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The effects of reversible inactivation by lidocaine of ventral striatum and intralaminar nucleus of the thalamus in match to sample tasks with and without delays trained in the lever box

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THE EFFECTS OF REVERSIBLE INACTIVATION BY LIDOCAINE OF VENTRAL STRIATUM AND INTRALAMINAR NUCLEUS OF THE THALAMUS IN MATCH TO SAMPLE TASKS WITH AND WITHOUT DELAYS TRAINED IN THE LEVER BOX

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DEDICATION

This work is dedicated to my family, who has taught me life's most important lessons: to follow one's heart, to learn for the sake of learning, and to believe in oneself. I am grateful to them for a lifetime of guidance, warmth, unflagging support, and love.

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Three experiments systematically examined the effects of reversibly inactivating intralaminar nuclei and ventral striatum with infusions of lidocaine. The ventral striatum and the intralaminar nucleus were targeted based on deficits in accuracy and speed of responding in delayed conditional discrimination tasks associated with permanent lesions of these structures (Mair, Burk and Porter, 1998).

The present research showed that a 4.0 μl of 2% lidocaine caused impairments accuracy, but not in response speed, in a match to sample task trained in the lever box without delays when the internal cannula projects 0.5 mm from the guide cannula. One microliter of 2%, 4% or 8% lidocaine caused impairments in accuracy, but not in response speed, in a match to sample task trained with delays of 1.0, 3.0, 8.0 and 13.0 seconds, when the internal cannula projects 2.0 mm beyond the guide cannula. Taken together, we conclude that the specific parameters of lidocaine delivery have an impact on deficits in this task. This impact is likely to be dependent on the size of the inactivated area of the intralaminar nucleus and the ventral striatum.

INTRODUCTION

Relevant Anatomy

Brief anatomy of amnesia. Neuroscientists have yet to agree on the functional roles played by discrete brain areas that govern memory. Although damage to individual brain areas may disrupt memory, it is not known precisely how these areas interact to form a neural *system* of memory. Subcortical amnesia is most often caused by damage to the basal forebrain, medial temporal lobe, or the thalamic region of the diencephalon (Markowitsch and Pritzel, 1985; Burk and Mair, 1999). Studies of severely amnestic patients often show that discrete, individual lesions to these areas generate distinguishable deficits but spare other abilities (Zola-Morgan and Squire, 1993). Vascular disruption, neoplasms and degenerative conditions are chief sources for damage to these three areas (Cummings, 1990).

The basal forebrain is susceptible to degenerative processes and to disruption of vascular beds. Parkinson's disease and Huntington's disease are both characterized by degeneration of basal forebrain structures that include the basal ganglia. In addition, amnesia often results from the disturbance of normal metabolism following occlusion or breach of the anterior communicating artery that also affects the basal ganglia (Cummings, 1989; DeLuca and Diamond, 1995). Alzheimer's disease is also often linked to basal forebrain damage in humans. Damage to the septum, diagonal band of Broca and the nucleus basalis affects memory as well as intellectual ability, personality, emotion, and

sensory-perceptive function (Muir, 1997; Perry, Haroutunian, Davis, Levy, Lantos, Eagger, et al, 1994).

Several types of memory tasks are affected by basal forebrain damage in humans, including delayed matching to position, delayed matching to sample, visual discrimination, and the acquisition and retention of spatial information (Muir, 1997; Givens and Sarter, 1997). Cognitive effects of injury to basal forebrain are considered largely due to the connections that basal forebrain makes to other memory-related areas such as the hippocampus, and cortex (Givens and Sarter, 1997).

Animal models of basal forebrain lesions have also contributed insight to the role of this region in memory. Lesion studies and pharmacological manipulations (especially on cholinergic systems), demonstrate that injury here disrupts discrimination tasks, conditional discrimination, passive av`oidance, and spatial tasks (See Collerton, 1986, for a review).

Medial temporal lobe damage is frequently caused by epileptic seizure-related degeneration or surgery performed to correct the seizure condition. Alternatively, Alzheimer's disease, or herpes encephalitis may cause medial temporal lobe amnesia (Markowitsch and Pritzel, 1985; Parkin, 1992, in Squire and Butters). Damage to the medial temporal lobe, including to the parahippocampal area, the hippocampus, perirhinal cortex and entorhinal cortex, may interfere with remembering trial-specific information over a prolonged time, such as during delayed conditional discrimination tasks or the acquisition of new information (Squire and Zola-Morgan, 1991). Additionally, insult to involvement of the hippocampus may impair memory for spatial information in both

humans and animals. In animals, hippocampal damage often disrupts performance in the Morris water maze, radial arm maze and spatial alternation tasks, under a variety of experimental conditions (Squire and Butters, 1993).

Amnesia may also follow bilateral disturbance (such as infarction) of the paramedian nuclei and other nuclei found inside the internal medullary lamina of thalamus (Bogousslavsky, Regli, and Uske, 198; von Cramon, Hebel and Schuri, 1985) or tumors at the base of the third ventricle (McEntee, Biber, Perl, and Benson, 1976). However, the main source of thalamic-related forgetting is a result of Korsakoff's syndrome. This condition caused by thiamin-deficiency, affects the periventricular structures in brainstem and diencephalon with possible involvement of mammillary bodies (Victor, Adams, and Collins, 1989).

Damage to the midline diencephalon, and especially to multiple nuclei of the thalamus in humans, may impair remembering information for temporal sequence and information related to space (Zola-Morgan and Squire, 1993). In animal models, rats with permanent prefrontal and intralaminar nucleus lesions also perform slower than controls in a standard task of forgetting in the lever box (Burk and Mair, 1998; Harrison and Mair, 1996; Porter and Mair, 1997; Young, Stevens, Converse, and Mair, 1996)).

Although many research studies confirm the effects of these discrete lesions, less agreement has been reached regarding effects of damage on a networked system of memory in the brain. Advances made in the 1970s and 1980s in anatomical staining techniques reveal that prefrontal cortex (which is connected with basal forebrain and medial temporal lobe), and intralaminar nuclei are heavily interconnected to each other,

and that these areas connect to the basal ganglia. Of particular interest to the field of memory research is the extensive anatomical and neurochemical connections between frontal cortex, intralaminar nucleus and the striatum (Berendse and Groenewegen, 1991; Groenewegen, Wright, and Uylings, 1997; Wright and Groenewegen, 1995). Despite these three chief areas implicated in clinical amnesia, medial temporal lobe, basal forebrain, and midline thalamus, "[t]he principal structure involved in most subcortical dementias is the striatum" (Cummings, 1989, p. 11).

Striatal anatomy. The mammalian basal ganglia contains the striatum, including the caudate and putamen, the internal and external portions of the globus pallidus, nucleus accumbens, and sometimes the substantia nigra, ventral tegmental area, and the subthalamic nucleus. These last three areas are occasionally grouped with the basal ganglia due to known relationships between these areas and striatum and pallidum (Albin, Young, & Penney, 1989; Marín, Smeets & González, 1998). Many researchers also include olfactory tubercle and the ventral portion of the nucleus accumbens as members of striatum based on the cytoarchitecture and connections of these areas to cortex and thalamus. Olfactory tubercle, portions of nucleus accumbens and ventromedial caudate-putamen comprise ventral striatum (Graybiel, 1990; Heimer, Switzer, and Van Hoesen, 1982; Heimer, Alheid, and Zaborszky, 1985; McGeorge and Faull, 1989; Parent, 1990).

Research in recent years has brought increased interest in the basal ganglia, especially with regard to changes in the anatomical conceptualization of the area. Formerly, the basal ganglia were thought to be restricted in projections to a few discrete areas of cortex via the ventrolateral thalamus. Now the basal ganglia are known to project

information to, and receive information from, the entire cortex. Connections arrive at cortex via several thalamic regions (Alexander, DeLong, & Strick, 1986; Alexander & Crutcher, 1990; DeLong, 1990; Parent, 1990; Berendse, Galis-de-Graf, & Groenewegen, 1992). The sheer volume of anatomical connections between basal ganglia and other areas of the brain warrants careful investigation; for instance, researchers agree that the physical circuitry between cortex, thalamus and striatum is among the most highly sophisticated and complex in the central nervous system (Goldman-Rakic & Selemon, 1990). It is useful, therefore, to consider the circuitry of the striatum in light of other areas of the brain to gain an understanding of the relationship between basal ganglia anatomy and higher cognitive functions such as memory.

Cortico-striatal circuits. Understanding the intricate relationship between cortex and striatum is enormously important for discovering striatal function. Cortical projections represent the major input to striatum and are highly organized on several levels. Generally, cortical fibers arise from a particular cortical layer (layers V and Va) and arrive at the striatum with topographical specificity (McGeorge and Faull, 1989). While the striatum receives projections from all areas of cortex, the termination fields in striatum are highly organized according to anterior-posterior, medial-lateral, and dorsal-ventral gradients. Termination fields of functionally distinct cortical areas are believed to correspond to functionally distinct striatal areas. Furthermore, these termination fields in striatum can be classified into three general, "longitudinally-oriented" regions (McGeorge and Faull, 1989, p. 503). McGeorge and Faull (1989) used retrograde tracing techniques to illustrate that the neocortex projects to caudate-putamen, mesocortex projects chiefly to medial and ventral caudate-putamen and to ventral striatum, and allocortex, or limbic-

related areas, project to ventral striatum. Several other neuronal tracing studies conclude that sensorimotor areas of cortex project to putamen in the monkey and lateral caudate-putamen in the rat, while association areas of cortex project to caudate nucleus in the monkey and medial caudate-putamen in the rat. Limbic and paralimbic areas of cortex, including orbitofrontal cortex, project to ventral striatum, including ventral nucleus accumbens, ventromedial caudate-putamen and olfactory tubercle (Afifi and Bergman, 1998; Alexander, DeLong and Strick, 1986; Kemp and Powell, 1970; Parent, 1990; Selemon and Goldman-Rakic, 1985; Wichman and DeLong, 1996).

Researchers agree that three general areas of striatum, dorsolateral, medial and ventral striatum, are involved in a wide range of functions and contain some overlapping cortical input (Berendse, Galis-de-Graf, and Groenewegen, 1992; Haber, Lynd-Balta, and Spooren, 1994; Kemp and Powell, 1970). Cortical information received at each of these regions of striatum is processed and projected back to the original areas of cortex (Afifi, and Bergman, 1998; DeLong, 1990; Selemon and Goldman-Rakic, 1985; Wright and Groenewegen, 1996). However, some question remains regarding which cortical layer receives the returning afferents (Groenewegen, Wright, and Uylings, 1997; Mair, Burk and Porter, 1998).

Any discussion of cortico-striatal circuits must necessarily lead to a discussion of related midline and intralaminar circuits. Like many others, Groenewegen, Wright and Uylings (1997) conclude that most cortical activity, including memory, is due in part to events in striatum. Their position is based on intricate reciprocal connections between cortex and striatum, as well as the striatum's modulating effects on midline and

intralaminar nuclei (Berendse and Groenewegen, 1991; Goldman-Rakic and Selamon, 1990; Groenewegen, et al., 1997; Nauta, Smith, Faull, Domesick, 1978, in Haber, Lynd-Balta, and Spooren 1994).

Thalamo-striatal circuits. A second major source of projections to striatum is provided by both intralaminar and midline nuclei of thalamus (Berendse and Groenewegen, 1990; Smith, Bennett, Bolam, Parent, and Sadikot, 1994). Generally, the intralaminar nuclei are divided into rostral (anterior) and caudal (posterior) groups. The rostral intralaminar group includes the central medial, central lateral, and paracentral nuclei, while the caudal group includes the parafasicular nucleus in rats and the analogous central médian-parafasicular nuclei in primates (Groenewegen and Berendse, 1994; Jones, 1985). Several elaborate tracing studies reveal that all intralaminar nuclei project massively to areas of the striatum and less heavily to many cortical areas, chiefly the "frontal, medial, and dorsolateral cortex" (Jones, 1985, p. 628; Parent, Bourassa, and Deschênes, 1996; Herkenham, 1979). Furthermore, all areas of cortex receive information from at least one intralaminar nucleus (Groenewegen and Berendse, 1994). Generally, the rostral group, including the central median, central lateral and paracentral nuclei, projects to anterior portions of the caudate and putamen in the primate. The caudal group, the parafasicular or center médian nucleus, projects to posterior portions of the caudate and to putamen (Jones, 1985).

Despite these anatomical subdivisions, it is important to note that Groenewegen and Berendse (1994) consider the two classes (rostral and caudal) of intralaminar nuclei

continuous. They support this position on the basis of similar patterns of reciprocal associations between the rostral and caudal intralaminar nuclei and other brain regions.

Like the intralaminar nuclei, midline nuclei have been subdivided into related groups. The dorsal components of the midline thalamus are the paratenial, paraventricular and intermediodorsal nuclei. The ventral midline thalamic components include the rhomboid nucleus (which is considered by some to belong to the intralaminar group) and the reuniens nucleus (Groenewegen and Berendse, 1994; Jones, 1985).

Historically, intralaminar and midline regions have been treated as segregated groups of nuclei; however, strong arguments in recent years from Groenewegen and Berendse (1994) and others propose that intralaminar nuclei and dorsal midline nuclei may be considered physiologically and functionally continuous. This idea is based on several studies that reveal distributions of projection fibers from the intralaminar nuclei resemble the distributions of projection fibers from the dorsal midline nuclei. The importance of these labeling studies is two-fold. First, it demonstrates physiologically the closure of the circuitry between cortex, striatum, and thalamus. Second, it provides a vehicle for discussing possible functional relationships between discrete regions of these areas. It is these functional relationships that address the usefulness of considering memory as a neural system rather than discrete "storage" sites (Squire and Butter, 1992).

Critical to the discussion of cortico-striatal and thalamo-striatal circuitry is the finding that midline and intralaminar nuclei project to areas of frontal cortex and to areas of ventral striatum that are connected to each other via cortico-striatal fibers. Retrograde labeling of midline nuclei reveals ipsilateral projections from at least two layers of chiefly

ventromedial cortex, such as medial orbital cortex, agranular insular areas, perirhinal, entorhinal, infralimbic, and ventral anterior cingulate cortex. Intralaminar nuclei injections of radioactive tracer labeled dorsal areas and lateral of prefrontal cortex such as lateral orbital cortex, dorsal prelimbic, and dorsal agranular insular areas. Sophisticated labeling studies reveal very little overlap of projection fibers between cortex and thalamus (Berendse and Groenewegen, 1991).

The specificity with which thalamic nuclei receive input from cortex corresponds to the specificity with which these same thalamic nuclei target three main areas of striatum. Midline nuclei of thalamus, which receive information from limbic areas of cortex, project to medial and ventral striatum. Intralaminar nuclei receiving input from sensory cortex project to lateral areas of striatum. Intralaminar nuclei receiving input from motor areas of cortex project to dorsolateral portions of cortex. All three areas of striatum are also directly innervated by corresponding cortical areas (Berendse, and Groenewegen, 1990; Berendse, and Groenewegen, 1991; Groenewegen, et al, 1997; Kuhnishio and Haber, 1994; McGeorge and Faull, 1989).

Principles of organization describing cortico-thalamo-striato-cortical circuitry.

Funneled information vs. segregated loops. Current research on the role of the basal ganglia in cognitive function draws chiefly on two perspectives to describe effects of damage to nuclei of the basal ganglia or to related areas. The older model proposes that the information entering the basal ganglia from cortex is "funneled," or highly integrated within the striatum. Information is further consolidated within the output nuclei of the

basal ganglia, which are responsible for the promotion and coordination of global events (Goldman-Rakic and Selemon, 1990; Alexander, DeLong and Strick, 1986). Alternatively, the currently accepted model of basal ganglia circuitry is based on sophisticated anatomical tracing studies that reveal several parallel and largely segregated circuits. These circuits maintain predictable and separated "loops" traveling between cortex, striatum, pallidum, thalamus and back to cortex (Alexander, Strick and DeLong, 1986; Haber, Lynd-Balta, and Spooren, 1994). Specifically, Alexander and his colleagues suggest five specific loops involved in the functional anatomy of the basal ganglia. Each loop is composed of a cortical, striatal, pallidum and thalamic level, and probably includes both closed and open segments.

Despite both a great deal of anatomical and behavioral evidence to support a model of five segregated loops, some research suggests that basal ganglia afferents are not as convergent as once believed. Recent evidence from Haber, Lynd-Balta, and Spooren (1994) suggests that some circuits between areas of striatum and cortex remain segregated while other circuits converge their information between cortex and striatum. Specifically, striatal afferents from sensorimotor cortex, association cortex, and subcortical and limbic areas remain segregated, as do efferents from striatum to the internal and external areas of pallidum. Conversely, these researchers argue that afferents destined for areas in the substantia nigra may transmit information that will be integrated rather than segregated. This idea is based upon tracing studies of several injections made to various areas of dorsal or ventral striatum or to areas of pallidum. Terminal fields in the substantia nigra for these injections revealed that efferents from these areas of striatum or pallidum were not strictly topographical. Labeled termination fields in substantia nigra did not depend

upon the geographic location of the striatal injection, but instead, upon presumed function of the injected striatal area. The function of an injected striatal area is presumed from the area's cortical efferents.

From these results, Haber, et al (1994) concluded that care must be taken to consider the convergence as well as divergence of efferents relative to striatal anatomy and function. Specifically, they conclude that the ventral striatum, by virtue of feedback loops with pallidum exert an extensive influence on many areas of striatum, as ventral striatum projects to a discrete area of pallidum, but that area of pallidum projects diffusely to areas of ventral striatum.

Bergman and his colleagues (1998) reiterate conclusions drawn by Haber after using "cross-correlation techniques" to determine the extent of simultaneous firing by neurons in undamaged, dopaminergic output nuclei of striatum (p. 32). In their study, Bergman, et al. investigated the potential for coincident action potentials in monkey pallidal neurons during a GO/NO GO task. This was accomplished by recording from multiple pallidal neurons during the task in normal and parkinsonian monkeys. They predicted that simultaneous firings in the pallidum would support an information sharing hypothesis while non-simultaneous firings of pallidal neurons would support a segregated circuitry view.

The results of this experiment reveal that simultaneous firings of basal ganglia neurons only occurred in dopamine-depleted (parkinsonian) monkeys, but never occurred in normal monkeys. The authors conclude that normal striato-pallidal circuits are largely segregated, but are careful to point out that segregated circuitry in the striatum and its

outputs does not preclude the possibility of funneling or information convergence in this area. They suggest that information may be shared within just one of the loops. Alternatively, information may be shared by individual neurons via common input from several striatal cells to one pallidal cell, or from common input from subthalamic nucleus projections.

Direct vs. indirect pathway. This organizing principle contributes information regarding the functional character and information-processing role of the parallel cortico-striato-thalamic loops described above (Beiser, Hua, and Houk, 1997). The model's foundation is that a direct pathway and an indirect pathway are contained in each of the five parallel loops that connect cortex, striatum and thalamus (Alexander and Crutcher, 1990). Each of the main parallel loops between these areas contains a direct, monosynaptic path of striatal efferent fibers, whose ultimate function is to disinhibit thalamic activity, and an indirect polysynaptic path of efferent fibers whose ultimate function is to inhibit thalamic activity (Wichman and DeLong, 1996).

The direct pathway contained within each loop is believed to be composed of chiefly GABAergic fibers which arise from the striatum and synapse onto internal globus pallidus or substantia nigra reticulata to thalamic nuclei (Goldman-Rakic and Selemon, 1990; Graybiel, 1995). The indirect pathway is responsible for countering the effects of the direct pathway. The indirect path travels from striatum to the external globus pallidus to subthalamic nucleus to internal globus pallidus and exerts a net excitatory effect upon thalamus (Alexander and Crutcher, 1990; Graybiel, 1995.)

Key to the discussion of direct and indirect paths is the role this perspective plays in symptoms of Parkinson's disease. Common to Parkinson's disease is one or more types of tremor, muscle rigidity, akinesia, bradykinesia, degradation of balance or impaired postural integrity (Stacy & Jankovic, 1992; Huber and Cummings, 1992; Knight, 1992). Many of the symptoms classified under these general headings seem to result from the disruption of retrieval or execution of motor programs related to the basal ganglia and associated areas (Stacy & Jankovic, 1992). Although patients may be able to perform motor sequences with difficulty, several experiments show that they are slower to do so, resulting in both impaired reaction and movement time (Bergman, et al, 1998; Knight, 1992).

It has been proposed that disruption of the balance between the direct and indirect pathways is responsible for the delayed motor program execution, diminished motor capability, and the inability to generate the desired motor sequence in Parkinson's patients. Specifically, the loss of dopamine neurons in Parkinson's patients from the substantia nigra is thought to cause the excitation of pallidum which causes the extreme inhibition of thalamus, and results in inappropriate motor movement (Beiser, et al, 1997). Likewise, Wichman and DeLong (1996) suggest that hypoactivity in the indirect pathway, combined with dopaminergic loss in the striatum is responsible for hyperkinetic motor behavior in Huntington's and Parkinson's disease patients.

Dysfunction Following Permanent Lesions to Striatum or Thalamus

Brief neuropsychology of Parkinson's Disease. Research on cognitive impairment has, until recently, been overshadowed by extensive research on motor dysfunction in

Parkinson's disease. (Goldman-Rakic & Selemon, 1990; Albin, Young & Penney, 1989). However, Pirozzolo, Hansch, Mortimer, Webster, and Kuskowski (c.f. Dubois and Pillon, 1992) report that ninety-three percent of people with Parkinson's disease experience predictable patterns of cognitive disruptions as a result of striatal damage. Moreover, experiments in rats, cats, and monkeys reveal cognitive deficits comparable to those in human patients following both permanent and transient damage to striatal and pallidal systems (Floresco, Seamans, & Phillips, 1997; Cook & Kesner, 1988; Dunnett, 1990; Packard, Cahill, and McGaugh, 1996).

Specifically, Parkinson's patients may experience a collection of cognitive impairments that include language difficulty, bradyphrenia, decreased attention, disrupted executive function, forgetting, the inability to integrate external sensory stimuli into useful cognitive sequences, or to switch successfully between sets of information (Dubois & Pillon, 1997). Despite agreement that cognitive deficits occur in Parkinson's disease patients and in animals with damage to the basal ganglia, the specific mechanisms governing the role of the basal ganglia and anatomically related areas in cognitive function is yet unclear (Bergman, et al, 1998). Cognitive dysfunction in Parkinson's Disease and in animals with damage to comparable areas provide important insight into a complicated array of functions potentially attributable to the striatum and its myriad connections.

Brief neuropsychology of Korsakoff's disease. Most of the available information regarding the role of intralaminar nuclei in remembering comes from studies of patients with Korsakoff's syndrome, a condition caused by severe thiamin deficiency and marked by both anterograde and retrograde amnesia. While the neuropathology associated with

Korsakoff's syndrome is not entirely agreed upon, researchers agree that midline portions of the thalamus are damaged in Korsakoff's, especially the mediodorsal nuclei, dorsal lateral nuclei and the pulvinar nuclei (Pepin, Auray-Pepin, 1993; Markowitsch, 1991; c.f. Abraham, Corballis, and White; Victor, Adams, and Collins, 1989).

Although the critical pathology of Korsakoff's disease is debated, several behavioral patterns related to Korsakoff's have emerged from careful study of human patients. Chief among the deficits seen in Korsakoff's patients is the inability to perform conditional discrimination tasks such as match to sample tasks and non-match to sample tasks (Squire, Zola-Morgan and Chen, 1988).

Animal models of striatal damage. According to many researchers, several higher cognitive events are mediated by striatum. These events include planning, habit formation (Divac and Oberg, 1979; Graybiel, 1995; White, 1997); learning and memory (Graybiel, 1995), motor planning, executive functions such as making decisions, sensory-perception events, attention, problem-solving, adaptation, and goal-seeking (Brown, Schnieder, and Lidsky, 1997); and context-specific set-shifting behavior (Hayes, Davidson, Keele, and Rafal, 1998).

Despite the range of cognitive functions attributed to it, some researchers have considered the role of striatum in remembering, pointing to patterns emerging from animal studies. Based on the anatomical connections between the midline and intralaminar nuclei and striatum, Burk and Mair (1999) compared the effects of permanent lesions to several areas of striatum in a delayed match to sample task and in a serial reversal task. These researchers found that the deficits caused by some striatal lesions appear similar to effects

of intralaminar lesions in identical tasks. Rats with permanent bilateral lesions to intralaminar nuclei are impaired in a delay-independent fashion in the match to sample task trained in an operant lever box, but are unimpaired in a serial reversal task. In addition, intralaminar lesions cause rats to respond slower than controls in this delayed match to sample task (Burk and Mair, 1998). An identical pattern of deficits was found for some groups of striatal lesions.

The match to sample task used in the Burk and Mair's (1999) striatal study involved training in standard operant chambers that were equipped with three levers; two levers were placed on the front wall, one on each side of a drinking port, and one in the middle of the back wall. Four retention intervals, 1.0, 3.0, 8.0 and 13.0 seconds, were counterbalanced on a trial-by-trial basis throughout each daily session. In addition to a control surgical group, excitotoxic lesions were made to medial or lateral caudate-putamen, to nucleus accumbens or to olfactory tubercle. Results showed that lateral caudate-putamen lesioned rats were not impaired on this task. Medial caudate-putamen and nucleus accumbens lesioned rats were impaired compared to controls; however, they performed significantly better than olfactory tubercle lesioned animals. Rats with olfactory tubercle lesions were dramatically impaired in their performance, both in their accuracy and in their response speed. Interestingly, no group was impaired in the serial reversal learning task. The pattern of impairments shown by the rats with olfactory tubercle damage corresponds with impairments following intralaminar damage. Thalamic lesions cause both inaccuracy and slow responding (Burk and Mair, 1998).

Several salient points emerge from this study. First, the authors found the effects of olfactory tubercle lesions to be behaviorally comparable to the deficits observed following intralaminar thalamic nuclei lesions in the same delayed match to sample task. In addition, neither lesion affected serial reversal learning. This implies that olfactory tubercle plays a role in remembering tested by the delayed match to sample task (conditional discrimination), but not by the serial reversal task (simple discrimination). Second, the authors suggest that, based on the histological analysis, consistent damage to the nearby ventral pallidum may explain part of the results. Third, anatomical connections, behavioral deficits following lesions to intralaminar thalamic nuclei and ventral striatum and hypotheses regarding the source of human amnesia all suggest thalamus and striatum are both involved in remembering.

Animal models of intralaminar thalamic nucleus damage. Several experiments have shown the damage to lateral internal medullary lamina, a portion of the thalamus that bounds the midline and intralaminar nuclei, disrupts performance in a variety of memory tasks. Damage to intralaminar nuclei invariably results from targeting the internal medullary lamina by virtue of the close physical contiguity of these areas. This is the case whether the damage arises from pyrithiamine-induced thiamin deficiency, radiofrequency lesions or excitotoxic lesions (Mair, Burk and Porter, 1998).

Specifically, rats with extensive intralaminar nuclei damage are impaired in conditional discrimination tasks such as delayed match to sample tasks in the operant box (Burk and Mair, 1998), continuous delayed non-match to sample tasks in an olfactory chamber (Zhang, Burk, Glode, Mair, 1998) and in the radial arm maze (Porter, Burk and

Mair, 1998). The extent of damage necessary to produce such deficits was directly addressed by Mair, Robinson, Koger, Fox, and Zhang (1992). These researchers subdivided the lateral internal medullary lamina lesion into anterior and posterior portions. They then compared the effects of discrete radiofrequency lesions to either anterior or posterior regions of the thalamus in a delayed non-matching to sample task. Data revealed that the complete rostral-caudal extent of the lateral internal medullary lamina must be disrupted to impair accuracy in this task.

Lidocaine Lesions

One serious issue to address in memory research is the considerable impact of recovery of function following permanent brain lesions. The brain is a plastic organ, with the capability of reacting at the cellular level to permanent neuronal damage. Recovery of function may refer either to physiological recovery of the brain or behavioral recovery of the organism during post-surgical testing for memory deficits. In physiological recovery, one or more brain areas may compensate for the lesioned area. Additionally, undamaged brain areas may compensate for areas that are collaterally damaged downstream from the primary lesion site. In behavioral recovery of function, the organism adapts its responses during post-surgical testing to solve cognitive problems using new or different behavioral strategies.

Both types of recovery may be addressed sufficiently in research using permanent lesions. However, reversible lesions provide opportunities to ask complementary questions about the function of brain areas while minimizing recovery of function issues.

In addition to minimizing recovery of function issues, the use of reversible lesions may provide insight into the role of neural systems of memory, rather than the role of discrete brain areas. This is because the entire circuit involved in solving memory tasks remains largely physiologically intact.

The use of reversible lesions also maximizes certain statistical features of experimental design. Because the critical areas for memory remain intact during reversible lesions, each subject acts as its own control subject, capitalizing on within-subject techniques for data analysis. The use of a within-subjects design has two immediate advantages not available in experiments using permanent lesion techniques. First, within-subjects designs shrink the possibility for heterogeneity of variance among subjects. Second, the related error term is reduced because any observed differences between treatment groups result from the comparison of the data generated from identical subjects, and not from the main effect of individual subjects. Within-subjects designs are therefore considered useful vehicles to increase statistical power and detect any real effect of treatments. (Maxwell and Delaney, 1990). While one disadvantage of using within-groups designs is the possibility of carryover effects between treatment sessions, this is not considered a major problem with reversible lesioning techniques, as the chief advantage of non-permanent lesions is their temporal transience.

One powerful reversible lesioning technique is the use of local anesthetics directly applied to brain areas involved in memory. In particular, lidocaine hydrochloride suspended in physiological saline is often used. Lidocaine's mechanism of action at the neuronal membrane is to disrupt the imbalance between the membrane-bound Na+ - K+

ATPase (responsible for an outward electrical current) and the inward electrical current responsible for maintaining an excitable state in the neuron (Butterworth and Strichartz, 1990). Specifically, local administration of lidocaine probably interrupts neuronal signal conduction by interfering with gating mechanisms at sodium channels (Butterworth and Strichartz, 1990).

Behavioral effects of lidocaine infused into intralaminar nucleus. While research into the specific role of the intralaminar nucleus in remembering is growing more common, a search of the relevant literature revealed that reversible lesions to this area have not been attempted. This is surprising given what is known about the intralaminar nucleus. First, researchers have determined that radiofrequency and excitotoxic lesions to this area disrupt performance in memory-related behavioral tasks (Mair, Burk and Porter, 1998). Second, the anatomical complexity of the area makes discrete, circumscribed lesions to individual nuclei difficult. Like other thalamic areas, intralaminar nuclei are both highly contiguous with each other, and highly interconnected to other areas of the brain (Mair, 1994). Permanent lesions to an area may cause collateral or unintended damage to other nuclei. Third, the sheer number of fibers of passage through intralaminar nuclei makes the lesion technique selected of critical importance. Radiofrequency lesions to intralaminar nuclei destroy fibers of passage. Excitotoxic lesions to these areas spare fibers of passage but may cause anterograde damage to the numerous intralaminar projection sites.

Lidocaine is a popular choice to generate reversible lesions in primate and rodent models of memory loss for several reasons. It is effective in miniscule doses, untoward side effects are negligible, and there is a literature base available addressing functional

spread of effects based on autoradiography and electrophysiological excitability of local neurons at the injection site (Malpeli, 1999).

One of the most important features of lidocaine as an inactivating agent is that its pharmacological effect is temporally constrained. Lidocaine locally inactivates neural tissue in a matter of seconds, although the diameter of area ultimately inactivated by the infused dose is influenced by several factors, and is a matter for some discussion. Use of a standard dose, 1 µl of 2% lidocaine for instance, is known to last between 15 and 60 minutes (Martin and Ghez, 1999, Sandkühler and Gebhart, 19991; Sandkühler, Maisch and Zimmerman, 1987). The temporary effects of lidocaine allow each animal to experience several experimental manipulations. Consistent effects following multiple sessions trained under the same experimental conditions establishes confidence in both the methods used and the functional role of the site inactivated. Moreover, effects following multiple sessions trained under differing conditions allows the researcher to assess the sensitivity of the site to various parameters (Horel, 1991).

Choice of Matching to Sample Tasks Trained With and Without Delays

A matching to sample task is an adaptable conditional discrimination task in which a subject is shown one sample stimulus in a trial and subsequently must chose this stimulus from a pair of stimuli presented. Four main reasons justify the use of the matching to sample task for animal models of memory disorders. First, this task is powerful in that it is both flexible and rule-oriented. The task's flexibility lies in that it may be adapted to a variety of contexts and environments to suit the purpose of the experiment. Successfully negotiating the MTS task indicates that post-surgical subjects retain some level of global

function, attention, sensory capability, motor control, and motivation. A match to sample task may be designed to assess remembering related to a specific sensory modality, for instance. Alternatively, the task may be adapted to maximize spatial arrangement or the spatial relations between objects by making the task match to position rather than match to sample (Dunnett, 1992, in Squire and Butter).

That the task itself is governed by a set of consistent and predictable rules tests whether post-surgical subjects are able to learn and maintain a sequence of rule-based behaviors. The match to sample task is appropriate for a range of subjects, and has often been used to test short-term or working memory of primates, rodents and birds (Dunnett, 1992, c.f. Squire and Butters; van Hest and Steckler, 1996).

Second, typical match to sample tasks consist of a number of individual trials massed over time separated from each other by a certain interval (the inter-trial interval). The use of many, sequential trials can identify the effects of proactive interference on performance. Massed trials with a relatively short inter-trial interval increase memory load and errors made later in a session may indicate a sensitivity to such interference (Roitblatt and Harley, 1988).

Third, the use of delays on a trial-by-trial basis within a MTS task is a common and efficient way to assess the effect of a retention interval on response behaviors that are identical to those tested in the no-delay MTS task. The use of a retention interval or a series of retention intervals within a MTS task goes a long way toward isolating the ability to remember stimuli over time. Subjects may perform comparably to controls when required to remember sample stimuli over brief intervals, but are progressively impaired at

longer retention intervals. This may indicate that subjects' performance is specifically related to a memory deficit, and the rate of forgetting can be visualized as a mathematical function (Dunnett, 1992). The use of a delay following the presentation of a sample to be remembered assesses behavior based on no-longer-present stimuli (Hest and Steckler, 1996).

Fourth, the consistency with which normal subjects are able to perform this test suggests that it is reliable over a number of sessions, each of which may be separated by a day or more. The performance of normal subjects notwithstanding, the match to sample task has the ability to demonstrate possible behavioral or physiological recovery of function in permanently brain damaged subjects if performance improves after running several sessions on the same task. In this case, nothing has changed about the task from one session to another session; however, any improvement in performance may be considered a sign of recovered function. The importance of the match to sample task is that it is used under a variety of circumstances to parcel out effects of interference, time and spatial arrangement. Moreover, damage to discrete sites in the brain generates specific patterns of spared and impaired behaviors within this task.

The match to sample tasks trained with and without delays is appropriate in the current study as it is identical to procedures used by Burk and Mair to test effects of intralaminar nucleus lesions (1998) and ventral striatal lesions (1999). Following the collection of treatment data in Experiment 2, rats were run on the match to sample task with delays in the Dry Condition to ascertain whether the experimental treatments were associated with permanent functional impairments.

EXPERIMENT 1

Introduction

Experiment 1 was designed to identify the dose of 2% lidocaine infused directly into ventral striatum or into intralaminar nuclei that would disrupt performance in a match to sample task trained without delays in an operant lever box. Previous research has shown that excitotoxic lesions of these sites disrupt performance of this task (Burk and Mair, 1998). We compared the effects of different volumes of the 2% lidocaine solution to the same volumes of saline. Volumes were tested in ascending order from 0.5 to 4.0 µl to identify potentially disruptive effects of the infusions. Infusions were made through cannulas implanted in either the ventral striatum or the intralaminar nucleus of thalamus in rats that had been trained to perform a match to sample task to criterion.

Method

Subjects. Thirty male Long-Evans rats (Charles River, Wilmington, MA) weighing between 350 and 400 grams and aged approximately eight weeks at the beginning of the study were used in Experiment 1. All were singly housed in a facility with a 12:12 light/dark cycle. Rats had *ad libitum* access to standard rat chow and 30 minutes access to water during periods of behavioral training. Water was available for 60 minutes on days when rats were not trained and behavioral training occurred during the light cycle.

All procedures related to the care and handling of the animals was approved by the Animal Care and Use Committee at the University of New Hampshire.

Apparatus. All rats were trained to criterion performance (90%) on a match to sample task in modular operant chambers (model ENV-007; MedAssociates of Georgia, VT) which were placed in sound attenuated boxes. Each of the four operant chambers housed a ventilating fan; a house light (model ENV-215M) centered on the back wall of the chamber near the ceiling, and three retractable levers (model ENV-112AM). One lever was in the center of the back wall; two levers flanked a drinking port in the center of the front wall. Within the drinking port was a water dipper (model ENV-202AM) and a photocell (model ENV-254) that detected responses of a rat to the port. All activity in the operant chambers was controlled and recorded by MedAsociates software interfaced with a 486 or Pentium computer.

General surgical implantation of cannulas. Upon reaching the performance criterion (90%) each rat was pseudorandomly assigned to one of the two treatment groups in a matching procedure. Twelve pairs of animals, matched by presurgical performance made up the ventral striatal lesion group and intralaminar lesion group. Six animals did not reach the presurgical performance criterion and were eliminated from the study. Under aseptic conditions, each experimental rat received a bilateral guide cannula implant (Plastics 1, Roanoke, VA) that targeted either the ventral striatum or the intralaminar nuclei. All rats were anesthetized for surgery with cocktail of ketamine (85 mg/kg) and xylazine (8.5 g/kg) given intramuscularly. Following anesthesia, rats were placed in the stereotaxic apparatus (Kopf Instruments, Tujunga, CA) with the incisor bar placed at 3.3 mm below the interaural line.

Holes 2 mm in diameter were bored on each side of the medial suture of the skull to accommodate the descent of either (1) four guide cannulas or (2) both the Hamilton microsyringe and two guide cannulas. Four holes were drilled in the case of the ventral striatal cannulas; two holes were drilled for the intralaminar nuclei implants. Dental cement (Cranioplaster, Plastics 1, Roanoke, VA) was applied in two layers to secure the cannula once implanted. **Table 1** provides coordinates for both the ventral striatum and the intralaminar treatments. **Figure 1** illustrates sites of typical lesions to these areas. Following implantation, dummy cannulas were placed in the guide cannulas to keep them patent, and the scalp incision was sutured. Each rat recovered in its home cage. Upon waking, (usually the following morning) each rat was injected subcutaneously with 1 mg/1 kg Butorphanol analgesic for alleviation of pain. All animals began behavioral testing between 10 and 14 days following surgery.

Table 1: Surgery Coordinates for Experiments 1 & 2

Treatment Group	AP	ML	DV
Ventral Striatum			
anterior cannula implants	+ 2.2[a]	± 1.9[b]	+ 4.0[c]
posterior cannula implants	+ 0.2[a]	± 1.9[b]	+ 4.5[c]
Intralaminar Thalamus			
anterior NMDA lesion sites	+ 7.2[c]	± 1.4 and ± 0.6[b]	+ 4.0[c]
posterior cannula implants	+ 5.7[c]	± 1.0[b]	+ 4.5[c]

a indicates mm from Bregma; b indicates mm from midline; c indicates mm from interaural line

Ventral striatum implants. Ventral striatum cannula implants (Plastics 1, Roanoke VA) were individually fashioned by filing the platforms of two individual double cannulas and cementing them together to create a four-pronged implant. The two anterior guide cannula shafts were 2 mm away from the posterior guide cannula shafts. In addition,

ventral striatal guide cannulas were cemented together at a slight offset so that the posterior pair of guide cannulas were 0.5 mm lower than the anterior pair of guide cannulas to follow the location of the rat ventral striatum. The two left guide cannulas were 3.8 mm distance from the implants' two right guide cannulas. Twenty-two gauge stainless steel tubing extended 9.5 mm below the plastic platform of the guide cannulas. After the implant was lowered to the appropriate depth, it was secured with dental cement and the scalp was sutured shut around the implant **Figure 2** shows the location of these implants.

It should be noted that the cementing of the two double cannulas together precluded the use of dust caps on the ventral striatum implants. Dummy cannulas were placed (and replaced if later removed by the animal) into the guide cannulas to ensure the shafts remained patent. Due to an error in filling the customized order for these long cannulas, once in place, the dummy cannulas extended 0.5 mm past the end of the guide cannulas for the ventral striatal rats.

Intralaminar nuclei lesion and implants. The second treatment group of rats received both a small excitotoxic lesion to anterior intralaminar nucleus and a two-pronged individual double guide cannula that targeted the posterior intralaminar nucleus of thalamus. After the skull was opened on each side of the midline, a 26-gauge needle mounted on a 10 µl Hamilton syringe was lowered into four targeted anterior intralaminar sites. Delivery of 0.1 µl NMDA (150 mM in buffered saline, 7.4 pH) was controlled by using a Kopf 5000 microinjector, and the syringe remained in place for one minute following the infusion of NMDA to allow diffusion of the excitotoxin. Following delivery of the excitotoxin, the posterior intralaminar implant was lowered into position and

secured with dental cement. A dummy cannula with tips flush with the guide cannula shafts was inserted and covered by a dust cap. The scalp was then closed and the rat returned to its home cage. Butorphanol was administered to alleviate pain upon the full recovery each rat in this study.

Behavioral training. Behavioral training for the current experiment was designed to match that described in Burk and Mair (1998). Prior to surgery, rats were shaped in four successive steps to perform a match to sample task in the operant chamber. In the first step, rats were trained to respond to a lever presented, that, when pressed, would raise the water dipper into the port located in the front of the operant chamber providing 0.1 ml of water as positive reinforcement for rats' responses. When rats' performance was reliable, the second step was trained. This second step required a press on the back lever followed by a press on the available front lever to raise the water dipper for positive reinforcement.

The third step in the series constituted training the rat to press levers with "errorless training." In this step, the back lever was presented at the beginning of a trial. A single press on this lever caused it to retract and the presentation of one of the front levers. A press on the front lever caused its retraction and gave the rat 3 seconds access to the water dipper in the port. The front lever presented on each trial alternated between the left and right positions to prevent side biases from developing. This task was considered "errorless" as only one front lever was presented at a time and a single press on that lever resulted in access to reinforcement.

Upon reaching reliable performance on the third task, the rats were trained on the standard match to sample task that was used for the remainder of Experiment 1. Each

trial in this final presurgical task began with the depression of the back lever. One press caused the back lever to retract and the appearance of one of the front levers randomly selected by the computer on a trial-by-trial basis. Presentation of this front lever constituted the sample stimulus. A single press on this lever caused its retraction and the reappearance of the back lever. A depression of the back lever caused the presentation of both of the front levers, constituting the choice phase of the trial. If the rat pressed the front lever identical to the one that had been presented during the sample phase, the rat received reinforcement and the computer recorded a correct choice. If, however, during the choice phase the rat depressed the front lever that had not been presented during the sample phase of the trial, no reinforcement was given and an error was recorded by the computer. The inter-trial interval for all three experiments was three seconds.

A match to sample session was terminated by the computer if 60 trials had been performed or if 75 minutes had passed. Rats were trained on this match to sample procedure until they reliably performed at least 85% of the session's completed trials correctly. Upon reaching reliable match to sample performance criterion, each rat was assigned to a treatment group using a random matching procedure. Rats were rank-ordered from highest- to lowest- performing animals during presurgical training, and divided between the two treatment groups to ensure balance of high- and medium-performing animals.

Post-surgical training. Animals were allowed to recover from surgery for 10 to 14 days, and then began training in the operant chambers on the same match to sample task trained presurgically. Post-surgical training on the match to sample task continued until animals reached at least 85% percent correct out of sixty possible trials in three

consecutive sessions. Upon reaching this criterion, animals underwent a series of progressive treatments of lidocaine throughout the post-surgical training of the match to sample task in the operant chamber.

Infusion apparatus and procedure. All infusions of sterile physiological saline or sterile 2% lidocaine (from J. A. Webster) (Packard, Cahill and McGaugh, 1994) were delivered via the introduction of internal (delivery) cannulas of a specified length. Internal cannulas projected 0.5 mm beyond the length of the guide cannula for both the ventral striatum and intralaminar groups (Floresco, Seamans, and Phillips, 1996; Poucet, Hermann, and Buhot, 1991; Poucet and Buhot, 1994). Each internal cannula was attached to about eight inches of polyethylene tubing (PE 50, standard wall, Plastics One, Roanoke, VA) connected to a Hamilton glass gas-tight 250 µl syringe (Fisher Scientific; Model 1725) fitted with a luer lock tip. Four Hamilton syringes mounted on a Harvard model syringe pump (Harvard Apparatus Model 927) allowed a rat to receive two simultaneous infusions to its guide cannulas. In the case of the ventral striatum group, infusions were first made to the anterior guide cannulas then to the posterior guide cannulas. Every effort was made to ensure the sterility and continuous flow of fluid in the internal cannulas.

When post-surgical performance criterion was reached, the rats underwent an infusion of either saline or lidocaine or ran without an infusion (Dry Condition). Prior to each infusion, each rat was wrapped snugly in a towel to prevent injury or interruption during delivery. All infusions regardless of final volume are made at a rate of 1 µl / 1 minute 24 seconds. Following the procedure, dummy cannulas were replaced and the rat was placed immediately in the lever box.

Five infusion volumes were used, in ascending order over 41 sessions (30 treatment sessions and 11 Dry Condition sessions). The infusion schedule is listed in **Table 2.** Each dose of saline or lidocaine was given for three consecutive sessions, one session per day. Before each new dose of saline or lidocaine began rats were trained in the task with no infusion, or in the "Dry" condition. In addition, after the third session of 4.0 µl lidocaine, all rats were run for two Dry sessions to test whether experimental treatments were associated with permanent functional impairments.

Table 2: Infusion Schedule for Experiment 1. All volumes are given in µl.

Session	0	1 – 3	4	5 – 7	8	9 - 11	12	13 - 15	16	17 – 19	20
Infusion	Dry	0.5 Saline	Dry	0.5 Lidocaine	Dry	1.0 Saline	Dry	1.0 Lidocaine	Dry	2.0 Saline	Dry

Session	21 – 23	24	25 – 27	28	29 – 31	32	33 - 35	36	37 – 39	40-41
Infusion	2.0 Lidocaine	Dry	3.0 Saline	Dry	3.0 Lidocaine	Dry	4.0 Saline	Dry	4.0 Lidocaine	Dry

Results

Experiment 1 compared the effects of 2% lidocaine and saline delivered at five volumes (0.5 µl, 1.0 µl, 2.0 µl, 3.0 µl and 4.0 µl). For each volume, response accuracy was compared for three sessions in which lidocaine was infused to three sessions in which the same volume of saline was infused. Each level of infusion volume was analyzed by paired t tests with one-tailed probability that compared accuracy following saline infusions to accuracy following lidocaine infusions.

Of the 12 rats that received intralaminar nucleus cannulas, 2 did not recover performance levels to criterion (> 85% correct). Data from the remaining 10 rats in this group were analyzed at each of the five levels of volume. The data shows that performance accuracy did not change substantially with increasing volumes of lidocaine

except at the highest volume infused (4.0 µl) (**Figure 3**). At the highest volume of lidocaine infused, the mean accuracy of intralaminar implanted rats declined from 94% correct to 91% correct. Equal volumes of saline had no effect on accuracy. Mean accuracy during saline treatments in the intralaminar implanted group remained between 96.2 % correct at the 0.5 µl volume and 96.4 % correct at the 4.0 µl volume. Each t test compared the performance following lidocaine infusion to performance following the same volume of saline. Results of these t tests are listed in **Table 3.**

These analyses show that performance for rats with intralaminar nucleus cannulas tends to be worse after lidocaine than performance after the same volume of saline.

Table 3. Results of Experiment 1 (Intralaminar rats) Comparing Performance after 2% Lidocaine Infusion to Performance after Saline Infusion at Progressive Volumes. Entries made in bold denote statistical significance.

TREATMENT	0.5 µl lidocaine / 0.5 µl saline	1.0 µl lidocaine / 1.0 µl saline	2.0µl lidocaine / 2.0 µl saline	3.0 µl lidocaine / 3.0 µl saline	4.0 µl lidocaine / 4.0µl saline
GROUP					
Intralaminar nucleus	t (9) = 1.422	t (9) = 1.983	t (9) = 1.608	t (9) = 2.149	t (9) = 4.086
	p = .0944	**p = .0393**	p = .0711	p = .0836	**p = .004**

Rats ran a total of three sessions at each infusion level followed by a session with no infusion (Dry Condition). The performance of intralaminar nucleus implanted animals during Dry Condition sessions was remarkably stable, ranging from 96.3% to 98.0% **(Figure 4)**. No difference was found between the saline and Dry conditions. This indicates that the volumes of lidocaine and saline infused had no permanent effect on performance accuracy of these animals.

Of the 12 rats that received ventral striatum cannulas, 5 did not recover performance to criteria (> 85%). The 7 rats remaining completed the 3 lowest infusions of

lidocaine and saline at 0.5 µl, 1.0 µl, and 2.0 µl infused volume. However, 2 of the rats in this group became sick before completing the 3.0 µl or 4.0 µl lidocaine and saline treatments. Paired t tests using one-tailed probability values reflect the difference between lidocaine and saline performances of 7 ventral striatal rats at 0.5 µl, 1.0 µl, and 2.0µl , and 5 ventral striatal rats at 3.0 µl and 4.0 µl volumes of lidocaine and saline.

Table 4. Results of Experiment 1 (Ventral Striatum rats) Comparing Performance after 2% Lidocaine Infusion to Performance after Saline Infusion at Progressive Volumes. Entries made in bold denote statistical significance.

TREATMENT	0.5 µl lidocaine / 0.5 µl saline	1.0 µl lidocaine / 1.0 µl saline	2.0µl lidocaine / 2.0 µl saline	3.0 µl lidocaine / 3.0 µl saline	4.0 µl lidocaine / 4.0µl saline
GROUP					
Ventral Striatum	t (6) = -0.801	t (6) = 1.695	t (6) = 3.73	t (4) = 1.471	t (4) = 5.472
	p = .2268	p = .0705	**p = .0048**	p = .1075	**p = .0027**
	n = 7	n = 7	n = 7	n = 5	n = 5

Inspection of the data shows that at all infusion volumes except the lowest (0.5µl), lidocaine infused into the ventral striatum resulted in poorer performance compared to that following saline (**Figure 3**). In addition, ventral striatal implanted rats improved across sessions during Dry Conditions (**Figure 4**).

From these results, it can be said that a disruptive dose of lidocaine is contained in 4.0 µl of 2% commercially prepared lidocaine hydrochloride infused to the intralaminar nucleus and ventral striatum.

EXPERIMENT 2

Introduction

Experiment 2 was designed to address several issues. First, we predicted that 3.0 µl volume of 2% lidocaine infused into the area of the intralaminar thalamic nuclei would disrupt performance when compared to performance preceded by saline infusions of the same size. Although this volume was not associated with a significant impairment in Experiment 1, it is in the range of treatments that were significant.

Second, we investigated the impact of lidocaine inactivation on delayed match to sample performance on a match to sample task at four retention intervals (delays) counterbalanced on a trial-by-trial basis. The retention intervals chosen for this experiment are identical to those used by Burk and Mair in their study of intralaminar lesions (1998). We predicted that the addition of retention intervals to the task would result in an overall decline in performance at longer retention intervals, but that lidocaine inactivation would generate a delay-independent pattern of impairment. This prediction is based on results of several studies that found that intralaminar lesions generate delay-independent deficits in different delayed conditional discrimination tasks. (See Mair, Burk and Porter, 1998).

Third, we investigated effects of lidocaine in intralaminar nucleus on response speed. Excitotoxic lesions to the intralaminar nucleus have been found to increase the

time to respond during individual trials in match to sample tasks trained in the lever box (Burk and Mair, 1998, 1999). We measured response speed as the time elapsed between the press on the back lever that activates the choice phase of the task and the press on one of the front levers that constitutes a choice. Response speed was thus measured from the end of the retention interval until a choice response was executed within each trial. Based on Burk and Mair's work, we hypothesized that lidocaine inactivations of intralaminar nuclei would increase time of responding just as permanent lesions to this area do.

Finally, Experiment 2 addressed the potential impact of treatment order effects present in Experiment 1. Lidocaine and saline conditions were counterbalanced across the eight daily sessions in Experiment 2. Thus, change in performance associated with lidocaine treatment would not be the result of treatment order effects but could stem from other changes. Improvement of performance over sessions may indicate 1) changes in metabolizing lidocaine at the site (Malpeli, 1999), 2) behavioral recovery of function while under the influence of lidocaine, 3) physiological recovery of function at the site of the damage caused by the cannulas (Sandkühler, Maisch, and Zimmerman, 1987), 4) behavioral recovery of function at the site of the damage caused by cannulas, or 5) overtraining (Perez-Ruiz and Prado-Alcala, 1989). Decreases in performance over sessions could be due to 1) carryover effects of lidocaine or 2) a cumulative effect of tissue displacement and damage following multiple or large infusions (Malpeli, 1999).

Only 5 rats with ventral striatal cannulas completed the highest volumes of Experiment 1 due to illness (2 rats), failure to train to post-surgical criterion (5 rats) and of them, only 2 were able to meet performance criteria (85% correct) on a match to sample task trained with delays (3 rats). Thus, there was not a sufficient number of

ventral striatum cannulated rats to provide the statistical power needed for Experiment 2. The remaining ventral striatal rats were trained with procedures like the intralaminar group. However, these results were not analyzed with inferential statistical tests.

Methods

Subjects. Rats used in Experiment 1 served as subjects in Experiment 2 provided there was no evidence of serious permanent damage or illness in individual animals. A total of 10 intralaminar nuclei implanted rats and 2 ventral striatal rats were included in Experiment 2.

Behavioral Apparatus and Training. Immediately after the final two days of the Dry condition in Experiment 1, rats were pretrained for Experiment 2 in the same apparatus used for Experiment 1. Pre-infusion training consisted of running the match to sample task with four delays randomly counterbalanced over trials. These delays were 1.0, 3.0, 8.0 and 13.0 seconds, as in Burk & Mair's (1998) study of permanent intralaminar lesions (1998). Rats in the present experiment were trained until they reached 85% correct out of 60 possible trials for three consecutive sessions with imposed delays.

The computer also recorded the time interval between (1) the first press on the back lever beginning a trial and the response at the sample lever presented; (2) the sample lever press and the second depression of the back lever; and (3) the second back lever press and the response made during the choice phase after the retention interval. This third recorded time constituted a measure of response speed.

Infusion Apparatus & Procedure. Infusions were conducted exactly as in Experiment 1 except that only one volume, 3.0 µl of saline or lidocaine, was infused throughout 8 consecutive sessions. Saline and lidocaine were infused at a rate of 1.0 µl over 1 min and 24 seconds. Commercially prepared 2% lidocaine was used. In Experiment 2, no Dry Condition sessions were trained except during the pretraining sessions ("Preinfusion condition") before infusions began and immediately following the close of all infusion sessions. Following the collection of treatment data in Experiment 2, rats were run on the match to sample task with delays in the Dry Condition to ascertain whether the experimental treatments were associated with permanent functional impairments.

To counterbalance any possible order effects, half of the rats were run in a schedule that was: Saline-Lidocaine-Lidocaine-Saline-Lidocaine-Saline-Saline-Lidocaine, and the other half had this order reversed. The infusion schedules are listed in **Table 5** below.

Table 5: Pattern "A" Order of Infusions for Experiment 2

Session	1	2	3	4	5	6	7	8
Infusion	3.0 Saline	3.0 Lidocaine	3.0 Lidocaine	3.0 Saline	3.0 Lidocaine	3.0 Saline	3.0 Saline	3.0 Lidocaine

Table 5: Pattern "B" Order of Infusions for Experiment 2

Session	1	2	3	4	5	6	7	8
Infusion	3.0 Lidocaine	3.0 Saline	3.0 Saline	3.0 Lidocaine	3.0 Saline	3.0 Lidocaine	3.0 Lidocaine	3.0 Saline

Results

Results of this experiment showed that rats performed equally well in this task whether they were infused with of 3.0 µl of 2% lidocaine or with 3.0 µl saline, $F(1,9) =$

3.069, p = .071. Performance accuracy was virtually identical for the saline condition (82.45 % correct) and the lidocaine condition (81.13% correct) (**Figure 5**). A 2 × 2 ANOVA (Treatment × Retention Interval) showed that, as expected, all rats performed worse at longer retention intervals in Experiment 2, F (3,27) = 27.318, p < .001. Mean percent correct for accuracy were 92% for the 1 second condition, 88% correct for the 3 second condition, 80% correct for the 8 second condition, and 71% correct for the 13 second condition. The interaction effect between Treatment and Retention Interval was not significant, F (6,54) = 0.673, p = 0.589, showing that rats treatment effects did not change over the four retention intervals.

Previous experiments have shown that rats with intralaminar nucleus lesions respond slower in this task. Data across conditions showed that during saline conditions, rats ran each trial in mean time of 2.92 seconds, while under the lidocaine condition, rats ran each trial in 2.70 seconds. A paired t test showed that there was no effect of treatment condition on response speed, **Figure 6,** t (9) = 0.750, p = 0.2356. This effect is somewhat surprising as previous studies report destruction of the intralaminar nucleus causes pronounced and irreversible slowing of responding in rats trained in the same delayed match to sample task in the lever box (Burk and Mair, 1998).

Three potential explanations can account for the lack of treatment effects on accuracy and on response speed in Experiment 2. The lack of a significant treatment effect at 3.0 µl lidocaine in Experiment 2 is consistent with the results of Experiment 1 which showed effects approaching significance at 3.0 µl lidocaine vs. saline (p = .0836 in Experiment 1). First, it is possible that the volume of lidocaine in the intralaminar nucleus

was associated with more consistent impairment. In Experiment 1, for instance, a dose of 4.0 µl of 2% lidocaine was required in intralaminar nucleus or in ventral striatum to disrupt accuracy. Alternatively, lidocaine used in Experiment 2 may have had diminished potency to disrupt neural conduction as the length of the blockade may be shortened after the 52 infusions of Experiments 1 and 2 combined (cf. Malpeli, 1999).

A second potential cause of the lack of treatment effects is that 3.0 µl is sufficient to disrupt performance, but the internal cannulas' projection of 0.5 mm past the tip of the guide cannula reduced the area of the exposed tissue. It is possible that the intralaminar nucleus only partially inactivated because the lidocaine did not diffuse a sufficient distance from the tips of the guide cannulas.

A third potential explanation for the failure to find a significant effect of treatment might be the result of physiological or behavioral recovery of the small excitotoxic lesion located in the anterior thalamus. This is a possibility for the physical damage of the cannula implant as well. Finally, Experiment 2 used a counterbalanced sequence of treatment conditions. It is possible that counterbalancing conditions contributed to the lack of treatment effects by avoiding potential carryover effects.

As predicted, all rats in Experiment 2 were less accurate as retention intervals lengthened between the presentation of the sample and the choice phase of the trial. This corresponds to results of similar experiments using rats with intralaminar lesions. Rats performed poorer on trials with longer delays in a delayed non-match to sample task in an operant chamber where delays were between 1.8 and 8.8 seconds (Young, Stevens, Converse and Mair, 1996); between 3 and 15 seconds (Robinson and Mair, 1992) and

between 3 and 12 seconds (Mair, Robinson, Koger, Fox, and Zhang, 1992); and delayed match to sample in the lever box where delays were identical to those used in Experiment 2 (Mair, Burk and Porter, 1998).

Retention intervals in Experiment 2 showed no greater impact on performance following lidocaine infusions than following saline infusions. This is somewhat surprising given the consistent finding that intralaminar nucleus damage causes delay-independent disruption in conditional discrimination tasks trained with delays (complete lesion, Young, et al, 1996; Burk and Mair, 1998). However, the present findings are in agreement with Mair, et al (1992) who compared effects of retention interval following partial thalamic lesions to effects following complete intralaminar lesions. The present data resemble Mair, et al's (1992) data from the animals with partial anterior or posterior radiofrequency lesions to intralaminar nucleus. These animals declined over retention intervals, but only complete lesions disrupted accuracy of performance in the delayed non-match to sample operant task. This suggests that rats in the present experiment did not experience complete inactivation of the intralaminar nucleus despite a small excitotoxic lesion to anterior intralaminar nuclei and 3.0 μl lidocaine infused to posterior intralaminar nucleus. This might be due to an insufficient volume of lidocaine used, failure of lidocaine to diffuse away from the cannula tips, the recovery of the anterior excitotoxic lesion, or a diminished effect of lidocaine following a large number of infusions.

Chicago Sky Blue and Two Preparations for Histological Examination of the Tissue.

Chicago sky blue infusion. Upon completion of behavioral testing, Chicago sky blue dye was infused into the rats' targeted brain areas in an attempt to determine the

physical spread of lidocaine away from the tips of the internal cannulas. Procedures for infusion of Chicago sky blue were identical to those used to infuse lidocaine during Experiment 1, except that the rats were placed under deep anesthesia with a cocktail of Ketamine and Rompun in preparation for sacrifice. Anesthetized rats received 2.0 µl of a 2.5% Chicago sky blue dye via bilateral infusion cannulas that were identical to the ones used to infuse lidocaine and saline during the Experiment 1(Ragozzino, Wilcox, Raso, Kesner, 1999). All equipment used to infuse lidocaine and saline was used to infuse the Chicago sky blue dye.

Following infusion with Chicago sky blue, each rat was perfused intracardially with 0.9% saline followed by 10% (volume/volume) paraformaldehyde in a buffered solution. Brains then were divided into two groups: those that were frozen extremely quickly and those that underwent standard histological preparation without quick-freezing. Brains frozen quickly were then compared to those that underwent regular histological preparation to determine whether the spread of the Chicago sky blue dye would differ between the techniques. It was proposed that flash freezing the brains would arrest the spread of Chicago sky blue so the coloration would represent a more accurate picture of the physical spread of lidocaine.

Histological preparation one: 'Flash freezing.' After perfusion and rapid removal from the skull, the brain was carefully blocked to include the cannula target site and enough tissue for slicing. The blocked brain was then placed coronal-surface up in a 5 ML microbeaker partially filled with Polyfreeze, a sucrose and water-based cryoprotectant. Then, the microbeaker was partially submerged in slurry of 95% ethanol and ground dry

ice. This caused the Polyfreeze-covered tissue to freeze rapidly, which avoided the formation of ice crystals and was thought to arrest the physical spread of Chicago sky blue. Flash frozen brains were stored immediately in an -80 degree Celsius freezer until slicing. Unfortunately, upon slicing these flash frozen brains, it became apparent that tissue not stored for 1 -3 days in 10% and then 20% paraformaldehyde solution are very fragile and are difficult to section. Alternate sections were stained with cresyl violet.

Histological preparation two: Standard perfusion protocol. Following perfusion and harvest of the brain, the tissue remained in a 10% glycerin/4% paraformaldehyde solution until they sunk. They were then preserved in a 20% glycerin/4% paraformaldehyde solution until sectioned for staining. Cresyl violet was used to stain every other section in the area of the cannulas. Sections were 30 mm thick and cut from the coronal plane of a frozen block (Givens and Olton, 1995).

Results of histological protocol comparison. Two of the ten ventral striatal implanted who died prematurely did not have their brains perfused. Their brains were harvested between five and 120 minutes after death. These brains have been sectioned and will be examined both for the placement of the cannulas and for a possible cause of death.

Although flash freezing is a convenient way to store brains by virtue of its rapid disposal of the tissue, procuring high quality sections proved difficult. The tissue and surrounding Polyfreeze remained different temperatures, complicating sectioning. Additionally, despite rigorous attention to maintaining temperature while sectioning, ice crystals appeared in the tissue. Standard perfusion methods and tissue preparation, on the

other hand, did not appear to cause additional diffusion of Chicago sky blue from the insertion point.

EXPERIMENT 3

Introduction

In Experiment 3, a naïve group of rats was trained to perform the same match to sample task with delays used in Experiment 2. This experiment was designed to explore the effect of a range of concentrations of lidocaine (2%, 4%, and 8% lidocaine) at a volume of 1.0 µl infused into the intralaminar nuclei. The range of concentrations for this study was selected to correspond to the doses of lidocaine infused at volumes in Experiment 1.

A second factor examined in Experiment 3 was the elimination of the excitotoxic lesion to the anterior nucleus present in the first two experiments. Results from Experiment 2 showed that rats performed equally well following lidocaine or saline treatments. It is possible that this lack of treatment effect was due to the physiological or behavioral recovery of the site of the small excitotoxic anterior thalamus lesion given to Experiment 2 rats.

To compensate for the elimination of the small lesion in anterior thalamus, the single bilateral cannulas in Experiment 3 were placed more anterior than those used in Experiment 1 and 2. In addition, internal cannulas used in Experiment 3 projected 2.0 mm beyond the tips of the guide cannulas in an attempt to expose more thalamic tissue to

infused lidocaine. Experiments 1 and 2 used internal cannulas that projected 0.5 mm beyond the guide cannulas.

Experiment 3 also investigated whether performance changed over multiple infusions of lidocaine. This was achieved by training rats for a total of 25 sessions divided into 5 blocks. Each of the five blocks consisted of four sessions of infusion followed by a session without an infusion ("Dry Condition"). For each rat, a block consisted of one session each of Saline, 2% lidocaine, 4% lidocaine and 8% lidocaine. Treatment conditions within the five blocks were scheduled in pseudorandom order so that every rat received each treatment in the first position, second position, third position, and fourth position. As there were five blocks, all rats ran one treatment twice in each position. Five blocks (and therefore five sessions at each treatment level) were considered necessary to test whether treatment effects changed after repeated administration.

Previous studies of lateral internal medullary lamina or intralaminar nuclear damage show that rats' accuracy and response speeds were adversely affected as a result of these lesions (Knoth and Mair, 1991; Burk and Mair, 1998). Based on these prior results, it was predicted that lidocaine blockade in the intralaminar nuclei would disrupt accuracy and increase response speed

Methods

Subjects. Twelve naïve male Long-Evans rats were used for Experiment 3. They were housed and handled exactly as rats were for Experiment 1 and 2.

Behavioral Apparatus and Training. The same match to sample task with four delays (1.0, 3.0, 8.0, and 13.0 seconds) was trained both presurgically and postsurgically

in Experiment 3. Upon reaching presurgical performance criteria (85%), all rats underwent implantation of bilateral thalamic cannulas. Following ten to fourteen days of recovery, rats began postsugical training in the operant lever boxes. Infusions began when rats reached 85% percent correct consistently.

Intralaminar nuclei implants. Bilateral cannulations of all rats in Experiment 3 targeted the middle intralaminar nucleus of thalamus (with respect to anterior-posterior directionals). The two-pronged double guide cannula implanted in each rat was shorter than that implanted in Experiment 1. The twenty-two gauge stainless steel tubing shafts were 5.5 mm from the platform, although they were still 2 mm distance apart as before (1 mm off midline bilaterally). Surgical coordinates for Experiment 3 are listed in **Table 6.** After the implant was lowered to the appropriate depth through two burr holes, it was secured with dental cement (Plastics 1, Roanoke, VA) and the scalp was sutured shut around the implant. A dummy cannula was inserted into the guide cannula and protected with a dust cap. As before, rats received 1 mg/kg Butorphanol as an analgesic upon awakening.

Table 6: Surgery Coordinates for Experiments 3

Treatment Group	AP	ML	DV
Intralaminar nucleus cannula implants	+ 6.2[a]	± 1.0[b]	+ 6.0[a]

[a] = coordinates with respect to the interaural line. [b] = coordinates with respect to the midline.

Infusion Apparatus and Procedure. A new Harvard Apparatus dual microsyringe pump (Model '11') was used for Experiment 3. Rats were infused once per day just prior to their session in the match to sample task with delays. Experiment 3 consisted of a total of 25 daily sessions divided into five Blocks. Each block comprised four pseudorandomly

balanced treatment conditions (one per session) followed by one Dry Condition session for which there was no infusion. The four treatment conditions were Saline, 2% lidocaine, 4% lidocaine, and 8% lidocaine. All lidocaine hydrochloride solutions were prepared in our lab with sterile saline as opposed to buying commercial premixed lidocaine hydrochloride. For every rat, each lidocaine treatment was given in each ordinal position of the blocks at least once and one treatment was given in one of the ordinal positions twice.

Due to programming constraints of the new microsyringe pump, infusions were made at a slightly higher rate than previously. However, this rate, 1 microliter/ 1 minute, is well documented in the literature (Fenton and Bures, 1994; Welsh and Harvey, 1991). In addition, the stainless steel tubing of internal cannulas in this study extended 2.0 mm (Chapman, Steinmetz, Sears, and Thompson, 1990) beyond the tips of the guide cannulas, rather than merely 0.5 mm beyond as they had in Experiments 1 and 2. This was done in order to expose a larger area of neural tissue to the volume of lidocaine infused.

Histological preparation of the tissue. One microliter of Chicago sky blue dye was infused through the internal cannulas at a rate of one microliter per minute to the intralaminar nucleus implants following deep anesthesia in the rats. All brains were perfused and sectioned as described above under the standard histological protocol with alternate sets of sections stained with cresyl violet. No brains were flash frozen in Experiment 3.

Results

As expected, accuracy in the delayed match to sample task declined with the increase in dose of lidocaine from 1 μl of 2% to 1 μl of 8% lidocaine infused into the

intralaminar nucleus Also as predicted, rats' performances declined with longer retention intervals (**Figure 7**). In addition, accuracy of rats in Experiment 3 improved across Blocks for saline and for all lidocaine treatments (**Figure 8**).

A repeated measures analysis of variance (ANOVA) for within subjects compared the effect of Block, Treatment, and Retention Interval. Descriptive statistics revealed that mean accuracy increased over blocks of sessions from 78% at Block 1 to 87% at Block 5. This constituted a significant main effect for Block as shown in the overall repeated measures ANOVA, $F(4,36) = 6.421$, $p = 0.008$. Simple contrasts for Block revealed that the means of Blocks 4 and 5 were significantly different from Block 1. This may reflect either a physiological or behavioral recovery of function following cannula implantation surgery or the effects of overtraining. While one of the purposes of conducting research with lidocaine is to minimize physiological or behavioral recovery of function, the damage generated by cannula implantation may by considered subject to the same recovery of function issues important in permanent lesion studies.

In addition to the main effect of Block, a significant main effect was found for Treatment condition. As the concentration of lidocaine increased from 0% lidocaine (saline) to 8% lidocaine, rats performed worse in the delayed match to sample task, $F(3,27) = 7.352$, $p = 0.008$). Simple contrasts revealed that all levels of lidocaine condition, 2%, 4% and 8% lidocaine, differed significantly from the rats' performance when they were infused with saline. The Dry Condition was purposely not included in the overall statistical analysis because it occurred at fixed intervals within the experiment, at the end of each block. Its inclusion was to determine the reversibility of lidocaine treatment (or possible carryover effects) following each level of lidocaine infusion. The purpose of the

study was to investigate the effect of concentration on deactivation of tissue. Therefore, the Dry Condition was treated separately from the other treatment conditions. A repeated measures ANOVA comparing the Dry Condition at each retention interval to the Saline at each retention interval was conducted to determine whether there was a significant difference in accuracy or in response speed in the Dry Condition compared to the Saline Condition. No significant difference was found either for percent correct for these two conditions.

A main effect was also found for Retention Interval, $F(3, 27) = 24.29$, $p < .001$, such that all rats performed worse at longer retention intervals. Simple contrasts showed that performance at delays of 3.0 , 8.0 , and 13.0 seconds were significantly different from performance at the 1.0 second, the shortest delay. Rats' accuracy declined from a mean of 87% following Saline infusions to a mean of 81% following the 8% Lidocaine infusions.

A significant interaction between Block and Treatment was found, $F(12,108) = 3.12$, $p = .022$. This suggests that rats were less impaired by the higher concentrations of lidocaine in some of the blocks. Inspection of the data shows that at the at 4% lidocaine conditions, performance did not follow any anticipated pattern, declining after the first block and rising after the second. Malpeli (1999), who administered several infusions of 2% lidocaine into lateral geniculate nucleus of thalamus in his experiments, offered one explanation for this result. Although he reports complete neural blockade at very small doses (100 nl of 2% lidocaine) in the thalamus, he notes that repeated infusions can shorten the duration of the blockade. He suggests that this is due to rapid dissipation of the drug, either through increased blood flow around the lateral geniculate or through extracelluar "channels" that are "open" following repeated infusions (p. 120). If this is the

case in the present study then repeated exposure to higher doses of lidocaine might cause the anesthetic blockade to last a shorter amount of time. This could account for the improvement of performance over session, and the lack of effect in Experiment 2 following several infusions of 3.0 µl of 2% lidocaine.

Pérez-Ruiz and Prado-Alcalá (1989) offer another explanation for the improved performance over blocks at the higher doses of lidocaine. These researchers note that lidocaine infused into striatum interferes with retention and the "maintenance" of operant behaviors (p. 599.) These authors argue that in tasks that require few trials for learning, such as operant tasks using negative reinforcement, rats are protected against the amnestic effect of lidocaine by virtue of the parameters of the task. In a study of atropine infusions to striatum, rats were amnestic for low but not for high intensity footshock (Prado-Alcalá and Cobos-Zapiaín, 1977, in Pérez-Ruiz and Prado-Alcalá, 1989). Similarly, infusions of lidocaine to anterior dorsal striatum caused amnesia for low but not for high intensity footshock. According to these authors, overtraining with positive reinforcement in spatial tasks and in continuous reinforcement tasks is analogous to the highly salient event of footshocks. With overtraining, these procedures are unimpaired in animals that receive local anesthetics or cholinergic agents (Prado-Alcalá and Cobos-Zapiaín, 1977, in Pérez-Ruiz and Prado-Alcalá, 1989). If this is the case, then it is possible that rats in Experiment 3 were overtrained, especially during later sessions, and were less susceptible to the effects of lidocaine.

It is possible that rather than the characteristics of a task or overtraining, rats adapt to the behavioral effects of lidocaine and are able to compensate in their performance. This may be considered an example of behavioral recovery of function. This is similar to

findings of Young, Stevens, Converse and Mair (1996) and Knoth and Mair (1991). These researchers, using rats with lesions to the lateral internal medullary lamina (which surrounds the intralaminar nuclei) found some improvements over sessions. Burk and Mair (1998) compared rats with the lateral internal medullary lamina lesions to those with intralaminar lesions in performance over sessions. They found a slight improvement of performance over sessions, but the difference was not large enough to be considered significant.

No other two-way interaction was significant, nor was the three-way interaction significant. All rats, regardless of the concentration of lidocaine infused, performed worst at the longest retention interval (13 seconds). This finding for a delay-independent impairment is in accordance with previous research regarding permanent lesions to intralaminar nuclei. In studies of permanent lesions to intralaminar nuclei, researchers showed that rats with pyrithiamine-induced thiamin deficiency (which causes thalamic lesions including to the intralaminar nucleus), (Knoth and Mair, 1992) or permanent lesions to bilateral intralaminar nuclei (Burk and Mair, 1998) are impaired in delayed match- and non-match to sample tasks in a delay-independent fashion. Thus, delay-independent deficits in the intralaminar nuclei implanted rats supports the hypothesis that lidocaine can be used as an alternative lesioning technique to generate deficits in the delayed match to sample.

Response Speed was measured as the latency between a press on the back lever at the end of the delay and a press on one of the two levers presented in the choice phase of a trial, after the retention interval. Median response speed was analyzed as a truer measure of the variable than the arithmetic average. The "Dry Condition" was again

compared to the Saline condition; no significant difference was determined, and Dry Condition was left out of further analyses.

A significant main effect of Block was determined, signifying the rats became faster with experience during training in post-surgical sessions, **Figure 9,** $F(1,4) = 9.625$, $p = .004$. Simple contrasts revealed that the average median response speed in Blocks 2, 3, 4, and 5 were significantly different from the average speed of the first Block. Overall, rats' mean speed during the first Block was 3.45 seconds, but improved at the second Block and remained stable through the end of the fifth Block. Examination of the relevant graphs of the data show that two animals performed much slower following the 8% lidocaine infusions during the first Block than at any other time in the experiment. The significant main effect of Block and response speed may be due to anomalous data in Block 1 rather than a real effect. Likewise, there was a significant main effect for Treatment with rats running slower after 8% lidocaine infusions (mean speed = 3.27 seconds), but running the same speed following Saline (mean = 2.62 seconds), 2% lidocaine (2.59 seconds) or 4% lidocaine infusions (2.64 seconds), , $F(1,3) = 4.893$, $p = 0.03$.

Histological analyses

All brains except one examined from Experiments 1, 2, and 3 showed that the cannulas hit their intended targets. The one rat whose intralaminar cannula was not correctly placed was a subject from Experiment 1 and 2. This cannula had been placed too posterior to Bregma to be included in the study. For all other brains, the squared-off shape of the damage caused by guide cannula tips was easily distinguishable from that of the narrower internal cannulas. The perimeter of all cannula damage was characterized by

glial scarring. In Experiments 1 and 2, damage from the tip of the internal cannula extended approximately 0.5 mm from the guide cannula reaching approximately the level of ventral mediodorsal nucleus of the thalamus. In Experiment 3, guide cannula damage stopped just dorsal to the thalamus in hippocampus. The projection of the internal cannula extended approximately 2.0 mm from the tip of the guide cannula as intended. This placement was also at the level of ventral mediodorsal nucleus of the thalamus. Typical cannula damage in the intralaminar nucleus for Experiments 1, 2 and 3 included injury to the centrolateral nucleus, central and lateral portions of mediodorsal nucleus, and lateral portions of centromedial nucleus.

All ventral striatal cannulas also hit their intended targets, with anterior and posterior placement approximately 2.0 mm apart. The 0.5 mm internal guide cannula extensions past the guide cannulas hit areas of striatum approximately 0.5 mm from the ventral surface of the brain.

Experiment 1 and 2 ended with 2.0 µl of 2.5% Chicago Blue infused into the internal cannulas with 0.5 mm projections in both ventral striatum and intralaminar nucleus implanted rats. Chicago Blue infusions here showed pale staining to neural tissue that extended approximately 0.5 mm radius from the tips of the cannulas. Experiment 3 ended with 1.0 µl of Chicago Blue infused into the internal cannulas with 2.0 mm projections into the intralaminar nucleus. Chicago Blue stained neural cells approximately 1.0 mm (radius) from the internal cannula tips.

GENERAL DISCUSSION

Three experiments systematically examined the effects of reversibly inactivating intralaminar nuclei and ventral striatum with infusions of lidocaine. The ventral striatum and the intralaminar nucleus were targeted based on deficits in accuracy and speed of responding in delayed conditional discrimination tasks associated with permanent lesions of these structures (Mair, Burk and Porter, 1998). The results of our experiments weigh upon two main issues. The first issue is the impact of lidocaine delivery on performance in the match to sample task. The second issue is the role of intralaminar nucleus and ventral striatum in remembering.

Reversible Inactivation by Lidocaine

Lidocaine is a powerful tool for investigating both the role of discrete brain areas and systems of memory. However, lidocaine's efficacy as an inactivating agent may vary based on experimental procedures followed. The three studies described here varied procedures systematically to investigate factors thought to influence extent of the inactivation produced by lidocaine treatment.

Experiment 1 varied the volumes infused of commercially prepared 2% lidocaine. We found that the extent of impairment increased as volumes increased from 0.5 µl to 4.0 µl. Experiment 3 compared the effects of different concentrations of lidocaine infused at a constant volume. These treatments were administered in a series of counterbalanced

blocks to test whether the effects of lidocaine change with repeated administration. Experiment 3 also changed the distance the internal cannula protruded from the guide cannula. We found that the extent of the impairment increased as the concentration increased from 2% to 8% infused lidocaine.

The lidocaine doses infused in Experiments 1 and 3 can be compared by multiplying the volume by the concentration of the lidocaine bolus infused. Experiment 1 manipulated volumes of a single concentration of lidocaine (2%) while Experiment 3 manipulated concentrations of a single volume of lidocaine (1 µl). **Table 7** shows that these manipulations generated a similar range of infused doses of lidocaine.

Table 7. Corresponding doses of lidocaine used in Experiment 1 and Experiment 3

EXPERIMENT 1		**EXPERIMENT 3**	
Volume	Concentration	Volume	Concentration
0.5 µl	2%	No corresponding dose	
1.0 µl	2%	1.0 µl	2%
2.0 µl	2%	1.0 µl	4%
3.0 µl	2%	No corresponding dose	
4.0 µl	2%	1.0 µl	8%

The amount of lidocaine delivered to the targeted area (dose) is dependent on several things: the tendency of lidocaine to diffuse from the cannula tip, the volume infused and concentration infused. The tendency of lidocaine to diffuse from the cannula tips can be measured as either physiological or functional inactivation of tissue. Physiological spread of lidocaine is most often determined with autoradiographic (tritiated lidocaine or 2-deoxyglucose absorption) (Martin, 1991; Martin and Ghez, 1999) or electrophysiological recording measures (Albert, and Madryga, 1980; Malpeli, 1999;

Sandkühler and Gebhart, 1991; Sandkühler, Maisch, and Zimmerman, 1987; Tehovnik and Sommer, 1997).

Recording from monkeys' dorsolateral cortex, Tehovnic and Sommer (1996) determined that physiological spread of lidocaine is described by the equation for the cube root law: **4/3 π (radius)^3**. Malpeli (1999), recording from monkey lateral geniculate nucleus, also reported that radius of lidocaine-affected tissue is a function of the volume's cube root. Both researchers have investigated a large range of infusion volumes (0.10 µl – 30 µl) in the monkey. Clearly, the comparable range of viable infusion volumes for rats must be considerably smaller. Typically, researchers find desired effects in rats using either 0.5 or 1.0 µl of their chosen concentration of lidocaine as their infusion volume (Floresco, Seamans, and Phillips, 1997; Poucet and Buhot, 1994; Pérez-Ruiz and Prado-Alcalá, 1988), although volumes of 3.0 µl and 4.0µl have been used in rat gray matter (Albert and Wong, 1978; Chapman, Steinmetz and Sears, 1990).

The physiological spread of 0.5 µl of 4% lidocaine in the rat gray matter has been given as 0.80 to 1.0 mm (radius) from the cannula tip in a teardrop shape (Sandkühler and Gebhart, 1991). Martin (1991) reported that 1.0 µl of 4% lidocaine spreads in an area with a radius of 1.4 mm from cannulas' tips in gray matter if measured by with electrophysiological apparatus. This radius of inactivation shrinks about 20 minutes after infusion. Martin also reports that the use of glucose absorption autoradiography leads to a smaller estimation of the affected area (1.0 mm radius). Finally, Martin claims that a region of hypometabolism for glucose surrounds the area of inactivated tissue with a radius of 3.0 mm, proposing that functional inactivation by lidocaine may be underestimated by the physiological spread of the agent.

It is possible that Martin's claims (1991) that 1 µl of 4% lidocaine inactivating a subcortical area with a radius of 1.4 mm is too high an estimate. Evidence for this is found in that Martin cites similar findings as those of Sandkühler and Gebhart (1987) who offer a much smaller radius of inactivation with the same lidocaine dose. Moreover, a detailed discussion by Tehovnic and Sommer (1996) applies the cube root law to several researchers' data and findings. Tehovnic and Sommer find that the cube root law estimates claims of lidocaine's physiological spread consistently for several researchers, but not for Martin (1991). Applying the cube root law to Martin's methods would result in an inactivation radius of 0.62 mm, not 1.4 mm (Tehovnik and Sommer, 1996).

According to Tehovnik and Sommer, both large and small infusions of 2% lidocaine follow the cube root law, which is the mathematical equation for the volume of a sphere. It may seem simplistic to claim that 2% lidocaine infused into areas of the brain, which are more or less densely populated with neurons, more or less vascularized, and more or less influenced by the concentration of lidocaine infused, will inactivate an area described by the volume of a sphere. However, their investigation demonstrates that the relationship of the spread of 2% lidocaine is described linearly by the volume of a sphere (the cube root law).

Other researchers have investigated functional inactivation by lidocaine empirically in gray matter. Welsh and Harvey (1991) found the functional spread of 1.0 µl of 2% lidocaine to be 1.4 mm in diameter, while Albert and Madryga (1980) found a functional inactivation of 4.0 µl 2% lidocaine infused slowly to be 0.5 mm radius. Albert and Madryga also found a region of hypometabolism with a radius of 0.9 mm. They take this as evidence that a large volume of lidocaine (4 µl) functionally inactivates a smaller region

than would be expected by the size of the volume, although it is not clear why this should be the case.

Despite differing estimates regarding the spread of lidocaine inactivation, a brief review by Floresco, Seamans, and Phillips (1997) concludes that 1 µl of 2% lidocaine infused into ventral striatum would affect a maximum area 1.5 mm in diameter. This is an increase of 20% larger than the figures provided by Tehovnic and Sommer's (1996) investigation of the cube root law. Our present research shows that the highest doses of lidocaine infused into intralaminar nuclei produced the worst performances. These findings are entirely consistent with predictions given by the cube root law.

The cube root law, $\mathbf{r = [(3 \times V) / 4 \times \pi)]^{1/3}}$ where r stands for radius and V stands for volume, generates consistent and electrophysiologically verified estimates of physiological spread for both large and small volumes of lidocaine (Tehovnic and Sommer, 1996). We have applied the cube root law to the volumes and concentrations of lidocaine used in the present experiments to estimate the area of the tissue inactivated by each dose of lidocaine.

If we accept Floresco's suggestion that 1.5 mm is the maximum diameter of the area inactivated by 1 µl of 2% lidocaine, based on several parameters of the infusion (volume, concentration, flow rate, target), then it can be assumed that lidocaine's distribution by diffusion is relative to the number of molecules of agent infused. Thus, the range of concentrations infused in Experiment 3 would have roughly the same area of inactivation as the corresponding volume of 2% lidocaine infused in Experiment 1.

Applications of the cube root law (**Table 8**) and Floresco's estimates (**Table 9**) to the volumes and concentrations used in our experiment are listed in below.

Table 8. Estimations of diameter of area inactivated by lidocaine infusions using the cube root law provided by Tehovnic and Sommer (1996).

Volume of 2% Lidocaine (Experiment 1)	Concentration of Lidocaine (Experiment 3)	Estimated Diameter of Inactivated Area (Experiments 1 & 3)
0.5 µl	No corresponding dose	0.9847 mm
1.0 µl	1 µl of 2% lidocaine	1.2407 mm
2.0 µl	1 µl of 4% lidocaine	1.5631 mm
3.0 µl	No corresponding dose	1.7894 mm
4.0 µl	1 µl of 8 % lidocaine	1.9694 mm

Table 9. Estimations of diameter of area inactivated by lidocaine infusions using the cube root law given above and Floresco's 1997 maximum estimate for 1.0 µl 2% lidocaine.

Volume of 2% Lidocaine (Experiment 1)	Concentration of Lidocaine (Experiment 3)	Estimated Diameter of Inactivated Area (Experiments 1 & 3)
0.5 µl	No corresponding dose	0.7937 x 1.5 = 1.191mm
1.0 µl	1 µl of 2% lidocaine	1.000 x 1.5 = 1.500 mm
2.0 µl	1 µl of 4% lidocaine	1.2599 x 1.5 = 1.88 mm
3.0 µl	No corresponding dose	1.4422 x 1.5 = 2.16 mm
4.0 µl	1 µl of 8 % lidocaine	1.5874 x 1.5 = 2.37mm

Estimations of the size of areas affected by lidocaine can be compared to the level of impairments seen in Experiments 1 and 3 to predict the extent of inactivations required to disrupt match to sample performance. According to the cube root law, the largest doses of lidocaine (4.0 µl of 2% and 1 µl of 8%) are estimated to inactivate an area roughly 2.0 mm in diameter. Maximum estimates given by Floresco, Seamans, and Phillips (1997) for infusions into ventral striatum predict our largest doses of lidocaine would spread in an area with a diameter of 2.37 mm around guide cannula tips. This is significant in that previous studies have found that the size of the area damaged by intralaminar lesions predicts the level of impairments. Mair, et al (1992) found that small

anterior or posterior intralaminar lesions, approximately 1.3 mm in diameter (Mair and Lacourse, 1992), had no effect on accuracy for responding in a delayed non-match to sample task trained in an enclosed operant chamber (Mair and Lacourse, 1992). The same study showed that larger lesions, which extended the length of the intralaminar nucleus (3.0 mm), produced significant deficits in performance. According to these results, large deficits in performance should follow the inactivation of an area that approaches the size of the entire anterior-posterior extent of the intralaminar nulcei. This is also consistent with the extent of damage caused by pyrithiamine-induced thiamin deficiency or excitotoxic lesions to intralaminar area that are associated with delayed match- or non-match to sample impairments (Burk and Mair, 1998; Knoth and Mair, 1992; Robinson and Mair, 1992).

Other researchers have demonstrated that the effects of lidocaine depend on diverse factors. Malpeli (1999) proposed that flow rate and the characteristics of the brain area targeted also influence the radius of inactivation. For instance, this researcher claims that a higher flow rate increases the spread of inactivation. Other researchers (Albert and Madryga, 1980) infused a large amount (4.0 μl) over a long time (15 minutes), presumably to increase the spread of lidocaine. In their discussion of the effects of lidocaine on nucleus accumbens in the rat, Seamans and Phillips (1994) justify their infusion of 1.0 μl of 2% lidocaine over 2 minutes, calling the flow rate optimal. The brain area targeted also influences the extent of lidocaine's effects. Malpeli (1999) states that higher amounts of myelination in the area will prolong the effects of lidocaine, and also increase the area of spread (See also Sandkühler and Gebhart, 1991). In addition, lidocaine weakly binds sodium channels, and is therefore cleared quickly from neural tissue in highly vascularized

areas or areas of high lymphatic activity (Martin, 1991; Sandkühler, Maisch, and Zimmerman, 1987). Finally, it has been claimed that the hypometabolic periphery surrounding inactivated tissue in cortex is due to diminished activity of cortico-cortical interneurons, not direct contact with lidocaine, according to Martin (1991). This idea has implications for the radius of functional inactivation in intralaminar nucleus and ventral striatum, brain areas that contain many fibers of passage rather than interneurons.

Despite some debate regarding the spread of the inactivation by lidocaine, most researchers would agree than 1.0 µl of 2% lidocaine spreads no more than 1.5 mm from the guide cannula tip (Seamans and Phillips; 1994). This may account for the consistent choice among researchers to infuse 0.5 - 1.0 µl of 2% lidocaine to study rats' behavior. We have found that infusions of 4 µl of 2% lidocaine or 1µl of 8% are needed to affect performance. This must be taken as evidence that relatively large areas of thalamus or ventral striatum must be inactivated to disrupt delayed match to sample tasks.

Rats in Experiment 1 tolerated 30 infusions, 3 at each of 5 saline volumes and 3 at each of the matching lidocaine volumes. Rats in Experiments 3 tolerated 20 infusions of one volume of varied lidocaine concentrations and saline. The consistent or improving performances on Dry Condition days interspersed throughout Experiment 1 and Experiment 3 show that it is unlikely that the large number of infusions or the ascending volume size in Experiment 1 caused any serious detriment to rats' performance. **Figure 10** shows that during Dry Conditions in both experiments, rats achieved a high percentage of correct trials, relative to their preinfusion performance, at all points throughout the experiments. The one exception to this is the Dry Condition run directly after Block 1 in Experiment 3. Data from this session are slightly lower than that of any of the subsequent

dry sessions. This may be the result of training with delays in the match to sample task. Alternatively, the first Block of varied concentrations of lidocaine may have had a cumulative effect on the performance of the Block 1's Dry Condition. If this is the case, it is important to note that rats recovered quickly, and improved over subsequent Blocks during Dry Conditions. Another explanation is related to damage produced by the 2.0 mm extentsion of the internal cannula beyond the guide cannula into the area of the intralaminar nuclei.

Figure 8 shows rats' performance in Experiment 3 improved over the study's 5 blocks of sessions. This gradual improvement of performance was seen following infusions of 2% and 8%, but not 4% lidocaine. The most improvement seen in Experiment 3 is consistent with Malpeli's (1999) idea that rats "tolerate" lidocaine over multiple sessions in that the duration of inactivation becomes shorter with successive infusions. However, parallel changes were also seen following infusions of saline. This must be taken as evidence that rats improved across Blocks.

Rats trained in Experiment 1 received 30 separate infusions, 6 at each of 5 different volumes. Rats continuing training in Experiment 2 received an additional 8 infusions. The administration of several infusions, some of them quite large compared to other studies, had the possibility of damaging tissue (Martin and Ghez ,1999; Sandkühler and Gebhart, 1991). The performance of rats across many sessions in Experiments 1 and 2, even following several large infused volumes, indicates that any injury experienced due to the infusion process, had little if any effect on performance. Rats in Experiment 3, also received numerous infusions without any sign of diminished delayed match to sample performance.

In addition to addressing issues of volume, concentration, and recovery of function, the delivery system for lidocaine was modified in Experiment 3. This was done to judge whether a longer projection of the internal cannula past the tip of the guide cannula would increase the effect of infused lidocaine. Rats implanted with intralaminar nucleus guide cannulas in Experiment 1 and 2 were infused using internal (delivery) cannulas that projected 0.5 mm beyond the tip of the guide cannulas. This is in accordance with similar experiments using lidocaine to inactivate rat brain areas. (Martin, 1991). In Experiment 3, however, the internal cannulas were lowered to the same depth as in the previous two experiments, but the internal cannulas projected 2.0 mm beyond the tips of the guide cannulas (**Figure** 7). It is possible that the longer projection of the internal cannulas would expose more tissue to lidocaine, as less lidocaine might diffuse into the guide cannula.

While this is a possibility, it is difficult to determine if this was the case. Current literature describes a range of projection lengths for lidocaine infusion. These lengths are typically from 0.5 mm to 1.5 mm. As infusions to different areas of the brain may have various effects, however, it is difficult to know which projection length is particularly useful for infusing the intralaminar nucleus or ventral striatum. For that reason, we used two lengths, 0.5 and 2.0 mm beyond the guide cannulas. In order to assess whether lidocaine might diffuse farther from the guide cannula tips with the longer projections, each rat was infused with 2.5% Chicago sky blue, a dye to mark the cannulas' placement, just prior to sacrifice (Givens and Olton, 1995). Rats in Experiments 1 and 2 were infused with an intermediate volume of Chicago sky blue (2.0 μl) and rats used in Experiment 3 were infused with the volume used for saline and lidocaine treatments (1.0 μl) of the dye.

This procedure was undertaken as a crude measure of the physiological spread of lidocaine. Despite efforts to qualify the diffusion of lidocaine from the two internal cannula projection lengths, it is difficult to state with precision whether longer projections of the internal cannulas contribute to an increased area of lidocaine exposure.

The Intralaminar Nucleus in Match to Sample Tasks Trained With and Without Delays

The current experiments were designed to use lidocaine to inactivate the intralaminar nucleus of the thalamus. Electrolytic and excitotoxic lesions to midline areas that include the intralaminar nuclei have been found to produce a consistent deficit in accuracy and response speed for a variety of tasks (Burk and Mair, 1998; Mair, Burk and Porter, 1998). Accuracy for trial-specific information in delayed conditional discrimination tasks requires that a stimulus be recalled after a certain amount of time has passed and in the absence of that trial stimuli. Researchers have shown that intralaminar-related deficits in conditional discrimination tasks are delay-independent; electrolytic and excitotoxic lesions to this area produce comparable impairments for accuracy at both short (1 second) and long (13 seconds) delays (Burk and Mair, 1998).

The results of Experiments 1 and 3 show that accuracy in a match to sample task is impaired in a dose-dependent fashion by lidocaine delivered to the intralaminar nucleus. As discussed in the previous section, these results are consistent with previous findings that thalamic lesions must involve substantial portions of the intralaminar nuclei to disrupt delayed match- or non-match to sample performance (Mair, Robinson, Koger, Fox, and Zhang; 1992)

Behavioral impairments associated with permanent damage to the intralaminar nuclei are confounded by changes in response speed. Burk and Mair (1998) found that excitotoxic intralaminar lesions caused increased latency to respond during the choice phase of the delayed match to sample task. Mair, et al (1992) and Mair and Lacourse (1992) found that animals with complete radiofrequency lateral internal medullary lamina lesions made fewer responses under 3 seconds from the end of the retention interval in spatial delayed non-match to sample tasks (although this finding was not a significant difference in Mair et al's (1992) study). Knoth and Mair (1991) and Robinson and Mair (1992) found similar results following pyrithiamine-induced thiamin deficiency.

In the present experiment, the median speed was analyzed on a trial-by-trial basis for Experiments 2 and 3. Lidocaine infusions in Experiment 2 had virtually no effect, and in Experiment 3 had minimal effect, on the latency to respond during the choice phase of the task (**Figure 9**). In Experiment 3, infusions of 8% lidocaine caused slower responses only in the first Block of sessions. By the second Block of sessions, all rats were responding at the same speed regardless of the lidocaine concentration infused. This finding is important as it confirms less direct evidence that accuracy in the delayed match to sample task is not due to the tendency of intralaminar rats to respond slowly at this task. The deficit may instead be due to a deficit in remembering information over time, remembering stimuli that are no longer present, or difficulty in performing conditional discrimination.

This implication is congruent with Burk and Mair's (1998) findings, and with research on the lateral internal medullary lamina. However, the current research may

potentially dissociate deficits of accuracy from speed of response. It is possible that lidocaine has allowed us to titrate the impairment to a point where only accuracy deficits are apparent. Previous studies' results have shown that intralaminar lesions cause profound deficits at even the shortest delays. However, these lesions do not disrupt a serial reversal learning task. In addition, Burk and Mair (1998) have previously shown that intralaminar lesions impair response speed on a serial reaction time task that does not require working memory. Burk and Mair's findings, together with the current results, indicate the effects of intralaminar inactivation on delayed match to sample task accuracy and speed are separable.

The Ventral Striatum in Match to Sample Tasks Trained Without Delays

The tips of the guide cannulas for the ventral striatum group in Experiment 1 were aimed at the site of the olfactory tubercle lesions used by Burk and Mair (1999). These researchers found that lesions to this site disrupted accuracy to near chance levels even at the shortest delays of a delayed match to sample task. Olfactory tubercle lesioned rats were significantly worse than control rats or rats in one of the three other experimental groups that received lateral or medial striatal lesions or nucleus accumbens lesions.

Burk and Mair (1999) also tested the striatal groups in a serial reversal learning task. All groups, including the olfactory tubercle group, were able to perform this task. This shows that excitotoxic lesions to the olfactory tubercle did not generate global memory deficits. The spared ability for serial reversal learning in ventral striatal rats is similar to the pattern of sparing seen in intralaminar nucleus lesioned rats (Burk and Mair,

1998). Arguments analogous with those used above may be used to maintain that the size of the inactivated area is critical to the level of impairments seen following inactivation by lidocaine. In order to inactivate an area of ventral striatum large enough to mimic the severe deficits seen by Burk and Mair (1999), we implanted two cannulas to each hemisphere of the brain. However, because the worst performance followed the highest volume of lidocaine infused, it is possible that the lower volumes did not inactivate large enough areas of ventral striatum.

Burk and Mair (1999) consider the possibility that deficits caused by olfactory tubercle lesions was partly the result of concomitant damage to the ventral pallidum. This is significant in light of potential pallidal involvement in basal forebrain disorders of memory (Burk and Mair, 1999). Specifically, the researchers outline the relationship between ventral striatal damage (including nucleus accumbens disruption) and parallel symptoms in midline thalamic and basal forebrain amnesia.

Another study examined the effects of excitotoxic nucleus accumbens lesions on a similar match to sample task using delays of 0 – 24 seconds (Reading and Dunnett, 1991). Results of that study showed bilateral nucleus accumbens lesions impaired accuracy in a delay-dependent manner. Lesioned rats performed comparably to controls on trials with no delay. Reading and Dunnett proposed that these effects are the result of side biases. The effects of bilateral nucleus accumbens lesions were also tested with a serial reversal learning task and in an extinction paradigm. Results showed that unlike Burk and Mair (1999), nucleus accumbens lesions disrupted serial reversal learning. The lesions also increased the number of trials to extinction. Reading and Dunnett (1991) interpret the results as an inability to switch testing paradigms caused by nucleus accumbens lesions.

Experiment 1 of the present study reflects the findings of Burk and Mair (1999) in that inactivation of ventral striatum caused poorer performance at all volumes of lidocaine compared with saline except at the 0.5 µl volume (**Figure 3**).

The relatively poorer performance of ventral striatum implanted rats in Experiment 1 at all but the lowest level of lidocaine may reflect an impairment in remembering. However, important alternative explanations are related to the role of nucleus accumbens in the perception of reward. Sensitivity to a change in reward value or to the properties of a reinforcer have been associated with nucleus accumbens lesions (Apicella, Ljungberg, Scarnati, and Schultz, 1990, Bowman and Brown, 1998; Robbins and Everitt, 1996; Salamone, 1994). Nevertheless, despite lower accuracy at all but the 0.5 µl lidocaine level, ventral striatum implanted rats completed nearly all trials of each saline and lidocaine session. This demonstrates that inactivation of the ventral striatum did not affect motivation or the perception of the value of the reinforcer to the point that it would affect the tendency to complete a number of trials.

The present results are consistent with the finding of Burk and Mair (1999) that ventral striatal lesions affected delayed match to sample at a minimal delays. Differences between our results and those of Reading and Dunnett (1991) may reflect several features of the task. First, Reading and Dunnett targeted the nucleus accumbens while our inactivation site targeted the olfactory tubercle. Olfactory tubercle may be critical for remembering information at a short delay. Second, the task used by Reading and Dunnett did not require that rats disengage from the panel where choice levers were presented, as Burk and Mair's (1999) task did. Reading and Dunnett's rats' may have remained physically oriented toward the correct levers' positions. Finally, there may be a difference

between effects of permanent lesions to this area compared to effects of temporary inactivation be lidocaine.

LIST OF REFERENCES

Afifi, A. K. and Bergman, R. A. (1998). Functional Neuroanatomy, Text and Atlas. McGraw-Hill Publishers: New York.

Albert, D. J. and Madryga, F. J. (1980). An examination of the functionally effective spread of 4 μl of slowly infused lidocaine. *Behavioral and Neural Biology, 29,* 378-384.

Albert, D. J. and Wong, R. C. K. (1978). Hyperactivity, muricide, and intraspecific aggression in the rat produced by local anesthetic into the lateral septum or surrounding areas. *Behavioral Biology, 18,* 211-226.

Albin, R. L., Young, A. B., & Penney, J. B. (1989). The functional anatomy of basal ganglia disorders. *Trends in Neuroscience, 12,* 366-375.

Alexander, G. E. and Crutcher, M. D. (1990). Functional architecture of basal ganglia circuits: neural substrates of parallel processing. *Trends in Neuroscience, 13,* 266-271.

Alexander, G. E., DeLong, M. R., Strick, P. L. (1986). Parallel organization of functionally segregated circuits linking basal ganglia and cortex. *Annual Review of Neuroscience, 9,* 357-381.

Ambrogi-Lorenzini, C.G, Baldi, E., Bucherelli, C., Sachetti, B., and Tassoni, G. (1999). Neural topography and chronology of memory consolidation: A review of functional inactivation findings. *Neurobiology of Learning and Memory, 71,* 1-18.

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

Beiser, D. G., Hua, S. E. and Houk, J. C. (1997). Network models of the basal ganglia. *Current Opinion in Neurobiology, 7,* 185-190.

Berendse, H. W., Galis-de Graaf, Y., Groenewegen, H. J. (1992). Topographical organization and relationship with ventral striatal compartments of the prefrontal corticostriatal projections in the rat.
The Journal of Comparative Neurology, 316, 314-347.

Berendse, H. W. and Groenewegen, H. J. (1990). Organization of the thalamostriatal projections in the rat, with special emphasis on the ventral striatum. *The Journal of Comparative Neurology, 299*, 187-228.

Berendse, H. W. and Groenewegen, H. J. (1991). Restricted cortical termination fields of the midline and intralaminar thalamic nuclei in the rat. *Neuroscience, 42*, 73-102.

Bergman, H., Feingold, A., Nini, A., Raz, A., Slovin, H., Abeles, M., Vaadia, E. (1998). Physiological aspects of information processing in the basal ganglia of normal and parkinsonian primates. *Trends in Neurosciences, 21*, 32-38.

Bogousslavsky, J., Regli, F., and Uske, A. (1988). Thalamic infarcts: Clinical syndromes, etiology and prognosis. *Neurology, 38*, 837-848.

Bowman, E.M. and Brown, V. J. (1998). Effects of excitotoxic lesions of the rat ventral striatum on the perception of reward cost. *Experimental Brain Research 123*, 439-48

Brasted, P. J., Debrossy, M. D., Robbins, T. W. and Dunnett, S. B. (1998). Striatal lesions produce distinctive impairments in reaction time performance in two different operant chambers. *Brain Research Bulletin, 46*, 487-493.

Brown, L. L., Schneider, J. S., Lidsky, T. I. (1997). Sensory and cognitive functions of the basal ganglia. *Current Opinion in Neurobiology, 7*, 157-163.

Burk, J. A. and Mair, R.G. (1998). Thalamic amnesia reconsidered: Excitotoxic lesions of the intralaminar nuclei, but not the mediodorsal nucleus disrupt place DMTS performance in the rat *(Rattus norvegicus). Behavioral Neuroscience, 112*, 54-67.

Burk, J.A. & Mair, R.G. (1999). Effects of dorsal and ventral striatal lesions on delayed matching to sample based on position cues: implications for thalamic amnesia. *Behavioral Neuroscience.*

Butterworth, J. F. And Strichartz, G. R. (1990). Molecular mechanisms of local anesthesia: A review. *Anesthesia, 72,* 711-734.

Chapman, P. F., Steinmetz, J. E., Sears, L. L. and Thompson, R. F. (1990). Effects of lidocaine injection in the interpositus nucleus and red nucleus on conditioned behavioral and neuronal responses. *Brain Research, 537,* 149-156.

Collerton, D. (1986). Cholinergic function and intellectual decline in Alzheimer's disease. *Neuroscience, 19,* 1-28.

Cook, D. & Kesner, R.P. (1988). Caudate nucleus and memory for egocentric localization. *Behavioral and Neural Biology, 49,* 332-343.

Cummings, J. L. (1990). *Subcortical Dementia.* Oxford University Press: New York.

DeLuca, J. and Diamond, B. J. (1995). Aneurysm of the anterior communicating artery: A review of neuroanatomical and neuropsychological sequelae. *Journal of Clinical and Experimental Neuropsychology, 17,* 100-121.

DeLong, M. (1990). Primate models of movement disorders. *Trends in Neuroscience 13,* 281-296.

Divac, I., and Oberg, R. G. E. (1979). Current conceptions of neostriatal functions. In *The Neostriatum,* I. Divac, and R. G E. (Eds.) Oberg, Pergamon, New York.

Dubois, B. and Pillon, B. (1997). Cognitive deficits in Parkinson's disease. *Journal of Neurology, 244.* 2-8.

Dunnett, S.B. (1990). Role of prefrontal cortex and striatal output systems in short-term memory deficits with ageing, basal forebrain lesions, and cholinergic-rich grafts. *Canadian Journal of Psychology, 44,.* 210-232.

Dunnett, S. B. (1992). Aging, memory, and cholinergic systems: studies using delayed-matching and delayed-non-matching task in rats. In L. R. Squire and N. Butters (Eds.) *Neuropsychology of Memory, 2nd Edition.* Guilford Press: New York.

Floresco, S.B., Seamans, J.K. and Phillips, A.G. (1997). Selective roles for hippocampal, prefrontal cortical and ventral striatal circuits in radial arm maze tasks with or without a delay. *The Journal of Neuroscience, 17,* 1880-1890.

Givens B. and Olton DS. (1995). Bi-directional modulation of scopolamine-induced working
memory impairments by muscarinic activation of the medial septal area. *Neurobiology of Learning and Memory, 63,* 269-76

Givens, B. and Sarter, M. (1997). Modulation of cognitive processes by transsynaptic activation of the basal forebrain. *Behavioral Brain Research, 84,* 1-22.

Goldman-Rakic, P.S. and Selemon, L.D. (1990). New frontiers in basal ganglia research. *Trends in Neuroscience, 13,* 241-258.

Graybiel, A.M. (1990). Neurotransmitters and neuromodulators in the basal ganglia. *Trends in Neuroscience, 13,* 244-254.

Graybiel, A. M. (1995). Building action repertoires: memory and learning functions of the basal ganglia. *Current Opinion in Neurobiology, 5,* 733-741.

Groenewegen, H. J, Berendse, H. W. (1994). The specificity of the 'nonspecific' midline and intralaminar thalamic nuclei. *Trends in Neuroscience, 17,* 52-7.

Groenewegen, H. J., Wright, C. I. And Uylings, H. B. M. (1997). The anatomical relationships of the prefrontal cortex with limbic structures and the basal ganglia. *Journal of Psychopharmacology, 11,* 99-106.

grosse Beilage E., Robinson J. K., Koger S. M., Fox G. D., Zhang Y.P. (1992) Delayed-non-matching-to-sample performance is impaired by extensive, but not by limited, lesions of the thalamus in the rat. *Behavioral Neuroscience, 106,* 646-56

Haber, S. N., Lynd-Balta, E., and Spooren, W. P. J. M. (1992). Integrative aspects of basal ganglia circuitry. In G. Percheron, J. S. McKenzie, and J. Féger (Eds.) *The Basal Ganglia IV, Volume 41: New Ideas and Data on Structure and Function,* Plenum Press: New York.

Harrison, L. M. and Mair, R. G. (1996). A comparison of the effects of frontal cortical and thalamic lesions on measures of spatial learning and memory in the rat. *Behavioural Brain Research, 75.* 41-49.

Hauber, W. and Schmidt, W. J. (1994). Differential effects of lesions of the dorsomedial and dorsolateral caudate-putamen on reaction time performance in rats. *Behavioural Brain Research, 60,* 211-215.

Hayes, A.E., Davidson M.C., Keele, S.W., Rafal, R.D. (1998). Toward a functional analysis of the basal ganglia. *Journal of Cognitive Neuroscience, 10,* 178-98

Heimer, L., Alheid, G.F., Zaborszky, L. (1985). Basal ganglia. In Paxinos, G. (Ed.) The Rat Nervous System, Volume II: The Forebrain and Midbrain. Sydney: Academic Press. pp. 37-86.

Heimer, L., Switzer, R.D., Van Hoesen, G.W. (1982). Ventral striatum and ventral pallidum: components of a motor system? Trends in Neuroscience, 5. 83-87.

Herkenham, M. (1979). The afferent and efferent connections of the ventromedial thalamic nucleus in the rat. *Journal of Comparative Neurology, 183,*487-517

Horel, J. A. (1991). Use of cold to reversibly supress local brain function in behaving animals. *Methods in Neuroscience, 7,* 110.

Kemp, J. M. and Powell, T. P. S. (1970). The cortico-striate projection in the monkey. *Brain, 93,* 525-546.

Knight, R.G. (1992). Parkinson's Disease. In The Neuropsychology of Degenerative Brain Disease. Hillsdale, NJ: Lawrence Earlbaum Associates.

Knoth, R. L. and Mair, R. G. (1991). Response latency and accuracy on pretrained non-match to sample tasks in rats recovered from pyrithiamine induced thiamin deficiency. *Behavioral Neuroscience, 105,* 375-385.

Kolb, B. (1990). Organization of the neocortex of the rat. In B.Kolb and R.C. Tees (Eds.) The Cerebral Cortex of the Rat. The MIT Press: Cambridge, MA.

Kuhnishio, K., and Haber, S. N. (1994). Primate cingulostriatal projections: limbic striatal versus sensorimotor striatal input. *Journal of Comparative Neuroscience, 350,* 337-356.

Jones, E. G. (1985). *The Thalamus.* Plenum Press: New York.

Marín, O., Smeets, W.J.A.J., González, A. (1998). Evolution in the basal ganglia in tetropods: a new perspective based on recent studies in amphibians. *Trends in Neuroscience, 21,* 487-494.

Mair, R. G. (1994). On the role of thalamic pathology in diencephalic amnesia. *Reviews in the Neurosciences, 5,* 105-140.

Mair, R. G., Burk, J. A. And Porter, M. C. (1998). Lesions of the frontal cortex, hippocampus, and intralaminar thalamic nuclei have distinct effects on remembering in rats. *Behavioral Neuroscience, 112,* 772-792.

Mair, R. G. and Lacourse, D. M. (1992) Radio-frequency lesions of the thalamus produce delayed-nonmatching -to-sample impairments comparable to pyrithiamine-induced encephalopathy in rats. *Behavioral Neuroscience, 106,* 634-45

Malpeli, J. G. (1999). Reversible inactivation of subcortical sites by drug injection. *Journal of Neuroscience Methods, 86,* 119-128.

Markowitsch, H. J. (1991). Memory disorders after diencephalic damage. In W. C. Abraham, M. Corballis, and K. G. White, (Eds.). *Memory Mechanisms: A Tribute to G. V. Goddard.* Lawrence Earlbaum Associates Publishers: Hilsdale, NJ.

Markowitsch, H. J. And Pritzel, M. (1985). The neuropathology of amnesia. *Progress in Neurobiology, 25,* 189-287.

Martin, J. H. (1991). Autoradiographic estimation of the extent of reversible inactivation produced by microinjection of lidocaine and muscimol in the rat. *Neuroscience Letters, 127,* 160-164.

Martin, J. H. and Ghez, C. (1999). Pharmacological inactivation in the analysis of the central control of movement. *Journal of Neuroscience Methods, 86,* 145-159.

Maxwell, S. E., Delaney, H. D. (1990). *Designing Experiments and Analyzing Data: A Model Comparison Perspective*. Wadsworth Publishing: Belmont, CA

McDonald, R.J. & White, N.M. (1994). Parallel information processing in the water maze: evidence for independent memory systems involving dorsal striatum and hippocampus. Behavioral and Neural Biology, 61, 260-270.

McEntee, W. J., Biber, M. P., Perl, D. P., Benson, D. F. (1976). Diencephalic amnesia: a reappraisal. *Journal of Neurology, Neurosurgery, and Psychiatry, 39,* 436-441.

McGeorge, A. J. and Faull, R. L. M. (1989). The organization of the projection from the cerebral cortex to the striatum in the rat. *Neuroscience, 29,* 503-537.

Muir, J. L. (1997). Acetylcholine, aging and Alzheimer's Disease, *Pharmacology, Biochemistry and Behavior, 56,* 687-696.

Nauta, W. J. H., Smith, G. P., Faull, R. L M., Domesick, D. B. (1978). Efferent connections and nigral afferents of the nucleus accumbens septi in the rat. *Neuroscience, 3,* 385-401.

Packard, M.G., Cahill, L. and McGaugh, J.l. (1994). Amygdala modulation of hippocampal-dependent and caudate nucleus-dependent memory processes. Procedings of the National Academy of Science (USA), 91. 8477-8481.

Packard, M.G. and McGaugh, J.L. (1996). Inactivation of hippocampus or caudate nucleus with lidocaine differentially affects expression of place and response learning. Neurobiology of Learning and Memory, 65, 65-72.

Parent, A. (1990). Extrinsic connections of the basal ganglia. *Trends in Neuroscience, 13,* 254-265.

Parent, A., Bourassa, J., and Deschenes, M. (1996) The thalamostriatal projection system in rodents. In C. Ohye, M. Kimura, and J. S. McKenzie (Eds.) *The Basal Ganglia V, Plenum Press: New York.*

Parkin, A. J. (1992). Functional significance of etiological factors in human amnesia. In L. R. Squire and N. Butters (Eds.) *Neuropsychology of Memory, 2nd Edition.* Guilford Press: New York

Percheron, G., McKenzie, J. S., and Féger. J. (1992) *The Basal Ganglia IV, Volume 41: New Ideas and Data on Structure and Function,* Plenum Press: New York.

Pepin, E. P. and Auray-Pepin, A. (1993). Selective dorsolateral frontal lobe dysfunction associated with diencephalic amnesia. *Neurology, 43,* 733-741.

Perry, E. K., Haroutunian, V., Davis, K. L. Levy, R., Lantos, P., Eagger, S., Honovar, M., Dean, A., Griffiths, M., McKeith, I. G., Perry, R. H. (1994). Neocortical cholinergic activities differentiate Lewy body dementia from classical Alzheimer's disease. *Neuroreport, 5,* 747-749.

Prado-Alcalá , R. A., and Cobos-Zapiaín, G. G. (1977). Learning deficits induced by cholinergic blockade of the caudate nucleus as a function of experience. *Brain Research, 138,* 190-196

Prado-Alcalá, R.A., Maldonado, M.G. and Váquez, G.H. (1979). Caudate nucleus lesions and passive avoidance: a quantitative study. *Boletín de Estudios . Biologica de Mexico, 30,* 211-215

Peréz-Ruiz, C. and Prado-Alcalá, R. A. (1989). Retrograde amnesia induced by lidocaine injection into the striatum: Protective effect of the negative reinforcer. *Brain Research Bulletin, 22,* 599-603.

Pirozzolo, F.J., Hansch, E.C., Mortimer, J.A., Webster, D.D., Kukowski, M.A. (1982). Dementia in Parkinson's disease. A neuropsychological analysis. Brain and Cognition, 1. 71-83. In Dubois, B.& Pillon, B. (1997). Cognitive deficits in Parkinson's disease. Journal of Neurology, 244. 2-8.

Porter, M.C. and Mair, R. G. (1997). The effects of frontal cortical lesions on remembering depend on procedural demands of tasks performed in the radial arm maze. *Behavioural Brain Research, 87,* 115-125.

Portney, L. G. and Watkins, M. P. (1993). Foundation of Clinical Research: Applications to Practice. Appleton Lange: Norwalk, CT.

Poucet, B. and Buhot, M-C. (1994). Effects of medial septum or unilateral hippocampus inactivations on reference and working spatial memory in rats. *Hippocampus, 4,* 315-321.

Reading P. J., and Dunnett, S.B. (1991). The effects of excitotoxic lesions of the nucleus accumbens on a matching to position task. *Behav Brain Research, 46,* 17-29.

Roitblatt, H. L. And Harley, H. E. (1988). Spatial delayed matching-to-sample performance by rats: learning, memory, and proactive interference. *Journal of Experimental Psychology, 14,* 71-82.

Robbins, T.W. Giardini, V. Jones, G.H. Reading, P. and Sahakian, BJ. (1990). Effects of dopamine depletion from the caudate-putamen and nucleus accumbens septi on the acquisition and performance of a conditional discrimination task. *Behavioural Brain Research, 38,* 243-261.

Robbins, T.W., and Everitt, B. J. (1996). Neurobehavioural mechanisms of reward and motivation. *Current Opinion in Neurobiology, 6,* 228-36.

Robinson, J. K. and Mair, R. G. (1992). MK-801 prevents brain lesions and delayed-non-matching-to-sample deficits produced by pyrithiamine-induced encephalopathy in rats. *Behavioral Neuroscience,106,* 623-33

Salamone, J. D. (1996). The involvement of nucleus accumbens dopamine in appetitive and aversive motivation. *Current Opinion in Neurobiology, 6,* 228-236.

Sandkühler, J. and Gebhart, G. F. (1991). Production of reversible local blockage of neuronal function. *Methods in the Neurosciences, 7,* 122-137.

Sandkühler, J., Maisch, B., and Zimmerman, M. (1987). The use of local anaesthetic microinjections to identify central pathways: a quantitative evaluation of the time course and extent of the neuronal block. *Experimental Brain Research, 68,* 168-78

Seamans, J.K. & Phillips, A.G. (1994). Selective memory impairments produced by transient lidocaine-induced lesions of the nucleus accumbens in rats. *Behavioral Neuroscience, 108,* 456-486.

Selemon, L.D. and Goldman,-Rakic, (1985). Longitudinal topography and interdigitation of corticostriatal projections in the rhesus monkey. *Journal of Neuroscience, 5,* 776-794.

Smith, Y., Bennett, B. D., Bolam, J. P., Parent, A., and Sadikot, A. F. (1994). Synaptic relationships between dopaminergic afferents and cortical or thalamic inputs in the sensorimotor territory of the striatum in the monkey. *The Journal of Comparative Neurology, 344,* 1-19.

Squire, L. R. and Butter, N. (1992). *Neuropsychology of Memory, 2nd Edition.* Guilford Press: New York

Squire, L. R.. Zola-Morgan, S. and Chen, K.S. (1988). Human amnesia and animal models of amnesia: Performance of amnestic patients on tests designed for the monkey. *Behavioral Neuroscience, 102,* 210-221.

Squire, L.R. and Zola-Morgan, S. (1991). The medial temporal lobe memory system. *Science, 253,* 1380-1386.

Stacy, M. & Jankovic, J. (1992). Clinical and Neurobehavioral Aspects of Parkinson's Disease. In S.J Huber & J.L Cummings (1992) (Eds.) Parkinson's Disease. New York: Oxford University Press. pp. 11-31.

Sutherland, R. J. and Rudy, J. W. (1988). Configural association theory: The role of hippocampal formation in learning, memory and amnesia. *Psychobiology, 17,* 129-144.

Tehovnik, E. J., and Sommer, M. A. (1997). Effective spread and timecourse of neural inactivation caused by lidocaine injection in monkey cerebral cortex. *Journal of Neuroscience Methods, 74,* 17-26.

van Hest, A., and Steckler, T. (1996). Effects of procedural parameters on response accuracy: lessons from delayed (non-) matching procedures in animals. *Cognitive Brain Research, 3,* 193-203.

Victor, M., Adams, R. D. and Collins, G. H. (1989). *The Wernicke-Korsakoff Syndrome and Related Neurological Disorders Dur to Alcoholism and Malnutrition.* F. A. Davis, Co: Philadelphia.

von Cramon, D. Y., Hebel, N. and Schuri, U. (1985). A contribution to the anatomical basis of thalamic amnesia. *Brain, 108,* 993-1008.

Welsh, J. P. and Harvey, J. A. (1991). Pavlovian conditioning in the rabbit during inactivation of the interpositus nucleus. *Journal of Physiology, (Lond). 444,* 459-80.

Wichman, T., DeLong, M.R. (1996). Functional and pathophysiological models of the basal ganglia. *Current Opinion in Neurobiology, 6,* 751-758.

Whishaw, I. Q. and Dunnett, S. B. (1985). Dopamine depletion, stimulation or blockade disrupts spatial navigation and locomotion dependent upon beacon or distal cues. *Behavioural Brain Research, 18,* 11-29.

White, N. M. (1997). Mnemonic functions of basal ganglia. *Current Opinions in Neurobiology, 7,* 164-169.

Wright, C. I. and Groenewegen, H. J. (1995). Patterns of convergence and segregation in the medial nucleus accumbens of the Rat: Relationships of prefrontal cortical, midline thalamic, and basal amygdaloid afferents. *The Journal of Comparative Neurology, 361,* 383-403.

Wright, C. I. and Groenewegen, H. J. (1996). Patterns of overlap and segregation between insular cortical, intermedial thalamic and basal amygdaloid afferents in he nucleus accumbens of the rat. *Neuroscience, 73,* 359-373.

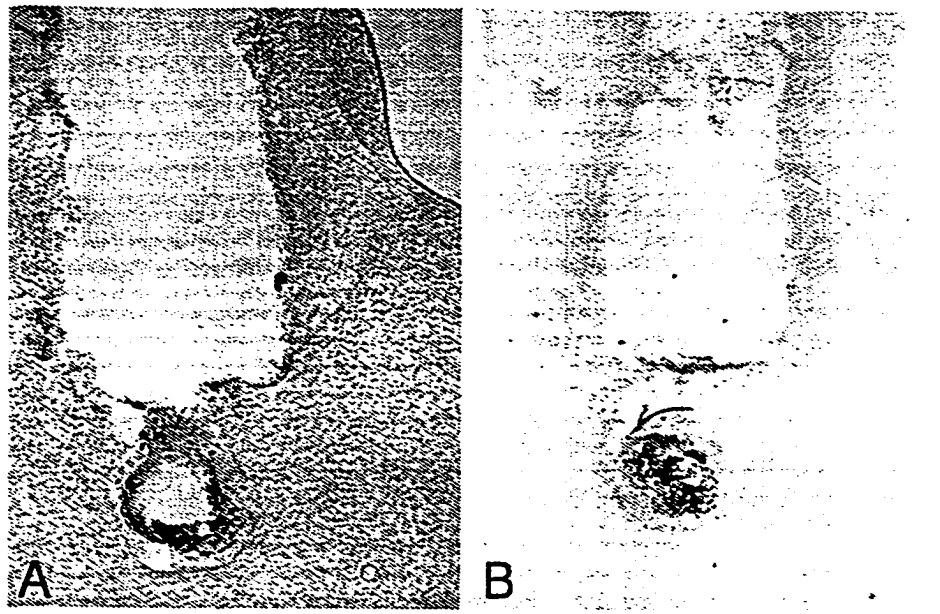
Young, H.A., Stevens, A. A., Converse, E. K., and Mair, R. G. (1996). A comparison of memory temporal decay in place memory tasks in rats with lesions affecting thalamus, frontal cortex, or the hippocampal system. *Behavioral Neuroscience, 110,* 1-17.

Zhang, Y.P. , Burk, J. A., Glode, B.M., and Mair, R. G. (1998). The effects of thalamic and olfactory cortical lesions on continuous olfactory discrimination DNMTS and olfactory discrimination in the rat. *Behavioral Neuroscience, 112,* 39-53.

Zola-Morgan, S. and Squire, L. R. (1993). Neuroanatomy of memory. *Annual Reviews in Neuroscience, 16,* 547-63.

APPENDIX

Figure 1 (top). Experiments 1 & 2. Typical cannula tip with 0.5 mm projection (cresyl violet) and spread of Chicago Blue from 0.5 mm projection of internal cannula (adjacent section) in ILn. Figure 1 (bottom). Experiment 3. Typical cannula tip with 2.0 mm projection (cresyl violet) and spread of Chicago Blue from 0.5 mm projection (adjacent section).



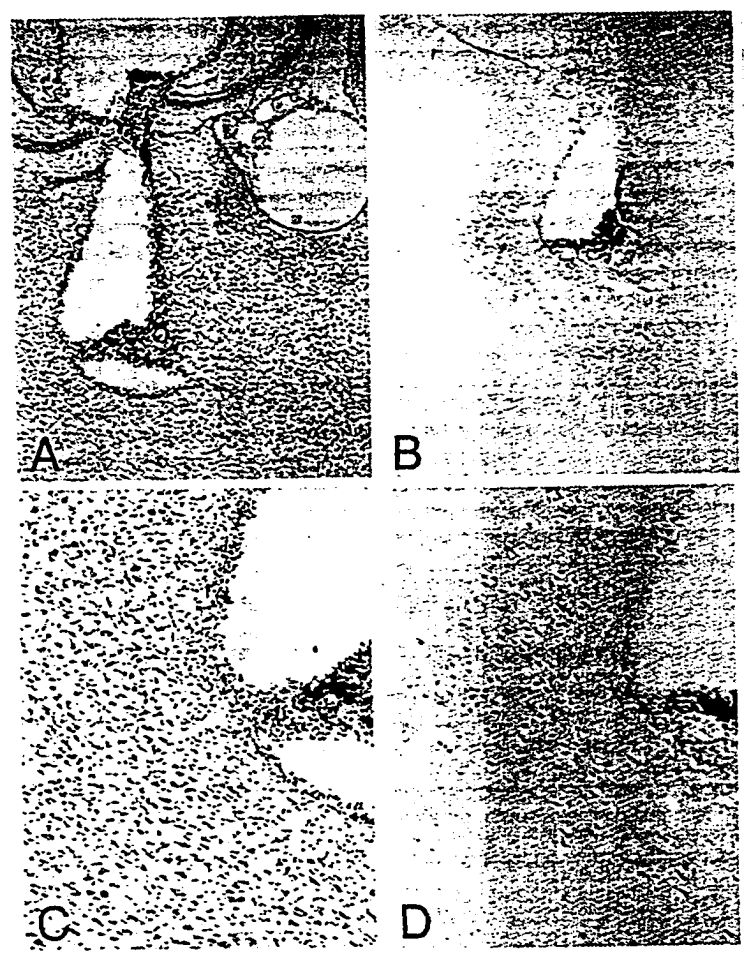


Figure 2A. Experiment 1. Typical cannula tips in anterior ventral striatum (cresyl violet). Figure 2B. Typical cannula tips in posterior ventral striatum (cresyl violet).

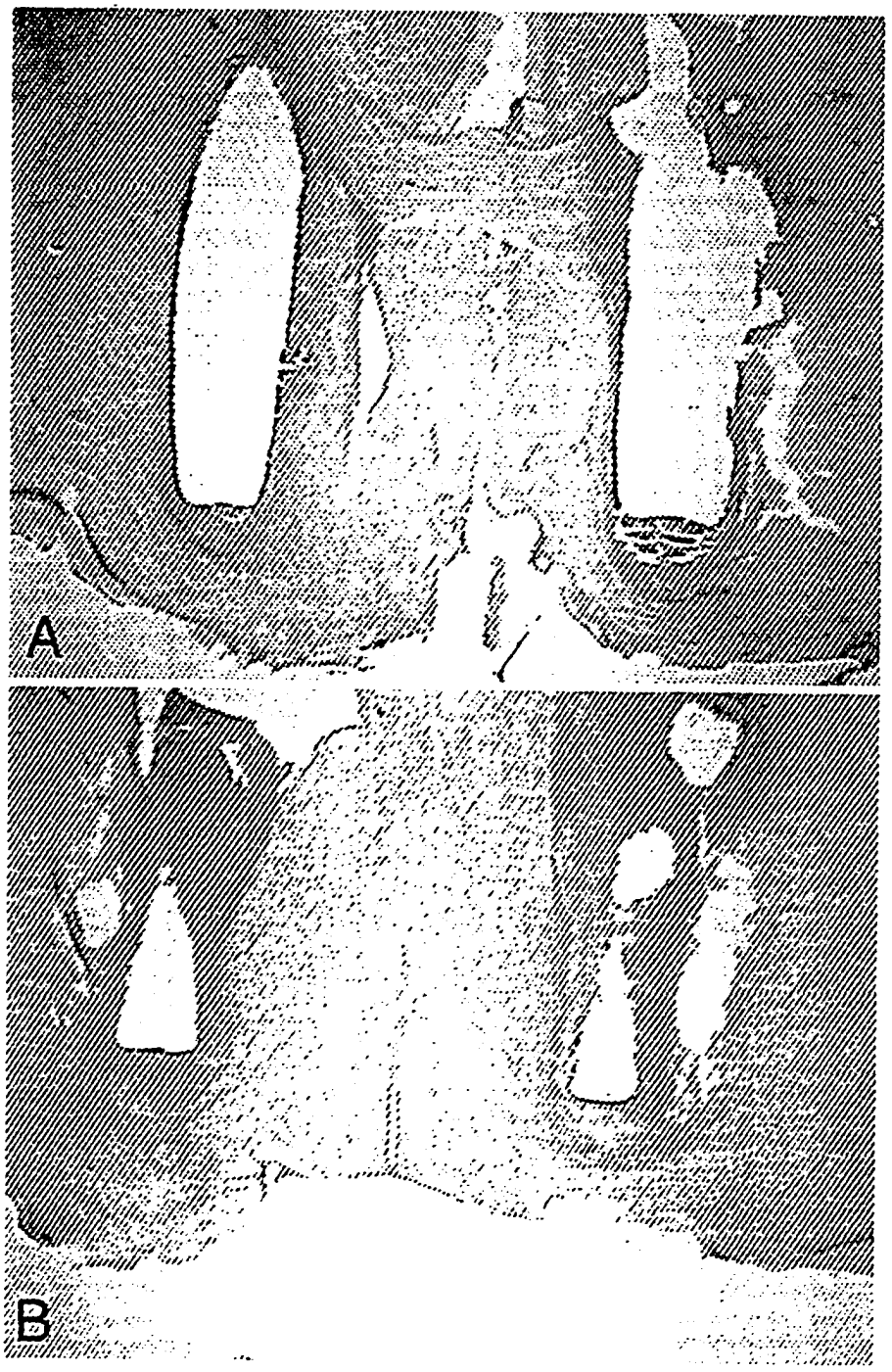
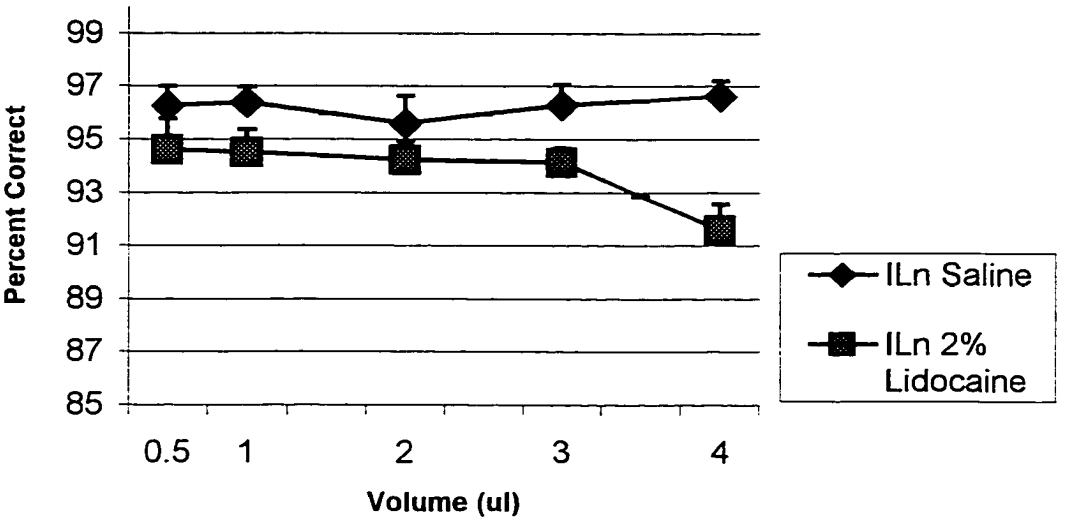


Figure 3. Experiment 1: Accuracy of intralaminar nucleus and ventral striatum implanted rats following ascending volumes of saline and 2% lidocaine



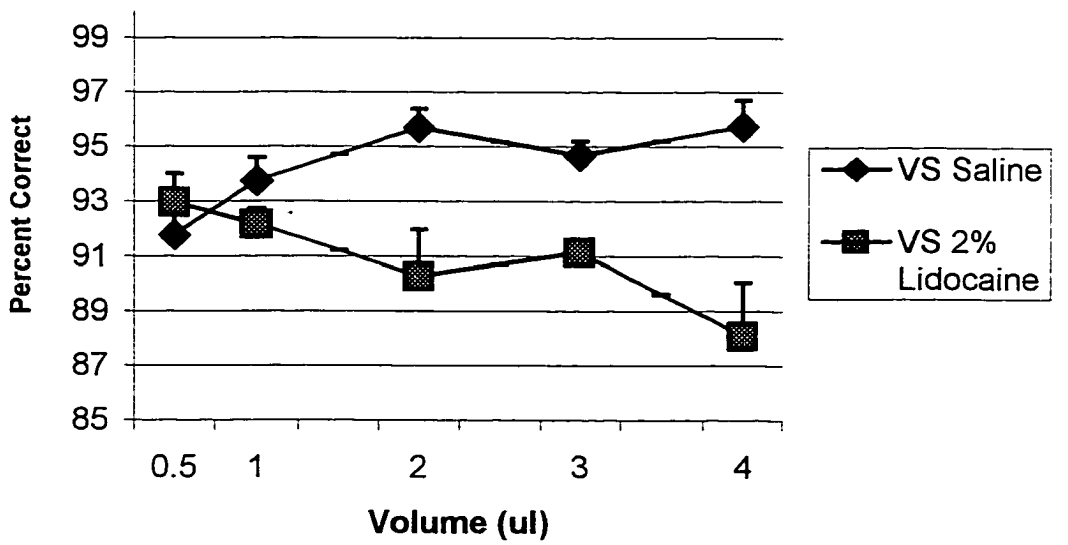


Figure 4. Experiment 1: Accuracy of intralaminar nucleus and ventral striatal implanted rats during Dry Conditions

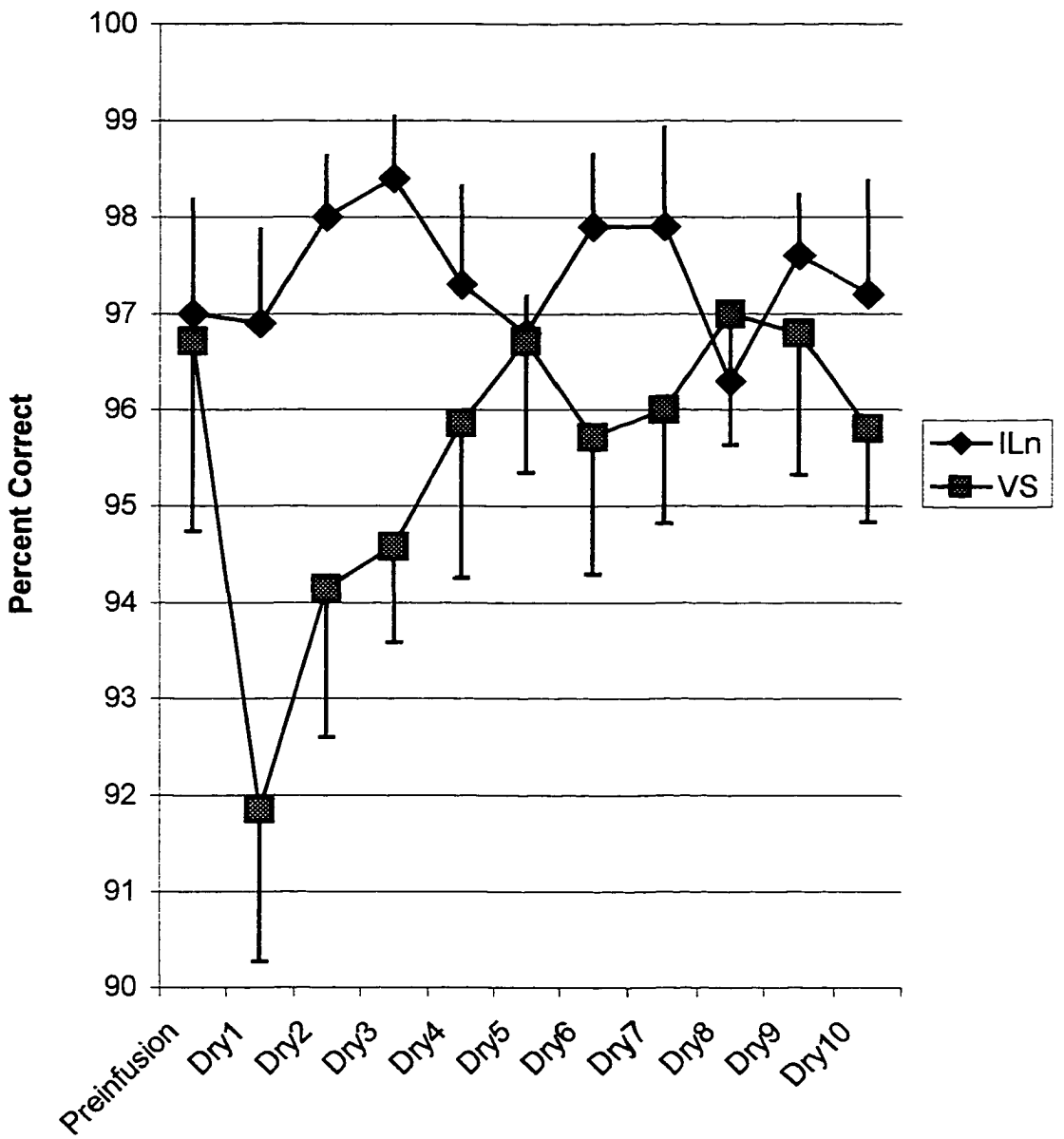


Figure 5. Experiment 2: Accuracy of intralaminar nucleus implanted rats, Dry vs. 3.0 ul of saline and 3.0 ul of 2% lidocaine

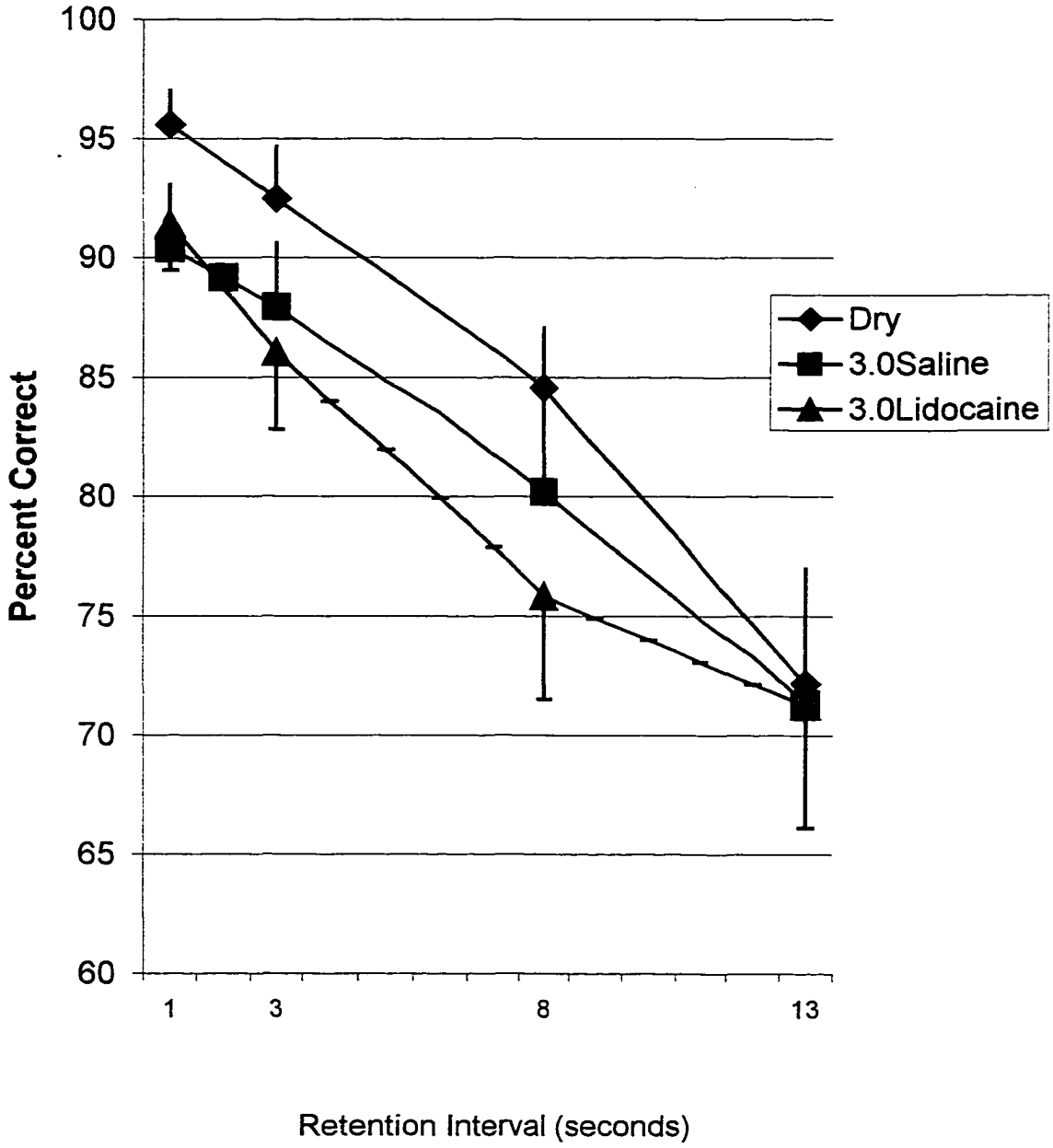


Figure 6. Exp 2: Median response speed of intralaminar nucleus implanted rats trained in dry conditions vs. infusions of 3.0 ul of saline and 3.0 ul of 2% lidocaine

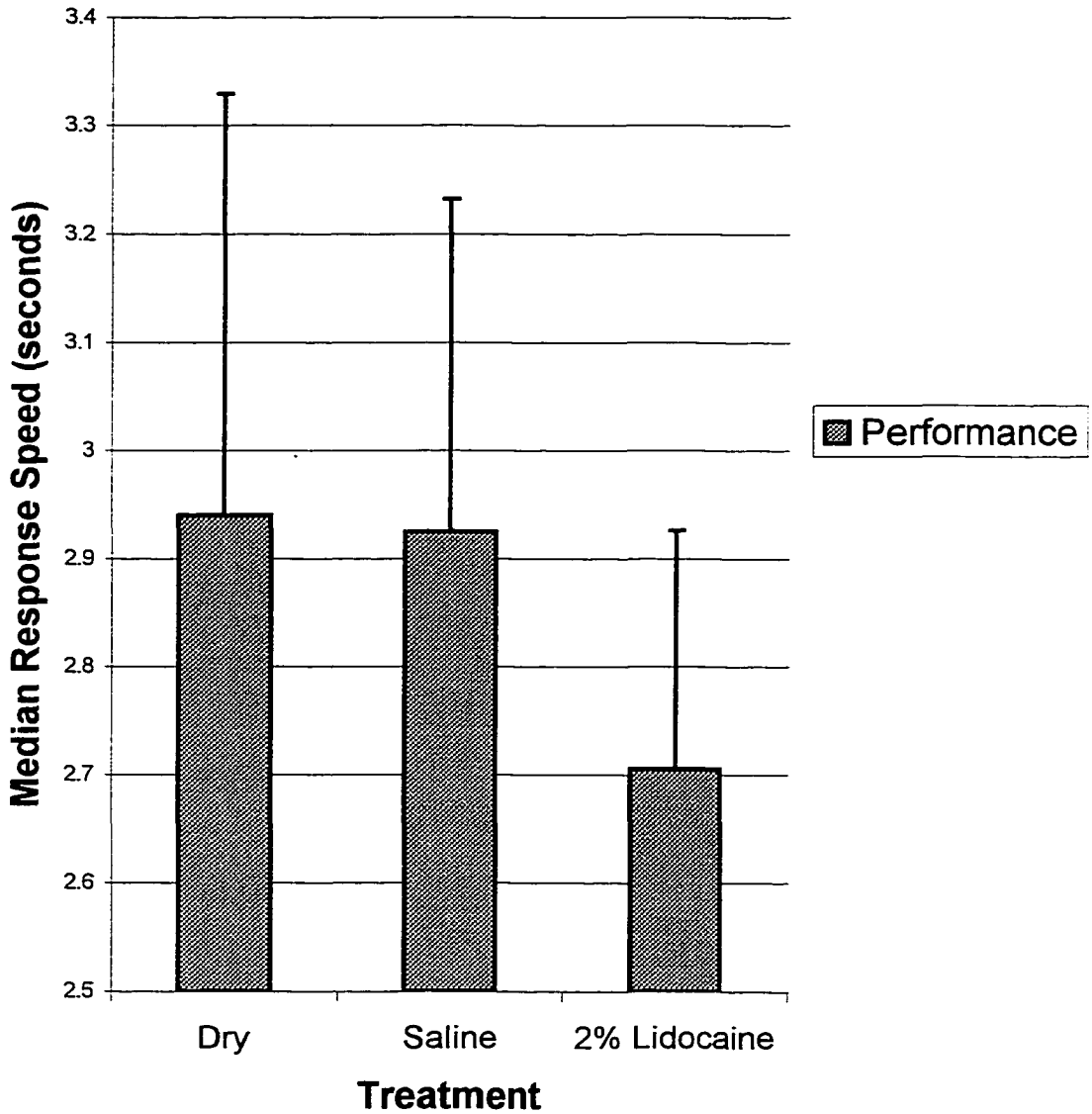


Figure 7. Experiment 3: Accuracy of intralaminar nucleus implanted rats following saline, 2%, 4%, and 8% lidocaine, at 4 delays

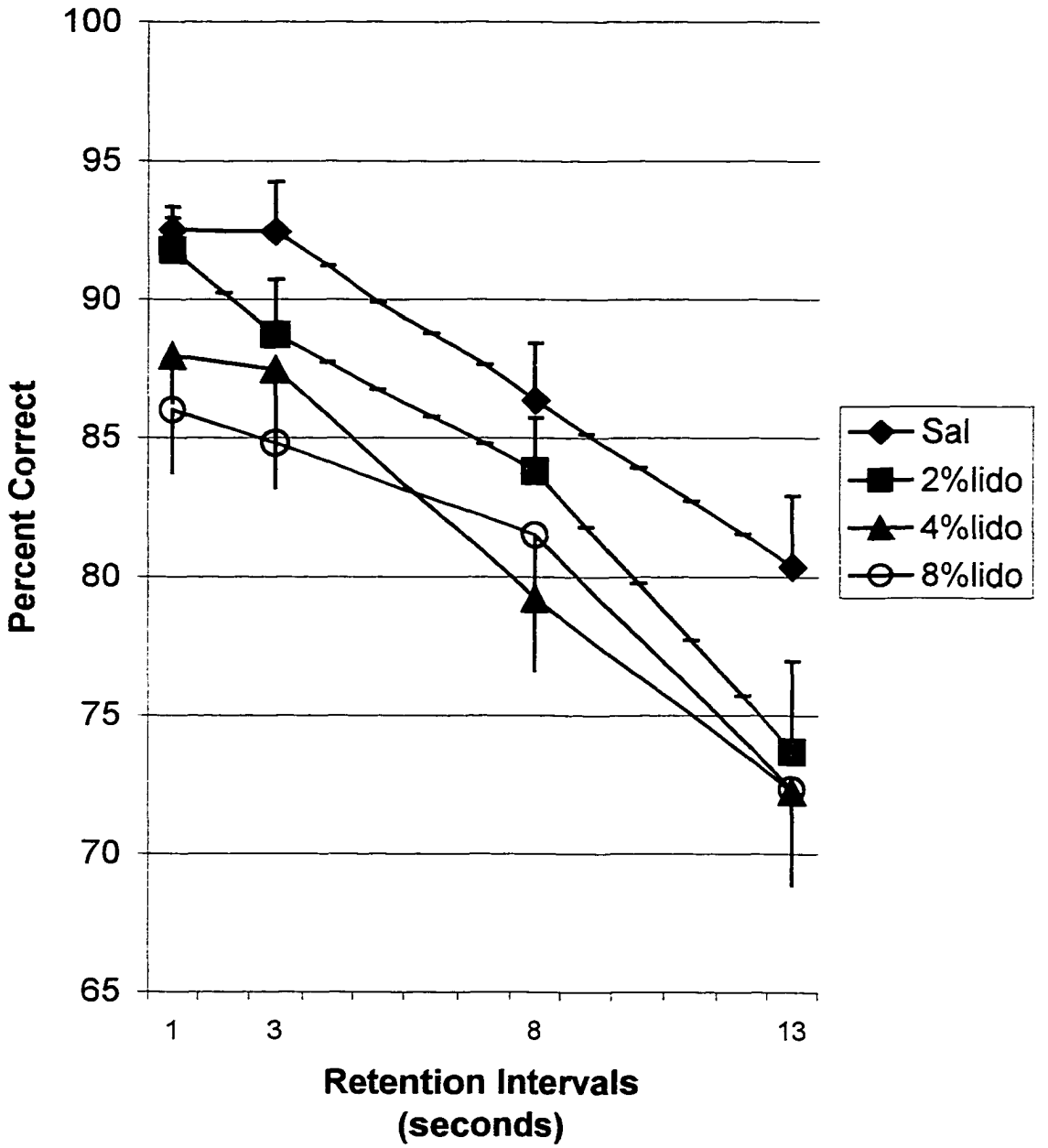
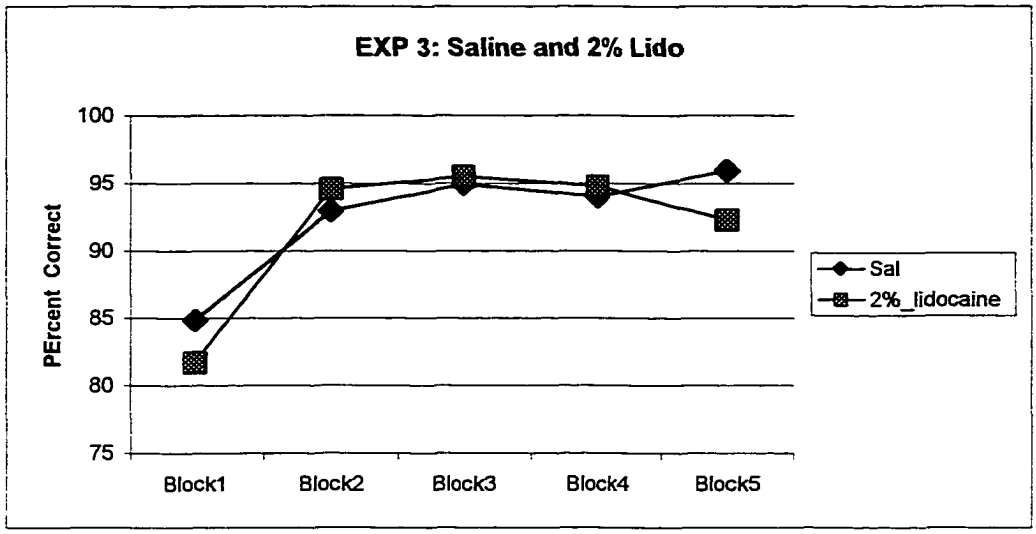
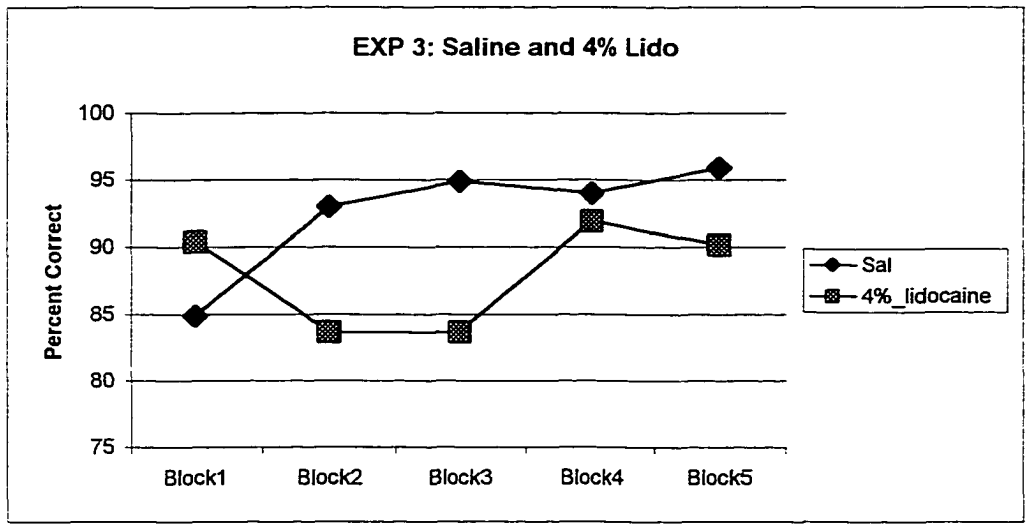


Figure 8. Experiment 3: Accuracy of intralaminar nucleus implanted rats at various concentrations of lidocaine





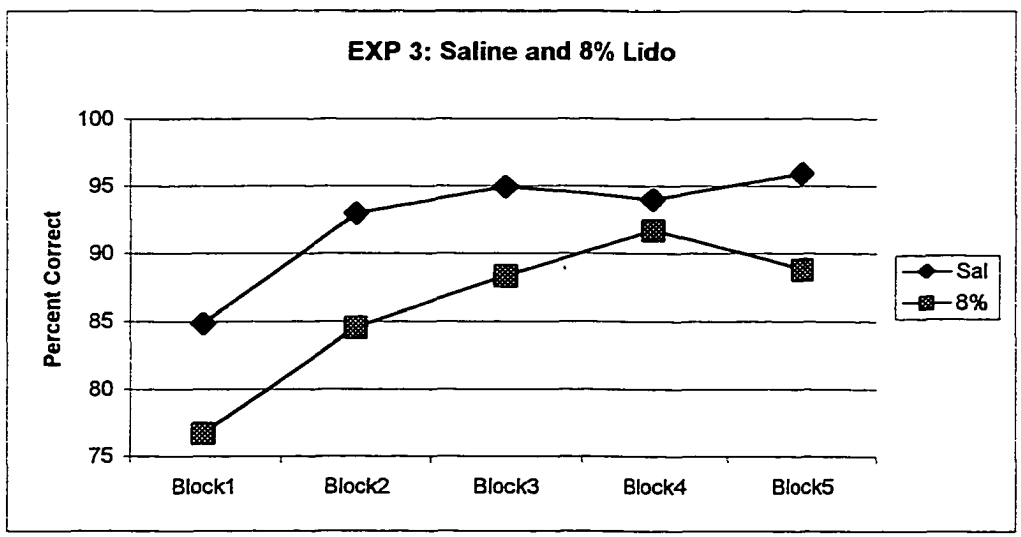


Figure 9: Experiment 3: Response speed of intralaminar implanted rats following infusions of various concentrations of lidocaine

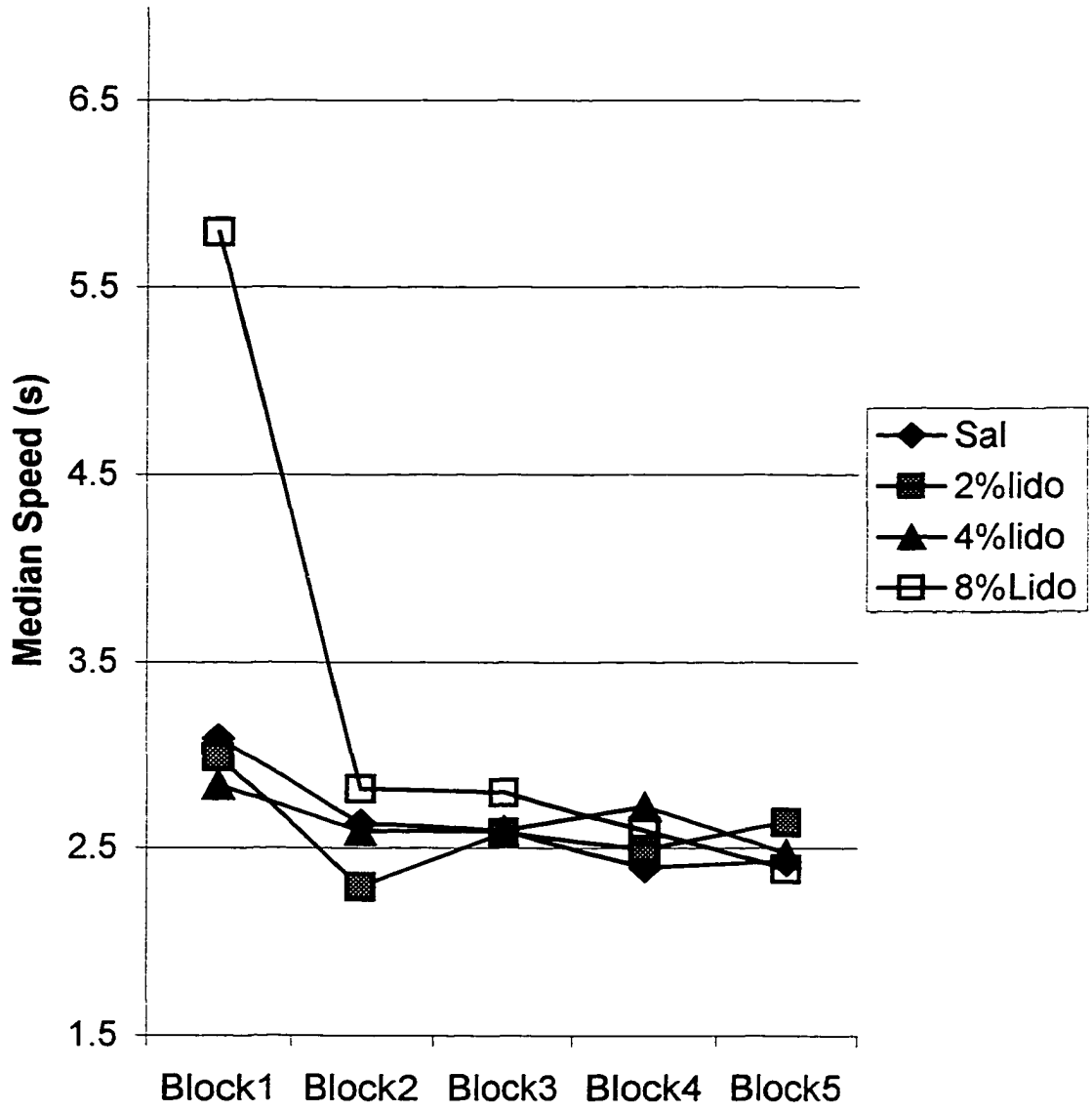


Figure 10. Experiment 3. Comparison of accuracy for intralaminar nucleus rats over dry conditions