



## Possible Mechanism of Placental Mesenchymal Stem Cell-Derived Neural Cell Transplantation on the Recovery of Neurogenic Bladder Function after Spinal Cord Injury

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### ABSTRACT

After spinal cord injury, more neurogenic bladder function is caused. The purpose of this article is to investigate the possible mechanism of placental mesenchymal stem cell-derived neural cell transplantation on the recovery of neurogenic bladder function after spinal cord injury. 50 SPF Wistar rats were selected to establish a spinal cord injury rat model and divided into experimental groups and in the control group, 25 animals in each group, the experimental group was transplanted with placental mesenchymal stem cell-derived nerve cells, and the urodynamics and TUNEL positive rate were compared. The results of the study showed that compared with the control group, the maximum bladder capacity and bladder compliance of the experimental group increased significantly ( $P < 0.01$ ), and the bladder basic pressure and urinary leakage pressure decreased ( $P < 0.05$ ). The values of these four items are 2.318ml, 28.892cm H<sub>2</sub>O, 46.34cm H<sub>2</sub>O, and 0.1389ml/cm H<sub>2</sub>O, respectively. It can be seen that the transplantation of neural cells derived from placental mesenchymal stem cells is of great significance for the recovery of neurogenic bladder function after spinal cord injury.

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### Introduction

Since the new century, various traumas have led to an increase in spinal cord injury (SCI), which has seriously affected the patients' daily lives and increased the family's financial burden (1).

Placental mesenchymal stem cell (MSC) nerve cell transplantation plays an important role in the subsequent treatment of neurogenic bladder function after spinal cord injury (2). This is the consensus of experts in the medical field. He injected green fluorescent-labeled MSC into the lumbar spine of spinal cord injured rats. It was observed that the animal model's movement and nerve function were restored to varying degrees, and some of the transplanted cells became nerve cells (3). Claudia can observe the regeneration of a large number of neurons and the regeneration of a large number of astrocytes after MSC injection in rats (4). Lu found that the brain-derived nerve application factor DNA was transfected into MSC through liposome transfection

enzyme, thereby improving the motor function of the lower limbs (5). Du transplanted undifferentiated MSC and observed a significant increase. Compared with the control group, the loss of axons and myelin is reduced, and scar formation is reduced. Importantly, the animal's nerve function has been improved (6). Sun transplanted MSCs into rats and found that the volume of the spinal cavity decreased and the white matter increased. Although no markers of neurons, glial cells or oligodendrocytes were detected, axons were found by electron microscopy (7). In short, the use of placental mesenchymal stem cells to prevent scar tissue hyperplasia, anti-nerve cell apoptosis, anti-inflammatory effect and secretion of nerve growth factor and other characteristics to solve the problem of spinal cord injury.

Urogenital diseases caused by the neurogenic bladder are usually serious complications in the late stage of SCI patients, and urinary tract dysfunction is caused by nerve damage to the lower urinary tract. Currently,

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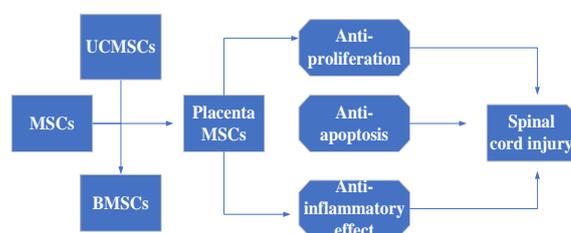
there are many studies in the medical field. A study by Pashmina showed that pelvic floor electrical stimulation therapy can effectively relieve urinary tract symptoms (8). Kumar reclassified patients with bladder dysfunction due to SCI based on the coordination of bladder detrusor, urethral sphincter and internal urethral pressure (9). Jung uses a simple self-made bladder volume measurement device to treat SCI neurogenic bladder patients and conducts bladder function training using guided intermittent catheters based on the measured bladder volume and residual urine volume (10). Tiwari's research found that the correlation between urodynamics and neuroimaging and ultrasound is unclear. Therefore, bladder management should not only rely on clinical evaluation of the bladder or separate neurological examination but should always include urodynamic examination (11). Hao used a portable ultrasound system to accurately measure the residual urine volume of patients with SCI and pointed out that two-dimensional ultrasound can be used to observe changes in the urinary system of patients with SCI, including the kidney, ureter, bladder, and prostate (12).

Sekido N has studied the loss of neurogenic bladder function in spinal cord injury (SCI) not only affects the quality of life but also places a heavy burden on patients' families and society (13).

This article mainly explores the possible mechanism of placental mesenchymal stem cell-derived neural cell transplantation. 50 SPF Wistar rats were selected to establish a spinal cord injury rat model. For each group of 25 animals, the experimental group underwent transplantation of placental mesenchymal stem cell-derived nerve cells and compared urodynamics and TUNEL positive rates. The results showed that the content of activated caspase-3 protein in the control group was  $0.439 \pm 0.091 \text{KD}$ , and the experimental group was  $0.235 \pm 0.089 \text{KD}$ . The innovation of this study is the first transplantation of neural cells derived from placental mesenchymal stem cells. Because the stem cells of the placenta are highly differentiated, it is easier to treat neurogenic bladder after spinal cord injury.

Mesenchymal stem cells are easy to isolate and obtain and have anti-scar tissue growth, anti-neuronal cell apoptosis and anti-inflammatory effects, but the functional treatment of stem cells is still unclear.

More and more studies can reduce the affected cavity and reduce the reaction of microglia/macrophages (14). Mesenchymal stem cells also secrete neurotrophic factors, such as interleukin 6, interleukin 7, and vascular endothelial growth factor. All these factors play an important role in repairing nerve damage. There are many ways to manage mesenchymal stem cells (15). For example, the antioxidant and anti-inflammatory effects are enhanced. During spinal cord injury, mesenchymal stem cells can be injected into the spinal cord using the subarachnoid space. Studies have shown that erythropoietin can accelerate this migration and repair spinal cord injury (16, 17). The function of mesenchymal stem cells and their effect on spinal cord injury are shown in Figure 1.



**Figure 1.** Classification and function of mesenchymal stem cells

Spinal cord injury is a complex pathophysiological process that can cascade (18). Two mechanisms can cause spinal cord injury. The first is the main damage. In other words, this is a direct nerve injury caused by a strong mechanical external force at the time of injury. Cell necrosis and death occur shortly after injury. The other is secondary injury (19). Many scholars believe that primary injury rarely leads to complete transection of the spinal cord, and secondary injury is the main obstacle affecting the recovery of nerve function. SCI undergoes a series of reactions, including the generation of oxygen free radicals, the proliferation of glial cells and infiltration of inflammatory cells, which significantly inhibits nerve regeneration in the damaged area (20). These processes influence and interact with each other to form a vicious circle. Currently, basic experimental and clinical treatments are used for secondary spinal cord injury (21). Some procedures can promote the regeneration of necrotic nerve cells, some treatment procedures can reduce the generation of free radicals

and inflammation, while other treatment procedures can reduce spinal cord edema reaction, increase local blood supply, minimize neuronal necrosis and promote the recovery of nerve function.

Therefore, in the current study, we tried to evaluate the possible mechanism of placental mesenchymal stem cell-derived neural cell transplantation on the recovery of neurogenic bladder function after spinal cord injury.

## Materials and Methods

### Establishment of Spinal Cord Injury Model

As shown in Table 1, fifty SPF Wistar rats (weight 196g-260g) were alternately placed in a quiet and ventilated environment for 12 hours at room temperature of 18°C-23°C and humidity of 50%-65%. Wistar rats gradually adapted to the environment. Establish a rat model of spinal cord injury, divided into an experimental group and a control group, 25 in each group; the experimental group undergoes placental mesenchymal stem cell-derived neural cell transplantation.

**Table 1.** Basic information of the subjects

Group	Temperature	Humidity	Experimental operation
Experimental group	18-23°C	50%-65%	Mesenchymal stem cell transplantation
Control group	18-23°C	50%-65%	X

Establishment of the experimental model: a longitudinal incision was made in the midline of the rat's back, the incision was about 4 cm long, and the skin and subcutaneous fascia were cut to expose the muscles in the surgical area. The key membrane extends to the position closest to the posterior midline and brings the spinous process of the thirteenth thoracic spine of the rat close to the level, while the blunt dissection of the paravertebral muscle exposes the rat spinous process, lamina and transverse process. Fix the spine with the fixing grooves and clips on both sides, and then place it under the homemade spinal cord injury strike device. Beat the affected head, and then squeeze the spinal cord for 2 minutes. After spinal cord injury, spinal cord edema and hyperemia can be seen, and the spinal cord becomes black.

### Experimental Instrument Materials

Inverted fluorescence microscope, ultra-pure water machine, electronic precision balance, ultra-clean

workbench, ultra-low temperature refrigerator, -80°C low-temperature refrigerator, -40°C refrigerator, real-time quantitative PCR instrument, basic fibroblast growth factor, anti-mouse GFAP, Nesting antibody.

### Experimental Testing Measures

Treatment within 6 hours after collecting the placenta: placenta indentation, washing the placenta and umbilical vein space with PBS solution containing double antibodies, and separating and culturing by blocking method (22). After 14 days of culture, count the number of colonies, fuse the cells to 70%-80%, and digest with a digestion solution rich in 1.25g/L trypsin-1g/L ethylenediaminetetraacetic acid (2.5-5.0) x 10<sup>3</sup>/L cm<sup>2</sup>, counted as P1 cells.

Cell transplantation was performed 9 days after spinal cord injury. After successful anesthesia with chloral hydrate, each layer of tissue was cut to isolate the exposed spinal cord injury, 10μl of cell suspension was drawn into the center of the injured area with a microsyringe, and each layer was sutured to the skin layer. During the perioperative period, it can enhance mouse care and reduce mortality (23). Cyclosporin at 10mg/kg/d was injected during transplantation. The mice were sacrificed 4 weeks after cell transplantation, and the spinal cord of the injured area was collected for dissection and examination. After inducing transplantation, human placental mesenchymal stem cells induced by nerve cell induction solution in the spinal cord injury area were injected into the cell population.

Urodynamic study: After 7 days of intervention, a urodynamic study was conducted on each group of rats, running parallel groups alternately, and recording the bladder pressure curve. Rats were injected intraperitoneally with 350%/kg 10% chloral hydrate (24). Take photos in the supine position after full anesthesia. The multi-channel physiological recorder is connected to the micro-infusion pump and the F3 catheter through a three-way tube. After emptying the bladder, insert the F3 catheter through the urethra, then gently introduce it into the rat bladder, and inject saline at 25-35°C. The bladder pressure increases as the volume increases (25). The basic bladder pressure is the initial pressure value after the catheter is inserted into the bladder. The maximum bladder pressure is the peak value in the pressure graph during the entire detection process. When fluid is first

discharged from the urethra, the perfusion rate is recorded as the maximum volume of the bladder, and the pressure value at this time is recorded as leakage, specifically.

$$C = \frac{\Delta V}{\Delta P} = \frac{1}{T} \quad (1)$$

### Observation Index

(1) The change of TUNEL positive rate in spinal cord tissue (26). The positive rate of apoptosis is the ratio of positive cells counted in the field to the total number of nuclei. (2) Analyze the change of caspase-3 expression in spinal cord tissue, analyze the gray value of target protein and internal reference  $\beta$ -actin, and use the ratio of the two gray values as the relative expression of the caspase-3 protein.

Use SPSS 17.0 software for processing. Express the measurement data as  $(x \pm s)$ , and perform the normality test. The normal distribution uses one-way analysis of variance, variance uses LSD method, and variance uses d T3 method. If it does not meet the normal distribution, then use rank independent test (K independent sample). Significance level  $\alpha = 0.05$ .

## Results and discussion

### Comparison of Protein Content between the Two Groups

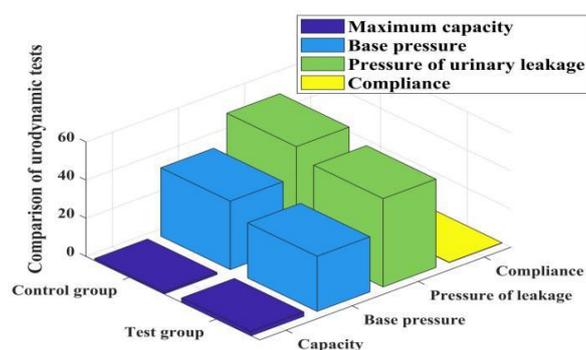
As shown in Table 2, the content of activated caspase-3 protein in the control group was  $0.439 \pm 0.091$  KD, and the experimental group was  $0.235 \pm 0.089$  KD. This indicates that the expression of caspase-3 is up-regulated after spinal cord injury, and its activity is enhanced. SCI inhibits the expression of apoptotic protein caspase-3 in spinal cord tissue, promotes spinal cord function remodeling and innervates the bladder and pelvic floor. The possible mechanism of transplantation of placental mesenchymal stem cells into the neurogenic bladder is to regulate and improve the pelvic floor muscle group. Coordination, inhibit excessive detrusor muscle activity and regulate bladder function.

**Table 2.** Comparison of Caspase-3 protein expression between the two groups

Group	Number of cases	Activated caspase-3
Experimental group	25	$0.235 \pm 0.089$
Control group	25	$0.439 \pm 0.091$

### Effect of Placental Mesenchymal Stem Cell-Derived Nerve Cell Transplantation on Neurogenic Bladder Function Recovery

The results of the study show that placental mesenchymal stem cell transplantation can effectively restore neurogenic bladder function. The urodynamic examination is now considered to be a diagnostic and therapeutic efficacy index for the neobladder after SCI. It can accurately and objectively reflect the functional status of the lower urinary tract while correlating subjective observation with bladder storage and urination functions. It provides a set of parameters for the diagnosis and evaluation of the neobladder. As shown in Figure 2, the maximum bladder capacity and bladder compliance of the control group were 1.087 ml and 35.874 cm H<sub>2</sub>O, respectively, and the bladder basic pressure and urinary leakage point pressure were 51.673 cm H<sub>2</sub>O and 0.0697 ml/cm H<sub>2</sub>O, respectively. Compared with the control group, the maximum bladder capacity and bladder compliance of the experimental group increased significantly ( $P < 0.01$ ), and the bladder basic pressure and urinary leakage pressure decreased ( $P < 0.05$ ). The values of these four items are 2.318ml, 28.892cm H<sub>2</sub>O, 46.34cm H<sub>2</sub>O, and 0.1389ml/cm H<sub>2</sub>O, respectively. This is because placental mesenchymal stem cell transplantation can increase the maximum bladder capacity and compliance of neurogenic bladder rats after spinal cord injury, reduce detrusor spasm, thereby reducing bladder pressure and lowering urine of patients with spinal cord injury.



**Figure 2.** Comparison of two groups of urodynamic tests

Studies have shown that the index of TUNEL positive rate can indicate the apoptosis of spinal cord tissue. As shown in Figure 3, the TUNNEL value of the control group fluctuated between 76.46% and

79.8%, while the TUNNEL value of the experimental group after placental mesenchymal stem cell transplantation decreased significantly, from the initial 78.5% to 52.56%. This reflects that placental mesenchymal stem cell transplantation can effectively reduce the apoptosis of spinal cord tissue and restore bladder function to a certain extent.

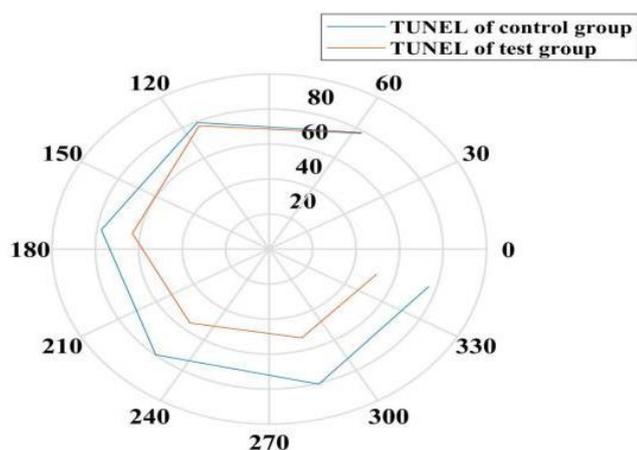


Figure 3. Comparison of TUNEL positive rate

### Therapeutic Mechanism of placental Mesenchymal Stem Cell-Derived Nerve Cell Transplantation on Neurogenic Bladder after Spinal Cord Injury

Mesenchymal stem cell transplantation improves the ultrastructure of detrusor muscle tissue, significantly reduces the rough endoplasmic reticulum and mitochondrial swelling, reduces thigh fibrils, and effectively improves the elasticity and compliance of detrusor muscles. As shown in Figure 4, the detrusor elasticity of the experimental group rose to 0.22HZ, and detrusor compliance increased by more than 83%. This shows that MSCs transplantation restores damaged bladder tissue cells to normal morphology, reduces inflammation and damage, maintains the normal morphological function of membrane epithelial tissue, and thus maintains the normal contractile function of detrusor muscles.

The results of this study show that placental mesenchymal stem cell transplantation can rapidly differentiate into nerve cells to compensate for the massive apoptosis of cells. As shown in Figure 5, the newly added cells in both parts were higher than the control group's 15%, 20%. This article believes that after transplantation with MSCs, the number of spinal cord cells has recovered to a certain extent. Therefore,

the neurogenic bladder has a certain function recovery.

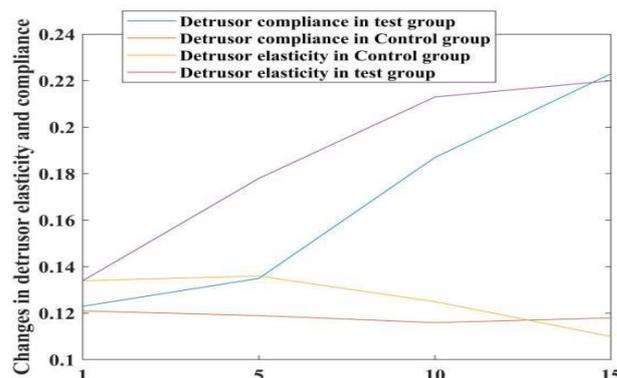


Figure 4. Comparison of compliance and elasticity of detrusor

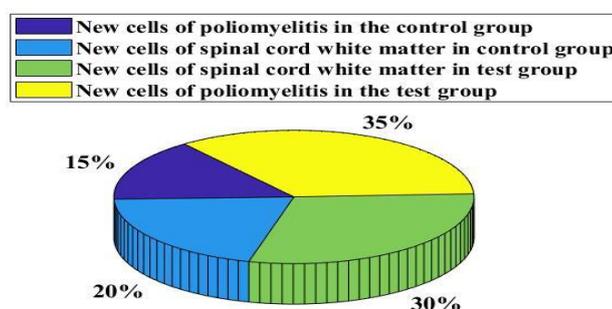


Figure 5. Comparison of newly added cells in the spinal cord

### Discussion

Urinary dysfunction caused by neurogenic bladder (NB) is currently the most difficult problem (27). Urinary dysfunction usually leads to difficulty in urination, prolonged urinary incontinence, repeated urinary tract infections, urinary tract stones and kidney accumulation, and even kidney failure in patients with SCI, usually accompanied by bladder pressure and urinary incontinence with frequent urination, urine urgent, a significant increase in urine leakage (27, 28).

Mesenchymal stem cells have a very powerful ability to differentiate neurons, making them resistant to apoptosis, repairing tissue damage and reducing the inflammatory response at the site of spinal cord injury (29). MSC reduces the detrusor muscle spasm by increasing the number of neurons in the spinal cord injury area and the maximum volume and compliance of the bladder, thereby reducing bladder pressure after

spinal cord compression and treating the neo-bladder (30).

At present, the main strategies mainly include preventing and reducing secondary injuries, improving the intrinsic capacity of nerve regeneration, eliminating nerve regeneration inhibitors, improving the regeneration of spinal cord nerve tissue and the good internal microenvironment of the injured spinal cord (31). At present, there are many methods to treat spinal cord injury, including improving spinal cord perfusion pressure, drug therapy, surgical treatment, limb function recovery, hyperbaric oxygen therapy, etc. (32). However, most of the treatment methods that have achieved significant clinical effects are currently not available. Most researches are limited to the animal test phase (33). Therefore, the repair and functional repair of spinal cord injury has always been a problem that plagues the medical community. Since the 20th century, with the increase in scientific research investment, the public's interest in SCI has continued to grow, science and technology have continued to develop, policies and regulations related to basic stem cell detection and clinical trials have been continuously improved, and basic research on the treatment of spinal cord stem cell injury has been increasing (31, 32). Including neural stem cells (NSC), the relative pursuit of stem cell transplantation technology is gradually increasing. The results of countless basic experiments completed by exogenous neural stem cells so far indicate that it is difficult to obtain a suitable source of exogenous neural stem cells, and there are related immune rejections and prone to tumor formation. And violation of related ethical issues, etc. (34).

Spinal cord injury (SCI) usually does not endanger the patient's life, but the main cause of death is complications caused by spinal cord injury (35). In patients with spinal cord injury, urinary dysfunction is extremely common. About 81% of patients have a certain degree of urinary system dysfunction within one year after the injury, and less than 1% of them can fully recover. Due to various factors or diseases, the connection between the bladder and the urination center is interrupted, and the urination control information of the brain cannot reach the bladder. Causing bladder dysfunction and urinary system damage, there are usually neurological diseases or traumas (36). Due to the different stages of spinal cord

injury, the clinical manifestations of the neurogenic bladder are also different (37). The main complications of the neurogenic bladder caused by spinal cord injury are urinary tract infection, deterioration of upper and lower urinary tract functions, and urinary calculi. It will cause various effects on the lower urinary tract, and also cause great difficulties for the treatment of urologists. Severe symptoms can lead to early chronic renal failure and death (35, 36). The neurogenic bladder will have different clinical manifestations for different nerve segments and the degree of injury. Although there are stem cell therapy, sacral cord electrical stimulation, pelvic floor muscle electrical stimulation, etc., the recovery and treatment of the neurogenic bladder is still a worldwide problem (37).

Stem cells are cells of origin with proliferation and differentiation potential and self-renewal and replication capabilities (2). In recent years, stem cells have become a popular direction in clinical treatment and basic experimental research due to their outstanding characteristics (36). Many scientists and researchers have devoted themselves to studying the directed differentiation of stem cells and their application in tissue engineering (2, 38). Stem cell research is a very large and complex research category. There are many discovered and potential stem cells used in treatment and repair (33). Stem cell researchers are engaged in efforts in many different directions, including seeking new sources and methods for pluripotent stem cells, inducing other stem cells to differentiate into pluripotent stem cells (iPSC), conducting research on stem cell intervention in the early stages of experimental animals or humans (38).

In this study, 50 SPF Wistar rats were selected to establish a rat model of spinal cord injury to simulate the bladder symptoms after spinal cord injury, with good clinical similarity. The experimental group was transplanted with placental mesenchymal stem cell-derived nerve cells to compare the urodynamic and TUNEL positive rates. At the same time, the site and degree of spinal cord injury can be effectively controlled in the model preparation. The results showed that the number of spinal cord cells recovered to a certain extent. The neurogenic bladder has a certain functional recovery. With the rapid development of science and technology, the methods

of treating neurogenic bladder have also increased. Each method has its advantages and disadvantages, and there is no unified treatment method. At the same time, we are also working hard to transform the results of basic science into clinical medicine in the field of treatment to benefit more patients.

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#### Interest conflict

The authors declare no conflict of interest.

#### Reference

1. Alizadeh A, Dyck SM, Karimi-Abdolrezaee S. Traumatic spinal cord injury: an overview of pathophysiology, models and acute injury mechanisms. *Front Neurol* 2019; 10: 282.
2. Azeez SH, Jafar SN, Azizaram Z, Fang L, Mawlood AH, Ercisli MF. Insulin-producing cells from bone marrow stem cells versus injectable insulin for the treatment of rats with type I diabetes. *Cell Mol Biomed Rep* 2021; 1(1): 42-51.
3. He Y, Jin X, Wang J et al. Umbilical cord-derived mesenchymal stem cell transplantation for treating elderly vascular dementia. *Cell Tissue Bank* 2017; 18(1): 53-59.
4. Cavallini C, Zannini C, Olivi E et al. Restoring in vivo-like membrane lipidomics promotes exosome trophic behavior from human placental mesenchymal stromal/stem cells. *Cell Transplant* 2018; 27(1): 55-69.
5. Lu Y, Wang Y, Zhou J. The biological characteristics and clinical practice of placental derived mesenchymal stromal cells. *Chin J Cardiol* 2016; 44(10): 909-912.
6. Du W, Li X, Chi Y et al. VCAM-1+ placenta chorionic villi-derived mesenchymal stem cells display potent pro-angiogenic activity. *Stem Cell Res Ther* 2016; 7(1): 1-13.
7. SUN B-H. Comparative cultivation of single cell derived placental mesenchymal stromal cells in vitro. *J Shanghai Jiaotong Univ* 2018: 1033-1038.
8. Payushina O. Hematopoietic microenvironment and the role of mesenchymal stromal cells in its organization. *Biol Bull Rev* 2015; 5(4): 383-393.
9. Kumar P, Gao K, Wang C et al. In utero transplantation of placenta-derived mesenchymal stromal cells for potential fetal treatment of hemophilia A. *Cell Transplant* 2018; 27(1): 130-139.
10. Jung J, Moon JW, Choi J-H, Lee YW, Park S-H, Kim GJ. Epigenetic alterations of IL-6/STAT3 signaling by placental stem cells promote hepatic regeneration in a rat model with CCl4-induced liver injury. *Int J Stem Cell* 2015; 8(1): 79.
11. Tiwari SK, Agarwal S, Seth B et al. Inhibitory effects of bisphenol-A on neural stem cells proliferation and differentiation in the rat brain are dependent on Wnt/ $\beta$ -catenin pathway. *Mol Neurobiol* 2015; 52(3): 1735-1757.
12. Hao G, Wang L, Chen D et al. Comparison of immunosuppressive effects between human placental MSCs derived from fetal and maternal origins on the rejection of allogenic skin grafts in mice. *Chin J Cell Mol Immunol* 2015; 31(5): 609-614.
13. Sekido N, Yoshino T, Takaoka E et al. Impact of adjuvant radiotherapy and reversibility of neurogenic bladder on bladder storage function and impact of urethral resistance on bladder emptying function after radical hysterectomy. *Open J Urol* 2017; 7(12): 252.
14. Sakakibara R, Uchida Y, Ishii K et al. Bladder recovery relates with increased mid-cingulate perfusion after shunt surgery in idiopathic normal-pressure hydrocephalus: a single-photon emission tomography study. *Int Urol Nephrol* 2016; 48(2): 169-174.
15. Zhu J, Cheng X, Wang Q, Zhou Y, Wang F, Zou L. Transplantation of endothelial progenitor cells for improving placental perfusion in preeclamptic rats. *Arch Gynecol Obstet* 2015; 291(5): 1113-1119.
16. Chen L, Zhang W-N, Zhang S-M, Yang Z-H, Zhang P. Effect of laparoscopic nerve-sparing radical hysterectomy on bladder function, intestinal function recovery and quality of sexual life in patients with cervical carcinoma. *Asian Pac J Cancer Prev* 2015; 15(24): 10971-10975.
17. Gao W, He X, Li Y, Wen J. The effects of FK1706 on nerve regeneration and bladder function recovery following an end-to-side neuroorrhaphy in rats. *Oncotarget* 2017; 8(55): 94345.
18. Yagi M, Kishigami S, Tanaka A et al. Derivation

- of ground-state female ES cells maintaining gamete-derived DNA methylation. *Nature* 2017; 548(7666): 224-227.
19. Bhansali S, Dutta P, Kumar V et al. Efficacy of autologous bone marrow-derived mesenchymal stem cell and mononuclear cell transplantation in type 2 diabetes mellitus: a randomized, placebo-controlled comparative study. *Stem Cells Dev* 2017; 26(7): 471-481.
  20. Shunmugavel A, Khan M, Hughes Jr FM, Purves JT, Singh A, Singh I. S-Nitrosoglutathione protects the spinal bladder: Novel therapeutic approach to post-spinal cord injury bladder remodeling. *NeuroUrol Urodyn* 2015; 34(6): 519-526.
  21. Ferrero SL, Brady TD, Dugan VP, Armstrong JE, Hubscher CH, Johnson RD. Effects of lateral funiculus sparing, spinal lesion level, and gender on recovery of bladder voiding reflexes and hematuria in rats. *J Neurotrauma* 2015; 32(3): 200-208.
  22. Tang B, Li X, Liu Y et al. The therapeutic effect of ICAM-1-overexpressing mesenchymal stem cells on acute graft-versus-host disease. *Cell Physiol Biochem* 2018; 46(6): 2624-2635.
  23. Li Y-H, Xu Y, Wu H-M, Yang J, Yang L-H, Yue-Meng W. Umbilical cord-derived mesenchymal stem cell transplantation in hepatitis B virus related acute-on-chronic liver failure treated with plasma exchange and entecavir: a 24-month prospective study. *Stem Cell Rev Rep* 2016; 12(6): 645-653.
  24. Okur SÇ, Erdoğan S, Demir CS, Günel G, Karaöz E. The effect of umbilical cord-derived mesenchymal stem cell transplantation in a patient with cerebral palsy: a case report. *Int J Stem Cell* 2018; 11(1): 141.
  25. Kubo K, Ohnishi S, Hosono H et al. Human amnion-derived mesenchymal stem cell transplantation ameliorates liver fibrosis in rats. *Transplant Direct* 2015; 1(4).
  26. Kawakubo K, Ohnishi S, Fujita H et al. Effect of fetal membrane-derived mesenchymal stem cell transplantation in rats with acute and chronic pancreatitis. *Pancreas* 2016; 45(5): 707-713.
  27. Gil-Tommee C, Vidal-Martinez G, Reyes CA et al. Parkinsonian GM2 synthase knockout mice lacking mature gangliosides develop urinary dysfunction and neurogenic bladder. *Exp Neurol* 2019; 311: 265-273.
  28. Aziziaran Z, Bilal I, Zhong Y, Mahmood AK, Roshandel MR. Protective effects of curcumin against naproxen-induced mitochondrial dysfunction in rat kidney tissue. *Cell Mol Biomed Rep* 2021; 1(1): 23-32.
  29. Alizadeh R, Bagher Z, Kamrava SK et al. Differentiation of human mesenchymal stem cells (MSC) to dopaminergic neurons: A comparison between Wharton's Jelly and olfactory mucosa as sources of MSCs. *J Chem Neuroanat* 2019; 96: 126-133.
  30. Andrzejewska A, Lukomska B, Janowski M. Concise review: mesenchymal stem cells: from roots to boost. *Stem Cells* 2019; 37(7): 855-864.
  31. Chen B, Li Y, Yu B et al. Reactivation of dormant relay pathways in injured spinal cord by KCC2 manipulations. *Cell* 2018; 174(3): 521-535. e513.
  32. Vinel C, Rosser G, Guglielmi L et al. Comparative epigenetic analysis of tumour initiating cells and syngeneic EPSC-derived neural stem cells in glioblastoma. *Nat Commun* 2021; 12(1): 1-20.
  33. Gazdic M, Volarevic V, Harrell CR et al. Stem cells therapy for spinal cord injury. *Int J Mol Sci* 2018; 19(4): 1039.
  34. Boukelmoune N, Chiu GS, Kavelaars A, Heijnen CJ. Mitochondrial transfer from mesenchymal stem cells to neural stem cells protects against the neurotoxic effects of cisplatin. *Acta Neuropathol Commun* 2018; 6(1): 1-13.
  35. Xia P, Gao X, Duan L, Zhang W, Sun Y-F. Mulberrin (Mul) reduces spinal cord injury (SCI)-induced apoptosis, inflammation and oxidative stress in rats via miRNA-337 by targeting Nrf-2. *Biomed Pharmacother* 2018; 107: 1480-1487.
  36. Vierck C. Mechanisms of below-level pain following spinal cord injury (SCI). *J Pain* 2020; 21(3-4): 262-280.
  37. Korupolu R, Stampas A, Gibbons C, Jimenez IH, Skelton F, Verduzco-Gutierrez M. COVID-19: Screening and triage challenges in people with disability due to Spinal Cord Injury. *Spinal Cord Ser Cases* 2020; 6(1): 1-4.
  38. Nagoshi N, Tsuji O, Nakamura M, Okano H. Cell therapy for spinal cord injury using induced pluripotent stem cells. *Regen Ther* 2019; 11: 75-80.