Pharmacology Biochemistry and Behavior, Vol. 44, pp. 565–572, 1993
Printed in the U.S.A.

0091-3057/93 $6.00 + .00

Ineffectiveness of GM1 and ORG2766 on Behavioural Recovery After Prefrontal Cortical Lesions in Adult Rats

J. M. DE BRABANDER,[1] J. P. C. DE BRUIN AND C. G. VAN EDEN

Netherlands Institute for Brain Research, Meibergdreef 33, 1105 AZ Amsterdam, The Netherlands

Received 1 April 1992

DE BRABANDER, J. M., J. P. C. DE BRUIN AND C. G. VAN EDEN. *Ineffectiveness of GM1 and ORG2766 on behavioural recovery after prefrontal cortical lesions in adult rats.* PHARMACOL BIOCHEM BEHAV 44(3) 565–572, 1993. – This study examines whether treatment with GM1 ganglioside or the corticotropin (ACTH)(4-9) analogue ORG2766 can facilitate the behavioural recovery of adult rats with medial prefrontal cortex (mPFC) lesions, as animals are impaired in their food hoarding and spatial delayed alternation performance following mPFC lesions. No ameliorating effects of GM1 treatment on performance of these behaviours were observed. Although treatment with ORG2766 somewhat improved the hoarding performance of lesioned animals, the intermediate amount of pellets hoarded was not significantly different from that of either sham-operated or vehicle-treated lesioned rats. No effect of ORG2766 treatment was observed in the spatial delayed alternation test. Further, no changes were detected in the mesocortical dopamine innervation, presumed to be involved in the neural mechanism of behavioural sparing, in response to either treatment.

Frontal cortex GM1 ganglioside ACTH(4-9) analogue ORG2766 Recovery Food hoarding Spatial delayed alternation Dopamine

DAMAGE to the CNS may be followed by compensation of the damage and even recovery of function (11,12,19, 28,29,31,32,42). Various factors are known to contribute to recovery of function, including some pharmacological compounds [for a review, see Stein and Sabel (49)]. Two such active agents are GM1 ganglioside and the corticotropin (ACTH)(4-9) analogue ORG2766. Studies carried out in several laboratories reported that administration of either GM1 or ORG2766 can exert beneficial effects after damage to the peripheral nervous system (9,16,46,54) or the CNS (17,20, 24,36,38–40,45,47,51,52,57,59,60).

It is known that damage to the prefrontal cortex (PFC) in adult animals results in impairment of several behavioural tasks (6,15,25). Such tasks may either be species typical, for example, the rat's food hoarding performance, or present across species and involve learning and memory, for example, spatial delayed alternation. However, behavioural sparing following PFC damage has been observed: Lesions inflicted on neonatal animals may be followed by complete sparing of function, as tests in adulthood have shown (2,6,14,26). Recovery has also been observed after medial PFC (mPFC) lesions in adults made serially in two stages but not when similar lesions were made in one stage (5,12,35). In previous studies, we reported that partial mPFC damage in early life results in complete functional sparing in adulthood. This sparing is accompanied by a pronounced increase in the mesocortical dopaminergic innervation in the remaining mPFC, ventral of the lesion site (7,8). Such an increased dopaminergic innervation was not observed in animals with mPFC lesions performed in adulthood. These animals showed behavioural deficits. These data on functional recovery following either neonatal mPFC damage or serial lesions in adulthood indicate that the prefrontal system has a capacity for functional compensation and that dopamine (DA) may be involved in the neural mechanism of behavioural sparing. However, for one-stage adult lesions such a capacity has not yet been reported.

In the present study, we examined whether recovery of this behaviour can be facilitated by administering either GM1 or ORG2766. Both GM1 and ORG2766 have been shown to have positive effects on neuronal repair after damage of dopaminergic systems in adulthood (18,30,43,51,52,57,59,60). Therefore, these two agents were used in the present study. Consequently, we investigated whether dopaminergic innervation increased as a result of treatment with either GM1 or ORG2766.

[1] To whom requests for reprints should be addressed.

METHOD

Animals

All 42 rats used in the present study were males of the random-bred albino Wistar strain (CPB-WU), obtained from Harlan TNO (Zeist, The Netherlands). Upon arrival, they were 7 weeks old, weighing approximately 190 g. They were housed in large Macrolon cages (75 × 55 × 23 cm) in groups of three or four. Food (standard rodent food pellets, Hope Farms, Woerden, The Netherlands) and water were available ad lib. Surgery was performed when animals were about 8 weeks old. One week later, food intake was reduced to 15 g pellets per animal per 24 h. This food deprivation schedule, continued throughout the whole testing period, is known to limit body weight of rats to approximately 85% of their free-feeding body weight. After a recovery period of 2 weeks, animals, now 10 weeks old, were tested behaviourally. Animals were kept at a 12 L : 12 D cycle (lights on from 1530–0330 h). All behavioural testing was performed during the dark period and with a radio softly playing to eliminate disturbing effects of background noises. The temperature was between 22 and 24°C and the relative humidity between 50 and 60%.

Surgery and Treatment

Each rat was randomly assigned to one of the following four groups: sham operates treated with vehicle (S, $n = 8$), lesioned rats treated with vehicle (L, $n = 11$), lesioned rats treated with GM1 ganglioside (L + GM1, $n = 12$), or lesioned rats treated with ORG2766 (L + ORG2766, $n = 11$). Because the vehicle and injection schedules were different in the GM1 and ORG2766 groups, both the S and L vehicle groups were divided into two distinct groups: They were injected either IM with the GM1 vehicle phosphate buffer (S, $n = 4$; L, $n = 6$) or SC with the ORG2766 vehicle saline (S, $n = 4$; L, $n = 5$). GM1 (Fidia Research Laboratories, Abano Terme, Italy) was dissolved in 0.01 M phosphate-buffered saline (pH 7.5) and injected 30 mg/kg IM beginning 3 days before surgery and continuing daily until day 14 postlesion. ORG2766 (Organon, Oss, The Netherlands) was injected SC, 2 μg per animal dissolved in 0.5 ml saline, beginning on the day of surgery and then seven times every other day. Surgery was performed when rats were 8 weeks old. Animals were anaesthetised with Hypnorm (0.1 ml/100 g body weight, IM; Janssen Pharmaceutica, Beerse, Belgium). The surgical procedures have been described in detail in De Brabander et al. (6). Briefly, in S, L, L + GM1, and L + ORG2766 animals the anterior sinus sagittalis superior was ligated. In L, L + GM1, and L + ORG2766 animals, the medial part of the PFC was bilaterally removed using a suction cannula. After the operation, animals were put back in their cages, each cage containing a combination of S, L, L + GM1, and L + ORG2766 animals.

Behavioural Testing

Two weeks after surgery, when administration of vehicle, GM1, or ORG2766 was finished, the behavioural testing procedure began. Animals were first tested in an open field, followed by tests for food hoarding and spatial delayed alternation. These tests were carried out in the same way as described previously in De Brabander et al. (6) and Stam et al. (48). They will, therefore, be described only briefly.

Open field. To measure locomotor and exploratory activity, rats were placed in a circular open field (diameter 82 cm) for 5 min. The following data were collected: frequency of line crossings and frequency of rearing.

Food hoarding. Food hoarding tests began the day following the open-field test and were run every other day for 4 testing days. The apparatus consisted of a home cage connected with a 50-cm long alley. At the far end of the alley, 50 normal food pellets (each weighing approximately 2 g) were placed. After a 30-min adaptation period in the home cage, animals were given the opportunity to transport the pellets from the alley to the home cage for 45 min. The following data were collected: weight and number of pellets hoarded and eaten.

Spatial delayed alternation. Following the food hoarding tests, rats (now 12 weeks old) were tested for their spatial delayed alternation performance in a T-maze. The T-maze consisted of a main alley with a start box and two side alleys. Animals were required to alternate between the side arms to get a reward (Noyes food pellet, 45 mg). After an adaptation and autoshaping period, the actual testing began. Each daily session consisted of 16 trials and sessions were run on 2 × 5 consecutive days with an intertrial interval of 0 s (ITI 0), followed by 2 × 5 consecutive days with an intertrial interval of 15 s (ITI 15). During this interval, animals were kept in a holding cage placed in line with the start box. The number of correct responses was recorded.

Anatomy and Dopamine Immunocytochemistry

After completion of behavioural testing, animals were anaesthetised with a high dose of sodium pentobarbital (0.1 ml/100 g body weight) and perfused with 5% glutaraldehyde in 0.05 M acetate buffer (pH 4.0). After perfusion, brains were removed from the skull and postfixed for 30 min. Brains were then immersed in 0.05 M Tris-buffered saline (TBS) containing 1% $Na_2S_2O_5$ (pH 7.2). Using a vibroslice, coronal serial sections (50 μm) were gathered for DA and Nissl staining. The Nissl sections were collected in TBS and the DA sections in TBS containing 1% $Na_2S_2O_5$. The DA sections were stained using an antibody against DA, raised in our institute (13), and subsequently incubated via the peroxidase-antiperoxidase (PAP) method (50), with 0.5% nickel to enhance the 3,3-diaminobenzidine (DAB; Sigma Chemical Co., St. Louis, MO) reaction. For Nissl staining, the sections were first mounted on glass slides and dried, then rinsed in a solution of ethanol and chloroform (1 : 1) to remove the lipids, thus brightening the stain (0.5% thionine).

Microphotographs were taken of cortical layers V and VI of the infralimbic area (IL; right hemisphere) of the DA sections of all animals at a standard level of 2.5 mm anterior of bregma (61). These microphotographs provided no information as to whether the section came from an S rat or one of the three L group rats. One S and one L animal were left out because the quality of the staining was suboptimal. The microphotographs were ranked according to the degree of DA innervation and given a corresponding rank number by three investigators. Rank 1 was given to the microphotograph of the section with the highest innervation. Thus, each photograph received three rank numbers; their median values were tested statistically (see the Statistics section).

Statistics

Data were analyzed using parametric tests of the SPSS^X package. For analyses of the number of line crossings (open-field test), the amount of pellets eaten (food hoarding test),

and the number of correct responses (spatial delayed alternation test), overall group effects were assessed with a multivariate analysis of variance (MANOVA) with repeated measurements. Acceptable statistical significance was set at $p \leq 0.05$. Subsequently, pair-wise group comparisons were performed again with a MANOVA with repeated measurements. Because the food hoarding scores did not fulfill the criterion for homogeneity (Box M-test), we analysed their rank order numbers instead of their hoarding scores, again with a MANOVA with repeated measurements (4).

The rearing frequency (open-field test) during the 5-min testing period was analysed with an analysis of variance (ANOVA) with group as factor. The degree of DA innervation was analysed by testing the median rank number of the microphotographs (vide supra) with an ANOVA with group as factor (4).

The criterion for considering the effects statistically significant was set at $p \leq 0.05$.

RESULTS

Location and Extent of Lesions

Using cytoarchitectonic criteria (27,53,56), the lesion location and extent of L, L + GM1, and L + ORG2766 animals were established. There was some variation in the extent of lesions; animals in which lesions involved not only the mPFC but also the genu of the corpus callosum and caudate putamen were excluded from further analysis. In all remaining animals, lesions were restricted to frontal area 2 (Fr2), dorsal anterior cingulate area (ACd), and variable parts of prelimbic (PL) and IL areas. This led to the following group sizes: L, n = 7; L + GM1, n = 8; and L + ORG2766, n = 8 animals. Reconstruction of the smallest and largest lesion of these three groups is represented in Fig. 1.

Behavioural Results

Open field. After placement in the open field, animals were actively walking around and rearing. In all four groups, the frequency of line crossings decreased in a similar fashion during the 5-min testing period (Fig. 2). No significant group difference was found. Total rearing frequency during the 5-min testing period did not differ significantly between groups.

Food hoarding. After a 30-min adaptation period in the home cage, the door to the alley was raised and animals were given the opportunity to collect food pellets for a 45-min period. On the first testing day, S animals hoarded about 16 g pellets and this amount increased steadily, to 43 g, on testing day 4 (Fig. 3). Statistical analyses revealed significant overall differences between the groups [MANOVA, $F(3, 27) = 4.12$, $p \leq 0.05$]. Hoarding performance of vehicle-treated lesioned animals over the 4 testing days was significantly impaired as compared to S animals [MANOVA, $F(1, 13) = 5.73$, $p \leq 0.05$]. Treatment with GM1 did not improve hoarding performance: L + GM1 animals hoarded amounts of pellets similar to the amounts hoarded by L animals and hoarded significantly fewer pellets than S animals [MANOVA, $F(1, 14) = 15.20$, $p \leq 0.01$]. Treatment with ORG2766 had some effect on the hoarding scores: The differences both between L + ORG2766 and L animals and between L + ORG2766 and S animals failed to be significant.

Spatial delayed alternation. In Fig. 4, the acquisition of spatial delayed alternation is depicted. In S animals, the number of correct responses during sessions with a delay of 0 s (ITI 0) increased rapidly. The task was made more difficult by lengthening the delay to 15 s (ITI 15), evident from a decrease in the percentage of correct responses on testing day 1 compared to testing day 10 of the ITI 0 sessions. After a few 15-s delay sessions, however, 80% of the responses was correct again. There were significant differences between the four groups during both ITI 0 and 15 sessions [MANOVA, $F(3, 27) = 9.20, p \leq 0.001$, and $F(3, 27) = 3.31, p \leq 0.001$, respectively]. L animals performed significantly worse than S animals during both ITI 0 [MANOVA, $F(1, 13) = 14.53$, $p \leq 0.01$] and ITI 15 [MANOVA, $F(1, 13) = 17.37$, $p \leq 0.01$]. During ITI 0 and 15, treatment with either GM1 or ORG2766 did not have any beneficial effect on the performance of L animals. Compared to S animals, both L + GM1 and L + ORG2766 animals performed significantly worse during ITI 0 [MANOVA, $F(1, 14) = 46.72$, $p \leq 0.001$, and $F(1, 14) = 15.97$, $p \leq 0.001$, respectively] and ITI 15 [MANOVA, $F(1, 14) = 20.85$, $p \leq 0.001$, and $F(1, 14) = 43.60$, $p \leq 0.001$, respectively].

Dopaminergic Innervation

In all animals, coronal sections of the frontal cortex were immunocytochemically stained for DA. In S animals, the distribution and morphology of DA fibres in the PFC was normal, as has been described previously (1,7,21,55). The DA fibre density in the PFC was considerably higher than that in other frontal cortical areas and within the mPFC the density in the basal cortical layers (V and VI) was higher than in the more superficial layers (I, II, and III). Further, a gradual decrease in density of DA fibres was observed from the ventral mPFC subareas PL/IL to the most dorsal subarea Fr2.

The dopaminergic innervation in the remaining mPFC of animals with lesions, irrespective of the kind of treatment (vehicle, GM1, or ORG2766), was comparable to the innervation in S animals (Fig. 5), except that in most animals no fibres were present in the direct vicinity of the lesion site.

Microphotographs of the basal layers (V and VI) taken from a standard level of the mPFC subarea IL of S, L, L + GM1, and L + ORG2766 animals were rated on the basis of the degree of DA innervation (Table 1). The median rank numbers obtained (see the Method section) are presented in Table 1. The means of these median rank numbers were 13.3, 15.2, 17.9, and 14.6 for S, L, L + GM1, and L + ORG2766 animals, respectively. There were no significant differences between these four groups.

DISCUSSION

The present study demonstrated that GM1 treatment of animals with mPFC lesions performed in adulthood does not result in improvement of animals' food hoarding or spatial delayed alternation behaviour. Although treatment with ORG2766 resulted in a slight improvement of food hoarding performance, hoarding scores were not significantly different from those of either sham-operated or vehicle-treated lesioned rats. Also, spatial delayed alternation performance was not improved by ORG2766 treatment. A change in locomotor or exploratory activity was not responsible for the impaired performance of animals with mPFC lesions, whether treated or not, because no differences between sham-operated and lesioned animals were observed in the open-field test. Also, the dopaminergic innervation in the remaining mPFC, known to increase considerably after neonatal mPFC lesions and a con-

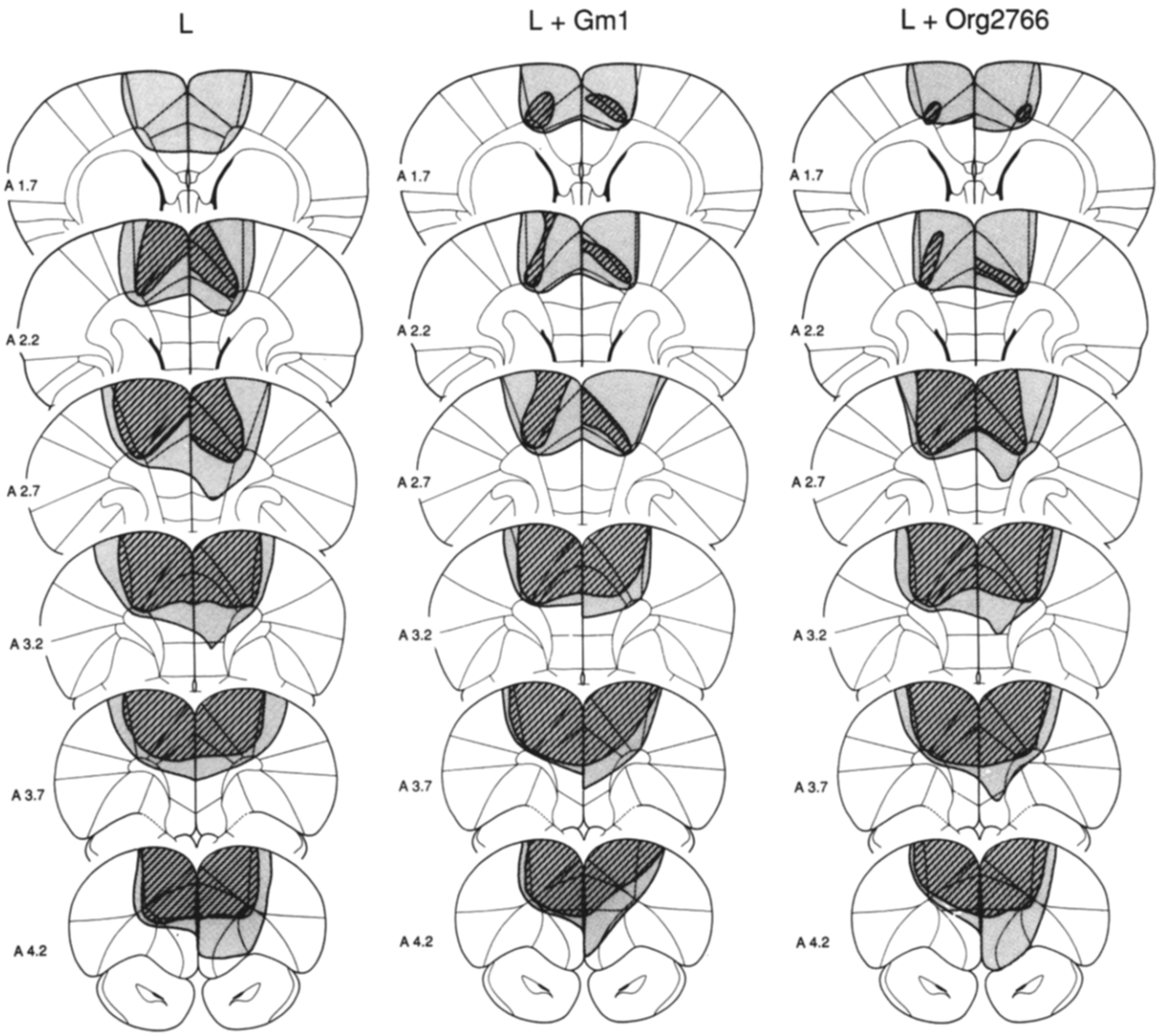

FIG. 1. Reconstruction [atlas of Paxinos and Watson (37)] of the smallest and largest medial prefrontal cortex (mPFC) lesions of animals with adult lesions treated with vehicle (L), GM1 (L + GM1), or ORG2766 (L + ORG2766). In all animals, the mPFC subareas frontal area 2, dorsal anterior cingulate area, and variable parts of the prelimbic and infralimbic areas were either removed or severely damaged.

comitant of behavioural sparing, did not change in response to mPFC lesions in adults. Neither GM1 nor ORG2766 treatment induced detectable changes in DA innervation.

Effects of GM1 or ORG2766 Treatment

Many studies have reported that after adult brain damage treatment with either GM1 or ORG2766 results in an improvement of behavioural performance (10,18,20,22,30,36,38–44,47,51,57,59,60). However, the present study and those of Butler et al. (3) and McDaniel et al. (33,34) failed to detect beneficial effects. It seems that several conditions are important for positive effects to occur in lesioned animals treated with GM1 or ORG2766, such as the treatment regimen, type of behaviour tested, and brain area damaged.

Thus, the beneficial effects on recovery of GM1 or ORG2766 may depend upon dose administered, duration and timing of treatment, and route of administration. In the literature, the dose used to achieve a beneficial effect of GM1 treatment varies between 10 and 30 mg/kg body weight (45). With respect to the duration and timing, it is known that treatment is most beneficial if started directly after surgery. Maximal recovery is achieved when animals are pretreated with GM1 (23). In an earlier study (unpublished), a dose of 20 mg/kg was used, with 1 day of pretreatment, followed by five daily injections and five injections every other day. However, no

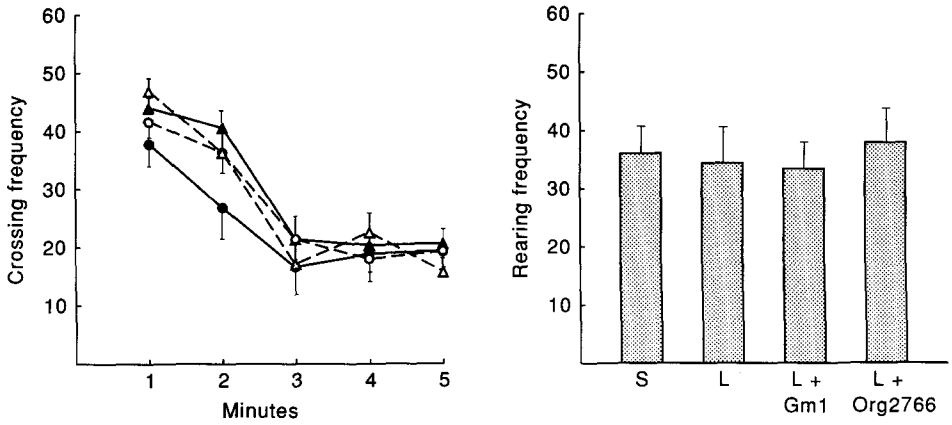

FIG. 2. Open-field activity of S (●), L (▲), L + GM1 (○), and L + ORG2766 (△) animals. Locomotor activity is plotted as the mean (SEM) frequency of line crossings per minute during the 5-min testing period. Rearing activity is depicted as mean (SEM) frequency per 5 min. There were no significant differences in locomotor or exploratory behaviours between the four groups.

improvement was observed in the animals' performance following GM1 treatment. This negative result was confirmed in the present study, although the dose of GM1 was higher (30 mg/kg), with a longer pretreatment period (3 days) and more frequent injections (daily until day 14 postlesion). The choice of these protocols was based upon the literature and made in consultation with Dr. S. Karpiak (Columbia University, New York) and Dr. G. Bonvento (Fidia Research Laboratories, Italy). When ORG2766 is administered SC, a dose of 1 or 2 μg per animal has been reported to exert beneficial effects (20,38,39,47). In the present study, we administered ORG2766 conforming to the literature and in consultation with Dr. B. Spruijt (State University Utrecht, The Netherlands). ORG2766 treatment began immediately after the mPFC lesion

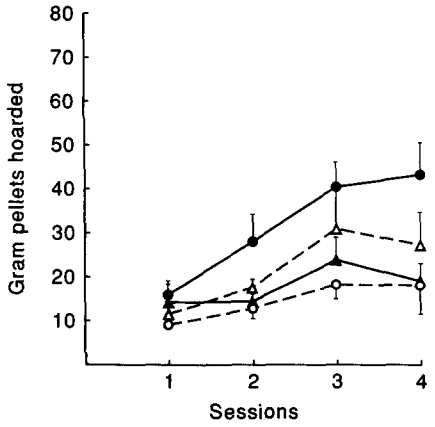

FIG. 3. Mean (SEM) food hoarding scores of S (●), L (▲), L + GM1 (○), and L + ORG2766 (△) animals during four sessions conducted every other day. L and L + GM1 animals hoarded significantly fewer pellets than S animals ($p \leq 0.05$ and $p \leq 0.01$, respectively). L + ORG2766 animals hoarded an intermediate amount of pellets; the difference in food hoarding between L + ORG2766 and L animals, as well as between L + ORG2766 and S animals, did not reach any statistical significance.

was made and ORG2766 was administered every other day until day 14 postlesion. Judged by the positive effects found in other studies, our negative findings are not likely to be explained by the injection schedule or the dose of either GM1 or ORG2766 used in the present study.

Another explanation of the ineffectiveness of GM1 or ORG2766 treatment could be the kind of task used. In the present study, animals were tested for their food hoarding ability, a species-typical task, and spatial delayed alternation behaviour, a learning task. Both GM1 and ORG2766 have been shown to exert beneficial effects on various learning tasks, including a spatial alternation task. No other studies are available in which a species-typical task was tested. Thus, it seems unlikely that in the present study the ineffectiveness of GM1 and ORG2766 treatment on spatial delayed alternation performance is due to the type of behavioural task.

Further, the brain area damaged could be of important relevance to the beneficial effects of GM1 or ORG2766 treatment. Negative results of GM1 treatment were obtained following lesions of the PFC (present study) and the visual cortex (3), and just like GM1 treatment ORG2766 treatment of animals with PFC lesions did not always result in recovery [the present study; (33,34)]. It is possible that after damage to these cortical regions it is more difficult to exert beneficial effects than after damage to other brain areas. However, this does not explain the discrepancy between the negative results of GM1 treatment in the present study and the positive ones in the study by Sabel et al. (44). Both studies investigated the effects of GM1 treatment on spatial (delayed) alternation performance in animals with medial frontal lesions. There are some differences in the procedure of the spatial alternation testing. Another noteworthy difference between the two studies is that in the present study animals already had experience with another task (food hoarding) before they were tested for their spatial (delayed) alternation performance. It has been reported, for example by Whishaw et al. (58), that postsurgical enrichment improves the performance of lesioned animals in learning tasks. Behavioural testing can be considered such an enrichment. In addition, preliminary (unpublished) data from our laboratory indicated a somewhat better spatial delayed alternation performance in mPFC-lesioned rats that had

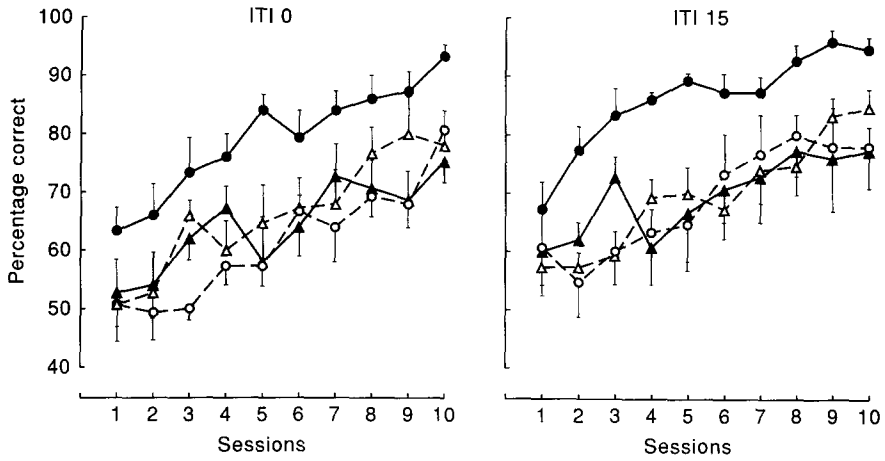

FIG. 4. Spatial delayed alternation performance of S (●), L (▲), L + GM1 (○), and L + ORG2766 (△) animals. Each session consisted of 16 trials and the mean (SEM) percentage of correct responses per session is presented. The first 10 sessions were performed with no delay between trials (ITI 0), followed by 10 sessions with a delay of 15 s. Both in sessions with ITI 0 and 15 the performance of lesioned animals was significantly impaired ($p \leq 0.01$ for L animals and $p \leq 0.001$ for L + GM1 and L + ORG2766 animals).

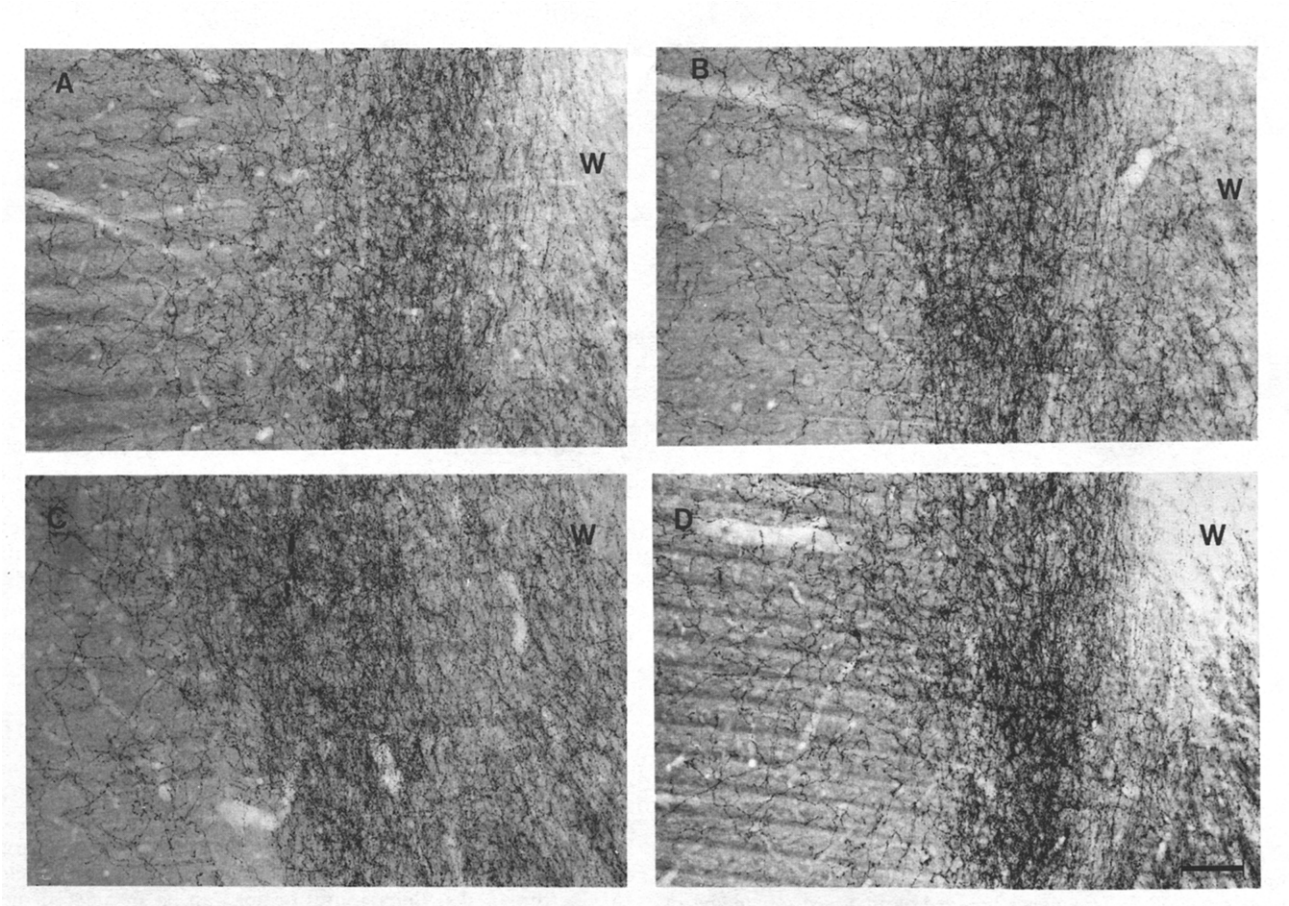

FIG. 5. Microphotographs of the dopaminergic innervation of the basal layers in the (remaining) medial prefrontal cortex subarea infralimbic area (right hemisphere) at a level of 2.5 mm anterior of bregma (61). No differences in DA innervation were detected between S, L, L + GM1, and L + ORG2766 animals. (A), S animal, median rank 16 (see Table 1); (B), L animal, median rank 13; (C), L+GM1 animal, median rank 12; (D), L + ORG2766 animal, median rank 16; (W), white matter. Bar is 0.08 mm.

TABLE 1

MEDIAN RANK NUMBERS OF THE DEGREE OF DA INNERVATION OF THE IL OF SHAM-OPERATED ANIMALS (S) AND ANIMALS WITH mPFC LESIONS TREATED WITH VEHICLE (L), GM1 (L+GM1), OR ORG2766 (L+ORG2766)

Treatment		Median Rank Number (for each animal) of the Three Investigators
S	(n = 7)	1 8 9 13 16 21 25
L	(n = 6)	2 10 11 13 26 29
L + GM1	(n = 8)	3 12 17 18 20 21 25 27
L + ORG2766	(n = 8)	4 6 11 16 19 21 22 23

Microphotographs were ranked by three investigators with the densest innervation rated as rank 1. Thus, each photograph received three rank numbers, of which the median rank number is presented. Testing these median rank numbers did not reveal any significant differences in DA innervation between the four groups. The means of these median rank numbers were 13.3, 15.2, 17.9, and 14.6 for S, L, L + GM1, and L + ORG2766 animals, respectively.

previously been tested for food hoarding. It is, therefore, possible that the spatial delayed alternation performance of lesioned animals of the present study had already improved and that GM1 (or ORG2766) could not ameliorate it any further.

It has been suggested that behavioural recovery, induced by GM1 or ORG2766 treatment following brain damage, could be the result of a reduction of the lesion-induced deterioration of cells and/or an enhancement of structural repair. Based upon previous studies, we hypothesised that following damage to the PFC the dopaminergic system is involved in the neural mechanism of behavioural recovery. In response to neonatal mPFC damage, sparing of function was observed concomitant with a large increase in dopaminergic innervation in the (remaining) mPFC, whereas there were no changes in DA innervation after mPFC lesions in adulthood. If following mPFC lesions in adult animals behavioural recovery had been induced by either GM1 or ORG2766, we would have expected an increase in the mesocortical dopaminergic innervation. However, the present study demonstrates that GM1 and ORG2766 induced neither recovery nor an increase in the dopaminergic innervation. Apparently, the repair processes resulting in functional recovery after mPFC damage in adults were not brought about by treatment with either GM1 or ORG2766, although these agents have been shown to exert positive influences on repair processes following damage to other parts of the CNS.

ACKNOWLEDGEMENTS

The authors thank Dr. G. Bonvento and Dr. A. Consolazione of Fidia Research Laboratories (Abano Terme, Italy), Dr. A. van Delft of Organon (Oss, The Netherlands), and Dr. S. Karpiak and Dr. B. Spruijt for contributions to present study and donation of GM1 ganglioside and ORG2766. Further, they thank Ellis Jansen and Alice Rinkens for technical assistance, Olga Pach for correcting the English, Gerben van der Meulen for photographs, and Henk Stoffels for drawing the figures. The comments of Harry Uylings and Dick Swaab are gratefully acknowledged.

REFERENCES

1. Berger, B.; Verney, C.; Febvret, A.; Vigny, A.; Helle, K. B. Postnatal ontogenesis of the dopaminergic innervation in the rat anterior cingulate cortex (Area 24). Immunocytochemical and catecholamine fluorescence histochemical analysis. Dev. Brain Res. 21:31–47; 1985.
2. Bowden, D. M.; Goldman, P. S.; Rosvold, H. E. Free behavior of rhesus monkeys following lesions of the dorsolateral and orbital prefrontal cortex in infancy. Exp. Brain Res. 12:265–274; 1971.
3. Butler, W. M.; Griesbach, E.; Labbe, R.; Stein, D. G. Gangliosides fail to enhance behavioral recovery after bilateral ablation of the visual cortex. J. Neurosci. Res. 17:404–409; 1987.
4. Conover, W. J.; Iman, R. L. On some alternative procedures using ranks for the analysis of experimental designs. Commun. Stat. Theor. Meth. A5(14):1349–1368; 1976.
5. Corwin, J.; Nonneman, A.; Goodlett, C. Limited sparing of function on spatial delayed alternation after two-stage lesions of prefrontal cortex in the rat. Physiol. Behav. 26:763–771; 1981.
6. De Brabander, J. M.; De Bruin, J. P. C.; Van Eden, C. G. Comparison of the effects of neonatal and adult medial prefrontal cortex lesions on food hoarding and spatial delayed alternation. Behav. Brain Res. 42:67–75; 1991.
7. De Brabander, J. M.; Van Eden, C. G.; De Bruin, J. P. C. Neuroanatomical correlates of sparing of function after neonatal medial prefrontal cortex lesions in rats. Brain Res. 568:24–34; 1991.
8. De Brabander, J. M.; Van Eden, C. G.; De Bruin, J. P. C.; Feenstra, M. G. P. Activation of mesocortical dopaminergic system in the rat in response to neonatal medial prefrontal cortex lesions. Concurrence with functional sparing. Brain Res. 581:1–9; 1992.
9. De Koning, P.; Gispen, W. H. A rationale for the use of melanocoticortins in neural injury. In: Stein, D. G.; Sabel, B. A., eds. Pharmacological approaches to the treatment of brain and spinal cord injury. New York: Plenum Press; 1988:233–258.
10. Fass, B.; Ramirez, J. J. Effects of ganglioside treatments on lesion-induced behavioral impairments and sprouting in the CNS. J. Neurosci. Res. 12:445–458; 1984.
11. Finger, S.; Almli, C. R. Brain damage and neuroplasticity: Mechanisms of recovery or development? Brain Res. Rev. 10:177–186; 1985.
12. Finger, S.; Walbran, B.; Stein, D. G. Brain damage and behavioral recovery: Serial lesion phenomena. Brain Res. 63:1–18; 1973.
13. Geffard, M.; Buijs, R. M.; Seguela, P.; Pool, C. W.; Le Moal, M. First demonstration of highly specific and sensitive antibodies against dopamine. Brain Res. 294:161–165; 1984.
14. Goldman, P. S.; Galkin, T. W. Prenatal removal of frontal association cortex in the fetal rhesus monkey: Anatomical and functional consequences in postnatal life. Brain Res. 152:451–485; 1978.
15. Goldman, P. S.; Rosvold, H. E.; Vest, B.; Galkin, T. Analysis of the delayed alternation deficit produced by dorsolateral prefrontal lesions in the rhesus monkey. J. Comp. Physiol. Psychol. 77:212–220; 1971.
16. Gorio, A. Muscle reinnervation: Motor neuron regenerative capacity. Enhancement by ganglioside treatment. Neuroscience 8: 417–430; 1983.
17. Gradowska, M.; Skup, M.; Kiedrowski, L.; Calzolari, S.; Oderfeld-Nowak, B. The effect of GM1 ganglioside on cholinergic and serotonergic systems in the rat hippocampus following partial denervation is dependent on the degree of fibre degeneration. Brain Res. 375:417–422; 1986.
18. Hadjiconstantinou, M.; Weihmuller, F. B.; Bruno, J. P.; Mariani, A. P.; Neff, N. H. Recovery of dopaminergic function following MPTP-induced neurodegeneration by exogenous GM1 ganglioside. In: Horrocks, L. A.; Neff, N. H.; Yates, A. J.; Hadjiconstantinou, M., eds. Trophic factors and the nervous system. New York: Raven Press; 1990:293–305.
19. Hicks, S. P.; D'Amato, C. J. Motor-sensory and visual behavior after hemispherectomy in newborn and mature rats. Exp. Neurol. 29:416–438; 1970.

20. Isaacson, R. L.; Poplawsky, A. An $ACTH_{4-9}$ analog (ORG 2766) speeds recovery from septal hyperemotionality in the rat. Behav. Neural Biol. 39:52–59; 1983.
21. Kalsbeek, A. P.; Voorn, P.; Buijs, R. M.; Uylings, H. B. M. Development of the dopaminergic innervation in the prefrontal cortex of the rat. J. Comp. Neurol. 269:58–72; 1988.
22. Karpiak, S. E. Ganglioside treatment improves recovery of alternation behavior after unilateral entorhinal cortex lesion. Exp. Neurol. 81:330–339; 1983.
23. Karpiak, S. E.; Mahadik, S. P. Reduction of cerebral edema with GM1 ganglioside. J. Neurosci. Res. 12:485–492; 1984.
24. Karpiak, S. E.; Mahadik, S. P. Enhanced cortical maturation: Gangliosides in CNS plasticity. In: Uylings, H. B. M.; Van Eden, C. G.; De Bruin, J. P. C.; Corner, M. A.; Feenstra, M. G. P., eds. The prefrontal cortex: Its structure, function and pathology, progress in brain research. vol. 85. Amsterdam: Elsevier; 1990: 299–309.
25. Kolb, B. Functions of the frontal cortex of the rat: A comparative review. Brain Res. Rev. 8:65–98; 1984.
26. Kolb, B.; Whishaw, I. Q. Plasticity in the neocortex: Mechanisms underlying recovery from early brain damage. Prog. Neurobiol. 32:235–276; 1989.
27. Krettek, J. E.; Price, J. L. The cortical projections of the mediodorsal nucleus and adjacent thalamic nuclei in the rat. J. Comp. Neurol. 171:157–192; 1977.
28. Land, P. W.; Lund, R. D. Development of the rats' uncrossed retinotectal pathway and its relation to plasticity studies. Science 205:698–700; 1979.
29. Laurence, S.; Stein, D. G. Recovery after brain damage and the concept of localization of function. In: Finger, S., ed. Recovery from brain damage. Research and theory. New York: Plenum Press; 1978:369–407.
30. Leon, A.; Toso, R. D.; Tresti, D.; Benvegnu, D.; Facci, L.; Krischner, G.; Tettamanti, G.; Toffano, G. Development and survival of neurons in dissociated fetal mesencephalic serum-free cultures: II. Modulatory effects of gangliosides. J. Neurosci. 8: 746–753; 1988.
31. Leong, S. K. Plasticity of cerebellar efferents after neonatal lesions in albino rats. Neurosci. Lett. 7:281–289; 1977.
32. Loesche, J.; Stewart, O. Behavioral correlates of denervation and reinnervation of the hippocampal formation of the rat: Recovery of alternation performance following unilateral entorhinal cortex lesions. Brain Res. Bull. 2:31–39; 1977.
33. McDaniel, W. F.; Davall, E. J.; Walker, P. E. ACTH(4-9) analog can retard spatial alternation learning in brain damaged and normal rats. Behav. Neural Biol. 52:271–278; 1989.
34. McDaniel, W. F.; Jones, P. D.; Weaver, T. L. Medial frontal lesions, postoperative treatment with ACTH(4-9) analog, and acquisition of a win-shift spatial strategy. Behav. Brain Res. 44: 107–112; 1991.
35. Nonneman, A. J.; Kolb, B. Functional recovery after serial ablation of prefrontal cortex in the rat. Physiol. Behav. 22:895–901; 1979.
36. Nyakas, C.; Veldhuis, H. D.; De Wied, D. Beneficial effect of chronic treatment with Org2766 and α-MSH on impaired reversal learning of rats with bilateral lesions of the parafascicular area. Brain Res. Bull. 15:257–265; 1985.
37. Paxinos, G.; Watson, C. The rat brain in stereotaxic coordinates. Sydney: Academic Press; 1982.
38. Pitsikas, N.; Spruijt, B. M.; Algeri, S.; Gispen, W. H. The ACTH/MSH (4-9) analog ORG2766 improves retrieval of information after a fimbria fornix transection. Peptides 11:911–914; 1990.
39. Pitsikas, N.; Spruijt, B. M.; Josephy, M.; Algeri, S.; Gispen, W. H. Effect of Org2766, an ACTH(4-9) analogue, on recovery after bilateral transection of the fimbria fornix in the rat. Pharmacol. Biochem. Behav. 38:931–934; 1991.
40. Poplawsky, A.; Isaacson, R. L. The GM1 ganglioside hastens the reduction of hyperemotionality after septal lesions. Behav. Neural Biol. 48:150–158; 1987.
41. Ramirez, J. J.; Fass-Holmes, B.; Karpiak, S. E.; Harshbarger, R.; Zengel, D.; Wright, P.; Valbuena, M. Enhanced recovery of learned alternation in ganglioside-treated rats after unilateral entorhinal cortex lesions. Behav. Brain Res. 43:99–101; 1991.
42. Ramirez, J. J.; Stein, D. G. Sparing and recovery of spatial alternation performance after entorhinal cortex lesions in rats. Behav. Brain Res. 13:53–61; 1984.
43. Sabel, B. A.; Dunbar, G. L.; Butler, W. M.; Stein, D. G. GM1 gangliosides stimulate neural reorganization and reduce rotational asymmetry after hemi-transections of the nigrostriatal pathway. Exp. Brain Res. 60:27–37; 1985.
44. Sabel, B. A.; Dunbar, G. L.; Stein, D. G. Gangliosides minimize behavioral deficits and enhance structural repair after brain injury. J. Neurosci. Res. 12:429–443; 1984.
45. Schengrund, C.-L. The role(s) of gangliosides in neural differentiation and repair: A perspective. Brain Res. Bull. 24:131–141; 1990.
46. Sparrow, J. R.; Grafstein, B. Sciatic nerve regeneration in ganglioside-treated rats. Exp. Neurol. 77:230–235; 1982.
47. Spruijt, B.; Pitsikas, N.; Algeri, S.; Gispen, W. H. ORG2766 improves performance of rats with unilateral lesions in the fimbria fornix in a spatial learning task. Brain Res. 527:192–197; 1990.
48. Stam, C. J.; De Bruin, J. P. C.; Van Haelst, A. M.; Van der Gugten, J.; Kalsbeek, A. Influence of the mesocortical dopaminergic system on activity, food hoarding, social-agonistic behavior and spatial delayed alternation in male rats. Behav. Neurosci. 103:24–35; 1989.
49. Stein, D. G.; Sabel, B. A. Pharmacological approaches to the treatment of brain and spinal cord injury. New York: Plenum Press; 1988.
50. Sternberger, L. A. Immunocytochemistry. Englewood Cliffs, NJ: Prentice-Hall; 1979.
51. Toffano, G.; Agnati, L. F.; Fuxe, K.; Aldinio, C.; Consolazione, A.; Valenti, G.; Savoini, G. Effect of GM1 ganglioside treatment on the recovery of dopaminergic nigrostriatal neurons after different types of lesion. Acta Physiol. Scand. 122:313–321; 1984.
52. Toffano, G.; Savoini, G. E.; Aporti, F.; Calsolari, S.; Consolazione, A.; Maura, G.; Marchi, M.; Raiteri, M.; Agnati, L. F. The functional recovery of damaged brain: The effect of GM1 monosialoganglioside. J. Neurosci. Res. 12:397–408; 1984.
53. Uylings, H. B. M.; Van Eden, C. G. Qualitative and quantitative comparison of the prefrontal cortex in rat and in primates, including humans. In: Uylings, H. B. M.; Van Eden, C. G.; De Bruin, J. P. C.; Corner, M. A.; Feenstra, M. G. P., eds. The prefrontal cortex: Its structure, function and pathology, progress in brain research. vol. 85. Amsterdam: Elsevier; 1990:31–62.
54. Van der Zee, C. E. E. M.; Brakkee, J. H.; Gispen, W. H. Putative neurotrophic factors and functional recovery from peripheral nerve damage in the rat. Br. J. Pharmacol. 103:1041–1046; 1991.
55. Van Eden, C. G.; Hoorneman, E. M. D.; Buijs, R. M.; Matthijssen, M. A. H.; Geffard, M.; Uylings, H. B. M. Immunocytochemical localization of dopamine in the prefrontal cortex of the rat at the light and electron microscopical level. Neuroscience 22: 849–862; 1987.
56. Van Eden, C. G.; Uylings, H. B. M. Cytoarchitectonical development of the prefrontal cortex in the rat. J. Comp. Neurol. 241: 253–267; 1985.
57. Vos, P. E. Post-lesion plasticity of dopaminergic systems in the rat brain; effects of the peptide ORG 2766. Doctoral dissertation, State University Utrecht, Utrecht, The Netherlands, 1991.
58. Whishaw, I. Q.; Zaborowski, J.-A.; Kolb, B. Postsurgical enrichment aids adult hemidecorticated rats on a spatial navigation task. Behav. Neural. Biol. 42:183–190; 1984.
59. Wolterink, G.; Van Ree, J. M. Functional recovery after destruction of dopamine systems in the nucleus accumbens of rats. III. Further analysis of the facilitating effect of the ACTH-(4-9) analog ORG 2766. Brain Res. 507:109–114; 1990.
60. Wolterink, G.; Van Zanten, E.; Kamsteeg, H.; Radhakishun, F. S.; Van Ree, J. M. Functional recovery after destruction of dopamine systems in the nucleus accumbens of rats. II. Facilitation by the $ACTH_{4-9}$ analog ORG2766. Brain Res. 507:92–100; 1990.
61. Zilles, K. The cortex of the rat. A stereotaxic atlas. Berlin: Springer; 1985.