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Effects of N, N-Dimethyltryptamine on Behavioural Habituation in the Rat[1]

SANDRA E. FILE

Pharmacology Department, The School of Pharmacy, University of London
England

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FILE, S. E. *Effects of n,n-dimethyltryptamine on behavioural habituation in the rat.* PHARMAC. BIOCHEM. BEHAV. 6(2) 163–168, 1977. – The processes involved in habituation and the various ways drugs might affect habituation are discussed. Exploration was measured in a holehoard and N,N-Dimethyltryptamine (DMT, 4 mg/kg) profoundly reduced the level of exploration, precluding any conclusions about the rate of habituation with this dose. However, both 2 and 4 mg/kg doses prevented the 24-hr retention of habituation of exploration. DMT (2 and 4 mg/kg) did not reduce the initial distraction to a tone stimulus, but the rate of habituation and its 24-hr retention was impaired.

Habituation Processes Drugs DMT Exploration Distraction Retention

HABITUATION is defined as the decrement of an unconditioned response to a stimulus as a result of repeated presentations of that stimulus. There are many ways in which drugs might interact with habituation, and a consideration of the processes involved in habituation should help with interpretation of drug results.

During the process of habituation two types of analysis of the stimulus take place. The first is an analysis of the physical features of the stimulus. Both the number of the stimulus parameters examined and the specificity with which they are coded may vary. For example, if an animal has habituated to an auditory stimulus this stimulus could be coded either as a high tone, or as a 9 kHz tone of 80 dB lasting for 5 sec. The second type of analysis that is made is in terms of the reinforcement value of the stimulus. Each stimulus presentation that does not signal any change in the probability of receiving reinforcement will decrease the probability of that stimulus being attended to. This classification of the stimulus as irrelevant could proceed in parallel with an analysis of its physical features.

Short-term habituation is the habituation that is retained for only a few minutes and is therefore only seen within a test session. Long-term habituation refers to the decrease in response which is retained for several days and can therefore be observed between sessions. Several workers have suggested that different neural mechanisms may be involved in short- and long-term habituation [4,11]. Certainly the two do have different characteristics. For example, within-session habituation occurs with fewer stimulus presentations at short stimulus presentation intervals, whereas long-term habituation requires fewer trials if there are longer intervals between stimulus presentations [6, 10, 15].

In studies of behavioural habituation three types of responses have been used – startle, distraction and exploration. For the first two situations the experimenter is able to control both the stimulus parameters and the rate of stimulus presentation, whereas these are determined largely by the subject in test situations where exploration is measured. The suggestion has been made [18] that different neuropharmacological mechanisms may be involved in the habituation of different response systems. Whilst it is plausible that habituation of exploration, startle and distraction may have different underlying mechanisms the reason for this may not necessarily be that different responses are involved. The test situations also differ in the manner in which the animal can sample the stimuli, so that the neuropharmacological differences may arise from differences in stimulus analysis.

Lysergic acid diethylamide (LSD) has been reported to impair the rate of habituation of the EEG arousal response [13], of an orienting response to a buzzer [12] and to increase the distracting effect of irrelevant stimuli [14,17]. Another psychotomimetic, N,N-Dimethyltryptamine (DMT), which has many similar effects to LSD [1, 2, 3 16] has been reported to impair retention of habituation of startle responses, although conclusions about its effect on the rate of habituation were impossible because of the reduction in startle amplitude [5]. In the present study the effects of DMT are examined in other behavioural situations. Exploration and its habituation were studied in a 4-hole holeboard, using the frequency and duration of

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head-dipping as measures of exploration [9]. Habituation of distraction was measured by engaging rats in a baseline activity of licking and measuring distraction to tone stimuli by the interruption in licking [6].

EXPERIMENT 1 HABITUATION OF EXPLORATION

METHOD

Animals

Eighty-four male hooded rats, 300–350 g, were housed in groups of six, in an 11 hr light-13 hr dark cycle (lights on at 0800 hr), in rooms maintained at a constant temperature of 25° C. They were allowed food and water ad lib.

Drugs

N,N-dimethyltryptamine (DMT) was dissolved in 1N HCl and then titrated back to pH 7.0 with dilute NaOH. The solution was diluted with deionised water to give a concentration of 1 mg/ml. The vehicle control solution was made up in a similar way. Injections were given intraperitoneally 10 min before test.

Apparatus

Testing took place in a holeboard [9] which was a wooden box with walls 45 cm high and a floor 55 × 55 cm. In the floor were four equally spaced holes, 3.8 cm in diameter. The floor of the holeboard was 12 cm above the base of the box and when objects were placed under the holes they came to 2 cm below the top of the holes. The objects were supported by glass funnels and were chosen to smell and feel different from each other. The objects used were a brass rod, a rolled piece of suede, a rubber bung and a cork. The illuminance on the floor of the holeboard was 25 scotopic lux, measured with a photocell calibrated with respect to the C.I.E. scotopic curve and the C.I.E. standard radiator (C.I.E. – Committée Internationale d'Eclairage).

Procedure

Forty-eight rats were randomly assigned to control, DMT (2 mg/kg) and DMT (4 mg/kg) groups to give 16 rats in each group. These animals were tested only between 10.00 and 12.00 hr with the test order randomised between groups and test conditions, because the level of head-dipping varies with the time of day [8]. These rats received one 10 min trial per day for 3 days, half of each group being tested in the absence of objects and half in their presence.

An additional 36 rats were randomly allocated to control, DMT (2 mg/kg) and DMT (4 mg/kg) groups to give 12 rats in each group. These animals were tested between 14.30 and 16.30 hr and received only 2 10 min trials. The trials were separated by 24 hr and objects were absent for the first trial and present for the second.

Each rat was placed in the centre of the holeboard and its behaviour recorded for 10 min. A head-dip was defined as occurring when the rat reached into the hole at least as far as its ears. The frequency of head-dips (number/10 min) and the duration of each dip (to the nearest sec) were scored. The scoring was divided into four 2.5 min periods to give a measure of within-session habituation. The number of rears made was also counted. At the end of each trial any boluses were removed and the floor was wiped with water and dried, to minimise any traces of the path taken.

RESULTS

Figure 1 shows the pattern of between-session habituation for control and DMT-injected rats, tested in the absence and in the presence of objects. DMT significantly reduced the duration of head-dipping, $F(2,42) = 8.86$, $p<0.001$, and retarded between-session habituation (drug × days interaction, for the duration of head-dipping $F(4,84) = 8.50$, for frequency $F = 7.61$, $p<0.001$), but this is difficult to interpret for the 4 mg/kg group because of their greatly reduced response level. However, the 2 mg/kg group showed impaired habituation in the presence of objects and this cannot be secondary to a reduced response level.

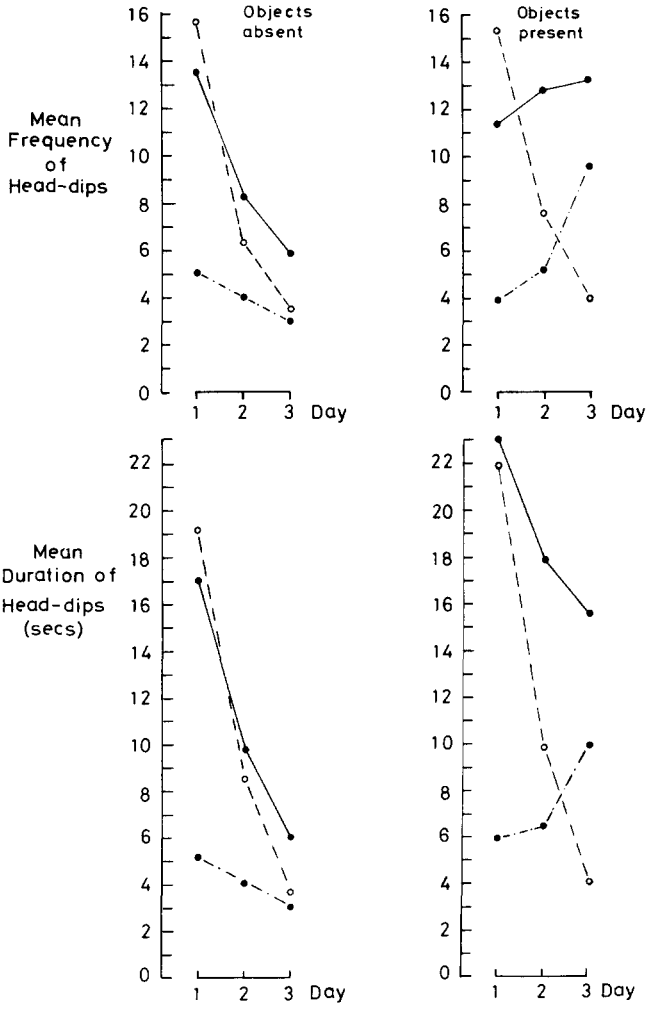

FIG. 1. Between-session habituation in the absence and in the presence of objects. Controls ○------○, DMT (2 mg/kg) ●————●, DMT (4 mg/kg) ●·-·-·-●. Frequency of head-dips = number/10 min trial.

Figure 2 shows the pattern of within-session habituation, in the presence of objects. There was an overall significant habituation within each session (for the duration $F(3,378) = 26.68$, for frequency $F = 56.28$, $p<0.001$), but this was slower in the drugged animals (for duration $F(3,378) = 5.62$, $p<0.001$, for frequency $F = 3.96$, $p<0.01$). However,

only the 4 mg/kg group shows profoundly disturbed pattern of within-session responding and once more this could well be due to their very low response level. What Fig. 2 does show is the very large spontaneous recovery between each day, shown by the drugged animals. The control animals show a significant decrease in the duration of head-dips made in the first trial period on days 1 and 2 ($t(7) = 2.78$, $p<0.025$), and on Days 2 and 3 ($t(7) = 2.90$, $p<0.025$), whereas the DMT (2 mg/kg) rats show no decrease from Day 1 to 2 and no significant decrease from Day 2 to 3 ($t(7) = 0.64$, $p>0.05$).

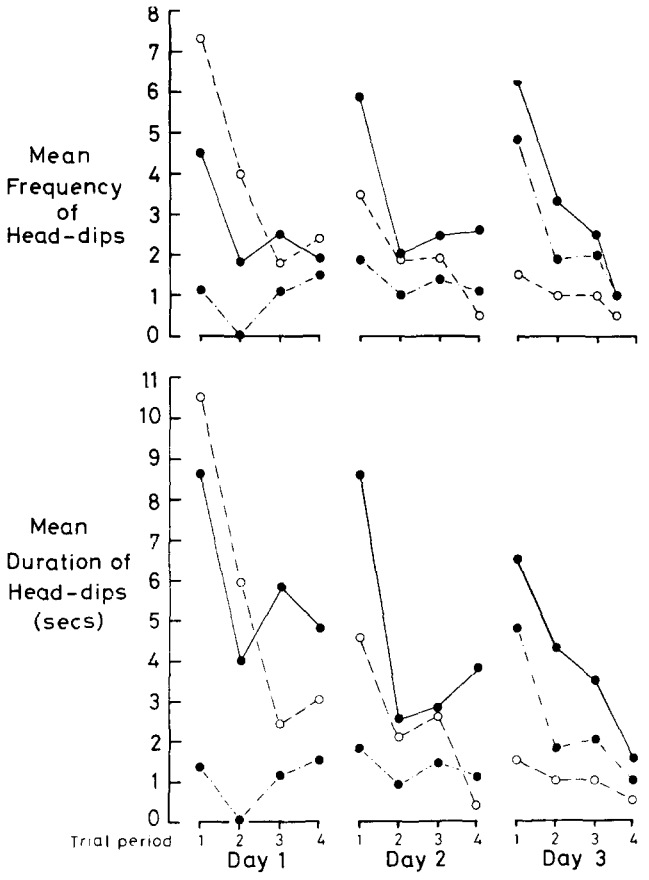

FIG. 2. Within-session habituation in the presence of objects. Controls ○------○, DMT (2 mg/kg) ●————●, DMT (4 mg/kg) ●-·-·-●. Frequency = number/10 min trial.

There was no objects × drug interaction (for duration F = 0.94, for frequency F = 0.37) suggesting that the DMT rats responded like the controls with increased head-dipping in the presence of objects. Figure 3 confirms that this is so, even for the 4 mg/kg group, since all groups showed an increased response when the objects were introduced on Day 2 ($t(11) = 1.78$, 3.10, 2.04 for control, 2 mg/kg and 4 mg/kg DMT respectively, $p<0.05$, one-tailed tests). All these rats were tested in the afternoon and show lower response levels than those tested in the morning; this is in agreement with previous results [8] and suggests that DMT does not alter diurnal variations in exploration.

Table 1 shows the mean number, and the standard error of the mean, of rears made on Day 1 in the presence and absence of objects for the control and drugged groups. Because of the unequal variances between the groups the data were subjected to a $\sqrt{x+1}$ transform before analysis. All the rats (both control and drugged) made fewer rears in the presence of objects, $F(1,42) = 5.13$, $p<0.05$, and in both the objects absent and in the objects present condition DMT reduced the number of rears, $F(2,42) = 36.92$, $p<0.001$.

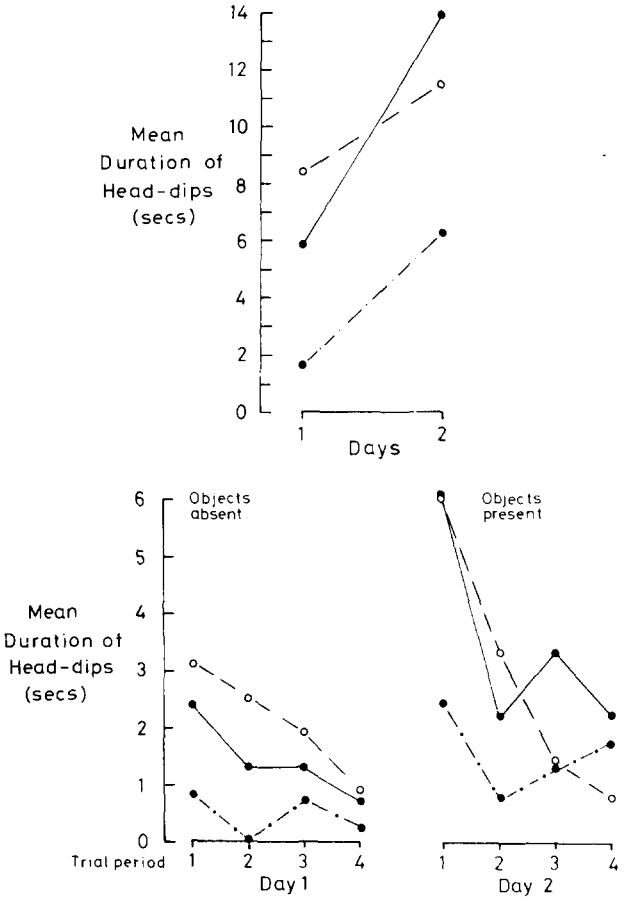

FIG. 3. Mean duration of head-dipping on Day 1 (objects absent) and Day 2 (objects present). Controls○------○, DMT (2 mg/kg) ●————●, DMT (4 mg/kg) ●-·-·-●.

TABLE 1

MEAN NUMBER, AND SEM, OF REARS MADE ON DAY 1, IN THE ABSENCE AND IN THE PRESENCE OF OBJECTS, FOR CONTROL AND DMT INJECTED RATS

	Objects Absent	Objects Present
Control	30.87 ± 3.78	24.87 ± 3.76
DMT (2 mg/kg)	24.34 ± 2.27	18.91 ± 2.58
DMT (4 mg/kg)	6.67 ± 1.61	2.62 ± 1.28

An analysis of variance was conducted on these data. Fewer rears were made in the presence of objects than in their absence, $p<0.05$, and in both the test conditions DMT reduced the number of rears, $p<0.001$.

EXPERIMENT 2 HABITUATION OF DISTRACTION

METHOD

Animals

Seventy-three male hooded rats, 350 g weight, were housed as previously described. These rats were given an initial 48 hr period of water deprivation and thereafter received water only during or immediately following test, in sufficient quantity to maintain a steady body weight. Food was available ad lib.

Apparatus

The test chamber, 19 × 19 × 26.5 cm, was enclosed in an acoustically insulated box. A slit in the end wall gave access to a water spout, and a drinkometer recorded the rat's licking. Experimental events were automatically programmed and the tones were delivered via a loudspeaker positioned in the lid of the chamber at the water spout end. The tone used was 9 kHz, 80 dB (re 0.0002 dynes/cm^2) for 9 sec.

Drugs

The DMT and control solutions were prepared as in Experiment 1.

Procedure

Twenty-eight rats were allocated to the control group, 22 to DMT (2 mg/kg) and 23 to DMT (4 mg/kg). On the first test day no injections were given and each rat was given 20 min in the test chamber.

On the second day rats received IP injections 10 min before test. The rat's 200th lick switched on a control period of 9 sec and the number of licks made in this time (A) was counted. Following this period the next 200th lick switched on the tone for 9 sec and the number of licks (B) was again counted. Control and tone periods alternated with a mean interval between tones of 78 sec until criterion was reached. Criterion was taken as three successive tone presentations producing a ratio of $\frac{A-B}{A} \leq 0.10$. Any rat not reaching criterion on the first day was replaced in its cage and tested again the following day, after the same injection. Testing continued until criterion was reached or 40 tones had been presented. For each rat the mean distraction ratio to the first tone presentation, and the trials to habituate were scored.

The day following the one on which criterion was reached a retention test was given. Ten of the control rats were retested undrugged, 9 were tested following injections with DMT (2 mg/kg) and 9 following DMT (4 mg/kg). Similarly, the animals habituated under DMT were either retested following the same injection, or were retested undrugged. Once more the distraction ratio to the first tone presentation and the trials needed to rehabituate were scored.

RESULTS

The baseline licking rate for the control rats was 56.4 licks in 9 sec, for the DMT (2 mg/kg) group it was 55.4 licks and for the DMT (4 mg/kg) group it was 54.9. Thus any peripheral changes produced by DMT did not influence the rate of licking. Nor did DMT impair the total amount of water consumed becaused all rats maintained a steady weight during the course of the experiment.

The mean distraction ratio for the control rats was 0.64 ± 0.05 and that for the DMT (2 mg/kg) group was 0.54 ± 0.07, this difference was not significant ($t(48) = 1.24$). The mean distraction for the DMT (4 mg/kg) group was 0.73 ± 0.05, again not significantly different from the control score ($t(49) = 1.26$).

Although DMT did not affect the initial distraction it did impair habituation. The control rats took a mean of 12.96 trials to habituate, the DMT (2 mg/kg) group a mean of 19.64 and the DMT (4 mg/kg) rats a mean of 19.74, in both cases a significant increase ($t(48) = 2.10$, $t(49) = 2.24$ respectively, $p<0.05$).

Only 17 rats in the DMT (2 mg/kg) group reached criterion; of these 7 were retested undrugged and 10 were retested after 2 mg/kg DMT. Sixteen rats in the 4 mg/kg group reached criterion, 9 of these were retested undrugged and 7 were retested after 4 mg/kg DMT. The first row in Fig. 4 shows the distraction ratio and trials to rehabituate for rats given the retention test in the same state as their original habituation. From this it can be seen that the control rats showed good retention, but that both the drugged groups showed a significant return of distraction ($t(9) = 3.96$, $p<0.01$ for 2 mg/kg, $t(6) = 5.92$, $p<0.001$ for 4 mg/kg), and needed further trials to reach criterion again.

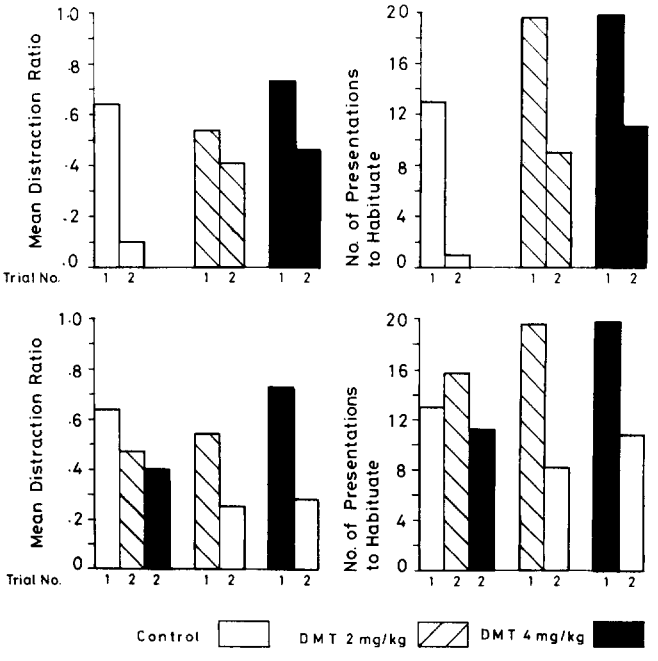

FIG. 4. Retention of habituation. Mean distraction ratio and number of stimulus presentations to reach criterion for Trial 1 and Trial 2.

The second row in Fig. 4 shows the performance of rats given a retention test in a different state from the one in which they were habituated. Comparing the first and second rows it can be seen that for the rats that were first habituated after injections of DMT there was little difference in their performance on the retention test whether they were tested drugged or not. This is particularly striking when the trials to rehabituate are compared. However, the

rats habituated undrugged showed a significant return of distraction when tested after DMT injections ($t(8) = 3.46$ for 2 mg/kg and $t(8) = 3.41$ for the 4 mg/kg group, $p<0.01$), which contrasts with the good retention when they were tested undrugged.

DISCUSSION

There are several ways in which drugs might impair habituation, but these have seldom been distinguished. Firstly, drugs may lead to a reduction in channel capacity. Thus there might be a loss in the specificity with which a stimulus parameter is analysed; a reduction in the number of parameters analysed; and/or a reduction in the number of stimuli analysed. The result would either be slower habituation or a loss of the specificity of stimulus coding or both. Such an impairment would not be restricted to habituation but should also be found in other tests of learning and discrimination.

Secondly, drugs may alter the stimulus salience by altering the drive state of the animal or by altering sensory thresholds so that, for example, a tone that normally produced only distraction might now appear intense enough to elicit a startle response [7].

Thirdly, a drug might impair an animal's ability to classify a stimulus as irrelevant, and so the stimulus salience would not decrease as a result of nonreinforced stimulus presentations.

Fourthly, drugs might disrupt the short- or long-term storage of information. This disruption might be common to short- or long-term memory in other test situations; or it could be specific to habituation, in that the nature of the information storage might depend on the type of learning involved.

Finally, a drug might act indirectly by altering the baseline response level, or by altering factors such as sleep patterns, arousal level, blood pressure, body temperature, to produce any of the consequences listed above.

In the light of this analysis what are the effects of DMT on behavioural habituation? Peripheral changes produced by DMT may have been responsible for the strange flattened posture adopted by the rats injected with 4 mg/kg DMT when they were placed in the holeboard. Whatever the reasons for this postural chnage this could have been one factor contributing to the profound reduction in the level of exploration produced by this dose, which highlights the difficulties of drawing any conclusions about habituation when the initial response level is low. From Fig. 2 it can be seen that when the level of head-dipping is low, whether this is in the control or drug groups, then no significant habituation can occur. It was not possible to assess the effects of DMT on startle habituation because it also profoundly reduced the amplitude of startle responses [5]. Davis [5] suggested that this could be due to the strange posture adopted by the DMT rats. An alternative reason for the reduction in startle response could be that DMT raises sensory thresholds; but the failure to find a reduced distraction to the tone stimuli suggests that this is unlikely. Peripheral changes from the drug did not alter either the baseline lick rate nor the total water consumed by the rats tested in the distraction task.

The main effect of DMT was its impairment of 24 hr retention of both habituation of exploration and of habituation of distraction. DMT also impaired 24 hr retention of habituation of startle [5], if an 8 sec interstimulus interval was used in habituation, but not if a 30 sec interstimulus interval was used. It was suggested that DMT retarded the transfer of information from a short-term to a long-term store, but that this impairment would not be found if the transfer could occur within a trial. An interstimulus interval of 30 sec was sufficient for this to occur for startle responses, but habituation of distraction was impaired with intervals greater than 60 sec. The complexity of the task might be critical to whether or not there is an impairment of retention since this was found in Experiment 1 for rats habituated with objects present, but not for those habituating in the absence of objects.

DMT impairs long-term storage of the three types of behavioural habituation so far investigated. But it would be necessary to examine its effects in a variety of learning situations in order to determine whether this impairment is specific to habituation.

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