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Housing conditions influence motor functions and exploratory behavior following focal damage of the rat brain

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The present study investigated influence of housing conditions on motor functions recovery and exploratory behavior following ouabain focal brain lesion in the rat. During 30 days post-surgery period rats were housed individually in standard cages (IS) or in groups in enriched environment (EE) and behaviorally tested. The EE lesioned rats showed enhanced recovery from motor impairments in walking beam task, comparing with IS animals. Contrarily, in the open field IS rats (both lesioned and control) traveled a longer distance, showed less habituation and spent less time resting at the home base than the EE animals. Unlike the EE lesioned animals, the lesioned IS rats, presented a tendency to hyperactivity in post-injury period. Turning tendency was significantly affected by unilateral brain lesion only in the EE rats. We can conclude that housing conditions distinctly affected the rat's behavior in classical laboratory tests.

Key words: focal brain damage, functional recovery, exploratory behavior, video-tracking system

INTRODUCTION

Any focal, severe damage of brain tissue is usually followed by structure-specific functional impairments. A variety of animal models and behavioral tests were developed to examine recovery following local brain lesions as well as the effects of experimental therapies on restoration of such impaired brain function (Johansson and Ohlsson 1996, Jolkkonen et al. 2003, Biernaskie et al. 2004, Leggio et al. 2005, Urakawa et al. 2007).

To study experimental therapeutic influences on functional recovery and eventual structural repair it would be desirable to use the animal models in which relatively slight brain lesion is able to induce significant, reproducible and long-lasting functional deficits easy to quantify in adequate behavioral tests. The problem many reports elucidate however is that rapid spontaneous functional recovery is detected in most of the animal models after focal brain injury.

In our experiments we decided to use chemical lesion caused by local ouabain injection. This model has been well characterized in our laboratory (Janowski et al. 2008). Ouabain is a competitive Na^+/K^+ ATPase inhibitor and after injection into the brain causes rapid cell death due to energy failure. It has been shown that at early stages the lesion induced by ouabain injection mimics changes observed following stroke (Veldhuis et al. 2003).

In this report we have manipulated with an injection site of previously established low dose of ouabain able to induce small (lacunar) lesion of deep brain structures (Janowski et al. 2008). Finally dorsolateral striatum was designed for further stereotactic injections due to localization of motor function circuits (Suvorov and Shuvaev 2004). Motor deficits were in focus of our interest because of relative feasibility for measuring dynamics of functional recovery by repeating assessments of motor tasks in various time points.

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The basal ganglia lesions are well known to influence locomotor activity in different ways so animals become hyper- or hypoactive or locomotion can remain unchanged. Thus, even if we expect that ouabain injection into striatum may induce measurable changes in distance traveled by rats in the open field, the characteristic patterns of animal exploratory behavior would be influenced also by other factors. Rodent exploratory behavior in the new arena has rather a complex structure (Tchernichovski and Golani 1995) which can not be described solely by the distance traveled. Basing our work on the existing literature, we expected that unilateral lesion with basal ganglia location might affect turning tendency of the animals with its direct influence on the open field test score. For these reasons in this study we have compared effects of housing conditions on the broad spectrum of different parameters used for estimation of the motor function impairments and the open field exploratory behavior dysfunctions evoked by ouabain injection into dorsolateral striatum in rats. The aim was to establish behavioral testing suitable to distinguish between benefits evoked by future experimental therapies in relation to spontaneous or supported by enriched environment functional recovery. The behavioral effects of an enriched environment social housing (EE) and an individual housing in standard cages (IS) has been compared. The EE conditions consisted of a large cage with various supplements encouraging physical activity, exploration and social interactions between animals. These conditions ensure rats to have opportunity for active rehabilitation for many hours every day of post-injury recovery period. The aim of IS housing design was to minimize spontaneous rehabilitation for most of the time of post-injury recovery period. To diminish strong negative effects of long-lasting social isolation on mental well-being of IS rats we allowed them to contact each other when handled during the post-surgery observation period.

METHODS

Animals and housing conditions

We used male Wistar rats about 250 g at the beginning of the experiment. The animals were kept in 12/12 h light/dark cycle. Two different housing conditions were employed. The first, called isolated standard (IS) - consists of the standard cages with one rat per cage. These rats were allowed to contact each other only when handled during the entire 30 day post-surgery observation period. The second one, an enriched environment (EE) – where animals were kept in groups of 7–8 rats per a large cage (70 × 41 × 56 cm) equipped with various supplements like platforms, ladders, branches, beams etc (Fig. 1). Four days before surgery all rats were adapted to the housing conditions and started to be handled.

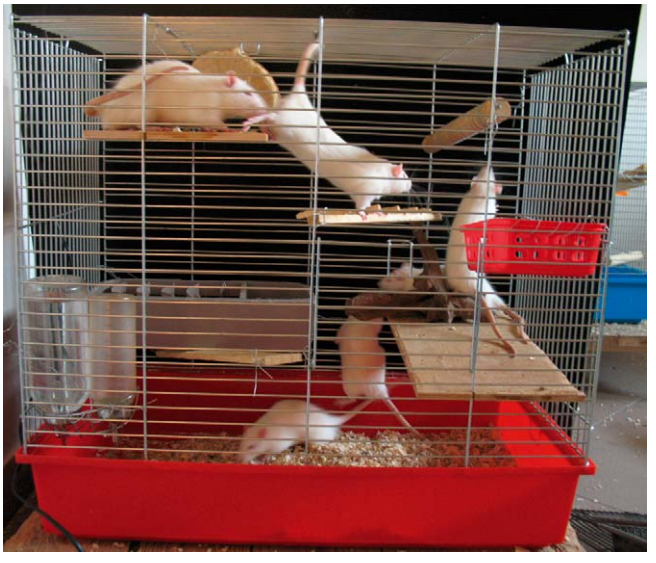

Fig. 1. Enriched environment cage

Surgery

The rats were anesthetized with 3.6% chloral hydrate (10 ml/kg b.w. given i.p.) and immobilized in a stereotactic apparatus (Stoelting). A small burr hole was drilled in the cranium over the right hemisphere. The needle (length 15 mm, gauge 33) connected with a 10 µl syringe (Hamilton) was lowered into the right striatum (coordinates A 0.5, L 3.8, V 4.7 mm). To minimize brain shift, a delay of 5 min between the needle insertion and the injection of the active substance was employed. One microliter of 5 nmole ouabain stock solution (Sigma) was then injected into the brain at a slow rate of 1 µl/min *via* a microinfusion pump (Stoelting) mounted on stereotactic apparatus. After injection, the needle was left *in situ* for 5 min to avoid leakage of the injected fluid through the needle tract. The needle was then withdrawn and the skin closed with a suture.

After the surgery lesioned rats together with controls were housed in either IS or EE conditions for 30 days observation period (lesioned IS n=8, control IS n=7, lesioned EE n=13, control EE n=8).

Table I

Point scale for evaluation of walking beam task	
7	Almost perfect walk
6	Irregular steps of both left and right limbs
5	Irregular, asymmetrical steps of contralateral limbs, including compensatory movements
4	Moderate impairment of contralateral hind limb, foot placed on the side of the beam, but close to the top
3	Significant impairment of contralateral hind limb, foot placed on the side of the beam much lower than the top, deep sleeps
2	Strong, significant impairment of both contralateral limbs
1	Able to shift along the beam, but with large difficulties
0	Unable to shift along the beam

Behavioral study

Walking beam task

To assess motor deficits caused by unilateral lesion we employed walking beam task in which rat walks along a narrow wooden beam (14 mm wide 80 cm long). Rats after short training were tested just before brain injury then at 2, 15 and 30 days following lesion the test was video recorded and then quality of the rat's gait was estimated using 8-point scale developed in our laboratory (Table I). The scale is sensitive to unilateral, consequent abnormalities in limbs movements.

Open field exploratory behavior

To examine changes in exploratory behavior 25 days following brain lesion rats were exposed individually to a new, black, circular arena of 100 cm in diameter and 30 cm high walls. The 30 min sessions were video recorded and analyzed using EthoVision video tracking system (Noldus). The xy coordinates obtained from EthoVision were analyzed using Software for the Exploration of Exploration (SEE) (Drai and Golani 2001) to obtain 37 parameters (endpoints) describing various aspects of rat's exploratory behavior.

'Szczurek' – video-tracking system developed in our laboratory

Another, simplified type of video tracking system has been developed in our laboratory and used to video-capture an animal moving freely in the arena. The system consists of a black arena intended for the use of white Wistar rats (the high contrast is required), an analogues monochrome video camera and a PC TV card.

At first a video file of the rat session in the open field is recorded and stored. A preliminary analysis starts with filtering video file with 'levels' filter to enhance contrast of white animal on the black background and saving as a bmp images sequence, using VirtualDub. Then an exact analysis takes place using the ('Szczurek') newly-developed software. Each bitmap is searched for white objects consisting of n pixels, where n is defined by the user prior to the analysis. Rectangles containing the white objects (rat) are drawn and xy coordinates of the rectangle center are fixed. This method allows the user to pinpoint the rat's body center regardless of, for example, its tail movements. The coordinates are stored in the csv. The file format allows the researcher to carry out an analysis using the SEE Workshop.

Validation of video-tracking system developed in our laboratory

The stored video files of the rat's sessions in the novel open field arena were analyzed using both: the Szczurek and the commercially available EthoVision. Then the xy coordinates were evaluated by means of the SEE Workshop (Drai and Golani 2001) and the results from all the experimental groups (lesioned IS n=4, control IS n=4, lesioned EE n=8, control EE n=5) were compared.

Statistical analysis

To find significant differences between the studied experimental groups we used non-parametric statis-

tics: Kruskal-Wallis ANOVA followed by the Whitney-Mann test. To compare the walking beam performance of the same rats at various testing time points, Friedman ANOVA and Wilcoxon match pairs test were used.

RESULTS

Recovery from motor impairments

Examined just after their undergoing brain injury, both groups of the rats (EE and IS) exhibited marked and significant impairments in the walking beam task. During the next 30 days of observation the animals partially recovered from this initial deficit. The functional restoration was much better pronounced in rats housed in the enriched environment than in those locked away in standard cages (Fig. 2). In spite of high spontaneous recovery rate after the brain injury, the EE lesioned rats still performed the walking beam task significantly worse than before surgery. Statistical significance was estimated using Friedmant ANOVA ($P<0.0001$) followed by the Wilcoxon matched pairs test for various time-points after the lesion ($P<0.01$ at 2 days, $P<0.01$ at 15 days and $P<0.05$ at 30 days). After the lesion the group of EE lesioned animals differed from the EE control significantly as far as statistics is concerned ($P<0.0001$, Kruskal-Wallis ANOVA) in all time points ($P<0.0001$ at 2 days, $P<0.0001$ at 15 days and $P<0.01$ at 30 days following brain lesion, Whitney-Mann test). Consequently, the walking beam task turned out to be very useful for evaluating the degree of impairment during the 30 days of recovery under the both housing conditions.

Exploratory behavior

We assessed the rat exploratory behavior during a 30 min session in the new open, circular arena after 25 days following the brain lesion. The 37 parameters (endpoints) evaluated by means of SEE (Drai and Golani 2001) and quantitatively describing the open field exploratory behavior were analyzed. We found some significant differences in the parameters describing the IS and EE rat behavior influenced by the housing conditions only and it being brain lesion free. However, the other endpoints turned out to be mainly affected by brain injury (Fig. 3, Table II). The parameters significantly influenced by brain damage or housing conditions can be grouped into

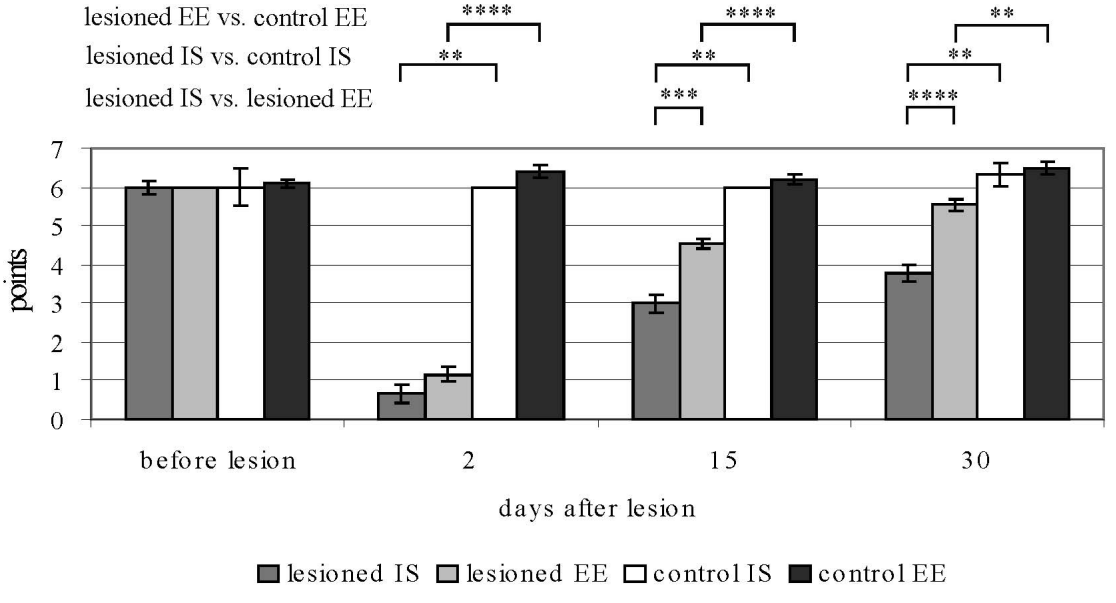

Fig. 2. Performance of walking beam task 2, 15 and 30 days after brain injury by lesioned and control rats kept singly in standard cages (IS) or socially in enriched environment (EE). Quality of stepping along the beam were quantified using 8-point scale developed in our laboratory (Table I), which is sensitive for lateralization of one side limbs abnormal movements. ** $P<0.01$; *** $P<0.001$; **** $P<0.0001$ in Whitney-Mann test.

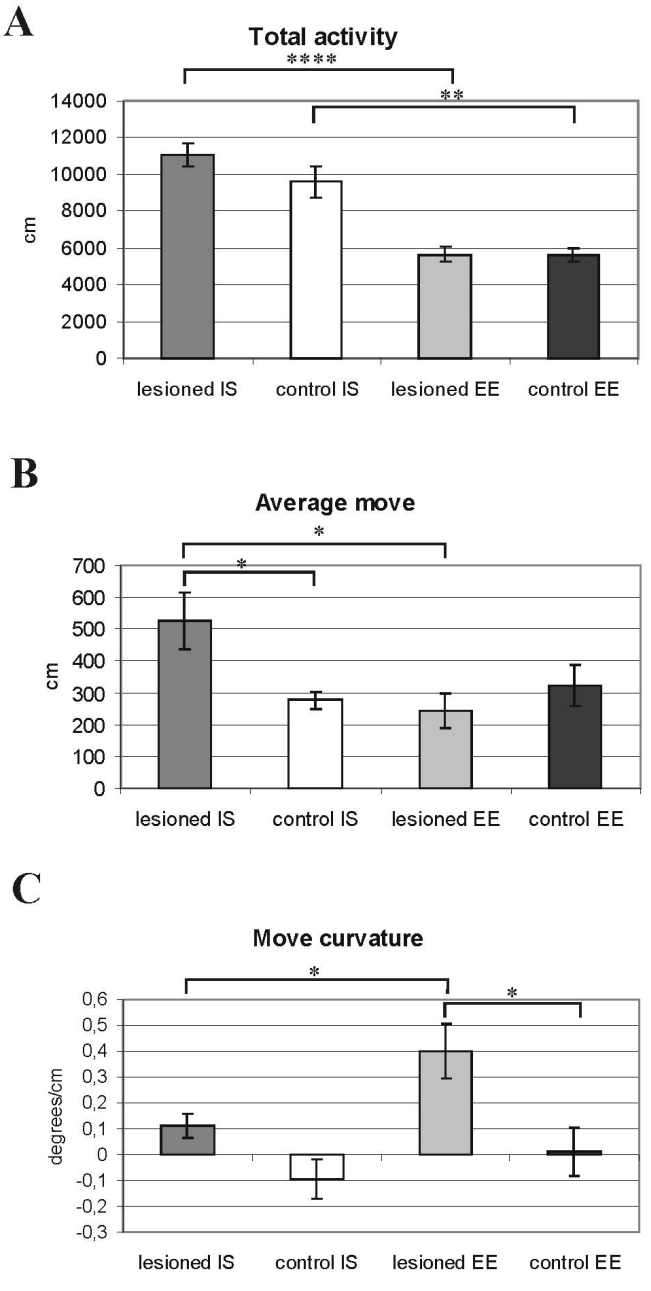

Fig. 3. Total activity during 30 min session in the new open arena as example of differences in exploratory behavior causing by housing conditions, no significant effect of the brain lesion (A). Average move length and curvature as examples of different effects of the brain lesion on open field behavior, depending on housing conditions following surgery (B and C). *P<0.05; **P<0.01; ****P<0.0001 Whitney-Mann test

those describing patterns of locomotor activity, turning tendency and home base behavior. Their definitions (italicized in text) are at the bottom of Table II.

Locomotor activity patterns

Locomotor activities expressed as the Total activity exhibited by the rats housed solely in standard cages was twice as high as those exhibited by the animals from the enriched environment. The Activity decrease parameter was also significantly higher in IS than in EE rats. However the parameters were not significantly affected by brain lesion.

The SEE software enable to automatic fragmentation of the rat path into progression and stay in place (lingering) segments using estimated velocity threshold of the animal (Drai et al. 2000). The parameters describing lingering segments – the Lingering speed and the Lingering activity – were largely influenced by the housing conditions. The parameters were also not affected by brain injury itself.

In contrast to the consistent effect of the housing condition on the open field rat behavior described above, the Stops frequency parameter was exclusively and significantly influenced by focal brain injury. The value of the parameter decreased in the IS lesioned rats whereas no significant differences were notified between the non-injured control IS and EE animals. Similarly the Average move did not show any significant influence of housing conditions on control rats. And as previously, the value of the parameter significantly increased due to the brain injury, exclusively in IS house group.

Turning tendency

The parameter Move curvature was used to determine the direction most often taken by the rat. This provided information about the balance between turns taken to the left against the ones taken to the right (note that in our experimental design left is contra-lateral side to the lesioned hemisphere). The lesioned EE rats significantly preferred the left turning giving positive values of estimated parameter. In the lesioned IS animals this endpoint was not affected significantly, however the rats showed a tendency to turn to the left more often. In control rats, kept under both the IS and EE housing conditions, the values of Move curvature were close to zero, meaning that animals did not show any turning preferences.

Table II

Parameters describing differences between open field exploratory behavior of lesioned and control rats housing in various conditions

Parameters	IS lesioned	IS control	EE lesioned	EE control	Kruskall-Wallis ANOVA *P* level	Whitney-Mann U test *P* level IS lesioned × control	EE lesioned × control	control IS × EE
				Locomotor activity patterns				
Total activity	11068 ± 591	9586 ± 819	5662 ± 372	5620 ± 344	0.0000	n. s.	n. s.	0.0022
Activity decrease	0.52 ± 0.03	0.48 ± 0.06	0.31 ± 0.03	0.26 ± 0.05	0.0005	n. s.	n. s.	0.0093
Lingering speed	1.18 ± 0.15	0.94 ± 0.07	0.56 ± 0.02	0.56 ± 0.06	0.0000	n. s.	n. s.	0.0022
Lingering activity	0.019 ± 0.005	0.031 ± 0.014	0.095 ± 0.015	0.083 ± 0.014	0.0003	n. s.	n. s.	0.0205
Average move	526 ± 88	276 ± 29	244 ± 54	323 ± 64	0.0179	0.0205	n. s.	n. s.
Stops frequency	0.0013 ± 0.0002	0.0023 ± 0.0003	0.0022 ± 0.0002	0.0024 ± 0.0003	0.0115	0.0093	n. s.	n. s.
				Turning tendency				
Move curvature	0.11 ± 0.05	-0.09 ± 0.08	0.40 ± 0.11	0.01 ± 0.9	0.0052	n. s.	0.0126	n. s.
				Home base behavior				
Diversity	20.2 ± 3.6	21.1 ± 4.2	10.4 ± 1.7	10.6 ± 4.2	0.0204	n. s.	n. s.	0.0401
Home base resting	0.70 ± 0.07	0.65 ± 0.10	0.86 ± 0.03	0.89 ± 0.06	0.0219	n. s.	n. s.	0.0289

Table contains selected parameters showing significant difference between analyzed groups. Presented parameters are endpoints (mean ± SE) obtained using SEE Workshop (Drai and Golani 2001). All showing parameters are statistically significant in Whitney-Mann test when compared lesioned IS and lesioned EE rats.
SEE Endpoints definitions:
Total activity (cm) – the activity of the rat during the entire session;
Activity decrease – the activity in the second half of the session divided by the activity in the first half of the session;
Lingering speed (s) – the total activity in 'lingering' segments divided by the total duration of 'lingering' segments;
Lingering activity – the proportion of the total activity of lingering segments, relative to the total activity of the entire session;
Average move (cm) – the median of the activity of progression segments, taken over all progression segments;
Stops frequency (segments/cm) – the number of lingering segments during the entire session, divided by the distance traveled during the entire session;
Move curvature (degree/cm) – the 'curvature' is the direction change between consecutive X,Y coordinates of the rat body center divided by the distance between those data points. This endpoint is the median of 'curvature' taken over all data points of progression segments.
Diversity (cm) – the time and space differentiation of lingering segments calculated during the entire session.
Home base resting – the proportion of time in lingering segments spent at the home base, relative to the total time of lingering segments.

Table III

Total distance traveled and activity decrease in second half of the session in experimental groups measured by Szczurek, the system developed in our laboratory, and by commercially available EthoVision (mean ± SE)

	Total activity		Activity decrease	
	Szczurek	EthoVision	Szczurek	EthoVision
lesioned IS (*n*=4)	11 945 ± 688	11 974 ± 698	0.53 ± 0.04	0.52 ± 0.05
control IS (*n*=4)	9 315 ± 506	9 411 ± 502	0.50 ± 0.08	0.51 ± 0.08
lesioned EE (*n*=8)	5 901 ± 529	5 956 ± 491	0.30 ± 0.05	0.33 ± 0.04
control EE (*n*=5)	5 782 ± 382	6 016 ± 378	0.28 ± 0.05	0.32 ± 0.05

Home base behavior

The home base is a place where the rat spends most of its time. It occasionally takes short trips and gets back to the home base again. The home base was detected automatically for each rat by the SEE software (Tchernichovski et al. 1998). The parameters like Home base resting as well as Diversity were significantly affected by housing conditions. The EE rats (both lesioned and control) spent more time at the home base and presented much less Diversity than the IS animals. There were no significant effects of lesion.

Validation of Szczurek – video-tracking system developed in our laboratory

To validate the developed method for detection of the animal body center movement which is used by our video-tracking system Szczurek, we analyzed the previously recorded video files with both: the Szczurek and EthoVison programs (IS lesioned *n*=4, IS control *n*=4, EE lesioned *n*=8, EE control *n*=5). The total distance traveled was estimated by means of the SEE Workshop using the raw data taken from both recording systems were compared. We found the Total activity values measured by Szczurek vs. EthoVision were comparable. That means that both systems are fit for estimating differences between experimental groups with comparable accuracy (Table III). We also confirmed that both the systems we compared were highly consistent with the other measured parameter, the Activity decrease (Table III).

DISCUSSION

Recovery from motor deficits

We found significant differences in recovery from motor deficits between IS and EE lesioned rats, subjected to the walking beam task between 15 and 30 days following brain injury. The long-lasting EE housing significantly enhanced the restoration of gait quality of contralateral limbs. The find is in agreement with other reports concerning the beneficial influence of enriched environment housing on recovery from brain lesion-induced impairments (Komitova et al. 2005, Hoffman et al. 2008). Similarly the control EE rats also performed the task better than the control IS animals however, using the point scale to estimate lesion induced abnormalities in movement, these differences were of little statistical significance.

Though the EE lesioned rats presented spontaneous functional recovery following the brain injury the point scale we used enabled us to detect a statistically significant difference between lesioned and control group up to 30 days following the brain damage. The discovery made it possible to compare different experimental treatments affecting the recovery in relation to housing conditions or rehabilitation intensity. The usefulness of the point scale developed for our experiments lies in its sensitivity to repeated abnormalities in either side limb stepping. In this case movement abnormalities do not only mean that one of the paws is inactive but also involves more subtle, consecutive changes in the contralateral limb stepping. This makes

it possible to discriminate between the partial compensation and functional recovery.

The enriched environment housing of intact rats was shown to improve the performance of various behavioral tasks as well as to influence directly the brain structure and neurochemistry (Benefiel et al. 2005). Exposure to EE increased levels of brain neurotrophins: nerve growth factor (NGF), brain derived neurotrophic factor (BDNF), neurofrophin 3 (NT-3) and other (Pham et al. 2002). This also up regulated the expression of immediate early genes: nerve growth factor induced gene A (NGFI-A), the activity regulated cytoskeletal protein (arc) gene (Pinaud 2004) with the promoting effect on neurogenesis in hippocampus (Segovia et al. 2006). Moreover, EE housing following brain injury increased the dendritic spine density (Johansson and Belichenko 2002) as well as extended total dendritic length in contralateral cortex (Macias 2008), enhanced expression of NGFI-A and NGFI-B, which correlated with improved performance of rotating pole test (Dahlqvist et al. 2003). A substantial diversity of the above findings as well as the existence of the other, contradictory observations (Gobbo and O'Mara 2004) suggest that the exact mechanism of enhanced recovery following focal brain damage induced by enriched environment housing remains still unclear.

Exploratory behavior

In the open field test we assessed the rat exploratory behavior in the new, empty arena. Significant influences of housing conditions on the various measured parameters were found in both the control and lesioned rats. Moreover, the brain lesion affected the open field behavior differently depending on housing conditions (Table II and Fig. 3). We carried out sophisticated statistical analysis using the SEE software (Software for Exploration of Exploration, Drai and Golani 2001) for handling the raw data obtained from video-tracking system (EthoVision, Noldus). In our experiments the SEE software enables the user to take automatic measurements of various aspects of exploratory behavior providing parameters (37 endpoints) characteristic for rodents. This software was previously used to study the effects of maternal deprivation (Shalev and Kafkafi 2002) and influence of amphetamine on the rat's exploratory behavior in the open field (Kafkafi et al. 2001) as well as to describe exploratory behavior of genetically modified mice (Kafkafi et al. 2003).

We found that the EE rats traveled a much shorter distance during the whole open field session than the IS animals. This effect can be observed in either lesioned or control groups. This may suggest that the emotional response to the new situation was much weaker in the case of the enriched environment than in the social isolation under the standard conditions. Congruent results concerning the influence of the enriched environment housing on the rat locomotor activity were reported (Van Waas and Soffié 1996, Varty et al. 2000, Elliott and Grundberg 2005).

The other parameter called Activity decrease was much less pronounced in the IS animals than in EE rats. It seems that undisturbed IS animals may display relatively weaker habituation to the new situation within 30min session. The fact that social isolation in standard cages may decrease habituation to the new environment is consistent with the results of Varty and coworkers (2000).

The rat exploration of the new open arena consists of a distinct movement forward (progression) punctuated by staying-about-place (lingering) segments in which the rat displays only slight movements such as head movements, stepping forward or backward, grooming, rearing etc. The SEE software enables the user to automatically discriminate between the progression segments and the rat lingering episodes using the estimated speed threshold value (Drai et al. 2000). We found some significant differences in parameters describing those two kinds of rat motion. The IS rats presented higher Lingering speed, but lower Lingering activity, than EE animals. The differences are likely due to a higher emotional response to the new environment in the group of the IS rats.

The IS housing conditions resulted in the lesion-induced increase of Average move with concomitant decrease of the Stops frequency parameter. The above find and the ability of the IS lesioned rats to travel longer distances combined, may suggest hyperactivity induced by the brain damage. This effect could not be observed in the EE rats. It is consistent with the results of Puurunen and the coworkers (1997) who found that the EE housing is able to reduce postischemic hyperactivity in lesioned Wistar rats.

The changes in locomotor activity pattern evoked by housing conditions described above tentatively may be explained by the differences in the emotional status of the tested rats. This would also suggest differences in the anxiety level between the experimental groups.

However in our experiments the parameters related to the wall/center behavior (one of the most common measure of animal anxiety in open field tests) did not show any significant differences.

It was often reported that rats after striatal unilateral lesion displayed spontaneous or apomorphine induced 'circling' behavior. In our experiments the rats performed apomorphine induced rotations at 1 month following deep brain structure injury (Janowski et al. 2008). We expected to see a bias in turning tendency induced here by explored dorsolateral striatal lesion. Consequently, results show that the EE lesioned rats turned in the contralateral direction more often than the non-lesioned controls. However in the group of the IS lesioned animals turning bias was much smaller and statistically insignificant. Here we used Move curvature parameter to measure the lesion-induced functional impairment, the result of the above experiment would have been hard to foresee and in contrast to many other reports in which the EE housing significantly improved recovery (Johansson and Ohlsson 1996, Puurunen et al. 1997, Dahlqvist et al. 2004, Gobbo and O'Mara 2004, Leggio et al. 2005, Urakawa et al. 2007, Hoffman et al. 2008). The explanation of this inconsistency might lead us to bring up results reported by Varty and coworkers (2000) who were able to reproduce the exact course of the rat movement forward in the open field test. They found that the rats that reared in the individual cages displayed greater willingness to move in straight line in consecutive parts of the open field session than the enriched environment reared animals. If the rat had chosen to move forward in straight line the desire to diverge either way might have flagged. But in our experiment we did not analyze thoroughly the shapes of progression segments so this hypothesis needs to be confirmed in the future.

Home base phenomenon – a place where the rat spends most of its time either staying about the place or taking short trips and returning to home base in the end (Tchernichovski et al. 1998) was the other aspect of the open field exploratory behavior to be investigated in our experiments. Home base resting successfully established within a single, time-limited session might be considered as a positive measure of the spatial working memory. The EE rats (both lesioned and controls) spent more time resting at the home base and exhibited a lower degree of time/space differentiation during the lingering episodes than the IS rats. The observation may suggest that the enriched environment must have directly improved spatial memory of the housed animals (Puurunen et al. 1997, Leggio et al. 2005).

A variety of measured aspects of animal exploratory behavior influenced by a particular experimental manipulation makes our model more suitable for testing effects of experimental therapies aimed to enhance functional recovery after the focal brain damage. Post-stroke impairments observed in human clinic are rather complex and affecting various aspects of everyday life (Dewey et al. 2007). The desired therapy intended for stroke patients should enhance the restoration of the impaired functions. That is why an animal model with a wide range of measurable impaired functions induced by the focal brain injury is so valuable in the experimental protective treatments.

Validation of Szczurek – video-tracking system developed in our laboratory

We compared the video-recordings of the same rats evaluated by Szczurek and commercially available EthoVision systems (Noldus). The average distance traveled by the rats coming from all the groups and estimated by both the video-tracking systems showed similar tendencies. The high compatibility between the two compared systems was also found in the other selected parameter, here referred to as the Activity decrease. As explained in Table II, this parameter proportionally determines the distance traveled by the animal in the second and the first half of the sessions.

The above observations allowed us to conclude that Szczurek, the video-tracking system developed in our laboratory, using a simple method to pinpoint the animal body center, may be a good alternative tool for the evaluation of locomotor activity measured as the total distance traveled by the subject animal. By using Szczurek it is also possible to obtain reliable information about the changes (decrease) in locomotor activity throughout the session.

CONCLUSIONS

We conclude that the rats housed in the enriched environment present enhanced recovery from the motor impairments following the focal brain damage compared with the animals housed solely in the standard cages. The degree of those deficits measured in

the walking beam task and evaluated by means of a developed point scale are easily noticeable and statistically significant up to 30 days of post-injury period in both housing conditions.

The open field exploratory behavior is distinctly affected by tested housing conditions. Significant differences between the IS and EE rats were found in a variety of parameters concerning locomotor activity patterns and home base behavior. The focal brain injury of the dorsolateral striatum in rat significantly affects certain parameters describing locomotor activity patterns and turning tendencies. The strong dependency of the lesion-induced effects from the housing conditions can be noticed in the majority of experimental settings.

This is very promising that we were able to find measurable functional deficits in both tested housing conditions and during relatively long observation period. It carries through comparison of the effect of a defined neuroprotective treatment in the relation to housing conditions or rehabilitation intensity.

The video-tracking system Szczurek can be recommended as a simple tool for reliable measure of distance traveled by rat. The software is available for free from the authors.

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The developed software – Szczurek – is available for free from the authors *via* e-mail (elago@autograf.pl, and_y@op.pl).

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