ELSEVIER

Pharmacology, Biochemistry and Behavior 67 (2000) 575–581

PHARMACOLOGY BIOCHEMISTRY AND BEHAVIOR

Ketamine blocks a taste recognition memory in fetal rats

G. Andrew Mickley*, Dawn R. Remmers-Roeber, Carrie Crouse, Rebecca Peluso

Department of Psychology, Baldwin-Wallace College, Carnegie Hall, 275 Eastland Road, Berea, OH 44017-2088, USA

Received 25 February 2000; received in revised form 14 July 2000; accepted 1 August 2000

Abstract

Decisions about novelty/familiarity are critical in determining whether or not information should be attended to, and possibly encoded, for long-term storage. We have reported that fetal and neonatal rats exhibit an increase in orofacial movements (e.g., perseverative mouthing and mouth movements, and licks) upon tasting saccharin (SAC), if it was experienced previously. E19 rat fetuses can acquire this taste recognition memory and retain it for at least 5 days (P3). In the current study, we sought to evaluate the role of *N*-methyl-D-aspartate (NMDA) receptors in establishing a taste recognition memory. Pregnant Sprague–Dawley rats received ketamine (NMDA receptor antagonist) (doses: 0, 50, or 100 mg/kg, ip). One-half hour later, we performed a reversible spinal block on each pregnant dam, and E19 fetuses received an oral injection of 10 μl, 0.3% SAC or water (control) while in utero. The uterus was replaced and the pups were later born via a normal vaginal delivery. On P3, all pups experienced oral lavage of 10 μl, 0.3% SAC, and motor responses were recorded. As expected, non-drugged control neonates tasting familiar SAC exhibited significantly more perseverative mouth movements, as well as total mouth movements and licks, than did pups tasting novel SAC. However, this taste recognition memory response was not observed in rats exposed to ketamine in utero. The data suggest that early non-associative taste memories may be disrupted by NMDA receptor blockade.

Keywords: Taste recognition memory; Fetus; Neonate; Rat; Novelty; Familiar; Memory; Saccharin; Development; Ketamine; NMDA

The ability to discriminate between novel and familiar objects is well documented in adult humans [10,46], as well as other animals [8,30]. Reactions to novelty have been viewed as preparatory ones, which facilitate the reception of information and allow the organism to choose an instantaneous response to be made within a particular context [30]. There is an incentive–motivational component of responses to novelty as the animal becomes more prepared to respond to significant stimuli in the environment [4,35]. Moreover, there is a memorial component to novelty responses, since novelty can only be specified in relation to an animal's past experience [30].

Animals must discriminate between a stimulus' novelty and familiarity in order to determine whether or not the information should be attended to, and possibly encoded, for long-term storage [49]. Making decisions about the novelty of a stimulus is presumably more conducive to survival than is automatically duplicating information that may be already available in the memory store. Determining and seeking out novelty is also an adaptive means by which animals avoid stimulus re-exposure and therefore maximize the amount of new (i.e., non-redundant) information available [22].

There is a growing literature suggesting that very young organisms may make discriminations between novel and familiar objects [10]. Because the gustatory and olfactory systems are fairly well developed late in gestation [44], researchers in several laboratories have been studying these systems as a means of assessing the ability of perinatal rats to detect and remember stimuli. For example, young rats can discriminate between a novel taste and a familiar taste that was first experienced in utero. Smotherman [40] exposed E20 rat fetuses to either apple juice or saline via amniotic fluid. When presented with a choice between apple juice and tap water, young-adult rats prenatally exposed to apple juice consumed more apple juice than control rats lacking the prenatal experience with this taste. Hepper [14] has

* Corresponding author. Tel.: +1-440-826-2194; fax: +1-440-826-8549.

E-mail address: amickley@bw.edu (G.A. Mickley).

0091-3057/00/$ – see front matter
PII: S0091-3057(00)00391-9

demonstrated that if pregnant dams eat garlic they produce offspring that exhibit an enhanced preference for the odor of garlic when tested 12 days after birth. Additional evidence suggests that pre and early postnatal exposure to citral (a lemon odor) induces rat pups to exhibit a preference for nipples painted with citral [33]. Likewise, recent reports from our laboratory indicate that neonates exhibit different orofacial motor responses to the taste of saccharin (SAC) depending on whether or not it was tasted in utero [25].

A variety of studies suggest that brain *N*-methyl-D-aspartate (NMDA) glutamate receptors are involved in memory formation (see Ref. [34] for review). This view has been strengthened by reports that long-term potentiation (a widely studied model of use-dependent changes in synaptic efficacy and information storage in the brain) is mediated by NMDA receptors [12]. NMDA receptor antagonists impair place learning in a water maze [29], and interfere with step-through passive avoidance learning [6]. Furthermore, prior administration of ketamine (a well-known non-competitive NMDA receptor antagonist [45]) blocks the formation of a conditioned taste aversion (CTA) in adult [1,49], neonatal [28], and fetal rats of age E19 or older [24]. These behavioral paradigms all involve associative learning of a conditioned stimulus–unconditioned stimulus pairing. The current paper extends these findings and investigates the extent to which ketamine can influence non-associative memory formation in fetal rats.

1. Method

1.1. Subject

The subjects were perinatal Sprague–Dawley rats (male and female) obtained from timed-pregnant female rats supplied by Zivic-Miller Laboratories (Zelienople, PA). The date of conception (i.e., the first day that a vaginal plug was detected) was designated as "embryonic day 0" (E0). Pregnant animals (from which our subjects were derived) were individually housed in plastic "shoe box" cages (44.45 cm long × 21.59 cm wide × 20.32 cm high). Home cage temperature was maintained at 23–26°C under a 12/12-h light:dark cycle (lights on at 0600 h).

1.2. Fetal injections

Pregnant rat dams carrying E19 fetuses were injected with either 0, 50, or 100 mg/kg, ip, ketamine HCl in saline vehicle. These doses were selected based on previous HPLC studies documenting the amount of ketamine present in the brain of fetuses 0.5 h after maternal injections with this NMDA receptor antagonist [29]. A maternal dose of 100 mg/kg, ip, ketamine produces fetal brain levels of approximately 14 μg/g. Similar brain levels have been shown to be sufficient to block the formation of CTAs in fetal and neonatal rats [24,28].

Casual observation of the dams, 30 min following ketamine injections, indicated that 50 mg/kg, ip, ketamine induces stereotypic head movements and either enhancement or little change in spontaneous locomotion. The higher dose of ketamine (100 mg/kg, ip) produced akinesia. These data are consistent with those from other labs indicating that low doses of ketamine frequently cause an increase in locomotor movements, whereas higher doses cause a reduction in movement and "catalepsy" [2,32]. Similar effects have been reported when bar-pressing behaviors were measured [20].

One-half hour after the ketamine injection, the dams were briefly anesthetized with isoflurane before they underwent a reversible spinal block procedure. A 30-gauge needle was used to inject lidocaine HCl 2% and epinephrine 1:100,000 (in a volume of 100 μl) between the first and second lumbar vertebrae. This procedure is effective in producing (a) a complete abdominal and hind limb paralysis, (b) consistently long periods of spinal anesthesia (>45 min), and (c) complete recovery after the anesthesia. There is no indication that litters are adversely affected by this procedure [42].

The analgesic dam was restrained in a plastic holding apparatus and her vision of the fetal injection procedure restricted. Both uterine horns were exposed through a midline laparotomy, and the hind legs and lower abdomen immersed in a warm bath (37.5 ± 1°C) containing isotonic saline (Locke's solution). Both horns of the uterus were exteriorized through the abdominal incision, and were allowed to float freely in the bath. The rat fetuses were seen through the semi-transparent walls of the uterus and positioned for accurate placement of injections. All fetuses, in a particular litter, received oral lavage with either SAC (10 μl, 0.3%) or H_2O (10 μl) via a 30-gauge needle. Following the injections, the uterus was replaced, the abdominal wall and the skin of the pregnant rat sutured, and the wounds infused with a local anesthetic (bupivicaine, 0.25%) in order to produce post-surgical analgesia.

Even et al. [9] have reported that steroids present in one amniotic sac may diffuse across the fetal membranes to other fetuses in the uterus. Although the injection was placed in the mouth of the fetus, SAC almost certainly also spilled into the amniotic fluid and may have moved into adjacent uterine compartments. If different pups in a litter had different oral injections, this could have confounded our conditioning procedure. For this reason, we did not mix different taste injections within litters. This procedure necessitates special data analysis techniques (see Statistical Analyses). The number of subjects/litters involved in this study were: 50 mg/kg ketamine [SAC pretreatment, $N=27/6$; water pretreatment, $N=17/5$]; 100 mg/kg ketamine [SAC pretreatment, $N=21/5$; water pretreatment, $N=26/6$]; saline controls [SAC pretreatment, $N=10/2$; water pretreatment, $N=25/9$].

Despite plans that called for equal *N*s/group, experiments like those we describe here are frequently influenced by

factors that make this difficult to achieve. In our laboratory, we have found that litter size can vary from 1 to 18. Infrequently, neonates appear very pale (suggesting poor oxygenation) or neglected (and dehydrated), and are excluded based on presumed health/viability problems. Approximately 20% of litters manipulated on E19 do not come to term. Note that the statistical analyses employed (see Statistical Analyses) adjust for unequal *N*s.

1.3. Behavioral testing

Rat pups were born via a normal vaginal delivery and were maintained with their dam until behavioral testing on P3. Twenty minutes before the behavioral test, pups were separated from the dam and placed with littermates in a small plastic container sitting on a warm ($38.5 \pm 0.5°C$) heating pad. This container was covered with gauze and maintained in a temperature-controlled incubator (ambient temperature $= 28 \pm 1°C$) until immediately before testing of the litter began. For the behavioral observations, neonates were placed in a warm (ambient temperature $= 28 \pm 1°C$), high-humidity chamber on a glass plate, warmed (via constantly circulating water) to $36 \pm 1°C$. All pups received oral lavage with 10 μl SAC through a blunt/smooth 18-gauge stainless steel infusion needle. Subjects were then placed (ventral side down) on the glass plate. Using a mirror, behavior was videotaped from below the animal for 1 min before (baseline), and after, oral injection.

1.4. Dependent variables

Rat behaviors were recorded on videotape and later reviewed and scored with the help of *The Observer* computer program developed by Noldus Information Technology. Using a modification of the methods described by Smotherman et al. [41], we sorted observed spontaneous neonatal behaviors into 12 exclusive and exhaustive categories. The scoring system is a reliable one. Within our laboratory, inter-rater reliability correlations have ranged from *r*s of .67–.99 with a mean of .91. Because they were the most sensitive indicators of taste recognition, this paper focuses on orofacial movements: (a) a combination of mouth movements and licks and (b) an index of perseverative mouthing. A mouth movement was defined as an opening and closing of the mouth. Each protrusion of the tongue was counted as a licking movement. The combined mouthing and licking statistic was computed for each subject by totaling the number of mouth movements and licks. The index of perseverative mouthing represented the number of times a mouth movement was followed by another mouth movement (within a 2-s period). For example, if there was a 2-s sequence of movements such as: "mouth, mouth, head, mouth, head, gape, mouth," this bout of behavior would receive a perseverative mouthing score of 3.

1.5. Statistical analyses

The data were analyzed via a two-way analysis of variance [ANOVA: Drug (0, 50, or 100 mg/kg ketamine) × Taste pretreatment (SAC or water)] using a linear model (SAS, SAS Institute, Cary, NC) compensating for unequal *N* values [16]. Since all the rats in a particular litter received the same conditioning treatment, we included litter as an independent, random, and nested factor (within the two pre-exposure treatments). This approach controls for litter effects and offers a direct statistical test of the significance of such effects [7,15]. In the current analysis, however, if an initial examination of the data failed to reveal a significant litter effect, a subsequent analysis was run without consideration of litter. Post-hoc comparisons of the SAC-pretreated and water-pretreated rats, within each drug dose, were conducted using the Duncan's multiple range test [16]. An $\alpha = .05$ was adopted throughout these tests.

2. Results

The current data indicate that perinatal rats can discriminate between a novel and familiar taste. On P3, control rats (i.e., those that did not receive ketamine on E19) previously treated with SAC, exhibited significantly more mouth movements and licks than rats previously treated with water on E19 (see Fig. 1). Similarly, perseverative mouth movements were much more prominent in neonates tasting familiar SAC (see Fig. 2). The ANOVA of the mouthing and licking data revealed a significant SAC

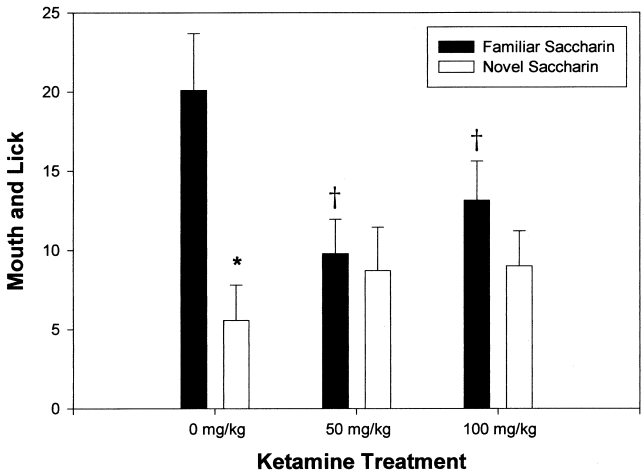

Fig. 1. Mean ± S.E.M. mouth and lick movements of P3 neonates following a taste of SAC. Rats that tasted SAC on E19 exhibited significantly ($*P \leq .05$) more mouth movements and licks (compared to control rats that received oral lavage of water on E19) when re-exposed to SAC on P3. This differential behavioral response to a novel vs. familiar taste was not observed in rats pretreated with ketamine. Compared to non-drugged animals, rats pretreated with ketamine significantly reduced mouth and lick movements following exposure to a familiar tastant ($^{\dagger}P < .05$). This phenomenon was not observed in rats tasting novel SAC.

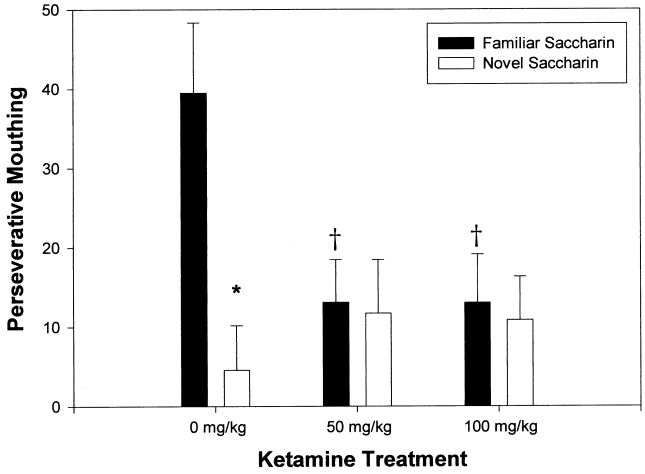

Fig. 2. Mean ± S.E.M. of perseverative mouth movements of P3 neonates following a taste of SAC. Rats that tasted SAC on E19 exhibited significantly ($*P \leq .05$) more perseverative mouthing (compared to control rats that received oral lavage of water on E19) when re-exposed to SAC on P3. This differential behavioral response to a novel vs. familiar taste was not observed in rats pretreated with ketamine. Compared to non-drugged animals, rats pretreated with ketamine significantly reduced perseverative mouth movements following exposure to a familiar tastant ($^{\dagger}P < .05$). This phenomenon was not observed in rats tasting novel SAC.

pretreatment effect [$F(1,120) = 9.49$; $P < .05$], and a significant Drug × Taste pretreatment interaction [$F(2,120) = 3.17$; $P < .05$]. Likewise, the ANOVA of the perseverative mouth movements following SAC drinking also showed a significant SAC pretreatment effect [$F(1,120) = 5.90$; $P < .05$], and a significant Drug × Taste pretreatment interaction [$F(2,120) = 3.78$; $P < .05$]. Post-hoc analyses indicated that the differential motor response to novel vs. familiar SAC was significant for the non-drugged control animals. However, ketamine-treated rats exhibited similar orofacial movements independent of their history with SAC (see Figs. 1 and 2). Thus, both 50 and 100 mg/kg doses of ketamine blocked this taste recognition memory.

Post-hoc analyses provided a clear indication that ketamine significantly reduced both perseverative mouthing, as well as combined mouth and lick movements following a familiar taste of SAC. This effect was prominent in animals treated with either 50 or 100 mg/kg ketamine. On the other hand, a trend for ketamine to increase these orofacial movements when SAC was novel did not achieve statistical significance. These data indicate a rather selective effect of ketamine on orofacial responses following exposure to a familiar tastant. At the same time, ketamine has a more limited effect on responses to a novel gustatory stimulus.

Baseline mouthing and licking movements were low (and not significantly different) for both groups of non-drugged control animals that were about to taste familiar (mean ± S.E.M. = 4.2 ± 1.32) or novel SAC (5.04 ± 1.01). Moreover, ketamine pretreatment did not significantly alter these levels of baseline orofacial movements.

3. Discussion

The data presented here indicate that P3 neonatal rats exhibited different orofacial movements depending on whether the taste of SAC was novel or familiar. The frequency of mouthing and licking responses is enhanced following lavage with familiar SAC and relatively suppressed after novel SAC. Ketamine administered before the initial taste manipulation (on E19) significantly reduced the differential response to novel vs. familiar SAC 5 days later.

The behaviors associated with this taste recognition memory may have ethological implications for the rat. In contrast with gaping mouth movements, which may function to expunge aversive tastes from the mouth, the mouthing and licking responses we recorded are consummatory and presumably aimed at ingestion [11]. Thus, few mouthing and licking movements may be associated with neophobia in reaction to a novel taste. Conversely, the enhanced mouthing and licking movements we recorded following lavage with a familiar taste may indicate recognition of the safety of the sweet solution — leading to ingestion.

The data presented here are consistent with the idea that the classification of a taste, as novel or familiar, may be based predominantly in the memory of the previous gustatory experience. NMDA receptor blockade with ketamine reduces the orofacial response to a previously experienced taste and reduces responding to a level that is similar to the initial reaction when the stimulus is novel. The baseline motor responding in ketamine-treated rats is not significantly different than that of non-drugged animals. Likewise, ketamine did not significantly alter orofacial motor responses to novel SAC. Thus, our data indicate that fetuses pretreated with ketamine on E19 are not generally hypoactive upon tasting SAC on P3. Rather, having had ketamine before initial exposure to SAC on E19, rats selectively reduce their ingestive-like mouth movements when a familiar taste is presented on P3. In essence, the neonates responded as if the taste was novel.

This taste recognition memory is a non-associative memory and has some similarity to the "priming" phenomenon that has been well studied in human subjects. Human priming often involves an increased facility for detecting or identifying words, or other stimuli, as a result of their prior presentation [43,47]. According to Vriezen et al. [48, p. 944], " . . . the mere presentation and processing of an item is sufficient to leave a trace in the perceptual representation system [37,38,47]. It is the reactivation of this trace on subsequent presentations that accounts for the repetition priming effect." This facilitative effect is presumably mediated by a neural memory system separate from that involved in performance on explicit or direct tests of recall and recognition [43].

The underlying mechanisms of the phenomenon described here have not been fully explored. Ketamine may block this taste recognition memory response by either

impairing the memory of SAC or by altering the original gustatory sensation. However, several lines of evidence suggest that NMDA receptors may have a limited role in taste sensation. Non-NMDA receptors are more abundant than NMDA receptors in taste buds [5]. Similarly, neuronal responses in the nucleus of the solitary tract (the second order neurons of the taste pathway) are completely blocked by the non-NMDA receptor antagonist 6-cyano-7-nitro-quinoxaline-2,3-dione (CNQX) but are not significantly affected by the NMDA receptor antagonist APV [39].

Ketamine, in particular, has minimal ability to modulate sweet taste sensations. Data from our laboratory indicate that rats normally prefer 0.3% over 0.6% SAC, and ketamine does not disrupt this pattern of consumption [26]. It should be noted that the ketamine doses (0.1, 1.0, or 10 mg/kg, ip) used in these sensory control experiments were lower than those employed in the current studies (50 or 100 mg/kg, ip). However, other reports indicate that ketamine doses ranging from 0.1 to 70 mg/kg, ip, can block a CTA in young rats [28]. Thus, it seems that doses of ketamine that fail to alter taste discriminations are still capable of blocking CTA formation. While not conclusive, this dissociation between the effects of ketamine on sensation and memory suggests that the drug's effects on conditioning are not always tied to drug-induced sensory changes.

Further information regarding NMDA receptor blockade and sensory experience may be obtained from the literature on gustatory habituation and neophobia. An intact gustatory capacity would be important to the demonstration of taste habituation. Our laboratory, and researchers in other laboratories, have shown that rats treated with <70 mg/kg of ketamine are capable of habituating to the taste of SAC [1,28]. Additional evidence indicating that blockade of NMDA receptors has a limited ability to alter gustatory sensation comes from experiments involving other NMDA receptor antagonists, phencyclidine (PCP) and MK801. Mastropaolo et al. [19] showed that an injection of PCP did not prevent rats from adjusting their drinking of SAC, depending on whether it had been previously associated with LiCl in a CTA paradigm. A similar failure to disrupt gustatory neophobic effects was observed when the NMDA receptor antagonist MK-801 was used [36].

Our evidence that ketamine does not significantly impair taste sensation in fetuses is also supported by the observation that ketamine (100 mg/kg administered through the maternal circulation — as in the current study) can actually enhance CTAs of E18 rat pups [23]. This age-dependent response to NMDA receptor blockade has been recently been explored in some depth [27]. Apparently, NMDA receptor blockade does not eliminate the ability of these younger fetuses to taste, since they can associate SAC and LiCl on E18, and then exhibit a CTA when tested over 2 weeks later [23]. It should be noted that ketamine's lack of ability to modulate taste might not predict its influence on other sensory systems [17,18,32]. However, if it is the case that ketamine has a limited ability to alter gustatory sensation, then the data reported here may reflect ketamine's ability to disrupt this non-associative taste recognition memory.

Alternatively, since fetuses were under the influence of ketamine on E19 but not during the behavioral test on P3, our findings may be influenced by state-dependent effects of the drug [31]. Experiments aimed at differentiating state-dependent effects from memory disruptions typically include a group of animals that are under the drug's influence both at the time of acquisition and testing. Ketamine has been used as a pediatric anesthetic [3,21] and has clear dose-dependent effects on spontaneous motor activity [2,20,32]. Thus, giving 50–100 mg/kg of ketamine to P3 neonates before behavioral testing would have significantly impaired the animal's ability to move and, therefore, to demonstrate a taste recognition memory. We have reported previously that ketamine impairs the formation of a CTA in neonatal rats [28]. Within the context of this study we controlled for state-dependent effects by administering lower doses of ketamine (i.e., ones that did not significantly retard movement) before behavioral testing. This procedure did not reveal state-dependent effects of ketamine. Our data are consistent with those of Welzl et al. [49], who did not detect state-dependent effects of ketamine (25 mg/kg) on gustatory learning in adult rats. However, some state-dependent effects of this same dose of ketamine have also been reported [1]. Since the ideal state-dependent control study (with equal doses of ketamine administered at times of training and test) could not be performed in the context of the current taste recognition memory experiment, the means by which ketamine blocks perinatal taste recognition memories remains uncertain.

The data presented here should be extended to determine the generalizability and robustness of ketamine's effects on memory. As described above, we have previously shown that ketamine can impair associative memory formation in E19 fetuses [24] and P0 neonates [28]. A ketamine dose as low as 0.1 mg/kg, ip, administered to neonates, can block formation of a CTA [28]. However, behavioral testing of these animals occurred when the animals were <2 weeks old. Future experiments should attempt to extend our work by measuring behavioral responses later in development when animals have a broader response repertoire. The effects of ketamine on both associative and non-associative memories should also be explored, as should the lowest portions of the dose–response curve. Finally, the use of behavioral measures not directly influenced by NMDA receptor blockade would allow further estimation of ketamine's state-dependent effects on learned responses.

NMDA receptor antagonists have a well-known ability to block the formation/expression of a variety of associative memory tasks in adult animals. Ketamine or MK-801 induces learning/performance deficits in mature animals learning a water maze [50], a delayed alternation task [13], or undergoing classical fear conditioning [51]. Further, ketamine administration (0.1, 10, or 70 mg/kg, ip) can block CTA formation in neonatal (P0) rats [28]. Our data extend

these findings and suggest that the acquisition/demonstration of non-associative memories, established in fetuses, may also be impaired by NMDA receptor blockade.

The apparent ability of fetuses to exhibit differential behavioral responses to a taste dependent on its novel, or familiar, characteristics reinforces the current concept of the fetus and neonate as sophisticated sensors and responders to the uterine and extra-uterine environment. Our data indicate that NMDA receptor blockade can disrupt this taste recognition memory response. Future studies will explore further the specific neural mechanisms by which this disruption is mediated.

Acknowledgments

The authors wish to thank David Revta and William Liggett for their assistance in building the neonatal testing unit. The following students and colleagues at Baldwin-Wallace College provided excellent technical support in these studies: Amy Booth, Kathryn Bryan, April Carter, Christine Dengler, Dominic Donnellan, Ben Girdler, Cynthia Kenmuir, Nadia Lelutiu, Colleen McMullen, Lora Pagel, Anaibelith Perez, Kristie Randall, Louie Rundo, Erin Simon, Tennille Lewis, Melissa Vanderkaay, Jessica Vensel, Bettina Weber, and Jessica Wolf. Partial presentations of these data were made at the 1999 meeting of the *International Behavioral Neuroscience Society.* Supported by NSF Award: 9514799.

References

[1] Aguado L, San Antonio A, Perez L, del Valle R, Gomez J. Effects of the NMDA receptor antagonist ketamine on flavor memory: conditioned aversion, latent inhibition, and habituation of neophobia. Behav Neural Biol 1994;61:271–81.

[2] Alessandri L, Battig K, Welzl H. Effects of ketamine on tunnel maze and water maze performance in the rat. Behav Neural Biol 1989;52:194–212.

[3] Bergman SA. Ketamine: review of its pharmacology and its use in pediatric anesthesia. Anesth Prog 1999;46:10–20.

[4] Blackburn JR, Pfaus JG, Phillips AG. Dopamine functions in appetitive and defensive behaviors. Prog Neurobiol 1992;39:247–79.

[5] Caicedo A, Kim K-N, Roper SD. Glutamate-induced cobalt uptake reveals non-NMDA receptors in rat taste cells. J Comp Neurol 2000;413:315–24.

[6] Danysz W, Wroblewski JT, Costa E. Learning impairment in rats by *N*-methyl-D-aspartate receptor antagonists. Neuropharmacology 1988;27:653–6.

[7] Denenberg VH. Statistics and experimental design for behavioral and biological researchers. New York: Wiley, 1976.

[8] Ennaceur A, Delacour J. A new one-trial test for neurobiological studies of memory in rats: 1. Behavioral data. Behav Brain Res 1988;31:47–59.

[9] Even MD, Dhar MG, vom Saal FS. Transport of steroids between fetuses via amniotic fluid in relation to the intrauterine position phenomenon in rats. J Reprod Fertil 1992;96:709–16.

[10] Fagan JF, Detterman DK. The Fagan test of infant intelligence: a technical summary. J Appl Dev Psychol 1992;13:173–93.

[11] Grill HJ, Norgren R. The taste reactivity test: I. Mimetic responses to gustatory stimuli in neurologically normal rats. Brain Res 1978;143:263–79.

[12] Harris EW, Ganong AH, Cotman CW. Long-term potentiation in the hippocampus involves activation of *N*-methyl-D-aspartate. Brain Res 1984;323:132–7.

[13] Hauber W, Schmidt WJ. Acquisition, but not retrieval of delayed alternation is impaired by ketamine. Res Commun Psychol Psychiatry Behav 1990;15:17–29.

[14] Hepper PG. Adaptive fetal learning: prenatal exposure to garlic affects postnatal preferences. Anim Behav 1988;36:935–6.

[15] Holson RR, Pearce B. Principles and pitfalls in the analysis of prenatal treatment effects in multiparous species. Neurotoxicol Teratol 1992;14:221–8.

[16] Kirk RE. Experimental design: procedures for the behavioral sciences. 2nd ed. Monterey, CA: Brooks/Cole, 1982.

[17] Krystal JH, Karper LP, Bennett A, D'Souza DC, Abi-Dargham A, Morrissey K, Abi-Saab D, Bremner JD, Bowers MB, Suckow RF, Stetson P, Heninger GR, Charney DS. Interactive effects of subanesthetic ketamine and subhypnotic lorazepam in humans. Psychopharmacology 1998;135:213–29.

[18] Krystal JH, Karper LP, Seibyl JP, Freeman GK, Delaney R, Bremner DJ, Heninger GR, Bowers MB, Charney DS. Subanesthetic effects of the noncompetitive NMDA antagonist, ketamine, in humans. Arch Gen Psychiatry 1994;51:199–214.

[19] Mastropaolo JP, Moskowitz KH, Dacanay RJ, Riley AL. Conditioned taste aversions as a behavioral baseline for drug discrimination learning: an assessment with phencyclidine. Pharmacol Biochem Behav 1989;32:1–8.

[20] Meliska CJ, Trevor AJ. Differential effects of ketamine on schedule-controlled responding and motility. Pharmacol Biochem Behav 1978;8:679–83.

[21] Menache L, Eifel PJ, Kennamer DL, Belli JA. Twice-daily anesthesia in infants receiving hyperfractionated irradiation. Int J Radiat Oncol Biol Phys 1990;18:625–9.

[22] Mickley GA, Ferguson JL, Nemeth TJ, Mulvihill MA, Alderks CE. Spontaneous perseverative turning in rats with radiation-induced hippocampal damage. Behav Neurosci 1989;103:722–30.

[23] Mickley GA, Lovelace JD, Farrell ST, Chang KS. The intensity of a fetal aversion is modulated by the anesthesia used during conditioning. Dev Brain Res 1995;85:119–27.

[24] Mickley GA, Remmers-Roeber DR, Crouse C, Peluso R. Ketamine blocks a taste-mediated conditioned motor response in perinatal rats. Pharmacol Biochem Behav 2000;66:547–52.

[25] Mickley GA, Remmers-Roeber DR, Crouse C, Walker C. Detection of novelty by perinatal rats. Physiol Behav 2000;70:217–25.

[26] Mickley GA, Remmers-Roeber DR, Crouse C, Walker C. *N*-methyl-D-aspartate (NMDA) receptor blockade and sensory experience (in review).

[27] Mickley GA, Remmers-Roeber DR, Dengler CM, Kenmuir CL, Crouse C. Paradoxical effects of ketamine on the memory of fetuses of different ages (in review).

[28] Mickley GA, Schaldach MA, Snyder KJ, Balogh SA, Len T, Neimanis K, Goulis P, Hug J, Sauchak K, Remmers-Roeber DR, Walker C, Yamamoto BK. Ketamine blocks a conditioned taste aversion (CTA) in neonatal rats. Physiol Behav 1998;64:381–90.

[29] Morris RGM, Anderson E, Lynch GS, Baudry M. Selective impairment of learning and blockade of long-term potentiation by an *N*-methyl-D-aspartate receptor antagonist, AP5. Nature 1986;319:774–6.

[30] O'Keefe J, Nadel L. The hippocampus as a cognitive map. Oxford, UK: Clarendon Press, 1978.

[31] Overton DA. State-dependent learning and drug discrimination. In: Iverson LL, Iversen SD, Snyder SH, editors. Handbook of psychopharmacology, vol. 18. New York: Plenum, 1983. pp. 59–127.

[32] Patel IM, Chapin JK. Ketamine effects on somatosensory cortical single neurons and on behavior in rats. Anesth Analg 1990;70:635–44.

[33] Pedersen PE, Blass EM. Prenatal and postnatal determinants of the first sucking episode in albino rats. Dev Psychobiol 1982;15:349–55.
[34] Reid IC, Morris RGM. *N*-Methyl-D-aspartate receptors and learning: a framework for classifying some recent studies. In: Meldrum BS, Moroni F, Simon RP, Woods JH, editors. Excitatory amino acids. New York: Raven Press, 1991. pp. 521–32.
[35] Robbins TW, Everitt BJ. Functions of dopamine in the dorsal and ventral striatum. Semin Neurosci 1992;4:119–27.
[36] Robinson GS, Crooks GB, Shinkman PG, Gallagher M. Behavioral effects of MK-801 mimic deficits associated with hippocampal damage. Psychobiology 1989;17:156–64.
[37] Schacter DL. Perceptual representation systems and implicit memory: toward a resolution of the multiple memory systems debate. In: Diamond A, editor. The development and neural bases of higher cognitive functions. Annals of the New York Academy of Sciences, vol. 608. New York: New York Academy of Sciences, 1990. pp. 543–71.
[38] Schacter DL. Priming and multiple memory systems: perceptual mechanisms of implicit memory. J Cognit Neurosci 1992;4:244–56.
[39] Smith DV, Li CS, Davis BJ. Excitatory and inhibitory modulation of taste responses in the hamster brainstem. Ann NY Acad Sci 1998;855:450–6.
[40] Smotherman WP. In utero chemosensory experience alters taste preferences and corticosterone responsiveness. Behav Neural Biol 1982;36:61–8.
[41] Smotherman WP, Richards LS, Robinson SR. Techniques for observing fetal behavior in utero: a comparison of chemomyelotomy and spinal transection. Dev Psychobiol 1984;17:661–74.
[42] Smotherman WP, Robinson SR. Dimensions of fetal investigation. In: Smotherman WP, Robinson SR, editors. Behavior of the fetus. Caldwell, NJ: Telford Press, 1988. pp. 19–34.
[43] Squire LR. Declarative and non-declarative memory: multiple brain systems supporting learning and memory. J Cognit Neurosci 1992;4:232–43.
[44] Teicher MH, Blass EM. First sucking response of the newborn albino rat: the roles of olfaction and amniotic fluid. Science 1977;198:635–6.
[45] Thompson AM, West DC, Lodge D. An *N*-methyl-D-aspartate receptor-mediated synapse in rat cerebral cortex: a site of action of ketamine? Nature 1985;313:479–81.
[46] Tulving E, Markowitsch HJ, Kapur S, Habib R, Houle S. Novelty encoding networks in the human brain: data from positron emission tomography studies. NeuroReport 1994;5:2525–8.
[47] Tulving E, Schacter DL. Priming and human memory systems. Science 1990;247:301–96.
[48] Vriezen ER, Moscovitch M, Bellos SA. Priming effects in semantic classification tasks. J Exp Psychol Learn Mem Cognit 1995;21:933–46.
[49] Welzl H, Alessandri B, Battig K. The formation of a new gustatory memory trace in rats is prevented by the noncompetitive NMDA antagonist ketamine. Psychobiology 1990;18:43–7.
[50] Wesierska M, Macias-Gonzalez R, Bures J. Differential effect of ketamine on the reference and working memory versions of the Morris water maze task. Behav Neurosci 1990;104:74–83.
[51] Xu X, Davis RE. *N*-Methyl-D-aspartate receptor antagonist MK-801 impairs learning but not memory fixation or expression of classical fear conditioning in goldfish (*Carassius auratus*). Behav Neurosci 1992;106:307–14.