

Importance of Behavioral Manipulations and Measures in Rat Models of Brain Damage and Brain Repair

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Abstract

The relevance of careful behavioral measures and manipulations in animal research on neural plasticity and brain damage has become increasingly clear. Recent research in adult rats indicates that an understanding of neural restructuring after brain damage requires an understanding of how it is influenced by postinjury behavioral experiences. Other research indicates that optimizing pharmacological and other treatments for brain damage may require their combination with rehabilitative training. Assessing the efficacy of a treatment approach in animal models requires the use of sensitive behavioral measures of functional outcome. In research on restorative plasticity after brain damage, procedures for handling and housing rats should promote the quality of behavioral measures and manipulations.

Key Words: dendritic growth; experience-dependent plasticity; learning; motor cortex; rehabilitative training; stroke; synaptogenesis

Introduction

Brain damage resulting from traumatic head injuries and strokes is a leading cause of death and long-term disability. The societal impact of brain damage, its monetary costs, and its impact on brain damage survivors and caregivers are enormous. Each year, approximately 1.5 million Americans sustain traumatic brain damage (Thurman et al. 1999) and more than 600 thousand have a new or recurrent stroke (AHA 2002). There are currently approximately 5.3 million survivors of traumatic brain damage and 4.6 million stroke survivors in the United States. Within the

upcoming decades, brain damage resulting from stroke is expected to become an even larger problem given the aging of our population and that the odds of having a stroke increase with age (e.g., Lakatta 2002; Williams 2001).

Stroke can cause a rapid and devastating amount of damage to the brain as a result of a cessation of the blood supply, resulting from a blockage or a rupture of vasculature. Secondary damage results from neuroinflammatory responses, excitotoxicity, mitochondrial dysfunction, disruptions of calcium homeostasis, and other severe alterations in normal cellular function (reviewed in Choi 1995; Martin 2001; Mattson et al. 2000; Sattler and Tymianski 2001). The loss of neurons and their connections (“synapses”) by these processes results in impairments in the functions previously controlled by the damaged region(s). For example, damage to the region of the cerebral cortex that controls motor function is associated with impairments in movement. Damage to regions of the posterior cortex leads to decrements in visual ability. Brain regions that are interconnected with the region of direct damage (e.g., connected cortical and subcortical regions) are also affected. Even neurons very remote from the site of the stroke must adapt to the degeneration of a portion of their synaptic input and/or output. If the extent of the damage is great, full recovery of behavioral function is unlikely.

In research aimed at improving stroke outcome, two major approaches are used for reducing the functional impairments incurred by stroke. The first approach is to try to reduce the damage resulting from the initial and secondary cascades of cell death after the stroke onset (e.g., Kermer et al. 1999; Phan et al. 2002). This approach has been a major focus of research since the mid-1980s and has resulted in the development of a few promising treatments, including treatment with tissue plasminogen activator (reviewed in Goldstein 2001). However, these treatments must be administered within a very limited time window (typically the first few hours) after the stroke onset to be effective, which excludes many people with strokes. Even of those undergoing acute stroke treatments, many will be left with sufficiently severe impairments to warrant additional treatment. The need to develop therapies that are effective when administered in the weeks to years after stroke clearly remains.

A second major research approach is to promote reorganization of remaining brain tissue and neural circuitry so that the lost function becomes side-stepped by the reorga-

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nized brain. Several promising areas of research along these lines include the provision of substances that promote the growth of new neural processes (“neurotropic factors” or “growth factors”), various other pharmacological approaches to enhance neural plasticity, and the insertion of neural progenitor cells into the brain (reviewed in Johansson 2000; Nishino and Borlongan 2000; Papadopoulos et al. 2002; Ren et al. 2001). However, such treatments are limited if the brain cannot be made to form new neural connections that are functionally beneficial. This problem would appear to be almost insurmountable at this time, given that neuroscientists are far from understanding most of the brain’s circuitry at the level of individual synapses; however, the problem seems less hopeless if one considers that the brain continuously modifies its neural circuitry in a functionally appropriate manner. It has apparently evolved to do so in response to changes in behavioral experience, including learning. This type of naturally occurring neural plasticity is, in our view, the key to making the brain reorganize in a functionally appropriate manner after strokes and other types of brain damage.

It is likely that one can capitalize on the brain’s neuroplastic responses to learning to promote neural reorganization and to improve behavioral function after brain damage. We briefly review the research supporting this idea below. A central component of this research in animals is the use of sensitive behavioral manipulations and measures. Thus, we conclude this review with a discussion of care and handling practices in promotion of this research in rats.

Experience-dependent Neural Plasticity

The brain undergoes continuous modification of its neural connections throughout the lifespan of an animal (reviewed in Black et al. 1997). During development, the appropriate types of experience during sensitive stages are required to shape the architecture of neural connections. In many mammals, including humans, synapses have been found to be overproduced in the developing cortex by approximately twice the number found in adults. The partial loss of synapses is driven by competitive processes (i.e., the selective maintenance of synapses that are effectively activated by behavioral experience and the loss of poorly activated ones). For example, experience with binocularly detected visual patterns is a critical force driving the connectivity in the visual cortex (reviewed in Crowley and Katz 2002). If this experience is not available during development, normal visual cortical development and visual function can be permanently lost. Similar experience-dependent processes have been found to occur during the development of other capacities (e.g., audition and somatosensation).

Behavioral experience continues to modify the brain throughout adulthood. There is a wealth of data now to indicate that adult animals, including humans, undergo brain changes as a result of learning. For example, learning new motor skills is associated with changes in the motor

cortex and cerebellum, including the growth neuronal dendrites (the sites of most synaptic inputs) (Kleim et al. 1997; Withers and Greenough 1989), the addition of synaptic connections (Black et al. 1990; Kleim et al. 2002), changes in patterns of neural activity (Kleim et al. 1998; Nudo et al. 1996; Plautz et al. 2000), increases in cortical thickness (Anderson et al. 2002), and, in humans, increased cortical activity as detected using functional magnetic resonance imaging (Elbert et al. 1995; Karni et al. 1998). Rats socially housed either as adults or as juveniles within complex environments (environments filled with toys and other objects to explore and manipulate) have a thicker cortex, increased synapse number per neuron, more vasculature, more glial (non-neuronal) cell processes, and increases in several plasticity-related molecules in the visual cortex compared with animals housed in pairs or individually in standard laboratory cages (reviewed in Black et al. 1997; Rosenzweig and Bennet 1996). For many morphological variables, social housing in standard laboratory cages results in effects that are intermediate between those of complex-housed and individually caged rats (e.g., Turner and Greenough 1985). Complex environment housing also promotes the survival of new neurons produced in the hippocampus of adult animals (mice and rats) (Kempermann et al. 1997; Nilsson et al. 1999). In later stages of life, behavioral experience can offset age-related losses of neural structure and function (e.g., Coq and Xerri 2001; Green et al. 1983; Nakamura et al. 1999).

Not all significant behavioral experiences promote neuronal growth. Although an extensive discussion of the topic is beyond the scope of this article, the negative side to the sensitivity of the brain to experience is its response to severe or chronic stress. One example of this response is the resulting loss of dendrites and suppression of neurogenesis in the hippocampus (reviewed in McEwen 2001).

Understanding experience-dependent neural plasticity is important for brain damage research in part because individuals that survive brain damage may undergo some of the most significant behavioral changes of their adult life. For example, individuals with sufficient damage to the sensory and motor cortex of the left hemisphere (a very common site of damage incurred by strokes) have impairments in movement and sensation of the right body side. It may be necessary for these individuals to learn to rely on the left hand to eat, write, and type. If walking is possible, they may develop whole body postural adjustments to accomplish it.

It is also important to understand how changes in behavior can be used to drive functional restructuring after brain damage. Several lines of recent research indicate that postinjury behavioral experiences are important contributors to neural restructuring after brain damage. The behavioral changes that contribute to brain reorganization after brain damage can be loosely grouped into two categories: (1) changes an animal develops in an effort to compensate for lesion-induced impairments, and (2) changes induced using rehabilitative training.

Neural Plasticity and Behavioral Compensation

It has long been appreciated that much of the recovery of function after brain damage is due to the development of compensatory behavioral strategies that circumvent impairments (e.g., Gazzaniga 1966; Gentile et al. 1978; Whishaw 2000). Our research indicates that the “normal” course of postinjury neuronal restructuring is also, in part, a consequence of such lesion-induced behavioral changes. This process is modeled in adult Long-Evans hooded rats that sustain unilateral damage to the forelimb representation region of the somatic-sensory and motor cortex (Figure 1A). This region is homologous to an area that is commonly

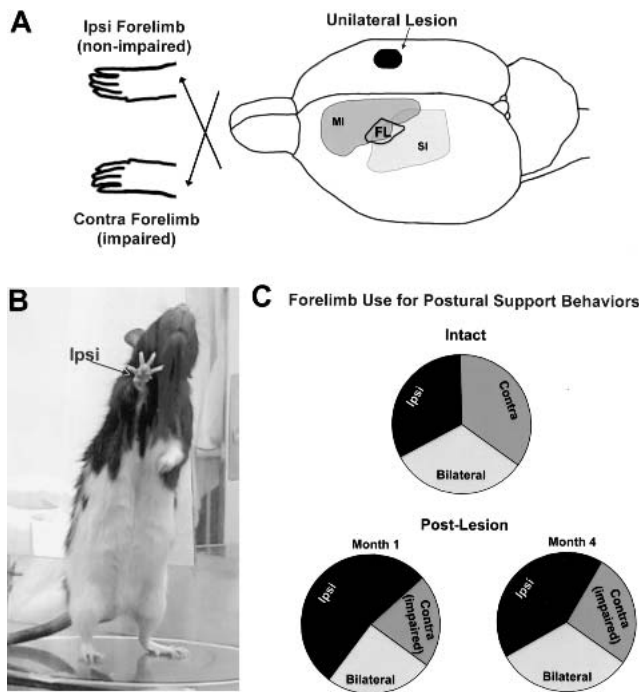


Figure 1 (A) Schematic representation of a rat brain showing the placement of unilateral lesions in the sensorimotor cortex. MI, primary motor cortex; SI, primary somatic-sensory cortex; FL, forelimb representation region. These lesions result in impairments in the forelimb opposite the lesions, and (B) the development of a compensatory reliance on the nonimpaired forelimb for the postural support behaviors used to explore vertical and horizontal surfaces. These postural support behaviors have been chosen for quantification because they are extremely common behaviors expressed by the rats whenever they are active. (C) Percentage of use of the forelimb ipsilateral and contralateral to the lesion and simultaneous use of both forelimbs (bilateral) for upright postural support behaviors. Intact animals typically have symmetrical use of the forelimbs. After the unilateral lesions, there is a major increase in the use of the ipsilateral (nonimpaired) forelimb, and this effect is very enduring. Data are from Gregory AD, Jones TA. 1998. Long-term synaptic-structural plasticity in the motor cortex opposite unilateral sensorimotor cortical damage in adult rats. Soc Neurosci Abstr 24:436.

damaged by strokes affecting the middle cerebral artery in humans. Damage to this region in rats results in impairments in the forelimb opposite the lesion on a variety of tests of sensorimotor function. Rats are still capable of using the impaired forelimb (e.g., for grooming and eating); however, sensitive behavioral measures indicate impaired responsiveness to somatic sensory stimulation and impairments in the use of this limb for postural support and coordinated forelimb placement during locomotion (e.g., Barth et al. 1990; Schallert et al. 1997, 2000).

In addition to lesion-induced impairments, another striking behavioral effect of the lesions is the development of compensatory behavioral changes (Jones and Schallert 1992). Rats “spontaneously” develop a hyper-reliance on the nonimpaired forelimb (ipsilateral to the lesion) for normal postural support behaviors, such as forelimb use for upright exploratory behaviors around the homecage (Figure 1, B and C). These compensatory behaviors are so effective in permitting exploratory behaviors in the brain damaged rats that within a few days after the injury, observers not extremely familiar with rat postural-motor behaviors would probably perceive the animals to be normal.

These unilateral lesions also result in remarkable neuroanatomical changes in the motor cortex opposite and homotopic to the lesions. Over time after the lesions, there is a sequence of changes in the neuronal and non-neuronal (glial) cells in this region (Figure 2). Early after the lesions,

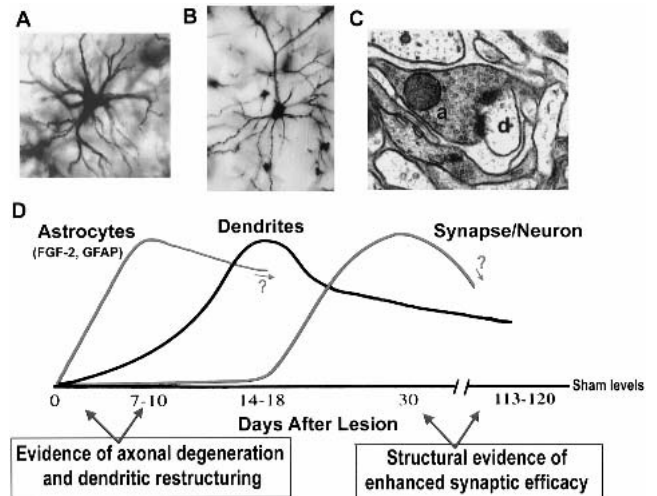


Figure 2 Approximate time course of morphological changes in the motor cortex opposite the lesions. (A) Astrocyte (a subtype of glial cell) immunostained with glial fibrillary acidic protein (GFAP). (B) Dendritic processes of a pyramidal neuron in the motor cortex. (C) Synaptic connection including an axonal (a) and dendritic (d) process. (D) Unilateral lesions of the sensorimotor cortex result in a sequence of glial and neuronal changes, including increases in the astrocytic expression of the neurotrophic factor fibroblast growth factor-2 (FGF-2) GFAP increases in dendritic processes and increases in synapse number per neuron. See text for details.

there is a loss of axonal processes (neuronal output presumably arising from the damaged cortex) (Jones 1999). Also around this time, there are reactive changes in a subtype of glial cells and astrocytes as well as increases in the presence of neurotropic factors and increases in proteins associated with neuronal plasticity (Dahms et al. 1999). At later time points, axonal processes, dendritic processes and synaptic connections undergo major growth within this region (Jones 1999; Jones and Schallert 1992; Jones et al. 1996). Synaptic connections also show structural changes that are characteristic of increases in their potency (i.e., the development of more perforated postsynaptic densities and multisynaptic boutons). Related research from other laboratories has revealed increases in synapse-associated proteins and neuronal growth-associated molecules (e.g., Cheng et al. 1997; McNeill et al. 1999; Stroemer et al. 1995) as well as changes in neuronal excitability (reviewed in Witte et al. 2000) in the cortex opposite unilateral lesions. Somewhat larger lesions of the sensorimotor cortex can cause the contralateral motor cortex to contribute new axonal projections to subcortical regions underlying the lesion (e.g., Carmichael and Chesselet 2002).

After unilateral lesions, animals begin to rely increasingly on the nonimpaired forelimb. To what extent does this increased reliance contribute to the neural and glial plasticity in the cortex opposite the lesion? Restricting the movements of the nonimpaired forelimb (using limb-restricting vests that limit forelimb movements to a pocket of space formed close to the torso) for the first 15 days after the lesions prevented the dendritic growth normally found in the cortex opposite the lesion (Jones and Schallert 1994). Animals that had the impaired forelimb restricted showed the normal dendritic growth. This indicated that use of the nonimpaired forelimb was necessary for the dendritic growth to occur in the motor cortex opposite the lesion. However, forced reliance on one forelimb in intact animals resulted in much more subtle changes in dendrites compared with those found after the lesions. Thus, the neuronal growth effects were not simply a result of the behavioral changes induced by the lesion.

These findings led to the hypothesis that the degenerative effects of the lesion (i.e., the loss of some axonal input and the resulting induction of growth-promoting processes) cause this region of cortex opposite the lesion to become especially responsive to behavioral changes. To test this hypothesis, we independently manipulated forelimb behavior and lesion-induced denervation of the motor cortex (Bury et al. 2000a). Denervation of connections to the motor cortex was induced by partial transections of the corpus callosum (the pathway taken by motor cortical fibers connecting the 2 hemispheres). To change forelimb behavior, some animals were placed in limb restricting (1-holed) vests to force reliance on a single forelimb. Animals with sham operations and control (2-holed) vests served as controls. It was found that animals forced to rely on one forelimb for 2 wk after corpus callosum transections had a major dendritic

growth in the motor cortex opposite the forced use limb. In contrast, transections and forced use alone each resulted in only subtle dendritic alterations in the motor cortex (Adkins et al. 2002). These findings support the hypothesis that partial denervation can enhance the propensity of neurons to grow new processes in response to behavioral changes. We also found that the surface density of astrocytic processes was greater as a result of combining transections and forced use than by either independent manipulation (Bury et al. 2000b). Thus, the interactive effects of lesion-induced degeneration and behavioral change are not specific to the neurons of the brain; these effects extend to glial cells.

Altogether these findings suggest that lesion-induced degenerative events promote an enhanced responsiveness to postinjury behavioral changes. The motor cortical region under investigation has been linked to the ability to acquire new motor skills in several species, including humans, as reviewed in the preceding section. Does the enhanced neural plasticity in the motor cortex opposite the lesions mean that rats are actually better at learning new motor skills with the nonimpaired forelimb? Our data support this hypothesis (Bury and Jones 2002). To address the question further, we used a very sensitive task of unilateral forelimb motor learning in rodents, a skilled reaching task (Figure 3). When we trained rats with unilateral lesions to use their nonimpaired forelimb on the task, we found much faster rates of acquisition on the task and greater asymptotic performance levels than we were able to obtain in intact animals. Thus, a positive side effect of some lesion-induced degenerative events may be that they promote the development of behavioral changes, including those that can be used to circumvent lesion-induced impairments.

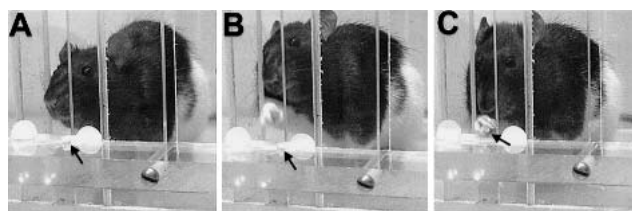


Figure 3 A rat in a unilateral skilled reaching task approaching (A), aiming (B), and retrieving (C) a small banana flavored food pellet placed in a shallow well. Incorrect aiming or grasping results in pellets that are missed, knocked from the well, or dropped. With well-handled rats, this task requires only moderate food restriction to ensure animals are not sated at the time of training and does not require food deprivation. Animals with unilateral lesions of the sensorimotor cortex are able to acquire this task faster with their nonimpaired forelimb than are intact animals. The reaching task was adapted from Peterson GM, Devine JV. 1963. Transfer on handedness in the rat resulting from small cortical lesions after limited forced practice. *J Comp Physio Psych* 56:752-756; and Miklyeva EI, Whishaw IQ. 1996. HemiParkinson analogue rats display active support in good limbs versus passive support in bad limbs on a skilled reaching task of variable height. *Behav Neurosci* 110:117-125.

Neural Plasticity and Rehabilitative Training

It has long been apparent that the experiences of an animal can influence behavioral recovery from brain injury. Exposure to complex environments before and after lesion induction in various brain regions in rats enhances functional outcome (e.g., Galani et al. 1997; Hamm et al. 1996; Held et al. 1985; Risedal et al. 2002; Whishaw et al. 1984). It has been suggested that an animal's experience in a complex environment aids behavioral compensation, or the use of alternative strategies to solve tasks, and not specifically the recovery of the lost functions (Finger and Stein 1982; Rose et al. 1987). Performance on tasks that are particularly dependent on the sensory modality most affected by the lesion (Bland and Cooper 1969) or tasks that are not influenced by learning (Rose et al. 1987, 1993) have not been found to be affected by complex environment exposure.

Given the previous findings suggesting that neocortical lesions make the motor cortex exceptionally responsive to forelimb behavioral changes, we asked whether the normal synaptic response to motor skills learning would be enhanced in the motor cortex opposite sensorimotor cortex lesions (Jones et al. 1999). We chose for this experiment a complex motor skills task, the acrobatic task (Figure 4),

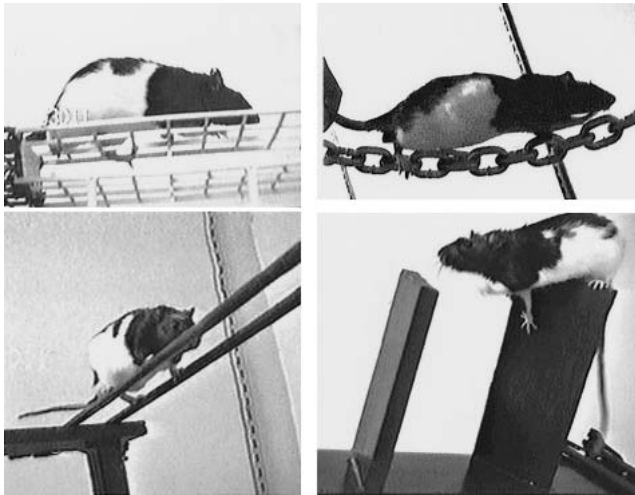


Figure 4 A subset of obstacles used in the acrobatic task. This task requires animals to learn many new coordinated and whole body movements to traverse the elevated obstacle course. Very tame male rats were guided through the task by the experimenter (typically by following the tip of a pen or finger). Exercised control animals were required to run back and forth in a simple straight alley. After unilateral lesions of the sensorimotor cortex, acrobatic training was found to improve forelimb function and increase synaptogenesis in the motor cortex opposite the lesion. The acrobatic task was adapted from Black JE, Isaacs KR, Anderson BJ, Alcantara AA, Greenough WT. 1990. Learning causes synaptogenesis, while motor activity causes angiogenesis, in cerebellar cortex of adult rats. *Proc Natl Acad Sci U S A* 87:5568-5572.

which had previously been demonstrated to result in synapse addition in the motor cortex of *intact* rats (Kleim et al. 1996). Rats with unilateral lesions or sham operations received 28 days of training on the task. As a control for handling and exercise effects, additional groups of lesion and sham-operated rats received simple locomotor exercise. Intact acrobatic rats as well as lesion-exercised controls had an approximate 21% increase in the number of synapses per neuron in the nondamaged motor cortex compared with sham-exercised rats. In rats with the combination of lesions and acrobatic training, the increase in the total number of synapses per neuron was approximately twice that of either manipulation alone. Acrobatic training also improved functional outcome on the Footfault test, a test of coordinated use of the forelimbs, improving accuracy with both the impaired and nonimpaired forelimb compared with exercised controls.

Recently, researchers have begun to investigate the effects of training focused on the impaired forelimb after unilateral cerebral injury. In rats (Castro-Alamancos and Borrell 1995), monkeys (reviewed in Nudo 1999), and humans (reviewed in Taub et al. 2002), forced training of the impaired forelimb/arm on motor tasks has been found to improve the function of this limb and to promote greater movement-associated activation in the remaining cortex of the injured hemisphere. Thus, behavioral manipulations can influence postinjury neural reorganization in the perilesion cortex. A potential issue with rehabilitative training focused on an impaired limb is that it appears to be possible to "overuse" impaired extremities. Fitting rats with vests that forced reliance on the impaired forelimb during the first week (but not later) after focal unilateral sensorimotor cortex lesions worsened the function of the impaired limb and increased the size of the lesion (Humm et al. 1998; Kozlowski et al. 1996; see also Farrell et al. 2001; Risedal et al. 1999). However, many other researchers have found that less intensive behavioral manipulations (e.g., the complex environmental manipulations summarized above) can be performed without worsening outcome. We have also found that acrobatic training after sensorimotor cortex lesions is associated with a minor reduction in the loss of perilesion cortical volume (Chu and Jones 2000). Thus, the detrimental overuse effect may require a substantial amount of activity with the affected limb, such as occurs via forced use or very intensive training, and it may also require the performance of the manipulation during early postlesion time points.

Some postinjury restorative therapies have been found to be rendered effective or to be optimized by their combination with behavioral manipulations. Amphetamine treatment combined with relevant physical therapy can result in remarkable improvements in training-related function in rats, cats, and humans (reviewed in Feeney 1997; Goldstein 2000). Amphetamine administered in the absence of behavioral training does not improve functional outcome. This effect has been found after several types and loci of brain damage in animal models. The behavioral efficacy of grafts

of fetal tissue into damage regions has also been found to be dependent on postinjury behavioral experiences. Housing rats in a complex environment has been found to promote the functional efficacy of grafts following ischemic cortical damage (Mattsson et al. 1997; see also Zeng et al. 1999) and hippocampal denervating lesions (Kelche et al. 1988).

Implications for Animal Housing and Handling Procedures

Central to the research summarized in the preceding sections is the need to detect behavioral and brain changes *sensitively* and to perform behavioral manipulations and measures that are not confounded by stress and emotional responses of the rats. With rehabilitative training, as with other therapies for brain damage, sensitive behavioral measures of outcome are a requirement to make determinations of the functional efficacy of the therapy (see Cenci et al. 2002, for a fuller treatment of this subject). Anatomical evidence of sparing of cells does not necessarily relate to significant behavioral improvements (e.g., Farrell et al. 2001). Sensitive behavioral measures and manipulations require care, housing, and handling procedures that minimize the stress and emotional responses of the animals and that preferably also promote the development and maintenance of relatively normal brains. In our laboratory, standard operating procedures for dealing with animals emphasize the need for well-handled rats that are extremely comfortable in their interactions with experimenters.

Example of Rat Handling Practices in a Behavioral Neuroscience Research Laboratory

If one were to stop by an animal room in our laboratory, they might find the following: The light levels in the room are dimmer than those in the outer hallway, and a radio is playing softly in the background. A graduate student is removing adult rats one by one from their cages and performing a test called the Footfault test. Each test lasts about 2 min. Another student is recording the data. The technician on the other side of the room is removing young rats one by one and placing them on his bent arm. He pets them for a few moments cradled against his laboratory coat. He then lifts them, gently rocks them, touches their forepaws and tickles their lower abdomens, places them on a laboratory bench and picks them up again, and repeats this sequence a few times. He is acclimating the rats to sensations they will experience during future behavioral tests and injections. In several cages, rats are tugging and shredding slips of paper that have been inserted into their cage tops.

Animal Handling and Housing Procedures Promoting Research in Neural Plasticity and Behavior

In our laboratory, we have found that an efficient way to produce very tame adult (4- to 6-mo-old) rats is by gentle

biweekly handling beginning shortly after weaning. Each episode of handling is brief, typically less than 1 min, and the production of a colony of tame rats thus requires only a minor time commitment. The rats are handled in a manner that accustoms them to the hand-holds, movements, and sensations they will experience in future behavioral tasks. Some procedures are experiment specific. For example, when we wished to have rats follow the tip of a pen through an obstacle course (Figure 4), it appeared to help to spend a bit of time playing tug-of-war using strips of paper inserted through their cage tops. Once a rat is tamed, only occasional handling appears to be necessary to maintain its tameness in adulthood. Anecdotally, it is easier to maintain tameness in animals that are socially housed. It is possible to tame old singly housed animals that have never previously been handled, but it requires much more effort.

Experimenters also, in a sense, need taming in their interactions with the rats. Experimenters who are comfortable with rats and very familiar with rat behavior are less likely to handle the animals in a manner that will cause subtle distressful reactions. We also suspect that they are more likely to obtain sensitive behavioral data and to detect potentially confounding events, including health problems and emotional reactions in the rats. In our laboratory, we have found it typically easy to produce “tame” experimenters by having them initially handle juvenile (~1 mo of age) rats and work their way up to adults.

The experiences of the rats in our laboratory are likely to be more complex than rats that are singly housed in individually vented biocontainment cages and never handled except for one or two injections. Adult rats are housed in pairs whenever possible. Exposure to auditory stimulation (other than the vocalizations of other rats) is used to reduce emotional responses to the sounds accompanying behavioral testing. (If animals are never exposed to patterned noise that is more complex than the sounds produced by ventilation, they may also fail to develop normal auditory systems.) Rats in some experiments are also given other minor variations of experience, such as paper to shred or occasional food treats. The extensive interactions with the experimenters are likely to be a major source of complexity. These practices are intended to enhance the quality of the behavioral and brain morphological variables, especially to minimize potentially confounding effects of emotionality and stress, rather than being intentional enhancements of the complexity of the experiences of rats. A movement toward considerably more complex “standard” housing might strengthen the external validity of much of the research in neural plasticity, learning, and brain damage, including our own.

Conclusion

We believe that procedures for promoting normal brains and minimizing stress during testing should be given equal priority to laboratory regulations concerning hygiene, cage

size, personal protection, and biosafety when research questions warrant such promotion. These procedures are, after all, likely to be in the best interest of quality, both of a laboratory rat's behavioral experiences and of the brain and behavioral data obtained. Behavioral manipulations and measures of functional outcome are likely to become even more common in future research on brain damage given the increasing appreciation that treatment approaches (e.g., supply of neural progenitors and pharmacological manipulations) are likely optimized in combination with rehabilitative training. Careful biocontainment and biosafety practices are essential for many lines of research. However, if these practices are applied in a manner that limits the richness of sensory experience and the development of close comfortable interactions between rodents and experimenters, they could interfere with the progress of research in neural plasticity and brain damage and ultimately result in delays for those awaiting more effective treatments for stroke and traumatic brain injury.

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