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Involvement of NMDA and AMPA/KA receptors in the nucleus accumbens core in instrumental learning guided by reward-predictive cues

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Abstract

The use of reward-predictive cues to guide behavior critically involves the nucleus accumbens. However, little is known regarding the role of ionotropic glutamate receptors in the core subregion of the nucleus accumbens (AcbC) in instrumental learning guided by reward-predictive cues. Here we examined the effects of an intra-AcbC blockade of NMDA and AMPA/KA receptors on the acquisition of an instrumental response in a reaction time (RT) task in rats. In this task, discriminative cues signaled in advance the upcoming reward magnitude (5 or 1 food pellet) associated with a lever release. During early acquisition (days 1–6) rats received daily bilateral injections of either the NMDA receptor antagonist AP5 (5.0 μg per side, $n = 14$), the AMPA/KA receptor antagonist CNQX (2.5 μg per side, $n = 14$) or vehicle (0.5 μL per side, $n = 19$). No treatment was given during late acquisition (days 7–12). The main result was that rats which received intra-AcbC injections of AP5 or CNQX during early acquisition exhibited a general RT increase of responses to high and low reward. However, treatment with AP5 and CNQX did not interfere with discriminative guidance of RTs by cue-associated reward magnitudes, i.e. during acquisition RTs of responses to expected high reward became significantly faster than RTs of responses to expected low reward. Our findings suggest that NMDA and AMPA/KA receptors in the AcbC play a critical role in invigorating responding during instrumental learning, but seem less important in guiding responding according to reward-predictive cues.

Introduction

The expectancy of reward is an important factor in guiding instrumental behavior (Gold, 2003). This notion is supported by findings that expected reward magnitudes determine the speed of instrumental responses. In rats, reaction times (RTs) of conditioned responses were shortest to the instructive cue predictive of the highest food reward (Brown & Bowman, 1995; Hauber *et al*., 2000, 2001). Likewise, in primates, RTs of conditioned reaching or saccadic eye movements decreased with increasing attractiveness of rewards predicted by instructive cues (Hollerman *et al*., 1998; Kawagoe *et al*., 1998).

The nucleus accumbens (Acb) represents an interface between limbic and motor structures (Mogenson *et al*., 1980) and plays a critical role in the acquisition and expression of instrumental responses to rewarding stimuli (e.g. Ikemoto & Panksepp, 1999; Cardinal *et al*., 2002; Kelley, 2004). Electrophysiological studies demonstrate that reward-predictive cues induced reward-related activations in striatal neurons (Apicella *et al*., 1991; Schultz *et al*., 1992; Kawagoe *et al*., 1998; Carelli *et al*., 2000; Setlow *et al*., 2003; Nicola *et al*., 2004). Notably, these neurons represent information about the nature of the expected behavioral outcome, because task-related neuronal activations were influenced by the types of upcoming reward (Hollerman & Schultz, 1998; Schultz *et al*., 2003). Furthermore, behavioral studies revealed that the Acb subserves instrumental behaviors elicited by cues predicting natural or drug reward (Everitt & Wolf, 2002; Kelley & Berridge, 2002). Structures such as amygdala and prefrontal cortex which encode the incentive value of reward-predictive cues (Thorpe *et al*., 1983; Schoenbaum & Eichenbaum, 1995; Rolls *et al*., 1996; Schoenbaum *et al*., 1999; Tremblay & Schultz, 1999) relay reward-related information via direct glutamatergic projections to medium spiny neurons in the Acb (Groenewegen *et al*., 1996). These neurons express both NMDA and non-NMDA receptors (Albin *et al*., 1992) and project to downstream structures such as the ventral pallidum that control adaptive motor behavior (Zahm, 2000).

NMDA and AMPA/KA receptors in the Acb play a critical role in reward-seeking under control of reward-associated cues (Di Ciano *et al*., 2001). For instance, we examined the contribution of intra-Acb AMPA/KA and NMDA receptors in reward-directed behavior using an RT task in which RTs of an instrumental response are a function of the expected food reward magnitude signaled in advance by discriminative cues (Hauber *et al*., 2000). In rats well trained on this task, an ionotropic glutamate receptor blockade in the Acb produced a general increase of RTs, but left RT guidance by expected reward magnitudes unimpaired, i.e. RTs were still shorter if the expected reward was high (Giertler *et al*., 2003). However, these earlier studies from our laboratory focused on the role of intra-Acb ionotropic glutamate receptors in performance rather than acquisition of instrumental behavior guided by reward-predictive cues. Here we investigated whether an NMDA or AMPA/KA receptor blockade in the core subregion of the ACB (AcbC) impaired acquisition of an instrumental response by naïve rats in the task described above.

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Materials and methods

Animals

Forty-seven male Sprague–Dawley rats (Charles River, Sulzfeld, Germany) were housed in groups of up to six animals in transparent plastic cages (36 × 52 × 25 cm; Ferplast, Nürnberg, Germany). Temperature (20 ± 2 °C) and humidity (50 ± 10%) were kept constant in the animal house. A reversed 12 : 12-h light–dark schedule was used (lights on between 19:00 and 07:00 h) with testing in the dark phase. Rats were given *ad libitum* access to water; food was restricted to 15 g per animal per day. On days without behavioral testing, rats received 15 g of standard laboratory maintenance chow (Altromin, Lage, Germany). On days with behavioral tests, rats received in the testing apparatus 9.5-g food pellets as reward (45-mg pellets, Bioserv, Frenchtown, USA). On these days, the amount of standard laboratory chow given was reduced to 5.5 g per animal. Rats weighed 200–250 g on arrival and 270–350 g at the time of surgery. All animal experiments were conducted according to the German Law on Animal Protection and were approved by the proper authorities in Stuttgart, Germany.

Surgery

For stereotaxic surgery, animals were anesthetized with sodium pentobarbital (50 mg/kg, i.p.) (Sigma-Aldrich, Taufkirchen, Germany) following pretreatment with atropine sulphate (0.05 mg/kg, i.p.) (Sigma-Aldrich) and secured in a Kopf stereotaxic apparatus (Kopf Instruments, Tujunga, USA). Bilateral 15.5-mm stainless steel guide cannulae with an outer diameter of 0.8 mm were aimed at the AcbC and implanted using standard stereotaxic procedures. The coordinates, with reference to the atlas of Pellegrino *et al*. (1981) (toothbar 5 mm above the interaural line), were: 3.4 mm anterior to bregma, ±1.9 mm lateral to midline, and 5.3 mm ventral from the skull. The guide cannulae were occluded by stainless steel stylets. Each rat was given at least 7 days to recover from surgery before the experiment was started.

Drug injection

On injection days, the stainless steel stylets were removed and bilateral injection cannulae with an outer diameter of 0.45 mm (length: 18 mm) were lowered at the final site of injection (−7.8 mm from skull) and attached via polyvinylchloride tubing to microliter syringes controlled by a syringe pump (Med Associates, St Albans, VT, USA). The competitive AMPA/KA receptor antagonist 6-cyano-7-nitroquinoxaline-2,3-dione (CNQX, disodium salt; Biotrend, Köln, Germany) (2.5 μg in 0.5 μL saline) and the competitive NMDA receptor antagonist DL-2-amino-5-phosphonovaleric acid (AP5; Biotrend) (5 μg in 0.5 μL saline), as well as vehicle (0.5 μL saline) were delivered bilaterally over a 1-min interval. Injection cannulae were left in position for a further 1 min after injection to allow for diffusion. After injection, a rat was placed back and remained in its home cage for an additional 5 min before being placed in the test chamber.

Apparatus

Six operant test chambers (24 × 21 × 30 cm) (Med Associates) were used. Test chambers were placed in separate sound-attenuating cubicles with fans providing a constant low level of background noise. Each chamber was supplied with a retractable lever, and two stimulus lights, one above the retractable lever, the other above the food receptacle. Each food receptacle was equipped with an infrared head entry detector. The experiments were controlled online by a Windows 98™-based computer system equipped with SmartControl® Interfaces and the MedPC™-Software (Med Associates).

RT task

A modified version of a simple RT task used in previous studies (Hauber *et al*., 2000, 2001) was employed. The task demands conditioned lever release with instructive stimuli indicating the reward magnitude to be obtained after a subsequent imperative stimulus. Rats had to press the lever and to wait for the imperative stimulus which was provided by the stimulus light above the lever after a foreperiod of 0.3 s. The imperative stimulus signaled to the rats to release the lever quickly and to respond to the food receptacle in which the food pellets were delivered (45-mg pellets, Bioserv). On each correct trial, the rats received either one or five food pellets. The number of pellets for each trial was pseudo-randomly determined in advance and was signaled to the rats by two distinct brightness levels of the cue lights which provide the instructive stimuli. After the intertrial interval of 3 s, the instructive stimulus was turned on at the beginning of each trial 3 s before lever insertion and remained present until delivery of the food reward. To check for equal perception of instructive stimuli of the two different brightness levels, for 50% of the rats, a bright stimulus was associated with delivery of five pellets and a dim stimulus was associated with delivery of one pellet. For the other 50% of the rats, the opposite pattern was used.

RT, defined as latency from the onset of the imperative stimulus to lever release, and movement time (MT), defined as latency from lever release to photobeam disruption in the food receptacle, were recorded with an accuracy of < 10 ms. For a correct trial, animals had to release the lever within RT < 2 s after presentation of the imperative stimulus. Responses before onset of the imperative stimulus presentation were defined as 'early' responses, and responses with RT = 2 s were defined as 'late' responses. A daily individual session demanded 50 correct trials, i.e. 25 correct trials for each reward magnitude (one and five pellets). A scheme on the order of trial events is given in Fig. 1.

Experimental procedure

Preoperative habituation

In the first two sessions, subjects were habituated to the operant chamber with access to food pellets placed into the food receptacle. In the following five sessions, a habituation program with an fixed ratio (FR) 1 schedule commenced until a criterion of 20 consecutive lever responses was attained. Afterwards, rats were subjected to surgery. To habituate animals to handling during injections, all animals were exposed to the handling procedure before each habituation session.

Acquisition

After postsurgery recovery, all animals received a single intra-AcbC injection of vehicle (0.5 μL saline) to adapt them to injections. Three days later, the experiment was started with one daily session over 12 days to investigate acquisition of the RT task described in Fig. 1. On days 1–6, each rat received injections of vehicle ($n = 19$), AP5 ($n = 14$) or CNQX ($n = 14$) before the onset of behavioral testing. On days 7–12, all animals received a sham injection procedure including handling, insertion of injection cannulae dummies and operation of the injection pump (without running an injection) before the onset of behavioral testing.

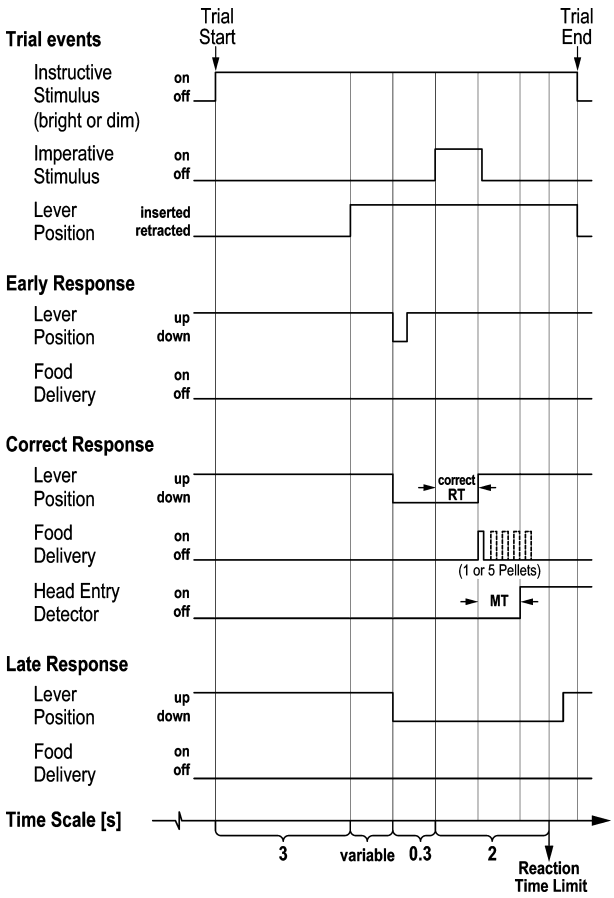


FIG. 1. Schematic representation of the order of trial events. At the beginning of a trial (after the intertrial interval of 3 s), the instructive stimulus delivered by a cue light above the food receptacle was turned on at one of two brightness levels, which were associated with different reward magnitudes (one or five pellets). After 3 s, the lever was inserted. Thereafter, the rat pressed the lever spontaneously. After the foreperiod of 0.3 s, the imperative stimulus provided by a cue light above the lever signaled the animal to release the lever in order to get the food reward. Responses with RT < 2 s were considered as being correct and were rewarded as indicated by the instructive stimulus. Early responses initiated before the onset of the imperative stimulus or late responses (RT = 2 s) caused the trial to be repeated with the identical foreperiod and reward magnitude.

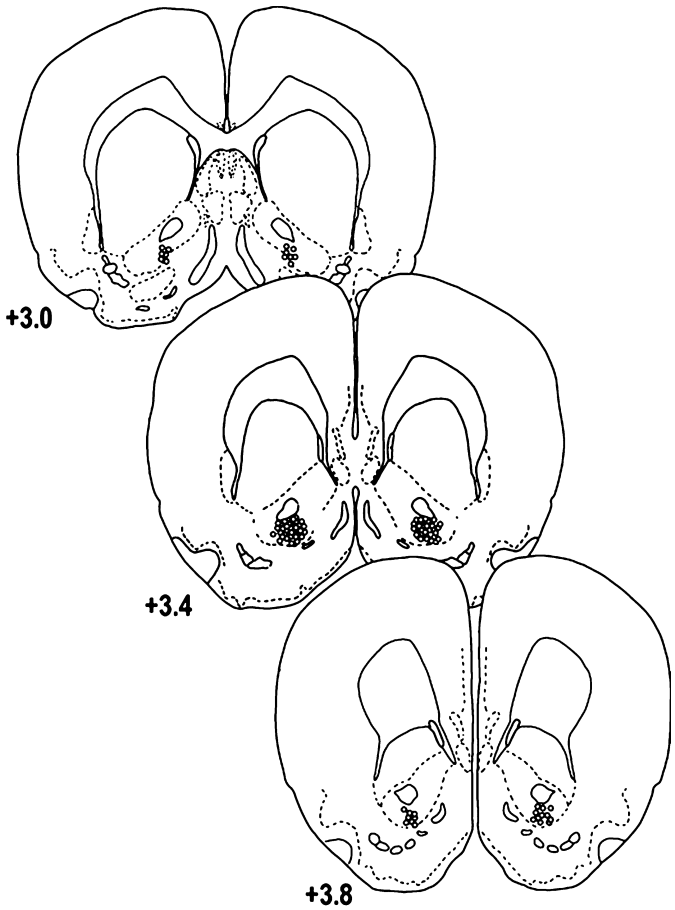


FIG. 2. Location of cannulae tips in the AcbC. The schematics depict the location of cannulae tips (•) in the AcbC for all rats. Plates are adaptations from the atlas of Pellegrino *et al.* (1981). Numbers beside each plate correspond to millimeters anterior to bregma.

Data analysis

Brightness levels of the instructive stimulus were perceived equally, as shown in earlier studies (e.g. Bohn *et al.*, 2003a): for a given reward magnitude level, mean accuracy and RT/MT values obtained with a bright or a dim stimulus did not differ significantly (data not shown). Therefore, response measures for a given reward magnitude obtained with bright and dim instructive stimuli were pooled.

Data were expressed as means ± standard error of the mean (SEM). Accuracy of performance was determined as a percentage from the proportion of correct trials from the overall number of trials (early + correct + late) necessary to reach the criterion of 25 correct responses for each stimulus–reward magnitude relationship [100 × correct responses/(early + correct + late responses)]. Furthermore, the number of early and late responses is given as a function of reward magnitude.

The calculations on RT and MT performance were conducted with data from correct trials (RT < 2 s). When averaging RT and MT data, a geometric mean was calculated for each rat for each session, as the geometric mean is less influenced by outlying data points than is the arithmetic mean. Overall, RT and MT means of responses associated with the high and low reward magnitude represent the arithmetic average of the geometric means of individual rats (Brasted *et al.*, 1997).

Treatment effects during initial acquisition (days 1–6) on accuracy of performance, early and late responses as well as on RTs and MTs of correct responses were assessed by between- subjects comparisons of groups treated with vehicle, AP5 and CNQX. Data were analysed using an analysis of variance (ANOVA) with treatment as between-subjects factor, reward magnitude and days as within-subjects (repeated measures) factors followed by planned contrast analyses. Accuracy of performance, early and late responses, RTs and MTs of correct responses during late acquisition (days 7–12) were compared using separate ANOVA with pretreatment (vehicle, AP5 and CNQX pretreatment on days 1–6) as between-subjects factor and reward magnitude and days as within-subjects (repeated measures) factors followed by planned contrast analyses. All statistical computations were carried out with STATISTICA^{TM} (version 5.5, StatSoft®, Inc., Tulsa, USA). The level of statistical significance (α-level) was set at $P < 0.05$.

Histology

After completion of behavioral testing, animals were killed with an overdose of sodium pentobarbital (150 mg/kg, i.p.) (Sigma-Aldrich) to control for correct placement of cannulae. Brains were rapidly removed, fixed in 10% formalin for 2.5 h and stored in 30% glucose.

Brain sections (30 μm) were cut with a cryostat (Reichert & Jung, Heidelberg, Germany), mounted on coated slides and stained with cresyl violet. Cannulae placements were verified with reference to the atlas of Pellegrino *et al*. (1981). In all animals, cannulae tip placements deviated less than 0.5 mm from target coordinates in the AcbC; no animal had to be excluded due to cannulae misplacement. The locations of cannulae tips for all rats are represented in Fig. 2. The exact amount of spread of drugs from the site of infusion in the AcbC is not known as studies with radiolabelled AP5 and CNQX would be necessary. It is likely that the behavioral effects determined here largely reflect drug actions within the AcbC, because core vs. shell microinfusions of AP5 (Di Ciano & Everitt, 2001) or CNQX (Wan & Swerdlow, 1996) in the same volume as used here induced distinct behavioral effects.

Results

Correct responses

Rats in all treatment groups rapidly learned the task within 1–6 days (Fig. 3). In line with previous studies with similar tasks (Brown & Bowman, 1995; Hauber *et al*., 2000; Hauber *et al*., 2001; Giertler *et al*., 2003) the per cent rate of correct responses gradually increased and eventually reached about 75–80%, i.e. rats needed approximately 60 responses to achieve the criterion of 50 correct responses per session, 25 for each reward magnitude. ANOVA on the rate of correct responses over days 1–6 indicated no main effect of treatment, but significant main effects of days ($F_{5,220} = 4.25$, $P < 0.002$) and reward magnitude ($F_{1,44} = 31{,}22$, $P < 0.001$) as well as a significant day × treatment interaction ($F_{10,220} = 2.80$, $P < 0.003$). Rats in all treatment groups tended to have lower correct response rates, if high reward is expected. This is largely due to an increased number of early responses for expected high reward (see below). Pretreatment during days 1–6 had no long-term effects on the accuracy of performance. A separate ANOVA on the rate of correct responses over days 7–22 revealed a significant main effect of reward magnitude only ($F_{1,44} = 39.81$, $P < 0.001$).

The number of early errors depended on the expected reward magnitude and tended to be higher, if a large reward was expected, as shown in Fig. 4. ANOVA on the number of early responses over days

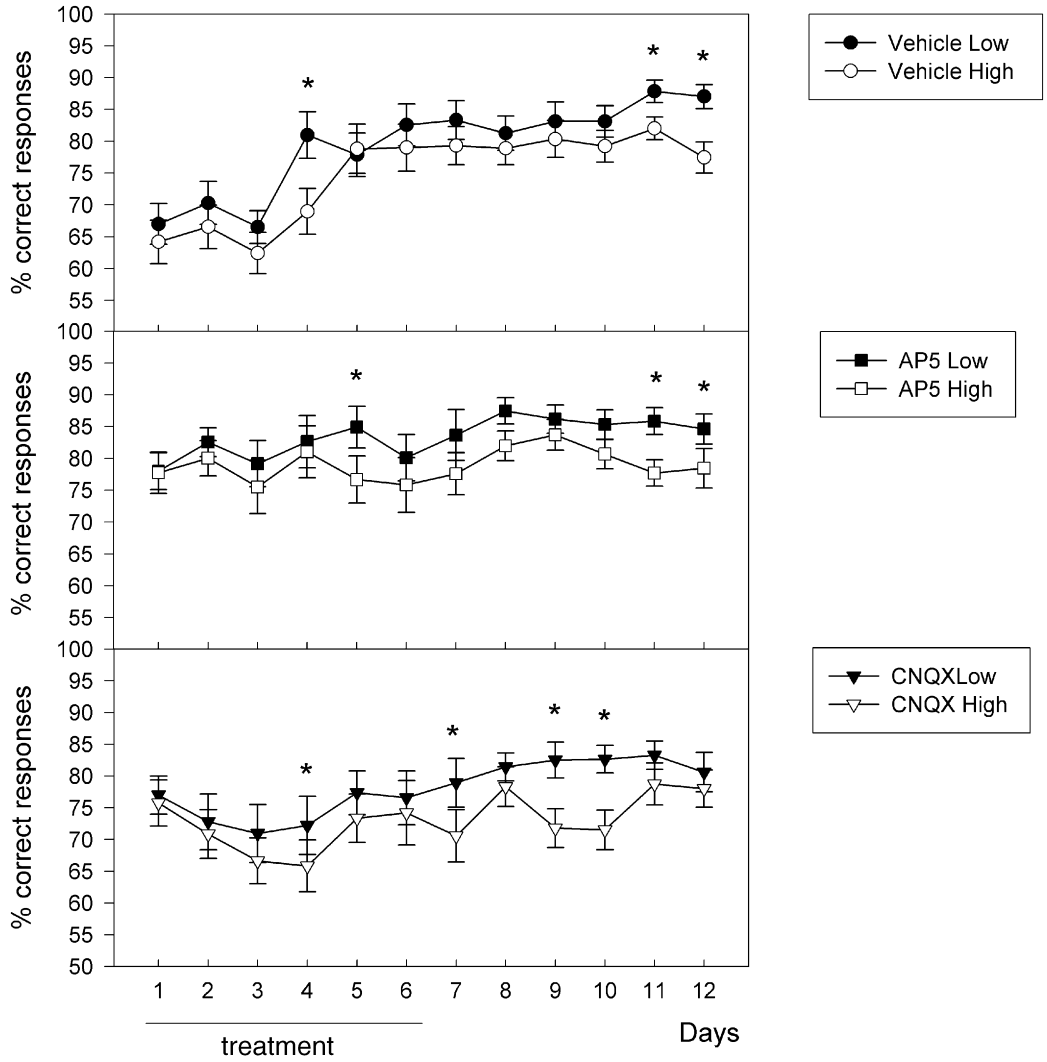


FIG. 3. Accuracy of performance measured as proportion of correct responses from the overall number of responses (early + correct + late) to reach criterion (50 correct responses) over 12 days of acquisition. Treatment groups received intra-AcbC injection of vehicle (0.5 μL, $n = 19$), AP5 (5 μg in 0.5 μL, $n = 14$) or CNQX (2.5 μg in 0.5 μL, $n = 14$) on days 1–6. No injections were given on days 7–12. Correct responses are shown as a function of expected reward (low: one pellet; high: five pellets). In all treatment groups, correct response rate was lower with expected high reward. Significant main effect of reward magnitude was further analysed by planned contrasts for each day (*$P < 0.05$).

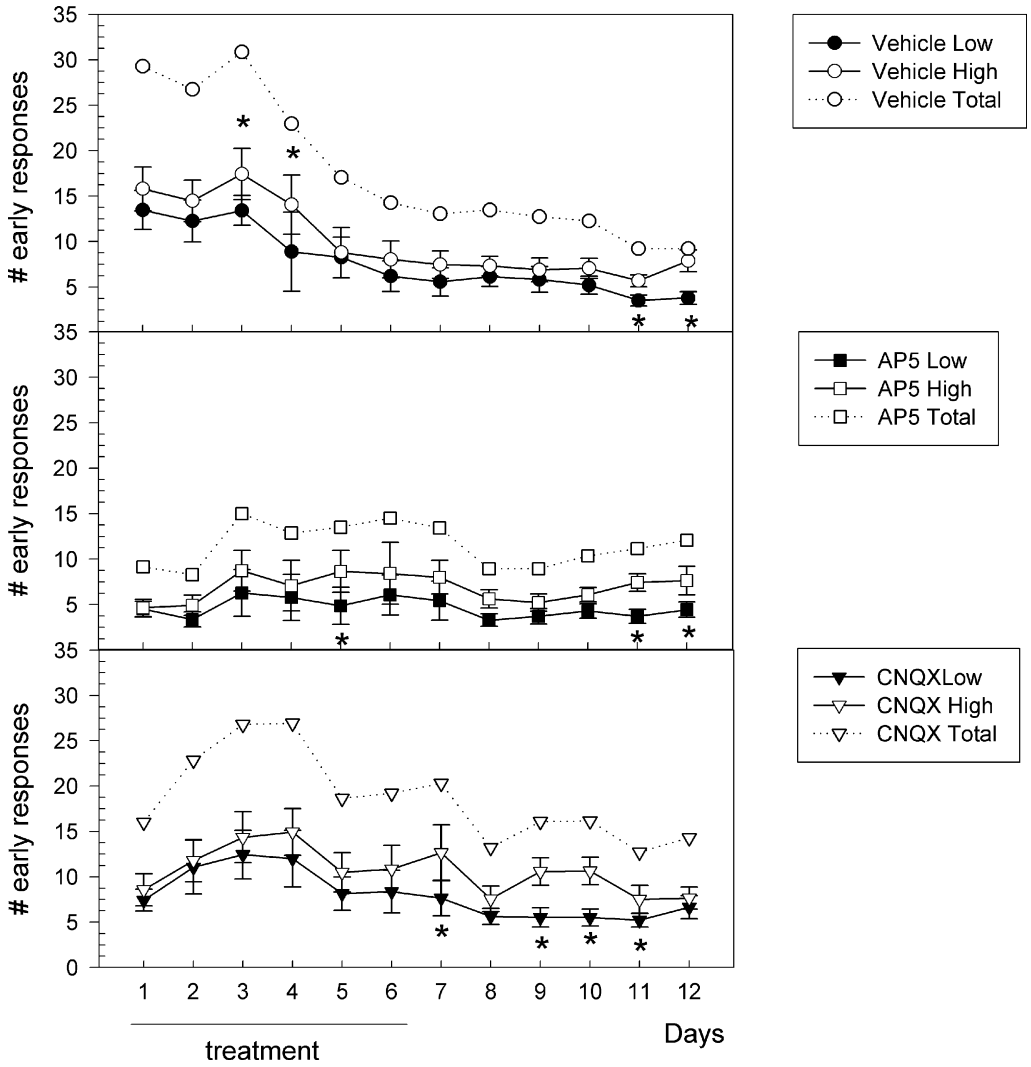


FIG. 4. Number of early responses over 12 days of acquisition. Treatment groups received intra-AcbC injection of vehicle (0.5 μL, $n = 19$), AP5 (5 μg in 0.5 μL, $n = 14$) or CNQX (2.5 μg in 0.5 μL, $n = 14$) on days 1–6. No injections were given on days 7–12. Early responses are shown in total and as a function of expected reward (low: one pellet; high: five pellets). In all treatment groups, number of early errors was higher for expected high reward. Significant main effect of reward magnitude was further analysed by planned contrasts for each day (*$P < 0.05$).

1–6 indicated no main effect of treatment, but a significant main effect of days ($F_{5,220} = 2.44$, $P < 0.04$) and reward magnitude ($F_{1,44} = 28.44$, $P < 0.001$) as well as a significant day × treatment interaction ($F_{10,220} = 2.07$, $P < 0.03$). The rate of early responses was particularly low in vehicle-treated animals on days 1–3, resulting in low rates of correct responses. As vehicle-treated animals had particularly short RTs on days 1–3 this reflects most probably their initial tendency to respond very quickly. Starting from day 4, RTs increased and vehicle-treated animals seemed to adapt their response speed to the RT corridor defined as being correct. In contrast, CNQX- and particularly AP5-treated animals exhibited lower numbers of early responses, possibly reflecting a slowing drug-induced response, as indicated by their longer RTs. ANOVA on the number of early responses over days 7–22 revealed a significant main effect of reward magnitude ($F_{1,44} = 46.93$, $P < 0.001$).

Late errors were somewhat more frequent with responses for low reward (Fig. 5). Their number was very low in vehicle and CNQX-treated animals, but prominent in AP5-treated animals. ANOVA on the rate of late responses over days 1–6 indicated main effects of treatment ($F_{2,44} = 17.87$, $P < 0.001$), days ($F_{5,220} = 4.54$, $P < 0.001$) and reward magnitude ($F_{1,44} = 4.54$, $P < 0.04$) as well as a significant day × treatment interaction ($F_{10,220} = 2.66$, $P < 0.005$). An ANOVA on the rate of correct responses over days 7–22 revealed a significant main effect of reward magnitude ($F_{1,44} = 19.92$, $P < 0.001$).

RT performance

As shown in Fig. 6A, RT performance of vehicle rats was guided by expected reward starting on day 2. Mean RT increase by expectation of high reward on days 1–12 was about 60 ms. Blockade of intra-AcbC NMDA or AMPA/KA receptors had prominent effects on RTs of responses to expected low and high reward. Infusion of AP5 and, less pronounced, CNQX increased RT during initial acquisition, but left their guidance by expected reward largely unaffected. Figure 6A shows RTs as a function of reward magnitude for each treatment separately to highlight reward expectation effects; in Fig. 6B the same RT data are depicted as a function of treatment separately for each reward magnitude to highlight drug effects. ANOVA on RT over days 1–6 revealed significant main effects of treatment ($F_{2,44} = 16.16$, $P < 0.0001$), days ($F_{5,220} = 15.17$, $P < 0.0001$) and reward

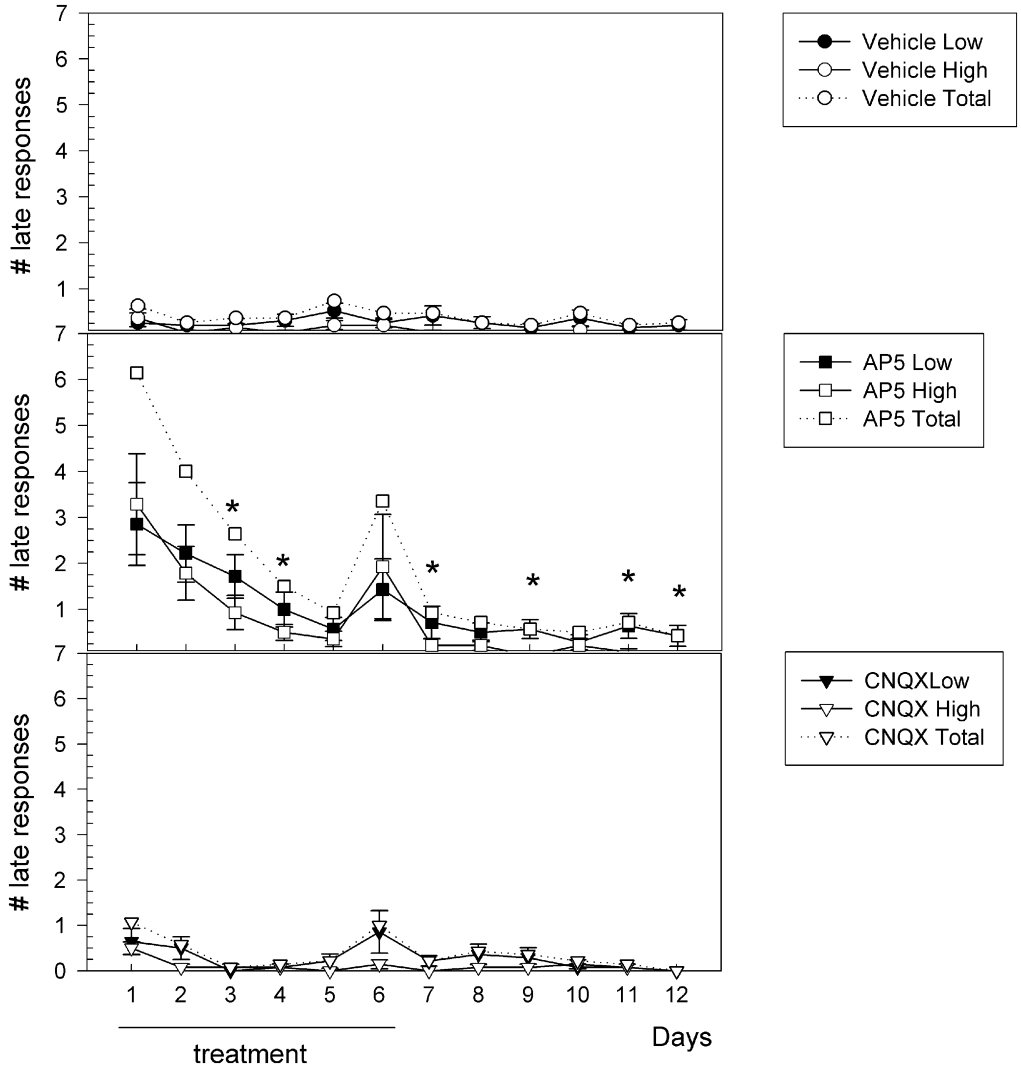


FIG. 5. Number of late responses over 12 days of acquisition. Treatment groups received intra-AcbC injection of vehicle (0.5 μL, $n = 19$), AP5 (5 μg in 0.5 μL, $n = 14$) or CNQX (2.5 μg in 0.5 μL, $n = 14$) on days 1–6. No injections were given on days 7–12. Late responses are shown in total and as a function of expected reward (low: one pellet; high: five pellets). In all treatment groups, number of late errors was higher for expected low reward. Significant main effect of reward magnitude was further analysed by planned contrasts for each day (*$P < 0.05$).

magnitude ($F_{1,44} = 32.58$, $P < 0.0001$) as well as significant treatment × day ($F_{10,220} = 10.45$, $P < 0.0001$) and reward magnitude × day ($F_{5,220} = 5.23$, $P < 0.0001$) interactions.

During late acquisition, RTs were also shorter for expected high reward. ANOVA on RT over days 7–12 indicated a significant main effect of reward magnitude ($F_{1,44} = 62.35$, $P < 0.0001$), but not of pretreatment and days.

MT performance

Figure 7A shows MTs as a function of reward magnitude separately for each treatment to highlight reward expectation effects; in Fig. 7B the same MT data are depicted as a function of treatment for each reward magnitude separately to highlight drug effects. MT gradually declined on days 1–6 in all treatment groups. As shown in Fig. 7A, in vehicle-treated animals MTs were significantly faster for expected high reward starting from day 2. This was in part also observed in CNQX-treated animals, but not in AP5-treated animals. Intra-AcbC blockade of NMDA or AMPA/KA receptors had no effects on MT (Fig. 7B).

ANOVA on MT over days 1–6 revealed significant main effects of days ($F_{5,220} = 58.63$, $P < 0.0001$) and reward magnitude ($F_{1,44} = 10.40$, $P < 0.003$). By contrast, no significant treatment effects or treatment–reward magnitude interactions were detected. ANOVA on MT over days 7–12 with pretreatment as between-subjects factor and reward magnitude and days as within-subjects (repeated measures) factors indicated significant main effects of days ($F_{5,220} = 6.33$, $P < 0.0001$) and reward magnitude ($F_{1,44} = 51.18$, $P < 0.0001$), but not of pretreatment. Furthermore, a significant pretreatment–reward magnitude interaction ($F_{2,44} = 4.816$, $P < 0.03$) was determined. The guidance of MT by reward magnitude during late acquisition was marked in vehicle-treated animals, but less pronounced in AP5- and CNQX-treated animals.

Correlational analysis

For a closer inspection of interrelations between response speed, reward magnitude and accuracy of performance, we performed a detailed correlational analysis of individual mean RTs for high and low reward from correct responses (from each session on days 1–6) and

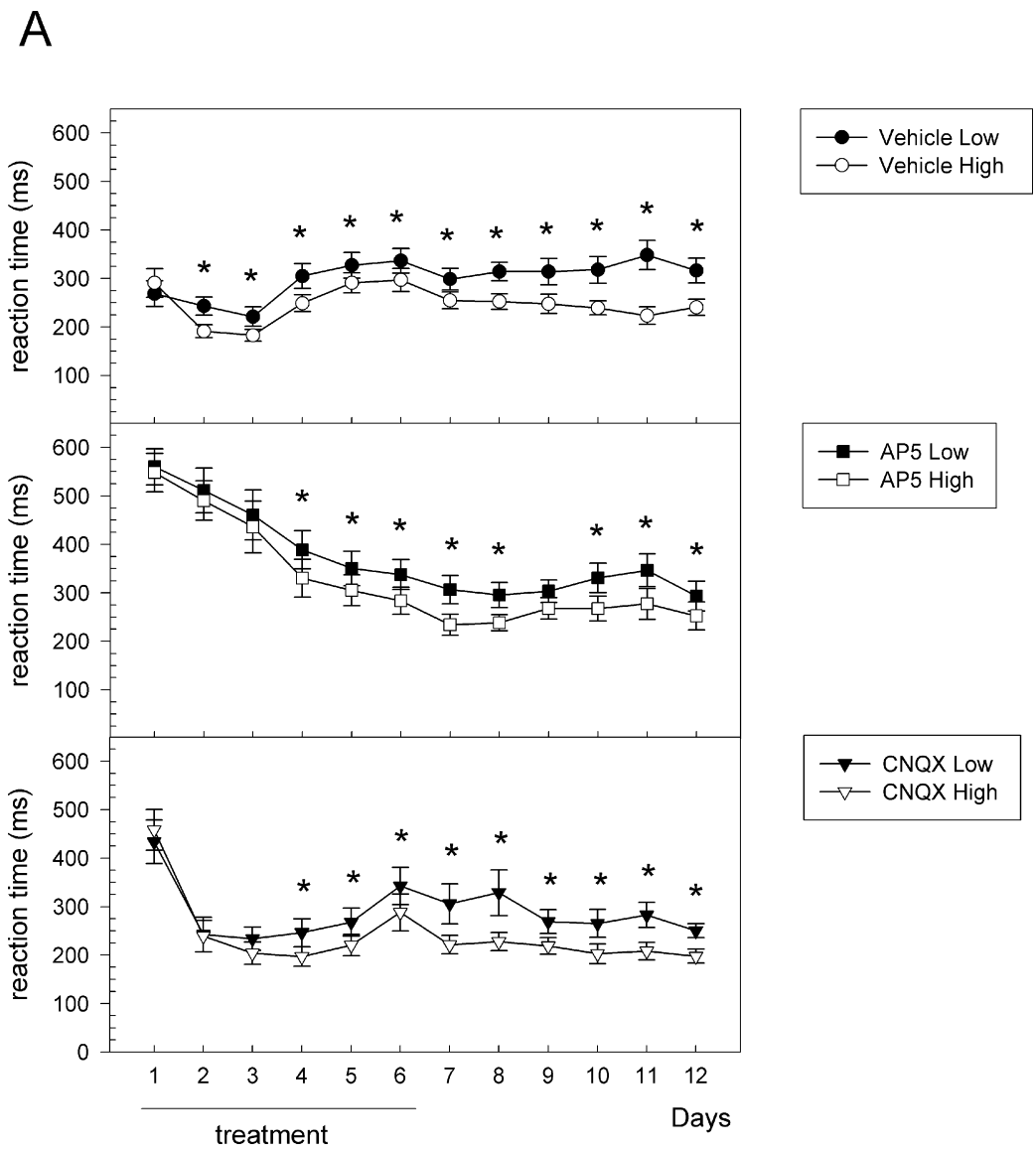


FIG. 6. RTs of correct responses. Treatment groups received intra-AcbC injection of vehicle (0.5 μL, $n = 19$), AP5 (5 μg in 0.5 μL, $n = 14$) or CNQX (2.5 μg in 0.5 μL, $n = 14$) on days 1–6. No injections were given on days 7–12. (A) RTs and their guidance by expected reward (low: one pellet; high: five pellets). In all treatment groups, RT was significantly guided by expected reward magnitudes; significant main effect of reward magnitude was further analysed by planned contrasts for each day (*$P < 0.05$). (B) The same RT data as in A are depicted as a function of treatment separately for expected low (top) and high (bottom) reward to highlight drug effects. Infusion of AP5 and CNQX increased RT on days 1–6; significant main effect of treatment was further analysed by planned contrasts for each day (*$P < 0.05$ vehicle vs. AP5; #$P < 0.05$ vehicle vs. CNQX).

associated rates of early, correct and late responses. Data revealed significant correlations in all treatment groups ($P < 0.05$, in each case) between individual mean RTs and the number of early responses, the proportion of correct responses as well as the number of late responses (Fig. 8). Correlation coefficients (Pearson's r) given in Fig. 8 indicate that correct response rates increased with longer RTs, while the number of early responses decreased with longer RTs. In addition, a weak positive correlation between the number of late responses and RT was found. In line with previous studies using comparable RT tasks (Hauber, 1996; Brasted *et al*., 1998), correlations between RTs and MTs were generally weak (data not shown).

Discussion

Here we examined the effects of an intra-AcbC blockade of NMDA and AMPA/KA receptors on acquisition of a RT task in which the upcoming reward magnitude (five vs. one food pellet) associated with an instrumental response was signaled in advance by discriminative cues. Results reveal that an intra-AcbC injection of AP5 or CNQX during early acquisition produced a general increase of RTs, but did not interfere with discriminative guidance of RTs by cue-associated reward magnitudes.

Role of AcbC glutamate receptors in discrimination of reward-predictive cues

In animals that received vehicle infusions on days 1–6, RTs were significantly guided by reward-predictive cues from day 2 onward, demonstrating rapid learning to discriminate cues and associated reward magnitudes. Surprisingly, in animals that were subjected to microinfusions of AP5 or CNQX, discrimination learning was largely

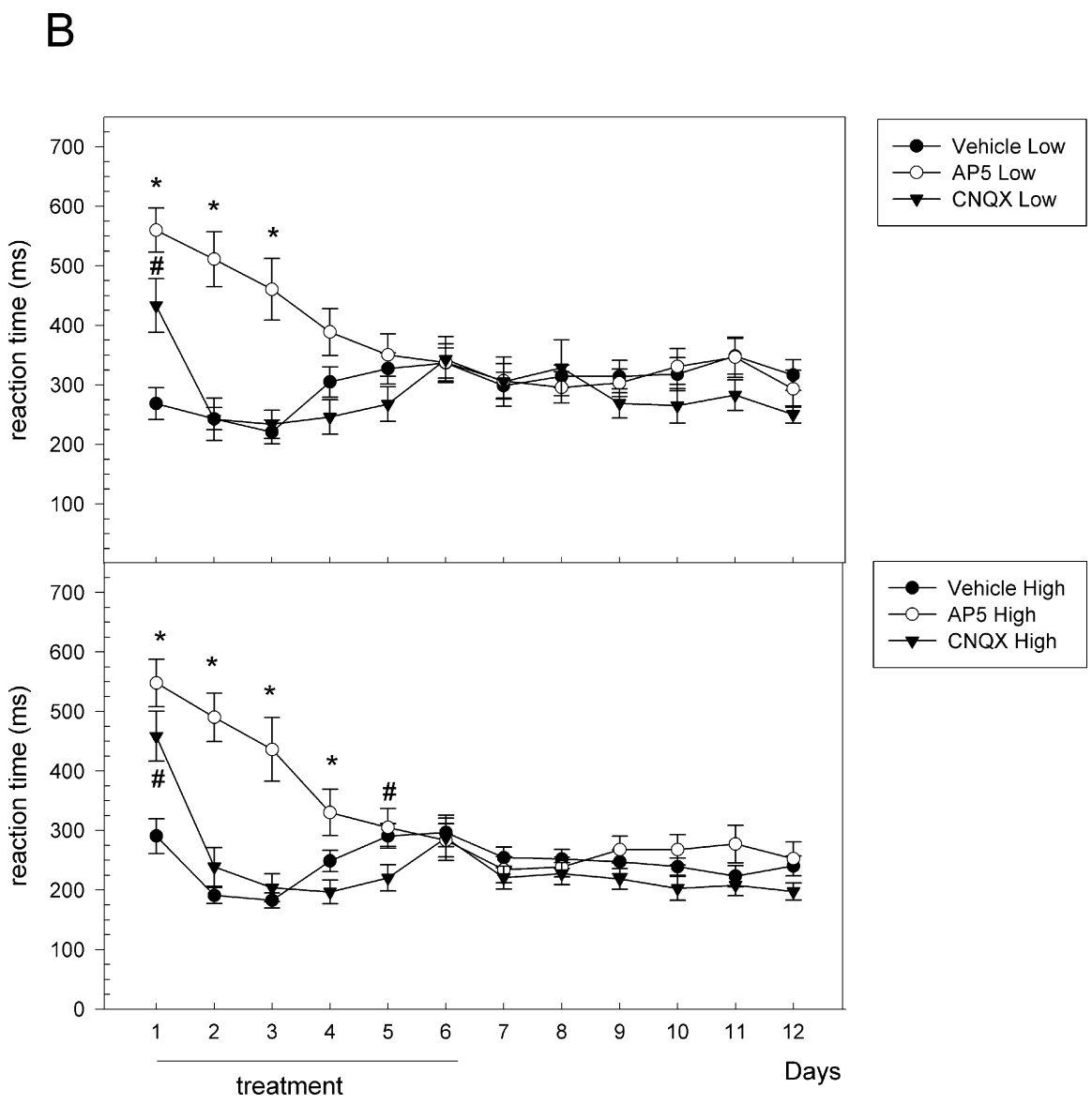


FIG. 6. Continued

intact on days 1–6 as RTs were guided by reward-predictive cues beginning on day 4.

The accuracy of responding was correlated with expected reward magnitudes in vehicle- and drug-treated animals. In general, short mean individual RTs per session were associated with a high number of early responses and, in turn, a low rate of correct responses. The tendency that fast responses were performed less accurately was particularly true if high reward was expected, thereby indicating reward-predictive cue effects.

Likewise, MTs were guided by expected reward magnitudes in vehicle- and CNQX-treated, but not in AP5-treated animals. The guidance of MTs by expected reward probably relies on different neural mechanisms as guidance of RTs, because MT and RT are not correlated significantly, as already shown in previous studies (Hauber, 1996; Brasted *et al*., 1998). Although MT is a sensitive parameter to control for drug-induced motor effects (Giertler *et al*., 2003), it is a less sensitive parameter for effects of reward expectancy in rats (Giertler *et al*., 2003) and primates (Hollerman *et al*., 1998). In our task, this is mainly due to the fact that MTs are influenced by a number of variables, such as body position relative to the manipulandum, which cause variability in MTs and interfere with reward expectancy effects. The observation that MT, accuracy of responding and RT guidance by expected reward were not markedly affected by either treatment indicates the absence of major sensorimotor impairments. However, we cannot rule out that subtle treatment-induced sensorimotor impairments contribute to the general increase of RTs.

Instrumental learning in tasks as used here is controlled by multiple mechanisms (e.g. Colwill & Rescorla, 1990; Baxter & Murray, 2002; Cardinal *et al*., 2002). Although not tested explicitly, different RTs of responses associated with high vs. low reward magnitude might reflect guidance of instrumental responding by stimulus–reward magnitude associations. Hence, one possibility to interpret intact guidance of RTs in AP5- and CNQX-treated animals on days 1–6 is that learning of stimulus–reward magnitude associations does not involve intra-AcbC NMDA or AMPA/KA receptor stimulation. Alternatively, shorter RTs of responses for expected high reward could reflect stronger stimulus–response representations not incorporating the outcome. If so, this process would not require an intra-AcbC NMDA and AMPA/KA

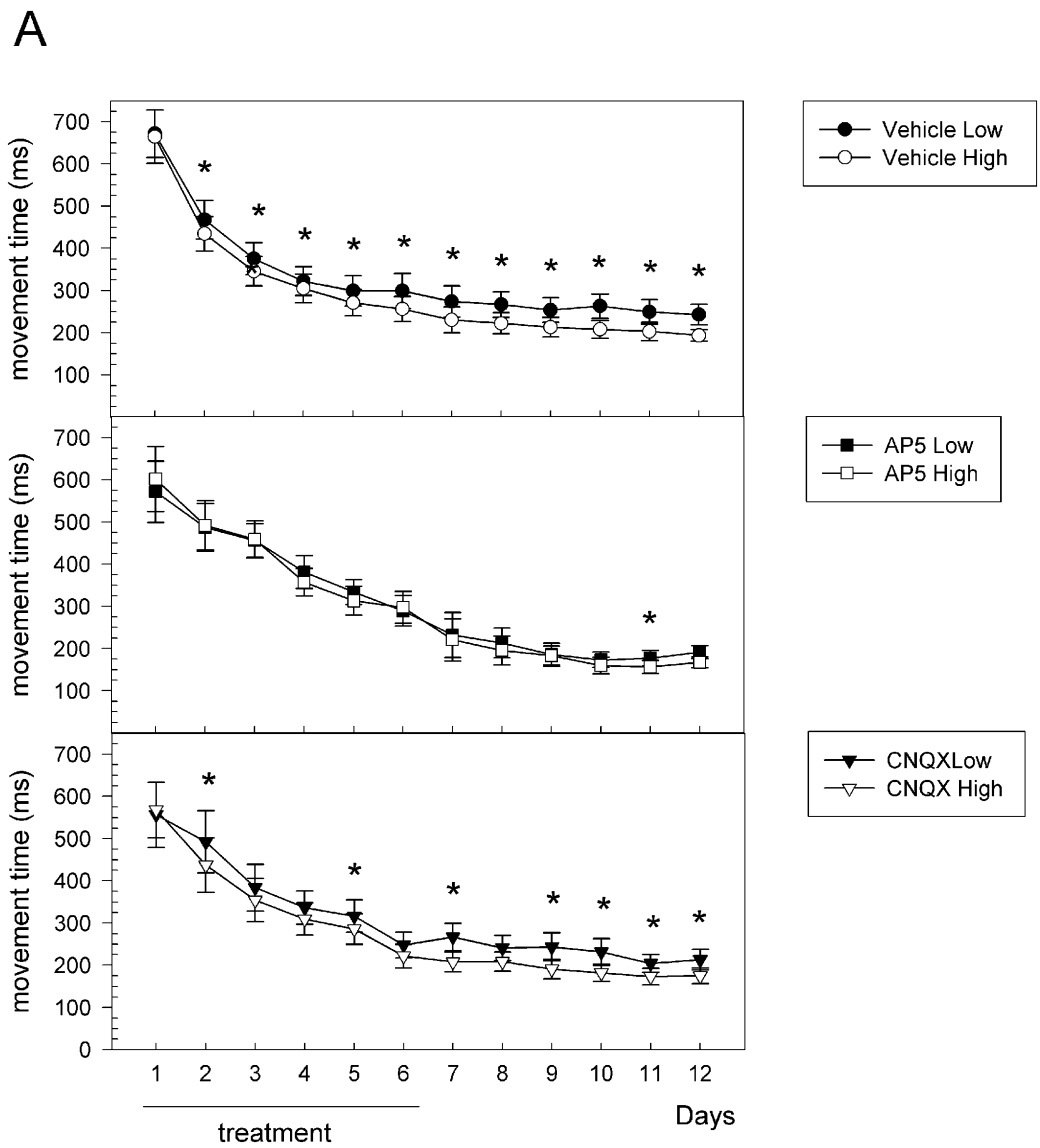


FIG. 7. MT of correct responses. Treatment groups received intra-AcbC injection of vehicle (0.5 μL, $n = 19$), AP5 (5 μg in 0.5 μL, $n = 14$) or CNQX (2.5 μg in 0.5 μL, $n = 14$) on days 1–6. No injections were given on days 7–12. (A) MTs and their guidance by expected reward (low: one pellet; high: five pellets). In all treatment groups, MT was significantly guided by expected reward magnitudes; significant main effect of reward magnitude was further analysed by planned contrasts for each day (*$P < 0.05$). (B) The same MT data as in A are depicted as a function of treatment separately for expected low (top) and high (bottom) reward to highlight drug effects. Infusion of AP5 and CNQX had no significant effects on MT.

stimulation. However, it is unlikely that instrumental responding tested here is not guided by the outcome as animals rapidly learn a reversal of original stimulus–reward magnitude associations (J. Schweimer and W. Hauber, unpublished results). In addition, response control involving stimulus–response associations probably relies on more extended instrumental learning (Adams, 1982) and might contribute to later stages of acquisition and performance.

Regardless of the precise associative mechanisms involved, an intra-AcbC stimulation of NMDA or AMPA/KA receptors seems not to be essential to learn the significance of reward-predictive cues and to adapt an instrumental response accordingly. However, we cannot exclude that a combined intra-AcbC blockade of ionotropic glutamate receptors or higher drug doses might interfere with guidance of RT by expected reward magnitudes. Furthermore, in a similar task with variable presentation of imperative cues, we previously showed that intra-Acb NMDA receptor blockade impaired RT guidance by expected reward magnitudes (Hauber *et al.*, 2000). Thus, particular features such as stimulus predictability might determine the extent to which the Acb is essential for intact discrimination learning in different tasks (Reading *et al.*, 1991; Burk & Mair, 2001). In addition, we have to take into account that in tasks as used here (Brown & Bowman, 1995; Cromwell & Schultz, 2003) correct identification of reward magnitude-predictive cues was not instrumental for accurate responding, i.e. reward magnitude information is not essential for accurate decision-making, but speeds responding for high reward.

Our interpretation that an intra-AcbC stimulation of NMDA or AMPA/KA receptors might be not critical to learn the significance of reward-predictive cues and to adapt instrumental behavior corresponds with the notion that the Acb is not required for goal-directed action (Balleine & Killcross, 1994; Balleine & Dickinson, 1998; Cardinal *et al.*, 2002; de Borchgrave *et al.*, 2002) and many aspects of instrumental learning (Cardinal & Everitt, 2004). For instance, a

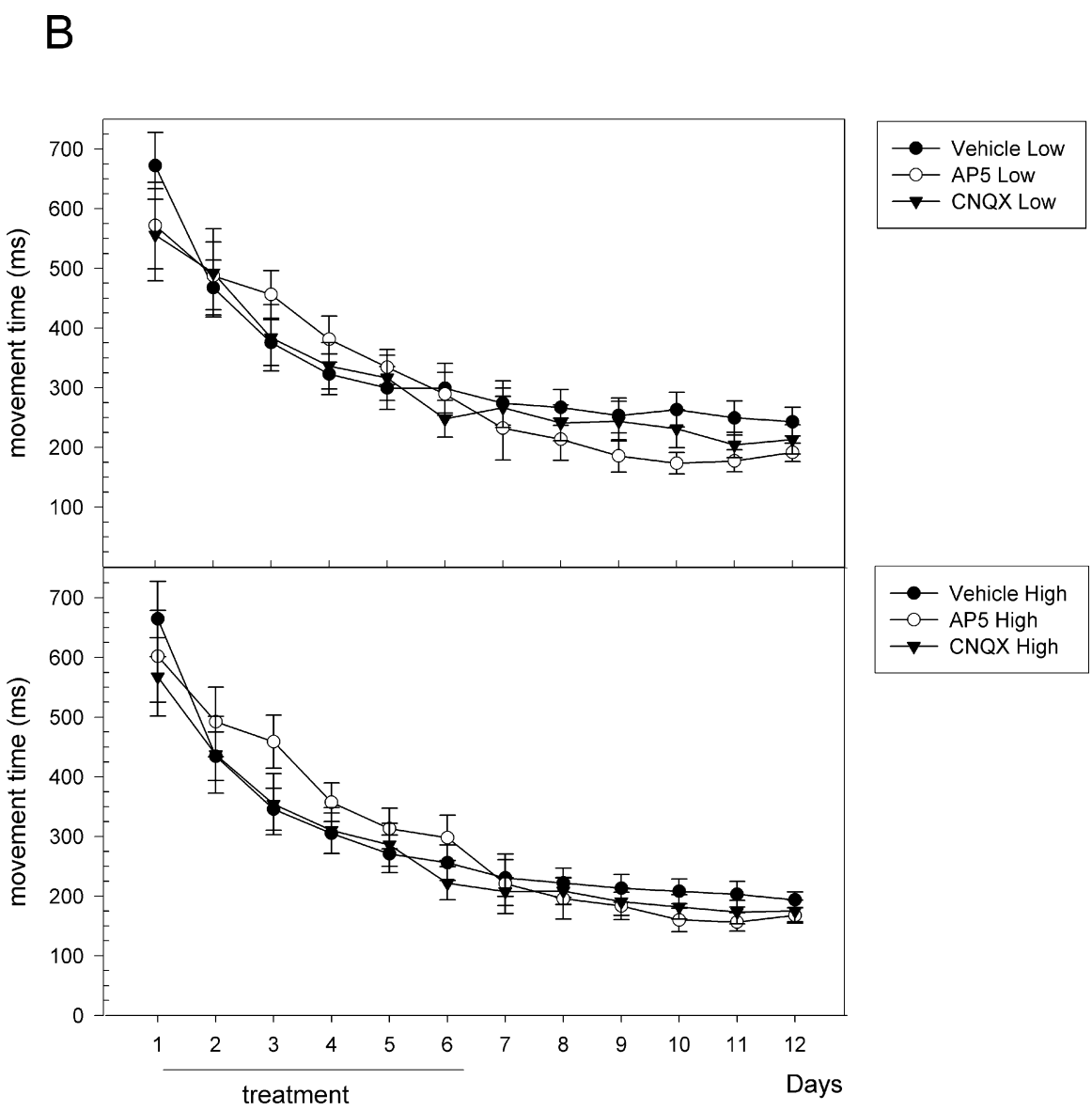
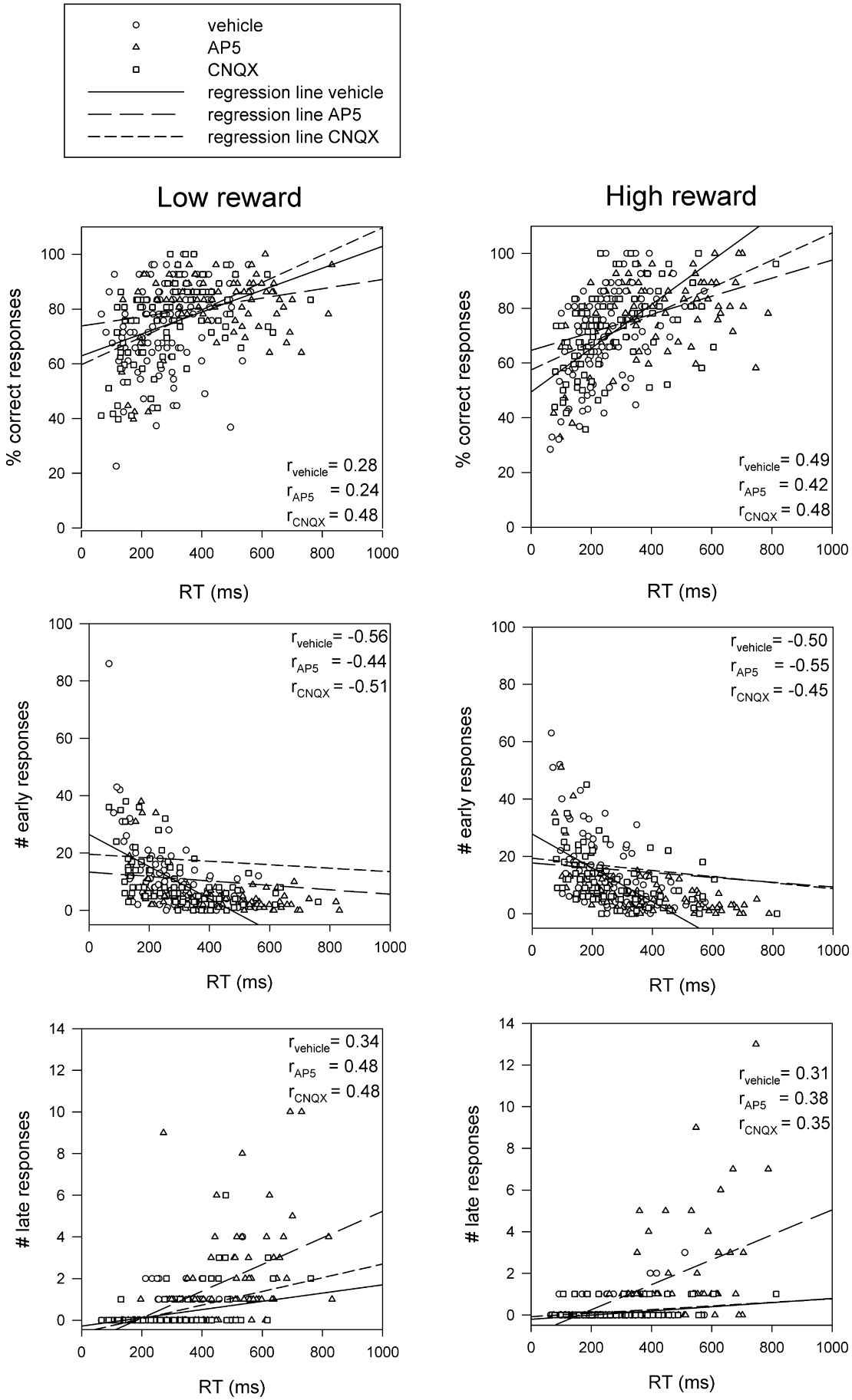


FIG. 7. Continued

combined blockade of intra-Acb NMDA and AMPA/KA receptors did not impair behavioral responding to a discriminative reward-predictive stimulus (Yun *et al*., 2004). However, this and many other studies (e.g. Giertler *et al*., 2003) focused on performance of a previously learned instrumental response. Relatively few studies have investigated effects of AcbC manipulations on instrumental learning guided by predictive cues. During learning of a go, no-go discrimination task, rats with lesions of the Acb failed to show normal changes of response latencies to odors predictive of either appetitive or aversive outcomes, but their choice behavior was unimpaired (Schoenbaum & Setlow, 2003). The latter finding suggests that some aspects of instrumental learning are intact after permanent Acb inactivation. Likewise, our present study implies that during instrumental learning, behavior can be adapted to information about outcome-predictive cues in the absence of intra-AcbC ionotropic glutamate receptor stimulation. Consequently, cue-induced reward expectancy activity displayed by subsets of Acb neurons (e.g. Schultz *et al*., 2003; Setlow *et al*., 2003; Nicola *et al*., 2004; Yun *et al*., 2004), which probably relies on intact intra-AcbC glutamate transmission, seems not to be essential for guiding instrumental responses according to reward magnitude-predictive cues. It appears that other neural circuits guide instrumental action according to expected reward magnitudes, if AcbC function is compromised by an ionotropic glutamate receptor blockade. On the other hand, there is consistent evidence for an involvement of the AcbC in instrumental learning as intra-AcbC microinfusion of AP5, post-trial infusions of a selective protein kinase A inhibitor or a protein synthesis inhibitor blocked instrumental responding on a variable ratio (VR) 2 schedule (Kelley

FIG. 8. Interrelations between response speed, reward magnitude and accuracy of performance based on a correlational analysis of individual mean RTs for high and low reward from correct responses (from each session on days 1–6) and associated rates of early, correct and late responses. Simple linear correlations between RT and the proportion of correct responses, the number of early and late responses in vehicle- (black), AP5- (red) and CNQC (green)-treated animals for low and high reward are depicted. Correlation coefficients: Pearson's *r*.


vehicle
AP5
CNQX
regression line vehicle
regression line AP5
regression line CNQX
Low reward
High reward
% correct responses
RT (ms)
$r_{vehicle}$ = 0.28
r_{AP5} = 0.24
r_{CNQX} = 0.48
$r_{vehicle}$ = 0.49
r_{AP5} = 0.42
r_{CNQX} = 0.48
early responses
$r_{vehicle}$ = -0.56
r_{AP5} = -0.44
r_{CNQX} = -0.51
$r_{vehicle}$ = -0.50
r_{AP5} = -0.55
r_{CNQX} = -0.45
late responses
$r_{vehicle}$ = 0.34
r_{AP5} = 0.48
r_{CNQX} = 0.48
$r_{vehicle}$ = 0.31
r_{AP5} = 0.38
r_{CNQX} = 0.35

et al., 1997; Baldwin *et al*., 2002; Hernandez *et al*., 2002). The reasons why intra-AcbC NMDA receptors contribute to instrumental learning in a VR schedule but not to learning of cue-evoked instrumental responding are not clear. Task-related differences may account for the observed discrepancies, as, for instance, we used a non-rate measure of response vigor, i.e. RT. Furthermore, intra-AcbC glutamate receptors seem to be differentially involved in both tasks as an intra-AcbC NMDA receptor antagonism blocked acquisition, but not performance, of instrumental responding on a VR schedule (Kelley *et al*., 1997), while the same manipulation interfered both with acquisition (this study) and with performance (Giertler *et al*., 2003) of instrumental responding guided by reward-predictive stimuli.

Role of AcbC glutamate receptors in invigorating instrumental responding

Vehicle-treated rats exhibited an initial decay of RTs on days 1–3 followed by a moderate increase on days 4–6 probably reflecting an adaptation of RTs to the time corridor defined as being correct. By contrast, AP5-treated animals showed a strong RT increase of responses to expected low and high reward on days 1–4 as well as higher numbers of late responses, in particular, if low reward was expected. In CNQX-treated animals, there was an increase in RTs of responses to expected low and high reward on day 1. It is unlikely that sensorimotor impairments account for RT increases, because the accuracy of responding, RT guidance by expected reward and MTs were not markedly affected by either treatment. Thus, stimulation of intra-AcbC NMDA and AMPA/KA receptors seems to play a prominent role in invigorating instrumental responses regardless of the expected reward size associated with the particular action. Whether the distinct persistence (3 vs. 1 day) of AP5 and CNQX effects on RTs during acquisition reflects a differential involvement of intra-AcbC NMDA and AMPA/KA receptors to invigorate instrumental responding is difficult to assess. For instance, it is not known whether the concentrations of CNQX and AP5 infused into the AcbC are equipotent or whether the efficacy of adaptive mechanisms to compensate for subsequent daily intra-AcbC NMDA and AMPA/KA receptors are similar.

We previously observed a general increase of RTs, but intact RT guidance by expected reward magnitudes in well-trained animals subjected to an intra-Acb infusion of AP5 and CNQX (Giertler *et al*., 2003). Thus, the Acb seems to modulate the vigor of responding guided by reward-predictive cues during acquisition as well as performance. Previous studies also demonstrated that Acb inactivation primarily affects the vigor of instrumental responding. In a go, no-go odor discrimination task, rats with lesions of the Acb failed to show normal changes in response latency during discrimination learning, despite intact choice behavior (Schoenbaum & Setlow, 2003). Likewise, combined intra-Acb administration of an NMDA and an AMPA/KA receptor antagonist increased response latency, but did not reduce instrumental responding to a reward-predictive cue (Yun *et al*., 2004).

The mechanisms affected by an intra-AcbC NMDA or AMPA/KA receptor blockade accounting for the general increase of RTs, but preserved RT guidance by expected reward magnitudes seen here are difficult to assess. It is well known that intra-AcbC NMDA receptors contribute to appetitive instrumental learning and mediate Pavlovian influences on behavior (Kelley *et al*., 1997; Smith-Roe & Kelley, 2000; Di Ciano *et al*., 2001; Baldwin *et al*., 2002; Kelley & Berridge, 2002), but their role in instrumental learning guided by cues predictive of natural reward has not yet been addressed in detail. The incentive motivation theory holds that the AcbC mediates Pavlovian-conditioned appetitive states in motor performance (Robbins *et al*., 1989; Balleine & Killcross, 1994; Berridge & Robinson, 1998; Cardinal *et al*., 2002), in particular Pavlovian influences to strengthen instrumental behavior as shown in Pavlovian-to-instrumental transfer tasks (Hall *et al*., 2001; Holland & Gallagher, 2003). According to this view, cues others than instructive cues, e.g. non-contingent contextual stimuli in the operant box, might serve in our task as Pavlovian stimuli predictive of an appetitive outcome and produce a motivational arousal thereby speeding RT of responses to high and low reward. If so, the general increase of RT observed here could reflect a reduced invigorating impact of such Pavlovian stimuli caused by an intra-AcbC NMDA and, to a lesser extent, an AMPA/KA receptor blockade. On the other hand, the Acb neurons encode movement-preparatory and outcome-related information (Schultz *et al*., 2003) and outcome devaluation can increase instrumental response latencies (Sage & Knowlton, 2000). Thus, another plausible interpretation could be that an intra-AcbC blockade of ionotropic glutamate receptors interferes with stimulus–outcome associations and the use of such information to invigorate responding.

Notably, in rats with an intra-AcbC NMDA or AMPA/KA receptor we found no evidence for an increased impulsivity as shown by Cardinal *et al*. (2001) to occur after Acb lesions. In our task, rats treated with AP5 or CNQX responded slower for expected high and low reward and exhibited an increased number of late responses, in particular, if low reward was expected. However, we used identical delays for high and low reward; thus the task might not be sensitive for impulsive choices that are characterized by a high preference for immediate, low rewards over large, delayed rewards.

Role of OFC and Acb in instrumental responding guided by reward-predictive cues

The orbital prefrontal cortex (OFC) is considered to be a another key component of a limbic cortico-striatal circuitry through which information on the motivational significance of stimuli mediates the selection and execution of reward-directed behavioral responses (Schoenbaum & Setlow, 2001). In line with this notion, OFC lesions impaired guidance of instrumental behavior after a reversal of stimulus-reward magnitude contingencies in the same task as used here (Bohn *et al*., 2003b). Likewise, an intra-OFC blockade of NMDA receptors inhibited learning of a reversal of previously acquired stimulus-reward magnitude contingencies and produced a general shortening of RT as well as an increased number of early responses (Bohn *et al*., 2003b).

The AcbC receives direct glutamatergic input from structures such as the OFC and the basolateral amygdala, which encode the learned motivational significance of cues (Thorpe *et al*., 1983; Schoenbaum & Eichenbaum, 1995; Rolls *et al*., 1996; Schoenbaum *et al*., 1999; Tremblay & Schultz, 1999). Thus, intra-AcbC NMDA and AMPA/KA receptors are likely to play a key role in guiding instrumental responding to reward-predictive cues. However, the disparate behavioral effects induced by an intra-OFC and AcbC NMDA receptor blockade in the task used here tentatively suggests that instrumental learning guided by reward-predictive cues may not necessarily require serial OFC–AcbC processing. Yet, this notion is preliminary and requires direct experimental support using disconnection lesions. Nevertheless, instrumental responding guided by outcome-predictive stimuli was largely intact after AcbC inactivation in our as well as other tasks (Schoenbaum & Setlow, 2003; Yun *et al*., 2004). Therefore, neural connections mediating instrumental responding

must exist within the limbic cortico-striatal circuit that bypass the AcbC.

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Abbreviations

Acb, nucleus accumbens; AcbC, core subregion of the nucleus accumbens; CNQX, 6-cyano-7-nitroquinoxaline-2,3-dione; MT, movement time; OFC, orbital prefrontal cortex; RT, reaction time; VR, variable ratio.

References

Adams, C.D. (1982) Variations in the sensitivity of instrumental responding to reinforcer devaluation. *Q. J. Exp. Psychol.*, **34**, 77–98.

Albin, R.L., Marcowiec, R.L., Hollingsworth, Z.R., Dure, L.S., Penney, J.B. & Young, A.B. (1992) Excitatory amino acid binding sites in the basal ganglia of the rat: a quantitative autoradiographic study. *Neuroscience*, **46**, 35–48.

Apicella, P., Ljungberg, T., Scarnati, E. & Schultz, W. (1991) Responses to reward in monkey dorsal and ventral striatum. *Exp. Brain Res.*, **85**, 491–500.

Baldwin, A.E., Sadeghian, K. & Kelley, A.E. (2002) Appetitive instrumental learning requires coincident activation of NMDA and dopamine D1 receptors within the medial prefrontal cortex. *J. Neurosci.*, **22**, 1063–1071.

Balleine, B.W. & Dickinson, A. (1998) Goal-directed instrumental action: contingency and incentive learning and their cortical substrates. *Neuropharmacology*, **37**, 407–419.

Balleine, B. & Killcross, S. (1994) Effects of ibotenic acid lesions of the nucleus accumbens on instumental action. *Behav. Brain Res.*, **65**, 181–193.

Baxter, M.G. & Murray, E.A. (2002) The amygdala and reward. *Nat. Rev. Neurosci.*, **3**, 563–573.

Berridge, K.C. & Robinson, T.E. (1998) What is the role of dopamine in reward: hedonic impact, reward learning, or incentive salience? *Brain Res. Rev.*, **28**, 309–369.

Bohn, I., Giertler, C. & Hauber, W. (2003a) NMDA receptors in the rat orbital prefrontal cortex and guidance of instrumental behaviour under reversal conditions. *Cereb. Cortex*, **13**, 968–976.

Bohn, I., Giertler, C. & Hauber, W. (2003b) Orbital prefrontal cortex and guidance of instrumental behaviour in rats under reversal conditions. *Behav. Brain Res.*, **143**, 49–56.

de Borchgrave, R., Rawlins, J.N., Dickinson, A. & Balleine, B.W. (2002) Effects of cytotoxic nucleus accumbens lesions on instrumental conditioning in rats. *Exp. Brain Res.*, **144**, 50–68.

Brasted, P.J., Dobrossy, M.D., Robbins, T.W. & Dunnett, S.B. (1998) Striatal lesions produce distinctive impairments in reaction time performance in two different operant chambers. *Brain Res. Bull.*, **46**, 487–493.

Brasted, P.J., Humby, T., Dunnett, S.B. & Robbins, T.W. (1997) Unilateral lesions of the dorsal striatum in rats disrupt responding in egocentric space. *J. Neurosci.*, **17**, 8919–8926.

Brown, V.J. & Bowman, E.M. (1995) Discriminative cues indicating reward magnitude continue to determine reaction time of rats following lesions of the nucleus accumbens. *Eur. J. Neurosci.*, **7**, 2479–2485.

Burk, J.A. & Mair, R.G. (2001) Effects of dorsal and ventral striatal lesions on delayed matching trained with retractable levers. *Behav. Brain Res.*, **122**, 67–78.

Cardinal, R.N. & Everitt, B.J. (2004) Neural and psychological mechanisms underlying appetitive learning: links to drug addiction. *Curr. Opin. Neurobiol.*, **14**, 156–162.

Cardinal, R.N., Parkinson, J.A., Hall, J. & Everitt, B.J. (2002) Emotion and motivation: the role of the amygdala, ventral striatum, and prefrontal cortex. *Neurosci. Biobehav. Rev.*, **26**, 321–352.

Cardinal, R.N., Pennicott, D.R., Sugathapala, C.L., Robbins, T.W. & Everitt, B.J. (2001) Impulsive choice induced in rats by lesions of the nucleus accumbens core. *Science*, **292**, 2499–2501.

Carelli, R.M., Ijames, S.G. & Crumling, A.J. (2000) Evidence that separate neural circuits in the nucleus accumbens encode cocaine versus natural (water and food) reward. *J. Neurosci.*, **20**, 4255–4266.

Colwill, R.M. & Rescorla, R.A. (1990) Evidence for the hierarchical structure of instrumental learning. *Anim. Learn. Behav.*, **18**, 71–82.

Cromwell, H.C. & Schultz, W. (2003) Effects of expectations for different reward magnitudes on neuronal activity in primate striatum. *J. Neurophysiol.*, **89**, 2823–2838.

Di Ciano, P., Cardinal, R.N., Cowell, R.A., Little, S.J. & Everitt, B.J. (2001) Differential involvement of NMDA, AMPA/kainate, and dopamine receptors in the nucleus accumbens core in the acquisition and performance of pavlovian approach behavior. *J. Neurosci.*, **21**, 9471–9477.

Di Ciano, P. & Everitt, B.J. (2001) Dissociable effects of antagonism of NMDA and AMPA/KA receptors in the nucleus accumbens core and shell on cocaine-seeking behavior. *Neuropsychopharmacology*, **25**, 341–360.

Everitt, B.J. & Wolf, M.E. (2002) Psychomotor stimulant addiction: a neural systems perspective. *J. Neurosci.*, **22**, 3312–3320.

Giertler, C., Bohn, I. & Hauber, W. (2003) The rat nucleus accumbens is involved in guiding of instrumental responses by stimuli predicting reward magnitude. *Eur. J. Neurosci.*, **18**, 1993–1996.

Gold, J.I. (2003) Linking reward expectation to behavior in the basal ganglia. *Trends Neurosci.*, **26**, 12–14.

Groenewegen, H.J., Wright, C.I. & Beijer, A.V.J. (1996) The nucleus accumbens: gateway for limbic structures to reach the motor system? In Holstege, G., Bandler, R. & Saper, C.B. (Eds), *Emotional Motor System*. Elsevier Science Publications, Amsterdam, pp. 485–511.

Hall, J., Parkinson, J.A., Connor, T.M., Dickinson, A. & Everitt, B.J. (2001) Involvement of the central nucleus of the amygdala and nucleus accumbens core in mediating Pavlovian influences on instrumental behaviour. *Eur. J. Neurosci.*, **13**, 1984–1992.

Hauber, W. (1996) Impairments of movement initiation and execution induced by a blockade of dopamine D1 or D2 receptors are reversed by a blockade of N-methyl-D-aspartate receptors. *Neuroscience*, **73**, 121–130.

Hauber, W., Bohn, I. & Giertler, C. (2000) NMDA, but not dopamine D2, receptors in the rat nucleus accumbens are involved in guidance of instrumental behavior by stimuli predicting reward magnitude. *J. Neurosci.*, **20**, 6282–6288.

Hauber, W., Giertler, C. & Bohn, I. (2001) NMDA and dopamine D2 receptors in the caudate-putamen are not involved in control of motor readiness in rats. *Psychopharmacology (Berlin)*, **155**, 43–51.

Hernandez, P.J., Sadeghian, K. & Kelley, A.E. (2002) Early consolidation of instrumental learning requires protein synthesis in the nucleus accumbens. *Nat. Neurosci.*, **5**, 1327–1331.

Holland, P.C. & Gallagher, M. (2003) Double dissociation of the effects of lesions of basolateral and central amygdala on conditioned stimulus-potentiated feeding and Pavlovian-instrumental transfer. *Eur. J. Neurosci.*, **17**, 1680–1694.

Hollerman, J.R. & Schultz, W. (1998) Dopamine neurons report an error in the temporal prediction of reward during learning. *Nature Neurosci.*, **1**, 304–309.

Hollerman, J.R., Tremblay, L. & Schultz, W. (1998) Influence of reward expectation on behaviour-related neuronal activity in primate striatum. *J. Neurophysiol.*, **80**, 947–963.

Ikemoto, S. & Panksepp, J. (1999) The role of nucleus accumbens dopamine in motivated behavior: a unifying interpretation with special reference to reward-seeking. *Brain Res. Rev.*, **31**, 6–41.

Kawagoe, R., Takikawa, Y. & Hikosaka, O. (1998) Expectation of reward modulates cognitive signals in the basal ganglia. *Nature Neurosci.*, **1**, 411–416.

Kelley, A.E. (2004) Memory and addiction; shared neural circuitry and molecular mechanisms. *Neuron*, **44**, 161–179.

Kelley, A.E. & Berridge, K.C. (2002) The neuroscience of natural rewards: relevance to addictive drugs. *J. Neurosci.*, **22**, 3306–3311.

Kelley, A.E., Smith-Roe, S.L. & Holahan, M.R. (1997) Response-reinforcement learning is dependent on N-methyl-D-aspartate receptor activation in the nucleus accumbens core. *Proc. Natl Acad. Sci. USA*, **94**, 12174–12179.

Mogenson, G.J., Jones, D.L. & Yim, C.Y. (1980) From motivation to action: functional interface between the limbic system and the motor system. *Prog. Neurobiol.*, **14**, 69–97.

Nicola, S.M., Yun, I.A., Wakabayashi, K.T. & Fields, H.L. (2004) Cue-evoked firing of nucleus accumbens neurons encodes motivational significance during a discriminative stimulus task. *J. Neurophysiol.*, **91**, 1840–1865.

Pellegrino, L.J., Pellegrino, A.S. & Cushmann, A.J. (1981) *A Stereotaxic Atlas of the Rat Brain*. Plenum Press, New York.

Reading, P.J., Dunnett, S.B. & Robbins, T.W. (1991) Dissociable roles of the ventral, medial and lateral striatum on the acquisition and performance of a complex visual stimulus-response habit. *Behav. Brain Res.*, **45**, 147–161.

Robbins, T.W., Cador, M., Taylor, J.R. & Everitt, B.J. (1989) Limbic–striatal interactions in reward-related processes. *Neurosci. Biobehav. Rev.*, **13**, 155–162.

Rolls, E.T., Critchley, H.D., Mason, R. & Wakeman, E.A. (1996) Orbitofrontal cortex neurons: role in olfactory and visual association learning. *J. Neurophysiol.*, **75**, 1970–1981.

Sage, J.R. & Knowlton, B.J. (2000) Effects of US devaluation on win-stay and win-shift radial maze performance in rats. *Behav. Neurosci.*, **114**, 295–306.

Schoenbaum, G., Chiba, A.A. & Gallagher, M. (1999) Neural encoding in orbitofrontal cortex and basolateral amygdala during olfactory discrimination learning. *J. Neurosci.*, **19**, 1876–1884.

Schoenbaum, G. & Eichenbaum, H. (1995) Information coding in the rodent prefrontal cortex. II. Ensemble activity in orbitofrontal cortex. *J. Neurophysiol.*, **74**, 751–762.

Schoenbaum, G. & Setlow, B. (2001) Integrating orbitofrontal cortex into prefrontal theory: common processing themes across species and subdivisions. *Learn. Mem.*, **8**, 134–147.

Schoenbaum, G. & Setlow, B. (2003) Lesions of nucleus accumbens disrupt learning about aversive outcomes. *J. Neurosci.*, **23**, 9833–9841.

Schultz, W., Apicella, P., Scarnati, E. & Ljungberg, T. (1992) Neuronal activity in monkey ventral striatum related to the expectation of reward. *J. Neurosci.*, **12**, 4595–4610.

Schultz, W., Tremblay, L. & Hollerman, J.R. (2003) Changes in behavior-related neuronal activity in the striatum during learning. *Trends Neurosci.*, **26**, 321–328.

Setlow, B., Schoenbaum, G. & Gallagher, M. (2003) Neural encoding in ventral striatum during olfactory discrimination learning. *Neuron*, **38**, 625–636.

Smith-Roe, S.L. & Kelley, A.E. (2000) Coincident activation of NMDA and dopamine D1 receptors within the nucleus accumbens core is required for appetitive instrumental learning. *J. Neurosci.*, **20**, 7737–7742.

Thorpe, S.J., Rolls, E.T. & Maddison, S. (1983) The orbitofrontal cortex: neuronal activity in the behaving monkey. *Exp. Brain Res.*, **49**, 93–115.

Tremblay, L. & Schultz, W. (1999) Relative reward preference in primate orbitofrontal cortex. *Nature*, **398**, 704–708.

Wan, F.-J. & Swerdlow, N.R. (1996) Sensorimotor gating in rats is regulated by different dopamine – glutamate interactions in the nucleus accumbens core and shell subregions. *Brain Res.*, **722**, 168–176.

Yun, I.A., Nicola, S.M. & Fields, H.L. (2004) Contrasting effects of dopamine and glutamate receptor antagonist injection in the nucleus accumbens suggest a neural mechanism underlying cue-evoked goal-directed behavior. *Eur. J. Neurosci.*, **20**, 249–263.

Zahm, D.S. (2000) An integrative neuroanatomical perspective on some subcortical substrates of adaptive responding with emphasis on the nucleus accumbens. *Neurosci. Biobehav. Rev.*, **24**, 85–105.