.22$, but the overall decline in responding over successive flavour trials was significant, $F(4,12) = 6.16$, $p < 0.01$. The flavours × trials interaction was also significant, $F(4,12) = 8.76$, $p < 0.01$, which confirms the statistical reliability of the greater suppression of responding for the amphetamine-paired flavours as compared with the saline-paired flavours.

The estimated amounts consumed of the reinforcers are shown in Fig. 3B. It can be seen that suppression of fluid intake developed with the amphetamine-paired flavours, but not with the saline-paired flavours. The changes over trials in bar-pressing (Fig. 3A) and in fluid consumption (Fig. 3B) were very similar; the correlations (r) between the two indices were 0.85 and 0.94 for drug- and saline-paired flavours respectively (df 18, $p < 0.001$ in each case). However, these very high correlations must be interpreted cautiously since, with fixed-ratio schedules, the number of presentations of the reinforcer is directly proportional to the number of responses.

The time-course of responding within sessions is shown in Fig. 4, where the 15 min sessions have been split into five 3-min segments. It can be seen that on the first occasion that flavours were presented, responding was maintained throughout the sessions. After flavour-injection pairings, responding in the first 3-min segment was little affected; during the remaining four segments, responding for the amphetamine-paired flavours was drastically suppressed (Fig. 4A), but responding for the saline-paired flavours was well maintained (Fig. 4B). Thus, the mean results for all four rats were consistent with the sample cumulative records shown in Figs. 1 and 2. Two-factor analyses of variance with repeated measures on both factors [27] confirmed the statistical significance of the findings described above. For amphetamine-paired flavours (Fig. 4A), differences between trials and between 3-min segments

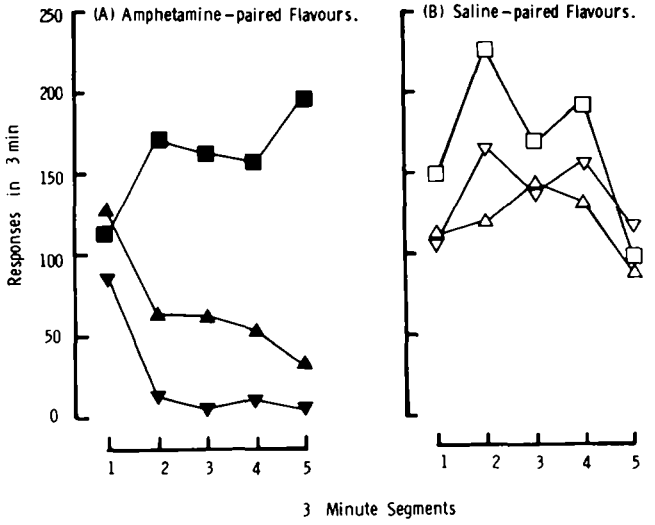

FIG. 4. Mean numbers of bar-pressing responses (FR40) for flavoured liquid reinforcers during successive 3-min segments of a 15 min session. Prior to pairing with amphetamine at 1 mg/kg, responding for the flavoured reinforcers was maintained through all five segments (■); the same flavours suppressed responding after one (▲) and three (▼) pairings with amphetamine. Flavours paired with saline did not suppress responding (□, △, ▽; first, second and fourth presentations respectively). Results for the third and fifth presentations of all flavours have been omitted for clarity, but were included in the statistical analyses (see text).

were both significant, $F(4,12) = 9.74$ and 6.22; $p<0.01$ in both cases, and there was also a significant trials x segments interaction, $F(16,48) = 3.21$, $p<0.01$. For saline-paired flavours (Fig. 4B), neither the effects due to trials or segments, $F(4,12) = 2.24$ and 2.88 respectively, nor the segments x trials interaction, $F(16,48)< 1$, were statistically significant.

DISCUSSION

Rats failed to emit an operant response (bar-press) for a flavoured reinforcer when its presentation had been followed by injections of amphetamine. Even a single flavour-amphetamine pairing produced some decrement in performance, and this was consistent with previous evidence for an aversive property of amphetamine in fluid intake paradigms [2, 3, 6, 19, 24]. Evidence for generalization of aversion between the two flavours used in the present study was minimal, although possibly deserving further study (cf. Fig. 3, Trials 2 and 3). Insofar as the present results involve only one schedule of reinforcement and one dose of amphetamine, they should be considered preliminary, but it is difficult in the face of such data to attribute the contrasting reinforcing and aversive properties of amphetamine merely to the class of response required by an experimental procedure. It remains possible that the changes in operant responding observed in the present experiments were mediated indirectly through the suppression of drinking. However, loss of water reinforcement cannot alone account for the findings since behaviour maintained by intermittent reinforcement is very resistant to extinction [11]. Whether it be direct or indirect, the power of flavour-drug pairings to influence operant behaviour is obvious, and for the reasons given below it seems worthy of more extensive study.

Previous studies with large doses of apomorphine [1] or lithium [20] have also shown that flavour-drug pairings can suppress operant behaviour. In one study, there was some evidence that the effect on the operant was less than that on the consummatory response [1], but several factors could account for that observation. For example, the flavour-injection pairings were not counter-balanced and, therefore, the unconditioned palatabilities of the flavours could have influenced the results. On the other hand, studies of conditioned suppression by auditory stimuli paired with foot-shock have not, to date, established that operant responses are more sensitive than consummatory responses to that type of manipulation [9]. Thus, the results of auditory-shock, flavour-drug and flavour-shock [16] pairings do not always support hypotheses that postulate a dichotomy of responses with respect to ease of conditioning with different cues and consequences.

The present experiments can be grouped with earlier studies which have also failed to isolate factors which may be critically related to a reinforcing rather than an aversive property of amphetamine [2, 5, 10, 14, 19]. To resolve the matter, it may be necessary to take into account the possibility that the superficially straightforward CTA paradigm involves features of both instrumental and classical conditioning [12]. The presentation of stimuli (flavours) is contingent upon responding (drinking or bar-pressing), a typical feature of instrumental conditioning. However, presentation of the unconditioned stimulus (drug) is not contingent upon responding but is merely related to it in time, a typical feature of classical conditioning procedures. This aspect of CTA may be relevant to the action of amphetamine [25].

There is evidence that after pairings with programmed administrations of amphetamine, sensory stimuli can suppress operant behaviour [26], whereas stimuli paired with self-administered amphetamine can acquire positively reinforcing properties [13]. However, it is difficult to maintain the argument that whether amphetamine is obtained by programmed or self-administration may be critical, since there is evidence both for positive reinforcement from programmed administrations [8] and for aversion in certain self-administration experiments [24,28]. The weight which can be given to these comparisons is limited by the absence of a systematic study with amphetamine similar to that carried out with electric shock; visual stimuli paired with programmed shocks acquired aversive properties, but the same visual stimuli paired with the same number and temporal pattern of response-produced shocks did not become aversive [21]. Thus, in experiments where sensory stimuli are paired with either a standard aversive stimulus (shock) or with amphetamine, the outcome may depend on the way these events are programmed to occur in relation to behaviour. In future analyses of CTA, it may be necessary to consider more carefully the role of such contingencies, rather than simply the class of response or the modality of the discriminative or conditioned stimuli.

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REFERENCES

1. Best, P. J., M. Best and R. H. Ahlers. Transfer of discriminated taste aversion to a leverpressing task. *Psychon. Sci.* **25**: 281–282, 1971.
2. Booth, D. A., G. D. D'Mello, C. W. T. Pilcher and I. P. Stolerman. Paradoxical aversive property of dexamphetamine. *Br. J. Pharmac.* **57**: 424–425P, 1976.
3. Cappell, H. and A. E. Le Blanc. Conditioned aversion to saccharin by single administrations of mescaline and *d*-amphetamine. *Psychopharmacologia* **22**: 352–356, 1971.
4. Cappell, H. and A. E. Le Blanc. Punishment of saccharin drinking by amphetamine in rats and its reversal by chlordiazepoxide. *J. comp. physiol. Psychol.* **85**: 97–104, 1973.
5. Cappell, H. and A. E. Le Blanc. Conditioned aversion by psychoactive drugs: Does it have significance for an understanding of drug dependence? *Addictive Behaviour* **1**: 55–64, 1975.
6. Carey, R. J. Long-term aversion to a saccharin solution induced by repeated amphetamine injections. *Pharmac. Biochem. Behav.* **1**: 265–270, 1973.
7. Cruse, D. B., W. Vitulli and M. Dertke. Discriminative and reinforcing properties of two types of food pellets. *J. exp. Analysis Behav.* **9**: 293–303, 1966.
8. Davis, W. M. and S. G. Smith. Blocking effect of α-methyltyrosine on amphetamine based reinforcement. *J. Pharm. Pharmac.* **25**: 174–177, 1973.
9. DeCosta, M. J. and J. J. B. Ayres. Suppression of operant vs. consummatory behavior. *J. exp. Analysis Behav.* **16**: 133–142, 1971.
10. D'Mello, G. D., I. P. Stolerman, D. A. Booth and C. W. T. Pilcher. Factors influencing flavour aversions conditioned with amphetamine in rats. *Pharmac. Biochem. Behav.* **7**: 185–190, 1977.
11. Ferster, C. B. and B. F. Skinner. *Schedules of Reinforcement.* New York: Appleton-Century-Crofts, 1957.
12. Garcia, J., J. C. Clark and W. G. Hankins. Natural responses to scheduled rewards. In: *Perspectives in Ethology*, edited by P. P. G. Bateson and P. H. Klopfer. London: Plenum Press, 1973, pp. 1–41.
13. Goldberg, S. R., R. T. Kelleher and W. H. Morse. Second-order schedules of drug injection. *Fedn Proc.* **34**: 1771–1776, 1975.
14. Goudie, A. J., E. W. Thornton and T. J. Wheeler. Drug pretreatment effects in drug induced taste aversions: Effects of dose and duration of pretreatment. *Pharmac. Biochem. Behav.* **4**: 629–633, 1976.
15. Holman, E. W. Some conditions for the dissociation of consummatory and instrumental behavior in the rat. *Learn. Motivat.* **6**: 358–366, 1975.
16. Krane, R. V. and A. R. Wagner. Taste aversion learning with a delayed shock US; implications for the "generality of the laws of learning". *J. comp. physiol. Psychol.* **88**: 882–889, 1975.
17. Le Magnen, J. Peripheral and systemic actions of food in the caloric regulation of intake. *Ann. N.Y. Acad. Sci. U.S.A.* **157**: 1126–1156, 1969.
18. Lovett, D. and D. A. Booth. Four effects of exogenous insulin on food intake. *Q. J exp. Psychol.* **22**: 406–419, 1970.
19. Martin, J. C. and E. H. Ellinwood. Conditioned aversion in spatial paradigms following methamphetamine injection. *Psychopharmacologia* **36**: 323–335, 1974.
20. Morrison, G. R. and R. Collyer. Taste-mediated conditioned aversion to an exteroceptive stimulus following LiCl poisoning. *J. comp. physiol. Psychol.* **86**: 51–55, 1974.
21. Orme-Johnson, D. W. and M. Yarczower. Conditioned suppression, punishment and aversion. *J. exp. Analysis Behav.* **21**: 57–74, 1974.
22. Pickens, R. and W. C. Harris. Self-administration of *d*-amphetamine by rats. *Psychopharmacologia* **12**: 158–163, 1968.
23. Seligman, M. E. P. and J. L. Hager. *Biological Boundaries of Learning.* New York: Appleton-Century-Crofts, 1972.
24. Stolerman, I. P., R. Kumar and H. Steinberg. Development of morphine dependence in rats: Lack of effect of previous ingestion of other drugs. *Psychopharmacologia* **20**: 321–336, 1971.
25. Vogel, J. R. and B. A. Nathan. Learned taste aversion induced by hypnotic drugs. *Pharmac. Biochem. Behav.* **3**: 189–194, 1975.
26. Whitney, G. D. and J. G. Trost. Response disruption following amphetamine self- and programmed administration. In: *Drug Dependence*, edited by R. T. Harris, W. M. McIsaac and C. R. Schuster. Austin: University of Texas, 1970, pp. 198–213.
27. Winer, B. J. *Statistical Principles in Experimental Design.* First edition, London: McGraw-Hill, 1962.
28. Wise, R. A., R. A. Yokel and H. DeWit. Both positive reinforcement and conditioned aversion from amphetamine and apomorphine in rats. *Science* **191**: 1273–1275, 1976.