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BRIEF COMMUNICATION

Effect of Dopamine-Receptor Blockade on Stimulation-Induced Feeding

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STREATHER, A AND M A BOZARTH *Effect of dopamine-receptor blockade on stimulation-induced feeding* PHARMACOL BIOCHEM BEHAV 27(3) 521–524, 1987 —The effect of pimozide on stimulation-induced feeding was tested in food satiated rats Pimozide produced a dose-dependent decrease in the number of animals eating during electrical stimulation of the lateral hypothalamus A quantal dose-response analysis yielded an ED50 of 0 323 mg/kg for pimozide Because this dose is within the range of pimozide doses found to be effective in disrupting feeding in other tests, it seems likely that the neural substrate mediating stimulation-induced feeding is similar to that involved in deprivation-induced feeding

Dopamine ED50 Feeding Lateral hypothalamus Pimozide Probit analysis Quantal dose-response Stimulation-induced feeding

THERE is considerable evidence that a dopaminergic mechanism may be involved in the regulation of feeding behavior (see [18] for a review) Dopamine-depleting lesions of the nigro-striatal pathway produce aphagia and adipsia [12] Neuroleptics, which block dopamine receptors, disrupt lever pressing for food in food deprived animals [21] Neuroleptics also attenuate feeding in a discrete trial free-feeding test which involves a simple consummatory response [20] Although neuroleptics can produce sedation and catalepsy [6], specific experimental designs have been developed that can eliminate motor-impairment explanations of their effect on feeding (e g , [20,21], see also [18])

Electrical stimulation of the lateral hypothalamus can elicit feeding in food satiated animals [3, 4, 11, 13, 14] The characteristics of this stimulation-induced feeding are very similar to natural feeding, although some differences exist [17] A previous report has shown that stimulation-induced feeding is disrupted by neuroleptic treatment, but only a single effective drug dose was tested [9] The present study examined the effect of a neuroleptic (pimozide) that has been used extensively in other studies involving the role of dopamine in feeding behavior Furthermore, by examining a range of doses, an accurate determination of the effective ness of the compound can be made This permits comparisons with other behavioral measures assessing the influence of neuroleptic treatment on feeding If similar neural mechanisms are involved in deprivation-induced feeding and stimulation-induced feeding, then the same neuroleptic should be equally effective in inhibiting feeding in both cases

METHOD

Subjects

Twenty-five male, Long-Evans rats, weighing 300–360 g, were implanted with chronically indwelling, stainless steel electrodes aimed at the lateral hypothalamus The monopolar electrodes were insulated with Formvar except at the cross section of the tips A wire wrapped around three stainless steel skull screws served as the stimulation ground The electrodes were implanted 0 8 mm posterior to bregma, 1 5 mm lateral to the mid-sagittal suture, and 8 7 mm ventral to the surface of the skull, the upper incisor bar was 3 2 mm above the interaural line Surgery was performed under sodium pentobarbital (60 mg/kg, IP) anesthesia, and atropine sulfate (0 4 mg/kg, IP) was used to decrease mucosal secre-

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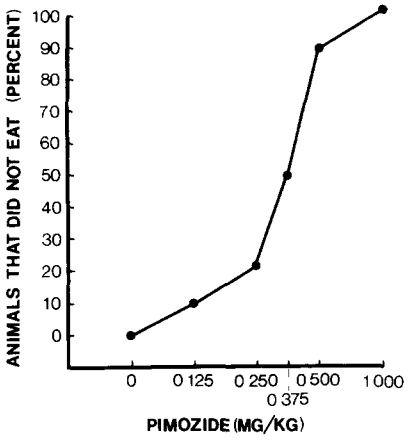

FIG 1 The effect of pimozide on stimulation-induced feeding The quantal dose-response analysis shows the percentage of animals inhibited at each dose of pimozide (ED50=0 323 mg/kg, n=10)

tions A single injection of penicillin G (30,000 units, IM) was administered prophylactically following surgery The animals were individually housed with a 12 hr light/dark cycle of illumination Food and water were freely available in the home cage

Following the completion of behavioral testing, each subject was deeply anesthetized with sodium pentobarbital (100 mg/kg, IP) and intracardially perfused with physiological saline followed by a 10% formalin solution The brains were removed and stored in a 10% formalin solution for at least 3 days Next, the brains were frozen and sliced into 40 micron sections on a coronal plane The brain sections were then stained with thionin and the location of the electrode tips determined under 10× magnification All electrode placements just dorsolateral to the fornix This zone has been previously reported to be very effective in producing stimulation-induced feeding [16,19]

Threshold Determinations

After a minimum of 7 days recovery from surgery, the animals were screened for stimulation-induced feeding Rats were tested in a 24×35×35 cm box with the floor covered with Purina rat chow A constant current source provided 60 Hz sine wave stimulation at various intensities Free movement of the subject during behavioral testing was maintained by using light, flexible electrical lead connecting the rat's electrode to an electrical commutator Animals had free access to food and water during all phases of the experiment

Electrical stimulation was administered at a preselected intensity for 20 seconds followed by 20 seconds of no stimulation The initial current intensity was 2 μA and each successive stimulation period was preceded by a 1 μA increase in the stimulation intensity until feeding was observed Testing was terminated and the animal dropped from further testing if aversive behaviors (e g , vocalization, jumping or escape attempts) had been elicited by the stimulation

Animals that exhibited stimulation-induced feeding were tested for a minimum of 10 daily training sessions to obtain a stable eating threshold for each subject A modified method of limits was used to determine thresholds The lowest current intensity that induced feeding during the preceding session was used as the starting stimulation intensity If the animal did not eat during the stimulation period, the current intensity was increased by 1 μA during successive stimulation periods When eating was observed, the current intensity was decreased by 1 μA on successive stimulation periods until eating was not elicited This procedure was repeated for a total of seven threshold determinations during each test session, and the mean of these measures was used as the stimulation threshold This 20-seconds-ON/20-seconds-OFF schedule of stimulation combined with stimulation intensities selected using a modified method of limits has been shown to be a fast, reliable procedure for estimating current thresholds for stimulation-induced feeding [16,19]

Drug Testing

Pimozide was dissolved in a tartaric acid vehicle (0 3% v/v) Injections were given intraperitoneally 4 hours before behavioral testing, this pretreatment time corresponds to the time after injections that pimozide has its strongest antidopaminergic effects [6] The pimozide concentration was adjusted to maintain a 1 ml/kg injection volume except at the highest dose which was injected using a 2 ml/kg injection volume Control injections consisted of the tartaric acid vehicle alone (1 ml/kg)

Drug injections were given over a 3-day cycle On the first day of each cycle, the animals were injected with the tartaric acid vehicle, and stimulation-induced feeding thresholds were determined On the second day, subjects received one of five doses of pimozide (0 125, 0 250, 0 375, 0 500, or 1 000 mg/kg, IP), and stimulation-induced feeding thresholds were again determined On the third day of each cycle, no injections or testing occurred This 3-day cycle was repeated five times so that each subject was tested under each dose of pimozide Drug doses were selected using a Latin-square design Following pimozide pretreatment, the maximum stimulation intensity tested was double the vehicle control threshold Stimulation intensities never exceeded 50 μA during any phase of the experiment because stimulation intensities above this level have been shown to affect subsequent responding for lower stimulation intensities [2]

RESULTS

Reliable stimulation-induced feeding was obtained in 10 of the 25 animals implanted with electrodes The mean current intensity thresholds ranged from 5 to 16 μA, and the standard deviation for each subject's threshold was below 1 0 μA and usually less than 0 5 μA The percentage of animals showing stimulation-induced feeding and the current intensity thresholds were within the ranges found by other investigators using electrode placements in the lateral hypothalamic area (e g , [13,16]) Stimulation thresholds remained stable across repeated vehicle testing as previously reported [17]

Pretreatment with pimozide produced dose-dependent effects on feeding At the higher doses feeding was totally suppressed At the lower pimozide doses, feeding thresholds were occasionally elevated, but this effect was not consistent across all of the subjects Because feeding tended to be either totally suppressed or not affected at all following pimozide pretreatment, a quantal dose-response analysis was performed on the percentage of animals eating at their threshold stimulation intensities following pimozide injections

Figure 1 shows that pimozide produced a dose-dependent

decrease in the percentage of animals eating during electrical stimulation, Cochran's Q-statistic $Q(5)=34\ 96$, $p<0\ 005$, see [15] More important than the simple demonstration that various pimozide doses inhibited feeding is an analysis of the nature of this effect A probit analysis with fiducial limits was performed according to the method of Finney [5] This analysis not only yields accurate estimates of the ED50 of a compound but also provides confidence intervals and a test of the goodness-of-fit for the data used for computing the ED50 The probit analysis revealed an ED50 of 0 323 mg/kg with a 95% confidence interval ranging from 0 245 to 0 421 The data conform well to the model as evidenced by the correlation coefficient for the probits ($r=0\ 907$, $p<0\ 05$) and by the chi square which tests the data for deviation from homogeneity, $\chi^2(2)=2\ 15$, $p>0\ 25$ The method of Litchfield and Wilcoxon [8], using the computation formula of Tallarida and Murray [10], produces similar values with an ED50 of 0 313 mg/kg and a 95% confidence interval of 0 218 to 0 448, $\chi^2(2)=0\ 65$, $p>0\ 5$

DISCUSSION

The present study clearly shows that pimozide inhibits stimulation-induced feeding, but the response was quantal in nature This was surprising because past work using this same experimental procedure has shown that increasing the current intensity can overcome the effects of aversive taste, stomach-loading, and satiation on stimulation-induced feeding [4, 11, 19] Furthermore, the response-inhibiting effects of pimozide on brain stimulation reward can also be offset by increasing the stimulation current intensity [1,7] Thus, the finding in the present study that increasing the current intensity failed to reinstate feeding in subjects affected by pimozide is not in accord with the earlier studies The fact that dopamine-receptor blockade inhibits feeding, however, is in agreement with an earlier study testing the effects of haloperidol on stimulation-induced feeding [9] That study reported that haloperidol significantly inhibited stimulation-induced feeding only at the highest dose tested The present study reveals a dose-dependent inhibition of feeding, although the effect was not manifest as a simple elevation of feeding thresholds The quantal nature of this effect suggests that there may be important differences in the mechanisms of central and peripheral manipulations that inhibit stimulation-induced feeding

The potential response-impairing effect of neuroleptics has been an area of considerable interest Some investigators have suggested that many of the effects of neuroleptics on behavior can be explained by a simple motor impairment and are not the result of blocking reward processes (see [18]) A number of studies, however, have ruled out simple response-impairment as an explanation of pimozide's effects on motivated behaviors (see [18] for a review) Because the ED50 of pimozide in this study is actually lower than the doses effective in studies where response-impairment has been clearly ruled out (i e , 0 323 vs 0 5 to 2 0 mg/kg, e g , [20–22]), it is unlikely that changes in the motor capacity of the subjects contribute to the inhibition of stimulation-induced eating This is even more unlikely when considering the relatively low response requirement for this type of task (i e , a simple consummatory response) compared with lever-pressing behavior which is more response demanding

Previous work has suggested that stimulation-induced feeding is very similar to natural feeding (see [17]) The present study supports this assertion by showing that pimozide is equally effective in inhibiting stimulation-induced feeding and deprivation-induced feeding, and this finding is consistent with the notion that a common neural substrate is involved in these feeding behaviors Furthermore, the pimozide doses effective in influencing feeding [20,21], brain stimulation reward [1], and intravenous amphetamine self-administration [22] all fall within the same range This suggests that a quantitatively similar dopamine-receptor population may underlie pimozide's effects on these diverse motivational events

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