

Review

# Stem Cell Transplantation for Enhancement of Learning and Memory in Adult Neurocognitive Disorders

Ben Waldau<sup>1\*</sup>

<sup>1\*</sup>Department of Neurological Surgery, University of Florida; Gainesville, FL; USA

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**ABSTRACT:** The role of adult hippocampal neurogenesis in learning and memory is still incompletely understood. Ablation of neurogenesis with different methods produced equivocal results with respect to working memory in Morris water maze and radial arm maze experiments. Therefore, it is remarkable that in the past few years several investigators have found a positive impact on working memory after adding stem or progenitor cells to the hippocampus in various disease models. The literature on stem cell transplantation for adult neurocognitive disorders is reviewed in this article and attempted to be reconciled with current research on adult hippocampal neurogenesis.

**Key words:** Neural stem cells; learning and memory; Alzheimer's disease; traumatic brain injury; stroke; aging

The exact mechanism by which adult hippocampal neurogenesis contributes to learning and memory is still unknown [1]. However, there is mounting evidence that addition of new stem or progenitor cells into the hippocampus is effective in enhancing learning and memory in various disease models. This article discusses the possible mechanism by which stem/progenitor cell transplantation may benefit learning and memory after reviewing the most recent data on adult neurogenesis and hippocampal stem cell transplantation.

## Learning and memory

Patients with bilateral hippocampal damage suffer from severe memory impairment [2]. Memory can be divided into declarative memory and non-declarative memory. Declarative memory involves the recall of facts and events [3] and may be the functional basis of language [4]. Non-declarative memory includes skills and habits, simple conditioning and priming. The hippocampus is required for declarative memory, whereas non-declarative memories do not require the hippocampus [5]. The hippocampus can encode associative memory. Temporal associative memory is

characterized by events that are associated with each other in time, but do not overlap [6], some forms of classical conditioning for example [7]. Specific neuronal unit clusters in CA1 and CA3 may increase their firing rate during classical conditioning even before a learned behavior is observed in the animal [8,9]. The increased frequency of firing results in monosynaptic augmentation of transmission, a process called long-term potentiation (LTP) [10]. Spatial associative memory is based on stable visual cues distributed in the surrounding environment which lead to the formation of a cognitive map [11]. Hippocampal single units, or place cells, fire when an animal is in a particular place [12].

## Neurocognitive tests of learning and memory in animal models

Hippocampal-dependent learning and memory can be tested in various experiments such as water maze [13], radial arm maze [14], conditional learning tasks [6], trace classical conditioning tasks [15,16] or contextual conditioning tasks [17, 18]. A short overview is given over the tests that are currently most commonly used

to evaluate hippocampal function in learning and memory after stem/progenitor cell transplantation.

Spatial associative memory is tested by mazes which come in all shapes and sizes [19]. They range from featureless arenas like the water maze to predetermined pathways such as the radial-arm maze. The most frequently used test is the Morris water maze [13]. Rats in the water maze use extra-maze cues find a hidden platform [20]. The navigational behavior is guided by learning the spatial relationships among a constellation of stimuli in an environment rather than by remembering one specific cue [21]. Rats need to be released from different locations in each trial in order to test for hippocampal-dependent learning [22]. The water maze environment may induce stress in tested animals which interferes with their learning and memory [23].

The radial arm maze (RAM) can be used to test short-term and long-term memory. In Jarrad's procedure, a subset of arms (4/8) is rewarded. Re-entry of the animal into arms that are never rewarded serves as a measure of long-term memory, whereas arms revisited in the same test indicate a deficit in short-term or working memory [24].

Different mazes may measure different subsets of hippocampal-dependent learning. For example, rats with hippocampal CA1 loss following global ischemia may show marked impairment in the water maze, but not in the radial arm maze [25, 26]. Furthermore, animals with hippocampal damage that have deficits in spatial memory tasks may have intact storage of non-spatial information [27-29].

The novel place recognition task (NPR) has also been shown to test the function of the hippocampus [30,31]. The NPR task uses the innate tendency of rats to explore novelty. First, rats are exposed to two identical objects in specific spatial locations within a test arena. Following a retention interval, one of the objects is moved to a new spatial location and rats are placed in the test arena again. Rats that remember the previous spatial arrangement of the objects will spend more time exploring the object that has been moved to the novel spatial location [32].

In contextual fear conditioning, a form of classical conditioning, there is an associative learning between a chamber and electrical footshocks that take place in that chamber. Temporal associative memory is tested in this task which is hippocampal-dependent. Memory is measured by assessing whether the animal shows freezing behavior. The type of learning that is measured by this test is thought to be based on a trisynaptic

pathway between entorhinal cortex, dentate gyrus, CA3 and CA1 [17, 18, 33].

### **How does neurogenesis in the hippocampus affect learning and memory?**

Learning and memory has been linked to neurogenesis in the hippocampus, but the exact mechanism by which hippocampal neurogenesis contributes to learning and memory is still unknown [1]. Testing has produced equivocal results concerning the impact of hippocampal neurogenesis on spatial learning. Some studies did not show a short-term effect of adult neurogenesis on the latency in Morris water maze experiments [34-39], others point towards a long-term effect [36, 39, 40] while one study showed enhanced spatial working memory in a radial-arm maze with suppression of neurogenesis [41]. The Morris water maze may not be the ideal test to examine the cognitive effects of adult hippocampal neurogenesis since the dentate gyrus may be bypassed in an entorhinal cortex to CA1 pathway [33, 42]. Rather than affecting the latency in water maze experiments, animals do not seem to be able to implement spatially precise search strategies after ablation of neurogenesis, especially when the location of the platform is changed [43]. Contextual fear conditioning has also failed to provide researchers with a straightforward answer concerning the role of neurogenesis in learning and memory. Several investigators did find a role of neurogenesis in the successful completion of this task [34, 44, 45], while others did not [35, 36, 46, 47]. Instead of directly storing information, hippocampal neurogenesis may be necessary for reorganization of memory and the transfer of information to non-hippocampal regions [48].

### **Neurocognitive disorders with a predominant effect on learning and memory**

Besides normal aging, many diseases can lead to a decline in learning and memory such as Alzheimer's disease, stroke, epilepsy or closed head injury. Many adult neurocognitive diseases have been studied with progenitor cell transplantation in recent years, a summary of which is given in the following.

### **Transplantation of stem cells for Alzheimer's disease**

Neural progenitor cells in the granule cell layer may be inhibited in their differentiation in

**Table 1: Studies investigating transplantation of stem/progenitor cells for neurocognitive enhancement in Alzheimer's disease**

Investigator	Animal model	Impaired animals transplanted with cells	Injected site	Graft	Cells injected per site	Behavioral test used	Time after injection	Result
Li et al. [60]	bilateral A $\beta$ injection into Hp	7	Hp	BMSC, BMSC-NGF, normal saline	200,000-300,000, 2 sites	MWM	8 days	improvement with BMSC and BMSC-NGF
Lee et al. [70]	APP/PS1 double heterozygous mutant mice	15	Hp	BM-MSCs, NIH 3T3 as control	10,000, 2 sites, biweekly	MWM	Approx. 1 month after first transplantation	BM-MSCs transplanted animals perform better than control transplanted rats
Tang et al. [54]	bilateral A $\beta$ injection into Hp	6	Hp	NPCs differentiated from mouse ES cells	1,000,000, 2 sites	MWM	4 weeks and 16 weeks	Rats grafted with NPCs had improved spatial learning
Moghadam et al. [57]	Ibotenic acid lesion of NBM	6	NBM	NPCs, PNPCs, ESCs, vehicle	200,000, 1 site	MWM	4 weeks	NPCs and PNPCs enhanced learning, ESCs impaired learning and formed tumors
Wang et al. [59]	Ibotenic acid lesion of NBM	10 (NPCs), 6 (ESCs)	frontal association cortex and barrel field of S1	NPCs, ESCs	4,000-20,000, 4 sites	8-arm radial maze	8 weeks	NPCs enhanced learning, ESCs impaired learning and formed tumors
Yamasaki et al. [50]	Transgenic model of neuronal injury	8 to 13	Hp	NSCs, vehicle	100,000, 2 sites	object recognition, place recognition	1 month and 3 months	NPCs significantly enhanced memory after 3 months, but not after 1 month
Xuan et al. [55]	Fimbria-fornix lesion	8	ventricle	NSCs, NSCs with BDNF	50,000, 1 site, biweekly	Y maze	4 weeks	NSCs and BDNF injections enhanced learning and memory
Xuan et al. [56]	Fimbria-fornix lesion	8	basal forebrain	NSCs, glia	50,000, 1 site	Y maze	4 weeks	NSCs enhance learning and memory more than glia
Blurton-Jones et al. [53]	3xTg-AD mice	18	Hp	NSCs, vehicle	100,000, 2 sites	MWM, context-dependent novel object recognition	1 month	BDNF is essential for NSC-induced cognitive rescue

BMSC: Bone marrow stromal cell, NGF: nerve growth factor, NSC: neural stem cell, AD: Alzheimer's disease, ESC: embryonic stem cell, BDNF: brain derived neurotrophic factor, BM-MSCs: Bone marrow – mesenchymal stem cells, NBM: Nucleus basalis Meynert, NPC: neural precursor cell, PNPC: primed neural precursor cell, Hp: hippocampus, MWM: Morris water maze

Alzheimer's disease [49]. Recently, transplantation of neural stem cells for the treatment of Alzheimer's disease has been the focus of several investigators (Table 1). A landmark study that showed functional improvement after grafting of neural stem cells was published by LaFerla et al. in 2007 [50]. In an elegant mouse model of inducible neuronal loss, LaFerla et al. showed that transplantation of neural stem cells into the hippocampus lead to an improvement in hippocampal-dependent place recognition, but not cortical-dependent object recognition. The group harvested neural stem cells from postnatal day 14-17 mice and injected 100,000

cells into each hippocampus. A significant effect on memory was only observed 3 months and not 1 month after transplantation. The authors suggested functional engraftment of transplanted stem cells since their differentiation and integration into the host neuronal circuitry is known to take more than a month [51,52], however, electrophysiological studies were not performed. The low power of the study (8-13 animals tested in each behavioral group) in combination with a p-value approaching 5 percent ( $p=0.0429$ ), warranted confirmation of findings with a larger group of subjects.

LaFerla et al. were able to reproduce their findings of cognitive improvement after stem cell transplantation 2 years later in a mouse model of Alzheimer's disease [53]. 3xTg-AD (n=18) and age-matched non-Tg mice (n=10) were injected with 100,000 murine neural stem cells into both hippocampi, controls were injected with vehicle. Behavioral testing was performed this time only one month after transplantation with water maze and a context-dependent recognition task. Injection of neural stem cells into 3xTg-AD mice rescued the learning and memory impairments in both behavioral experiments. The neurocognitive effects did not seem to be mediated by improvements in A $\beta$  or tau pathologies which the authors showed in quantitative analyses. Instead, the expression of brain-derived neurotrophic factor (BDNF) was found to be significantly elevated in the transplanted hosts. This elevation was thought to be mediated by the transplanted stem cells which were shown to still express BDNF five weeks after transplantation. Taking the experiments one step further, the authors injected only BDNF into the hippocampi of 3xTg-AD mice and controls and found a significant impact on memory, but not learning, in their water maze experiments. Furthermore, BDNF-knockout neural stem cells did not lead to an improvement in learning and memory. Thus, the authors showed that BDNF is crucial for a functional effect of neural stem cells grafts on learning and memory after transplantation into the hippocampus.

Tang et al. confirmed findings of improved spatial learning after hippocampal injection of neural precursor cells (NPCs) derived from mouse embryonic stem cells (ESCs) in an A $\beta$ -injection model of AD [54]. Lesioned rats that were treated with NPCs performed significantly better than sham surgery rats in the Morris water maze after 4 and 16 weeks of treatment.

Xuan *et al.* transplanted rat neonatal hippocampal stem cells with or without BDNF into the lateral ventricle of rats that had undergone unilateral lesioning of the fimbria-fornix pathway, a model of Alzheimer's disease [55]. Learning and memory was assessed with the Y-maze test. Lesion-only animals and animals treated with stem cells alone showed a significantly worse learning and memory performance than control animals. However, rats that received stem cells and BDNF into the lateral ventricle after the lesion preserved learning and memory compared to control animals. The clinical performance was linked

to the number of nerve growth factor receptor (NGFR)-positive neurons identified on the side of the lesion. In a second series of similar experiments, Xuan *et al.* compared the effects of transplantation of glial cells versus neural stem cells [56]. A significant increase in NGFR-positive neurons was only found after transplantation of neural stem cells and not glial cells. Since Xuan *et al.* showed that survival of nerve growth factor responsive neurons is linked to functional preservation of learning and memory; it is likely that the observed effects were caused by the secretion of neurotrophic factors by the grafts and BDNF and not by functional integration of the transplanted neural stem cells.

Moghadam et al. lesioned the nucleus basalis of Meynert in mice to create an Alzheimer Model for transplantation of neural stem cells derived from embryonic stem cells [57]. NPCs were induced by growing undifferentiated embryonic stem cells in Dulbecco's modified Eagle Medium (DMEM) and F12 supplemented with insulin, sodium selenite, transferrin, glutamine, non-essential amino acids and fibronectin [58]. A subgroup of the neural progenitor cells (primed neural progenitor cells or PNPCs) was primed in vitro towards a neuronal cholinergic fate with retinoic acid, Interleukin-6, leukemia inhibitory factor and nerve growth factor (NGF). Rats were tested with Morris Water maze four weeks after transplantation of cells into the nucleus basalis of Meynert. As expected, lesioned rats required more time to find the hidden platform compared to sham-lesioned and normal control groups. Performances of rats treated with primed neural progenitor cells and unprimed neural progenitor cells were similar. PNPC- and NPC-transplanted rats performed significantly better than vehicle-transplanted rats on the third and fourth day of water maze without however reaching the performance level of unimpaired rats. Rats transplanted with ESCs showed the worst performance of all groups and were found to have developed tumors on histological examination. Further staining showed that a large proportion of NPCs and PNPCs had differentiated into cholinergic neurons; however, priming of neural progenitor cells did not lead to a significant increase in cholinergic differentiation in vivo.

Wang et al. also lesioned the nucleus basalis of Meynert (NBM) to create a mouse dementia model and studied the neurocognitive effects of transplanting neural progenitor cells [59]. Mouse embryonic stem cell derived neurospheres were transplanted into the

frontal association cortex and barrel field of S1 cortex in C57BL/6 mice 4 weeks after lesioning the NBM with ibotenic acid. Jarrad's radial maze task was used to evaluate the mice 8 weeks after transplantation. The mean number of errors over 18 trials was taken as a performance measure for working memory. Mice that received only ibotenic acid performed significantly worse than controls. NPC transplantation rescued the working memory of the mice whereas embryonic stem cell transplanted rats fared much worse than all the other groups. Overall, Wang et al. results were consistent with Moghadam et al. Both investigators found that NPC transplantation improved learning and memory in a NBM model of dementia, whereas transplantation of ESCs worsened it.

Li et al. studied the effects of the transplantation of NGF-gene-modified bone marrow stromal cells on neurocognitive performance in a rat model of Alzheimer disease [60]. The disease model was created by injecting beta-amyloid protein bilaterally into the hippocampus. Bone marrow stem cells (BMSCs) were genetically engineered to secrete nerve growth factor since infusion of NGF has been shown to enhance functional recovery in animal models [61-66] and is tested in clinical trials in human Alzheimer's disease patients [67]. Lesioned animals that were transplanted with BMSCs performed significantly better than control Alzheimer's disease animals during 5 days of training in Morris water maze. Rats transplanted with NGF-secreting BMSCs showed the best performance. Histological analysis of transplanted animals indicated that neuronal numbers in hippocampal subfields were similar to normal control rats. This finding is consistent with a possible neuroprotective effect of NGF against A $\beta$ .

Inhibition of neuroinflammatory activity may be another mechanism which leads to neurocognitive improvement in animal models of Alzheimer's disease. BMSCs and neural progenitor cells have been shown to reduce A $\beta$  deposition, accelerate microglial activation [68] and attenuate inflammatory reactivity [69]. Therefore, Lee et al. examined A $\beta$  deposition after transplanting 15 APP/PS1 rats with bone marrow mesenchymal stem cells (BM-MSCs) into the hippocampus [70]. BM-MSCs-treated rats had significantly less A $\beta$ -40 and -42 in the hippocampus compared to controls. Moreover, the number of microglia significantly increased after the injection of BM-MSCs, and some microglia were found to contain A $\beta$ . Proinflammatory cytokines like TNF $\alpha$  were also found to be reduced after transplantation of BM-

MSCs. Instead, microglia were found to become activated over an alternative pathway via IL-4. Mice transplanted with BM-MSCs showed significantly better performance in water maze experiments than sham-surgery APP/PS1 rats.

Other successful transplantation strategies in models of Alzheimer's disease have focused on the grafting of mature cells which are genetically engineered to secrete neurotrophic factors [71,72].

### Transplantation of stem cells for aging

Several early studies have shown that transplantation of fetal grafts to the hippocampus or other targets may increase learning and memory in aged rats [73-77]. Fetal grafts which are transplanted with fibroblast growth factor 2 (FGF-2) show an increased survival presumably by the neurotrophic effect FGF-2 on neural progenitor cells [78]. Other studies have shown an improvement of cognitive function after viral-mediated NGF gene transfection [66,79,80] or after transplantation of mature cells engineered to express neurotrophic factors [72,81]. Only few studies have been published that examine the effect of intraparenchymal stem or progenitor cell transplantation on cognition in aged animals (Table 2).

Martinez-Serrano et al. transplanted conditionally immortalized neural progenitor cells (CINPs) into the nucleus basalis magnocellularis and septum of aged rats [82]. Rats that were functionally impaired before grafting significantly improved their performance in the Morris water maze after the transplantation of CINPs that were genetically engineered to secrete NGF, but not after transplantation with CINPs alone. Cells survived during the study period of 10 weeks and stably expressed NGF. Transplantation of conditionally immortalized neural progenitors that secreted NGF significantly increased the neuronal volume at the site of transplantation reflecting the neurotrophic effect of NGF.

Hodges et al. matched two groups of equally impaired aged rats and transplanted one group with the conditionally immortal Maudsley hippocampal stem cell line 36 (MHP36) [83]. MHP36 cells are conditionally immortalized by a temperature-sensitive oncogene which causes them to stop dividing when grafted [84]. Grafted and control rats were injected with Cyclosporin A for immunosuppression. Water maze testing showed that MHP36-transplanted rats were substantially superior to their matched impaired



**Table 2: Publications examining transplantation of stem/progenitor cells for enhancement of learning and memory in aging**

Investigator	Species and age	Aged animals transplanted with therapeutic graft	Injected site	Graft	Cells injected per site	Behavioral test used	Time after injection	Result
Martinez-Serrano et al. [82]	Rats, 22 to 23 months	7 or 8	NBM, septum	CINP cells	100,000 to 2,000,000	MWM	1 week and 1 month	NGF-CINP grafted animals, but not sham and control grafted animals showed reduced escape latencies
Fernandez et al. [86]	Rats, 20 to 22 months	10	Hp, striatum	rat femur bone marrow stem cells	300	MWM	5 weeks	rats improved after transplantation into Hp but not striatum
Hodges et al. [83]	Rats, 22 months old	9	frontal cortex, Hp, striatum	MHP36	7,500	MWM	6 to 8 weeks	Impaired aged rats were divided into 2 groups of equivalent performance, the transplanted group performed significantly better
Qu et al. [85]	Rats, 24 months old	6	lateral ventricle	HNSCs	100,000	MWM	4 weeks	improvement of cognitive scores

HNSC: human neural stem cell, MWM: Morris water maze, MHP36: Maudsley hippocampal stem cell line 36, CINP: conditionally immortalized neural progenitor cells, NBM: Nucleus basalis Meynert, NGF: nerve growth factor, Hp: hippocampus.

aged controls, and learned to find the platform as rapidly as unimpaired aged rats, although they were not as efficient as young controls. Qu et al. transplanted human neural progenitor cells into the lateral ventricle of 24-months-old rats and observed improvement of cognitive scores in the Morris water maze [85]. Finally, Fernandez et al. used bone marrow stem cells as a graft in the hippocampus of rats that were 22-24 months in age [86]. Aged rats showed a significant improvement in the Morris Water maze after transplantation of the bone marrow stem cells into the hippocampus, but not after transplantation into the striatum.

### Transplantation of neural stem cells into the hippocampus for other neurodegenerative diseases

Radiation inhibits endogenous neurogenesis [87-89]. Transplants of neural precursor cells were shown in the past to fail to differentiate into neurons in the irradiated hippocampus [89]. Acharya et al.,

nonetheless, were able to show that transplantation of human embryonic stem cells (hESCs) rescued cognitive impairment caused by irradiation [32]. Irradiated animals that received hESCs did not differ from normal controls in the novel place recognition task while animals that had only received irradiation were impaired. However, comparison of cognitive performance between irradiated animals that were transplanted with hESCs and irradiation-only animals did not show a significant difference. Transplanted hESCs did differentiate into neurons and glia and could still be seen 4 months after surgery. No formation of tumors was reported in their study.

Several investigators have focused on transplantation of neural stem cells into the ischemia-lesioned hippocampus (Table 3). Mochizuki et al. tested the effect of neural stem cell transplantation on spatial learning in the Morris water maze after the induction of hemispheric strokes in rats [90]. Strokes were generated by microsphere embolization, a method known to cause severe spatial learning deficits

**Table 3: Studies investigating transplantation of stem/progenitor cells for enhancement of learning and memory in other neurocognitive diseases besides Alzheimer's disease**

Disease	Investigator	Model	Number of impaired animals received therapeutic graft	Graft	Cells injected per site	Behavioral test used	Time after transplantation	Interpretation
Stroke	Mochizuki et al. [90]	Microsphere-induced cerebral embolism	9	NPCs from fetal rats	100,000	MWM	12 days after surgery	transplanted rats showed significantly better performance when testing at 14, 21 and 28 days after surgery
Stroke	Sinden et al. [93]	Transient 4-vessel occlusion	8 (12 weeks) and 12 (20 wks)	MHP36, NPCs from mice	50,000, 2 sites bilaterally	MWM	12 weeks and 20 weeks	Transplanted rats showed significantly better performance compared to the group with ischemia and sham grafts
Stroke	Toda et al. [94]	Transient 4-vessel occlusion	10	NSCs from adult hippocampus	75,000, bilaterally	MWM	3 weeks	transplanted rats with high number of surviving cells perform better than control in last 2 days of water maze
Radiation	Acharya et al. [32]	10 Gy to head of athymic nude rats	6	human ESCs	100,000, 4 sites bilaterally	NPR	4 months	hESCs rescue cognitive deficits
Stress	Menachem-Sidon et al. [96]	long-term isolation	8 to 19	NPCs from mice with overexpression of IL-1	6,000 spheres bilaterally	MWM, fear conditioning	4-5 weeks	Transplantation rescued memory impairments
Epilepsy	Waldau et al. [100]	Kainic acid model	6	NPCs from medial ganglionic eminence	100,000, 4 sites bilaterally	MWM	2 months	no effect
excitotoxic lesion	Virley et al. [99]	NMDA lesioning of hippocampus	4	MHP36 cells, CA1 field of fetal marmosets	96,000 to 100,000, 5 sites	Simple and conditional discriminations	6 weeks (MHP36), 12 wks (fetal grafts)	grafted animals were significantly superior to lesion-only animals
excitotoxic lesion	Jeltsch et al. [97]	colchicine injections into hippocampus	12	murine-derived NSCs	2,500, 3 sites bilaterally	MWM, radial arm maze	1 months (MWM, RAM), 9 months (Hebb-Williams maze)	cognitive improvement despite no histological evidence of graft
excitotoxic lesion	Srivastava et al. [98]	kainic acid injection in CA3 subfield	not published	NPCs, olfactory-ensheathing cells	125,000, unilateral	Y-maze	12 weeks	significant recovery in learning and memory when NPCs are transplanted with olfactory-ensheathing cells

Gy: Gray, NPC: neural precursor cell, NSC: neural stem cell, MHP36: Maudsley hippocampal stem cell line 36, MWM: Morris water maze, NPR: novel place recognition, IL-1: Interleukin-1, ESC: embryonic stem cell

[91]. As in other studies, the rescue of learning and memory may have been based on the secretion of BDNF by the transplanted neural stem cells [92]. The MHP36 stem cell line which later showed functional effects in aging [83] also improved learning and memory after stroke [93]. Toda et al. found improved performance in a rat stroke model after transplantation of neural stem cells as well, however, this improvement was only seen over the last 2 days of a 10 day testing period, and ischemic rats with a low number of surviving grafted cells performed more poorly than ischemic rats with no grafts [94].

Phillips et al. were able to demonstrate that neural stem cell transplantation can also lead to improvement in learning and memory in a rat model of traumatic brain injury [95]. In a stress model of chronic isolation, intrahippocampal transplantation of transgenic neural precursor cells overexpressing Interleukin-1 receptor antagonist prevented an isolation-induced decline in memory in mice [96]. Other investigators studied the effect of intrahippocampal stem cell transplantation after excitotoxic lesions of the hippocampus and found functional improvements in learning and memory [97-99]. No significant improvement on learning and memory could be found in a rat model of epilepsy after transplantation of neural progenitor grafts derived from the medial ganglionic eminence [100]. The lack of improvement in learning and memory may have been due to differentiation of grafts from the medial ganglionic eminence into GABAergic interneurons as opposed to hippocampal pyramidal neurons.

### **Mechanism of improvement of learning and memory**

Several mechanisms could have contributed to the improvement in learning and memory after stem cell transplantation across studies. Transplanted progenitor cells could have added to the endogenous pool of dentate gyrus progenitor cells or differentiated into functioning neurons in CA1 or CA3 areas. However, this mechanism is not supported by any data so far since none of the studies looked at electrophysiological integration of the stem cells into the host circuitry. Also, it has been shown that mature cells secreting neurotrophic factors are sufficient in improving learning and memory [71,72].

The most convincing evidence points towards the secretion of neurotrophic factors as the cause of

enhanced learning and memory in the studied disease models. LaFerla et al. [53], for example, have shown in their work that BDNF-knockout stem cells did not lead to an enhancement of learning and memory in a model of Alzheimer's disease. The question thus arises whether improved learning and memory is seen because of a BDNF-mediated increase in the production of endogenous progenitor cells or by enhancement of LTPs. Indeed, BDNF has been shown to enhance endogenous neurogenesis [101,102]. Most investigators studied a possible effect on learning and memory 4 weeks or more after transplantation which would have been enough time for additional adult-born dentate granule cells to form afferent and efferent connections with the local network.

However, since recent evidence in hippocampal neurogenesis research points towards an involvement of dentate gyrus neurogenesis in remote memory rather than short-term memory, it is interesting that stem cell transplantation improved short-term memory in several studies discussed previously. This finding argues against a mechanism of improvement due to integration of the stem cells into dentate gyrus circuitry or enhancement of endogenous neurogenesis, but rather points towards a direct effect of neurotrophic factors such as BDNF which has been shown to stabilize LTPs [103].

A growing body of literature suggests that progenitor cell transplantation enhances learning and memory in various disease models. Future research is needed to show whether the mechanism of improvement in learning and memory is based on stabilization and enhancement of LTPs by neurotrophic factors. In addition, our basic understanding of the effect of endogenous neurogenesis on learning and memory needs to be advanced in order to better understand to effects of progenitor cell transplantation into the diseased hippocampus.

### **References**

- [1] Deng W, Aimone JB, Gage FH (2010). New neurons and new memories: How does adult hippocampal neurogenesis affect learning and memory? *Nat Rev Neurosci*, 11:339-350.
- [2] Scoville WB, Milner B (1957). Loss of recent memory after bilateral hippocampal lesions. *J Neurol Neurosurg Psychiatry*, 20:11-21.
- [3] Eichenbaum H (1997). Declarative memory: Insights from cognitive neurobiology. *Annu Rev Psychol*, 48:547-572.



- [4] Ullman MT (2004). Contributions of memory circuits to language: The declarative/procedural model. *Cognition*, 92:231-270.
- [5] Squire LR (1992). Memory and the hippocampus: A synthesis from findings with rats, monkeys, and humans. *Psychol Rev*, 99:195-231.
- [6] Ross RT, Orr WB, Holland PC, Berger TW (1984). Hippampectomy disrupts acquisition and retention of learned conditional responding. *Behav Neurosci*, 98:211-225.
- [7] Thompson RF, Kim JJ (1996). Memory systems in the brain and localization of a memory. *Proc Natl Acad Sci U S A*, 93:13438-13444.
- [8] Berger TW, Alger B, Thompson RF (1976). Neuronal substrate of classical conditioning in the hippocampus. *Science*, 192:483-485.
- [9] Berger TW, Thompson RF (1978). Identification of pyramidal cells as the critical elements in hippocampal neuronal plasticity during learning. *Proc Natl Acad Sci U S A*, 75:1572-1576.
- [10] Lynch G, Baudry M (1984). The biochemistry of memory: A new and specific hypothesis. *Science*, 224:1057-1063.
- [11] Tolman EC (1948). Cognitive maps in rats and men. *Psychol Rev*, 55:189-208.
- [12] O'Keefe J, Speakman A (1987). Single unit activity in the rat hippocampus during a spatial memory task. *Exp Brain Res*, 68:1-27.
- [13] Morris R (1984). Developments of a water-maze procedure for studying spatial learning in the rat. *J Neurosci Methods*, 11:47-60.
- [14] Becker JT, Walker JA, Olton DS (1980). Neuroanatomical bases of spatial memory. *Brain Res*, 200:307-320.
- [15] Moyer JR, Jr., Deyo RA, Disterhoft JF (1990). Hippampectomy disrupts trace eye-blink conditioning in rabbits. *Behav Neurosci*, 104:243-252.
- [16] Solomon PR, Vander Schaaf ER, Thompson RF, Weisz DJ (1986). Hippocampus and trace conditioning of the rabbit's classically conditioned nictitating membrane response. *Behav Neurosci*, 100:729-744.
- [17] Kim JJ, Fanselow MS (1992). Modality-specific retrograde amnesia of fear. *Science*, 256:675-677.
- [18] Phillips RG, LeDoux JE (1992). Differential contribution of amygdala and hippocampus to cued and contextual fear conditioning. *Behav Neurosci*, 106:274-285.
- [19] Hodges H (1996). Maze procedures: The radial-arm and water maze compared. *Brain Res Cogn Brain Res*, 3:167-181.
- [20] Wallenstein GV, Eichenbaum H, Hasselmo ME (1998). The hippocampus as an associator of discontinuous events. *Trends Neurosci*, 21:317-323.
- [21] Nadel L, MacDonald L (1980). Hippocampus: Cognitive map or working memory? *Behav Neural Biol*, 29:405-409.
- [22] Eichenbaum H, Stewart C, Morris RG (1990). Hippocampal representation in place learning. *J Neurosci*, 10:3531-3542.
- [23] Selden NR, Cole BJ, Everitt BJ, Robbins TW (1990). Damage to ceruleo-cortical noradrenergic projections impairs locally cued but enhances spatially cued water maze acquisition. *Behav Brain Res*, 39:29-51.
- [24] Jarrard LE (1993). On the role of the hippocampus in learning and memory in the rat. *Behav Neural Biol*, 60:9-26.
- [25] Nunn JA, LePeillet E, Netto CA, Hodges H, Gray JA, Meldrum BS (1994). Global ischaemia: Hippocampal pathology and spatial deficits in the water maze. *Behav Brain Res*, 62:41-54.
- [26] Nunn J, Hodges H (1994). Cognitive deficits induced by global cerebral ischaemia: Relationship to brain damage and reversal by transplants. *Behav Brain Res*, 65:1-31.
- [27] Aggleton JP, Hunt PR, Rawlins JN (1986). The effects of hippocampal lesions upon spatial and non-spatial tests of working memory. *Behav Brain Res*, 19:133-146.
- [28] Kelsey JE, Vargas H (1993). Medial septal lesions disrupt spatial, but not nonspatial, working memory in rats. *Behav Neurosci*, 107:565-574.
- [29] Rothblat LA, Kromer LF (1991). Object recognition memory in the rat: The role of the hippocampus. *Behav Brain Res*, 42:25-32.
- [30] Mumby DG, Gaskin S, Glenn MJ, Schramek TE, Lehmann H (2002). Hippocampal damage and exploratory preferences in rats: Memory for objects, places, and contexts. *Learn Mem*, 9:49-57.
- [31] Save E, Buhot MC, Foreman N, Thinus-Blanc C (1992). Exploratory activity and response to a spatial change in rats with hippocampal or posterior parietal cortical lesions. *Behav Brain Res*, 47:113-127.
- [32] Acharya MM, Christie LA, Lan ML, Donovan PJ, Cotman CW, Fike JR, Limoli CL (2009). Rescue of radiation-induced cognitive impairment through cranial transplantation of human embryonic stem cells. *Proc Natl Acad Sci U S A*, 106:19150-19155.
- [33] Nakashiba T, Young JZ, McHugh TJ, Buhl DL, Tonegawa S (2008). Transgenic inhibition of synaptic transmission reveals role of ca3 output in hippocampal learning. *Science*, 319:1260-1264.
- [34] Saxe MD, Battaglia F, Wang JW, Malleret G, David DJ, Monckton JE, Garcia AD, Sofroniew MV, Kandel ER, Santarelli L, Hen R, Drew MR (2006). Ablation of hippocampal neurogenesis impairs contextual fear conditioning and synaptic plasticity in the dentate gyrus. *Proc Natl Acad Sci U S A*, 103:17501-17506.

- [35] Shors TJ, Townsend DA, Zhao M, Kozorovitskiy Y, Gould E (2002). Neurogenesis may relate to some but not all types of hippocampal-dependent learning. *Hippocampus*, 12:578-584.
- [36] Deng W, Saxe MD, Gallina IS, Gage FH (2009). Adult-born hippocampal dentate granule cells undergoing maturation modulate learning and memory in the brain. *J Neurosci*, 29:13532-13542.
- [37] Jessberger S, Clark RE, Broadbent NJ, Clemenson GD, Jr., Consiglio A, Lie DC, Squire LR, Gage FH (2009). Dentate gyrus-specific knockdown of adult neurogenesis impairs spatial and object recognition memory in adult rats. *Learn Mem*, 16:147-154.
- [38] Madsen TM, Kristjansen PE, Bolwig TG, Wortwein G (2003). Arrested neuronal proliferation and impaired hippocampal function following fractionated brain irradiation in the adult rat. *Neuroscience*, 119:635-642.
- [39] Snyder JS, Hong NS, McDonald RJ, Wojtowicz JM (2005). A role for adult neurogenesis in spatial long-term memory. *Neuroscience*, 130:843-852.
- [40] Imayoshi I, Sakamoto M, Ohtsuka T, Takao K, Miyakawa T, Yamaguchi M, Mori K, Ikeda T, Itoharu S, Kageyama R (2008). Roles of continuous neurogenesis in the structural and functional integrity of the adult forebrain. *Nat Neurosci*, 11:1153-1161.
- [41] Saxe MD, Malleret G, Vronskaya S, Mendez I, Garcia AD, Sofroniew MV, Kandel ER, Hen R (2007). Paradoxical influence of hippocampal neurogenesis on working memory. *Proc Natl Acad Sci U S A*, 104:4642-4646.
- [42] Brun VH, Leutgeb S, Wu HQ, Schwarcz R, Witter MP, Moser EI, Moser MB (2008). Impaired spatial representation in ca1 after lesion of direct input from entorhinal cortex. *Neuron*, 57:290-302.
- [43] Garthe A, Behr J, Kempermann G (2009). Adult-generated hippocampal neurons allow the flexible use of spatially precise learning strategies. *PLoS One*, 4:e5464.
- [44] Warner-Schmidt JL, Madsen TM, Duman RS (2008). Electroconvulsive seizure restores neurogenesis and hippocampus-dependent fear memory after disruption by irradiation. *Eur J Neurosci*, 27:1485-1493.
- [45] Winocur G, Wojtowicz JM, Sekeres M, Snyder JS, Wang S (2006). Inhibition of neurogenesis interferes with hippocampus-dependent memory function. *Hippocampus*, 16:296-304.
- [46] Dupret D, Revest JM, Koehl M, Ichas F, De Giorgi F, Costet P, Abrous DN, Piazza PV (2008). Spatial relational memory requires hippocampal adult neurogenesis. *PLoS One*, 3:e1959.
- [47] Zhang CL, Zou Y, He W, Gage FH, Evans RM (2008). A role for adult *tlx*-positive neural stem cells in learning and behaviour. *Nature*, 451:1004-1007.
- [48] Kitamura T, Saitoh Y, Takashima N, Murayama A, Niibori Y, Ageta H, Sekiguchi M, Sugiyama H, Inokuchi K (2009). Adult neurogenesis modulates the hippocampus-dependent period of associative fear memory. *Cell*, 139:814-827.
- [49] Waldau B, Shetty AK (2008). Behavior of neural stem cells in the alzheimer brain. *Cell Mol Life Sci*, 65:2372-2384.
- [50] Yamasaki TR, Blurton-Jones M, Morrisette DA, Kitazawa M, Oddo S, LaFerla FM (2007). Neural stem cells improve memory in an inducible mouse model of neuronal loss. *J Neurosci*, 27:11925-11933.
- [51] Auerbach JM, Eiden MV, McKay RD (2000). Transplanted cns stem cells form functional synapses in vivo. *Eur J Neurosci*, 12:1696-1704.
- [52] Englund U, Bjorklund A, Victorin K, Lindvall O, Kokaia M (2002). Grafted neural stem cells develop into functional pyramidal neurons and integrate into host cortical circuitry. *Proc Natl Acad Sci U S A*, 99:17089-17094.
- [53] Blurton-Jones M, Kitazawa M, Martinez-Coria H, Castello NA, Muller FJ, Loring JF, Yamasaki TR, Poon WW, Green KN, LaFerla FM (2009). Neural stem cells improve cognition via *bdnf* in a transgenic model of alzheimer disease. *Proc Natl Acad Sci U S A*, 106:13594-13599.
- [54] Tang J, Xu H, Fan X, Li D, Rancourt D, Zhou G, Li Z, Yang L (2008). Embryonic stem cell-derived neural precursor cells improve memory dysfunction in  $\text{abeta}(1-40)$  injured rats. *Neurosci Res*, 62:86-96.
- [55] Xuan AG, Long DH, Gu HG, Yang DD, Hong LP, Leng SL (2008). *Bdnf* improves the effects of neural stem cells on the rat model of alzheimer's disease with unilateral lesion of fimbria-fornix. *Neurosci Lett*, 440:331-335.
- [56] Xuan AG, Luo M, Ji WD, Long DH (2009). Effects of engrafted neural stem cells in alzheimer's disease rats. *Neurosci Lett*, 450:167-171.
- [57] Moghadam FH, Alaie H, Karbalaie K, Tanhaei S, Nasr Esfahani MH, Baharvand H (2009). Transplantation of primed or unprimed mouse embryonic stem cell-derived neural precursor cells improves cognitive function in alzheimerian rats. *Differentiation*, 78:59-68.
- [58] Okabe S, Forsberg-Nilsson K, Spiro AC, Segal M, McKay RD (1996). Development of neuronal precursor cells and functional postmitotic neurons from embryonic stem cells in vitro. *Mech Dev*, 59:89-102.
- [59] Wang Q, Matsumoto Y, Shindo T, Miyake K, Shindo A, Kawanishi M, Kawai N, Tamiya T, Nagao S (2006). Neural stem cells transplantation in cortex in a mouse model of alzheimer's disease. *J Med Invest*, 53:61-69.
- [60] Li LY, Li JT, Wu QY, Li J, Feng ZT, Liu S, Wang TH (2008). Transplantation of *ngf*-gene-modified

- bone marrow stromal cells into a rat model of alzheimer' disease. *J Mol Neurosci*, 34:157-163.
- [61] Zou L, Yuan X, Long Y, Shine HD, Yang K (2002). Improvement of spatial learning and memory after adenovirus-mediated transfer of the nerve growth factor gene to aged rat brain. *Hum Gene Ther*, 13:2173-2184.
- [62] Tuszynski MH (2000). Intraparenchymal ngf infusions rescue degenerating cholinergic neurons. *Cell Transplant*, 9:629-636.
- [63] Tuszynski MH, Smith DE, Roberts J, McKay H, Mufson E (1998). Targeted intraparenchymal delivery of human ngf by gene transfer to the primate basal forebrain for 3 months does not accelerate beta-amyloid plaque deposition. *Exp Neurol*, 154:573-582.
- [64] Niewiadomska G, Komorowski S, Baksalerska-Pazera M (2002). Amelioration of cholinergic neurons dysfunction in aged rats depends on the continuous supply of ngf. *Neurobiol Aging*, 23:601-613.
- [65] Ruberti F, Capsoni S, Comparini A, Di Daniel E, Franzot J, Gonfloni S, Rossi G, Berardi N, Cattaneo A (2000). Phenotypic knockout of nerve growth factor in adult transgenic mice reveals severe deficits in basal forebrain cholinergic neurons, cell death in the spleen, and skeletal muscle dystrophy. *J Neurosci*, 20:2589-2601.
- [66] Klein RL, Hirko AC, Meyers CA, Grimes JR, Muzyczka N, Meyer EM (2000). Ngf gene transfer to intrinsic basal forebrain neurons increases cholinergic cell size and protects from age-related, spatial memory deficits in middle-aged rats. *Brain Res*, 875:144-151.
- [67] Tuszynski MH, Thal L, Pay M, Salmon DP, U HS, Bakay R, Patel P, Blesch A, Vahlsing HL, Ho G, Tong G, Potkin SG, Fallon J, Hansen L, Mufson EJ, Kordower JH, Gall C, Conner J (2005). A phase 1 clinical trial of nerve growth factor gene therapy for alzheimer disease. *Nat Med*, 11:551-555.
- [68] Lee JK, Jin HK, Bae JS (2009). Bone marrow-derived mesenchymal stem cells reduce brain amyloid-beta deposition and accelerate the activation of microglia in an acutely induced alzheimer's disease mouse model. *Neurosci Lett*, 450:136-141.
- [69] Ryu JK, Cho T, Wang YT, McLarnon JG (2009). Neural progenitor cells attenuate inflammatory reactivity and neuronal loss in an animal model of inflamed ad brain. *J Neuroinflammation*, 6:39.
- [70] Lee JK, Jin HK, Endo S, Schuchman EH, Carter JE, Bae JS (2010). Intracerebral transplantation of bone marrow-derived mesenchymal stem cells reduces amyloid-beta deposition and rescues memory deficits in alzheimer's disease mice by modulation of immune responses. *Stem Cells*, 28:329-343.
- [71] Liu J, Zhang Z, Li JT, Zhu YH, Zhou HL, Liu S, Wang TH (2009). Effects of nt-4 gene modified fibroblasts transplanted into ad rats. *Neurosci Lett*, 466:1-5.
- [72] Dickinson-Anson H, Winkler J, Fisher LJ, Song HJ, Poo M, Gage FH (2003). Acetylcholine-secreting cells improve age-induced memory deficits. *Mol Ther*, 8:51-61.
- [73] Moron I, Ballesteros MA, Valouskova V, Gallo M (2001). Conditioned blocking is re-established by neurotransplantation in mature rats. *Neuroreport*, 12:2297-2301.
- [74] Lee MH, Rabe A (1998). Protective effects of fetal neocortical transplants on cognitive function and neuron size in rats with congenital micrencephaly. *Behav Brain Res*, 90:147-156.
- [75] Dunnett SB, Badman F, Rogers DC, Evenden JL, Iversen SD (1988). Cholinergic grafts in the neocortex or hippocampus of aged rats: Reduction of delay-dependent deficits in the delayed non-matching to position task. *Exp Neurol*, 102:57-64.
- [76] Gage FH, Bjorklund A (1986). Cholinergic septal grafts into the hippocampal formation improve spatial learning and memory in aged rats by an atropine-sensitive mechanism. *J Neurosci*, 6:2837-2847.
- [77] Gage FH, Bjorklund A, Stenevi U, Dunnett SB, Kelly PA (1984). Intrahippocampal septal grafts ameliorate learning impairments in aged rats. *Science*, 225:533-536.
- [78] Zaman V, Shetty AK (2003). Pretreatment of donor cells with fgf-2 enhances survival of fetal hippocampal ca3 cell transplants in the chronically lesioned young adult hippocampus. *Exp Neurol*, 183:11-24.
- [79] Martinez-Serrano A, Fischer W, Soderstrom S, Ebendal T, Bjorklund A (1996). Long-term functional recovery from age-induced spatial memory impairments by nerve growth factor gene transfer to the rat basal forebrain. *Proc Natl Acad Sci U S A*, 93:6355-6360.
- [80] Martinez-Serrano A, Bjorklund A (1998). Ex vivo nerve growth factor gene transfer to the basal forebrain in presymptomatic middle-aged rats prevents the development of cholinergic neuron atrophy and cognitive impairment during aging. *Proc Natl Acad Sci U S A*, 95:1858-1863.
- [81] Chen KS, Gage FH (1995). Somatic gene transfer of ngf to the aged brain: Behavioral and morphological amelioration. *J Neurosci*, 15:2819-2825.
- [82] Martinez-Serrano A, Fischer W, Bjorklund A (1995). Reversal of age-dependent cognitive impairments and cholinergic neuron atrophy by ngf-secreting neural progenitors grafted to the basal forebrain. *Neuron*, 15:473-484.

- [83] Hodges H, Veizovic T, Bray N, French SJ, Rashid TP, Chadwick A, Patel S, Gray JA (2000). Conditionally immortal neuroepithelial stem cell grafts reverse age-associated memory impairments in rats. *Neuroscience*, 101:945-955.
- [84] Gray JA, Grigoryan G, Virley D, Patel S, Sinden JD, Hodges H (2000). Conditionally immortalized, multipotential and multifunctional neural stem cell lines as an approach to clinical transplantation. *Cell Transplant*, 9:153-168.
- [85] Qu T, Brannen CL, Kim HM, Sugaya K (2001). Human neural stem cells improve cognitive function of aged brain. *Neuroreport*, 12:1127-1132.
- [86] Fernandez CI, Alberti E, Mendoza Y, Martinez L, Collazo J, Rosillo JC, Bauza JY (2004). Motor and cognitive recovery induced by bone marrow stem cells grafted to striatum and hippocampus of impaired aged rats: Functional and therapeutic considerations. *Ann N Y Acad Sci*, 1019:48-52.
- [87] Fike JR, Rola R, Limoli CL (2007). Radiation response of neural precursor cells. *Neurosurg Clin N Am*, 18:115-127, x.
- [88] Fike JR, Rosi S, Limoli CL (2009). Neural precursor cells and central nervous system radiation sensitivity. *Semin Radiat Oncol*, 19:122-132.
- [89] Monje ML, Mizumatsu S, Fike JR, Palmer TD (2002). Irradiation induces neural precursor-cell dysfunction. *Nat Med*, 8:955-962.
- [90] Mochizuki N, Takagi N, Onozato C, Moriyama Y, Takeo S, Tanonaka K (2008). Delayed injection of neural progenitor cells improved spatial learning dysfunction after cerebral ischemia. *Biochem Biophys Res Commun*, 368:151-156.
- [91] Date I, Takagi N, Takagi K, Kago T, Matsumoto K, Nakamura T, Takeo S (2004). Hepatocyte growth factor improved learning and memory dysfunction of microsphere-embolized rats. *J Neurosci Res*, 78:442-453.
- [92] Mochizuki N, Takagi N, Kurokawa K, Onozato C, Moriyama Y, Tanonaka K, Takeo S (2008). Injection of neural progenitor cells improved learning and memory dysfunction after cerebral ischemia. *Exp Neurol*, 211:194-202.
- [93] Sinden JD, Rashid-Doubell F, Kershaw TR, Nelson A, Chadwick A, Jat PS, Noble MD, Hodges H, Gray JA (1997). Recovery of spatial learning by grafts of a conditionally immortalized hippocampal neuroepithelial cell line into the ischaemia-lesioned hippocampus. *Neuroscience*, 81:599-608.
- [94] Toda H, Takahashi J, Iwakami N, Kimura T, Hoki S, Mozumi-Kitamura K, Ono S, Hashimoto N (2001). Grafting neural stem cells improved the impaired spatial recognition in ischemic rats. *Neurosci Lett*, 316:9-12.
- [95] Philips MF, Mattiasson G, Wieloch T, Bjorklund A, Johansson BB, Tomasevic G, Martinez-Serrano A, Lenzlinger PM, Sinson G, Grady MS, McIntosh TK (2001). Neuroprotective and behavioral efficacy of nerve growth factor-transfected hippocampal progenitor cell transplants after experimental traumatic brain injury. *J Neurosurg*, 94:765-774.
- [96] Ben Menachem-Zidon O, Goshen I, Kreisel T, Ben Menahem Y, Reinhartz E, Ben Hur T, Yirmiya R (2008). Intrahippocampal transplantation of transgenic neural precursor cells overexpressing interleukin-1 receptor antagonist blocks chronic isolation-induced impairment in memory and neurogenesis. *Neuropsychopharmacology*, 33:2251-2262.
- [97] Jeltsch H, Yee J, Aloy E, Marques Pereira P, Schimchowitsch S, Grandbarbe L, Caillard S, Mohier E, Cassel JC (2003). Transplantation of neurospheres after granule cell lesions in rats: Cognitive improvements despite no long-term immunodetection of grafted cells. *Behav Brain Res*, 143:177-191.
- [98] Srivastava N, Seth K, Khanna VK, Ansari RW, Agrawal AK (2009). Long-term functional restoration by neural progenitor cell transplantation in rat model of cognitive dysfunction: Co-transplantation with olfactory ensheathing cells for neurotrophic factor support. *Int J Dev Neurosci*, 27:103-110.
- [99] Virley D, Ridley RM, Sinden JD, Kershaw TR, Harland S, Rashid T, French S, Sowinski P, Gray JA, Lantos PL, Hodges H (1999). Primary ca1 and conditionally immortal mhp36 cell grafts restore conditional discrimination learning and recall in marmosets after excitotoxic lesions of the hippocampal ca1 field. *Brain*, 122 ( Pt 12):2321-2335.
- [100] Waldau B, Hattiangady B, Kuruba R, Shetty AK (2010). Medial ganglionic eminence-derived neural stem cell grafts ease spontaneous seizures and restore gdnf expression in a rat model of chronic temporal lobe epilepsy. *Stem Cells*, 2010 Jul;28(7):1153-64.
- [101] Henry RA, Hughes SM, Connor B (2007). Aav-mediated delivery of bdnf augments neurogenesis in the normal and quinolinic acid-lesioned adult rat brain. *Eur J Neurosci*, 25:3513-3525.
- [102] Taliatz D, Stall N, Dar DE, Zangen A (2010). Knockdown of brain-derived neurotrophic factor in specific brain sites precipitates behaviors associated with depression and reduces neurogenesis. *Mol Psychiatry*, 15:80-92.
- [103] Ji Y, Lu Y, Yang F, Shen W, Tang TT, Feng L, Duan S, Lu B (2010). Acute and gradual increases in bdnf concentration elicit distinct signaling and functions in neurons. *Nat Neurosci*, 13:302-309.