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BRIEF COMMUNICATION

Scopolamine Reverses Haloperidol-Attenuated Lever-Pressing for Water But Not Haloperidol-Attenuated Water Intake in the Rat

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LJUNGBERG, T. *Scopolamine reverses haloperidol-attenuated lever-pressing for water but not haloperidol-attenuated water intake in the rat.* PHARMACOL BIOCHEM BEHAV **29**(1) 205–208, 1988.—The operant lever-pressing response has previously (Ljungberg, *Pharmacol Biochem Behav* **27**: 341–350, 1987) been found to be inhibited by lower doses of haloperidol than the corresponding consummatory act, i.e., water intake. In the present study it was found that the attenuation of the lever-pressing response caused by the neuroleptic, but not the attenuation of the water intake, could be counteracted by scopolamine. The results support the notion that blockade of operant responding by low doses of neuroleptics are probably related to the extra-pyramidal side-effects of neuroleptics seen in the clinic, as both phenomena can be counteracted by anticholinergics. These results therefore conflict with the anhedonia hypothesis put forward as an explanation of the attenuating effects of neuroleptics in operant settings. The findings also have a clear bearing on the role of dopamine in feeding and drinking behavior, as the results implies that different aspects of the control of water intake (i.e., the operant vs. the consummatory phase) are governed by different mechanisms in the CNS.

Dopamine Haloperidol Operant behaviors Consummatory acts Anticholinergics Extrapyramidal side-effects Rats

ONE of the major problems with the clinically used antipsychotic drugs of the neuroleptic type is their tendency to induce extra-pyramidal side effects (EPS), such as acute dystonia and parkinsonism (see e.g. [4]). The induced EPS are routinely treated with anti-cholinergic drugs, which are considered to reduce the EPS more than the antipsychotic effect [4,8].

Several different types of animal models are used to screen for potential new antipsychotic drugs [12, 13, 17, 19, 24, 31] and it has been suggested that those effects of the neuroleptic drugs that can be counteracted by anticholinergics in animal models are more related to their propensity to induce EPS in the clinic than to their antipsychotic effect [3, 21, 27]. It has accordingly previously been shown that the catalepsy induced by neuroleptics, the antagonism of the apomorphine- and amphetamine-induced stereotyped behavior and the antagonism of the rotational behavior produced by dopamine agonists in unilateral 6-OHDA-lesioned animals can all be reversed by anticholinergics [2, 3, 9, 14, 18, 20–22, 25, 27, 28].

Neuroleptic drugs in low doses have also previously been shown to attenuate both positively and negatively reinforced operant behaviors, such as conditioned avoidance responses (CAR) and intra-cranial self-stimulation (ICSS), (see e.g., [5, 10, 15, 29]). The exact mechanism behind this effect is not known. Wise [29] has proposed that it may be caused by an attenuation of the ability of the animals to react to reinforcers, related to the antipsychotic effect of the neuroleptics. Alternatively, it may be caused by a motor deficit related to the EPS induced in the clinic (see discussion by [5, 10, 15, 29]): Reports showing that both the attenuation of CAR and ICSS caused by neuroleptics can be counteracted by anticholinergics [3, 6, 7, 11, 12, 21, 23, 25, 27, 28] support the latter suggestion.

In a recent publication [15] we have described the development of a new model system where the effects of neuroleptic drugs on both operant responding for water and water intake (i.e., the consummatory act) can be investigated in parallel experiments. We found that the neuroleptic drugs more potently attenuated the operant response (in this case lever-pressing) than the water intake (the corresponding consummatory act) similarly to how neuroleptic drugs at-

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tenuate the avoidance response more potently than the escape reaction in the CAR [1].

In order to further evaluate this new model we have, in the present study, investigated the ability of an anticholinergic drug, scopolamine, to counteract the decrease in the operant responding and the consumatory water intake caused by the dopamine-2 receptor blocker haloperidol [26]. This knowledge is of great importance both as an evaluation of this model to predict antipsychotic activity versus EPS and as a further investigation of the mechanisms behind the attenuation of operant responding caused by low doses of neuroleptics.

METHOD

Animals

The experiments were performed on 64 (30 and 34, respectively, in the two experimental paradigms described below) male Sprague Dawley rats (ALAB, Stockholm) which arrived at the animal colony at least one week prior to the start of the experiments. During the experiments, the animals were housed singly under conditions of controlled temperature and humidity on a 12 hour light/12 dark schedule (light on 7 a.m.–7 p.m.) with food ad lib. As well as getting water during the daily 45 minutes-long experimental session, the animals also had access to water for 15 minutes in their home cage 60 minutes after the end of their experimental session. During the rest of the day the animals had no access to water. With this water restriction schedule the animals are highly motivated to work during the experimental session but still can drink enough per day to gain in weight and to remain in good condition [15]. During weekends the animals had free access to water. The weight of the animals was 220–350 g.

Apparatus

All experiments were performed in slightly modified Skinner boxes (length=30 cm, width=20 cm, height=20 cm). The boxes were placed inside sound-protecting boxes equipped with one-way observation windows. Electric fans ventilated the boxes and provided a constant background noise in the boxes.

In the boxes where water intake was tested, the levers and the dipper mechanisms were removed and water nipples connected to a small water container were mounted in place of the dipper cups. The animal thus only needed to lick the nipple to obtain the water, not to perform or learn any operant response. The total amount of water consumed was monitored.

To test the ability to perform the lever-pressing response a specially developed lever was used which was fitted to one of the end walls, next to the centrally positioned dipper cup. The lever resembled a mill-wheel. The four wings were 4 cm long and 3 cm wide and made out of 5 mm black plastic. One lever-press was defined as one quarter of a turn of the wheel, which was signalled to the animal as a distinct click and with a sudden and transient drop in resistance, and resulted in the delivery of a "reward" (see below). The weight necessary to turn the lever was set to 20 g (for a further description of the lever, see [16]). A dipper of standard type, operated by a solenoid, delivered 0.05 ml water every time it was activated. The accumulated number of lever presses obtained during a session was printed out after every 5 minute period during the session.

Experimental Procedure

The data are calculated and presented as described in greater detail elsewhere (see [15]). In short, the animals were their own controls and each animal was used in only one of the two experimental paradigms. When they had reached a stable baseline response, they were injected with the drug vehicle alone for 1 to 2 days and then on the following day tested with the drug. The baseline response (called "control end value") for every animal was calculated by taking the median value of all the responses shown (total amount of water consumed or total number of lever-pressing responses) on each day of control injections. To get a measure of the effect of a drug treatment, we calculated the total responding after drug injection for every animal as a percentage of its own "control end value." The mean for a given treatment was then calculated and used as a measure of the effect of that treatment. We feel justified in calculating the mean in this way since the drugs were administered in a random order and since no effect was found as a result of the previous injections. No animal was tested with drug on more than three occasions, nor with the same dose(s) more than once and at least one week elapsed between each drug test. The different doses were given in a random order.

For control injections (shown as 100% in the figure) the mean total number of lever presses performed during a session was 346 (n=8) and the mean total amount of water consumed during a session was 16.3 ml (n=8).

Drug Treatment

Haloperidol (Leo, Sweden) was dissolved in 1% lactic acid and scopolamine-hydrochloride (Sigma) was dissolved in saline. The doses of haloperidol refer to the above mentioned form, while the doses of scopolamine refer to the base. Both drugs were injected subcutaneously in the flank in a volume of 1 ml/kg. Scopolamine was administered 60 and haloperidol 30 minutes before the start of the experiments.

The doses of haloperidol used (i.e., 0.1 mg/kg in the lever-pressing experiment and 0.4 mg/kg in the water intake experiment) were selected from a previous study [15] to cause a similar degree of inhibition in both the experimental paradigms.

Statistics

Because of the design of the experiment, with the main emphasis to test whether the inhibition caused by haloperidol could be fully counteracted by scopolamine, the degree of significance was tested for each treatment against control performance. The level of significance was tested using the Student's t-test for paired samples, corrected for multiple comparisons by the Bonferoni method [30]. By this method the p value of the Student t-test is adjusted according to the formula $p^*=p/m$, where p^* is the adjusted value, p is the p level set by the researcher (in this case 0.05; two-tailed), and m is the number of comparisons.

RESULTS

As described previously [15] haloperidol 0.1 mg/kg was found to significantly reduce the lever-pressing response ($p<0.05$; n=8) while a higher dose of haloperidol (0.4 mg/kg) was required to reduce the water intake to a similar degree ($p<0.05$; n=8), (see Fig. 1). Over a wide dose range scopolamine reduced both the lever-pressing response and

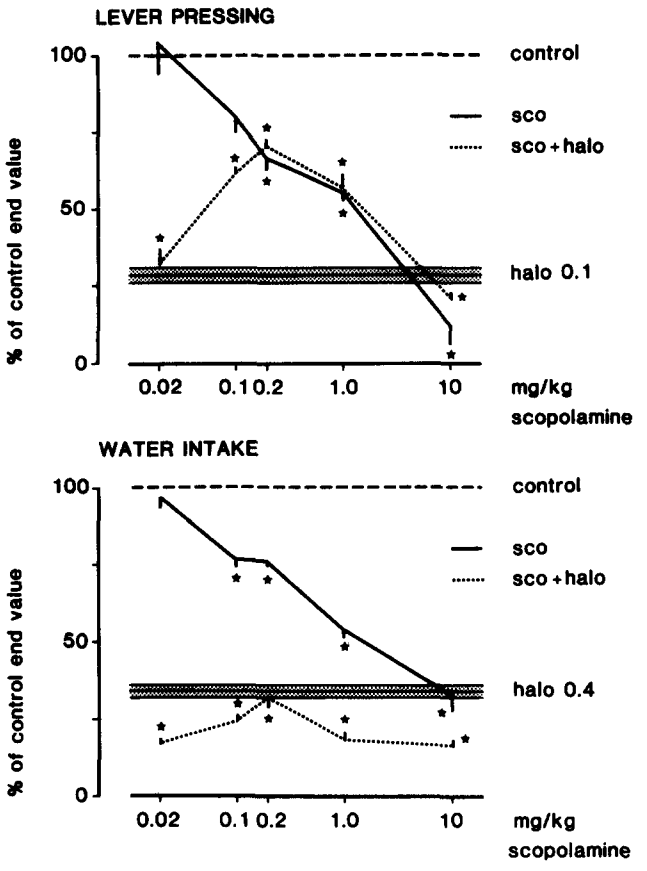

FIG. 1. Haloperidol (halo) 0.1 mg/kg (lever-pressing response; n=8) and 0.4 mg/kg (water intake; n=8) causes a significant reduction in the performance. The blockade of the lever-pressing response can be partially counteracted by scopolamine (sco), but not the blockade of the water intake. Scopolamine by itself causes an unspecific decrease in both the lever-pressing response and the water intake, thereby probably limiting the magnitude of the reversal of the haloperidol blockade. The data are shown as mean±S.E.M. and level of significance is tested against control performance using Student's *t*-test for paired samples, corrected for multiple comparisons by the Bonferoni method. Each data point for scopolamine and haloperidol plus scopolamine contains 5–6 animals.

the water intake to a similar degree (see Fig. 1), indicating a nonspecific effect [15].

Over a limited range of doses, scopolamine was found to partially counteract the blocking effect of haloperidol in the lever-pressing response but not in the water intake paradigm (see Fig. 1). It was not possible to restore the lever-pressing response to 100% of control values, probably because of the nonspecific effects of scopolamine itself as it was possible to reverse the effect only as far as the level produced by scopolamine alone (with the same time-curve as well; data not shown).

DISCUSSION

The results obtained in the present study are in good agreement with previous studies showing that pretreatment with anticholinergics can counteract the attenuation of operant responding caused by neuroleptics, as discussed in the introduction. One difference is, however, that both the blockade of the avoidance response and the escape reaction have been described to be counteracted by anticholinergics [27], while in the present study only the lever-pressing response was found to be reversed by scopolamine, not the unconditioned water intake.

As was also mentioned in the introduction, it is widely held that effects of neuroleptics in animal models that can be counteracted by anticholinergics reflect the ability of these drugs to induce EPS in the clinic rather than an antipsychotic potential. It has therefore been proposed that the attenuation of the CAR or ICSS by neuroleptics in animals is caused by a motor deficit, as both effects can be reversed by anticholinergics [7,27].

If blockade of operant responding is related to the antipsychotic effect in the clinic and blockade of consummatory water intake is related to the EPS, one would have expected the opposite effect of scopolamine to what was actually found, i.e., an effect on the consummatory, but not on the operant, behavior. Our data thus support the notion that the blockade of the operant responding is in some fashion more related to the propensity of neuroleptic drugs to induce EPS than to their antipsychotic effect.

The findings that the operant lever-pressing was counteracted by scopolamine while the unconditioned water intake was not could tentatively be explained by the fact that a higher dose of haloperidol was used to reduce the water intake. An alternative explanation might however be that this is instead a reflection of the fact that different aspects of the control of the food and water intake (i.e., the operant vs. the consummatory phase) is governed by different mechanisms in the CNS. Our additional findings that the neuroleptic dose-response characteristics ([15] and Ljungberg, in prep.) as well as the effects of chronic neuroleptic treatment (Ljungberg, in prep.) differs between the lever-pressing response and the water intake makes us favour the latter explanation.

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