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ORIGINAL ARTICLE

Clinical Response of 277 Patients with Spinal Cord Injury to Stem Cell Therapy in Iraq

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Background and Objectives: Spinal cord injury is a common neurological problem secondary to car accidents, war injuries and other causes, it may lead to varying degrees of neurological disablement, and apart from physiotherapy there is no available treatment to regain neurological function loss. Our aim is to find a new method using autologous hematopoietic stem cells to gain some of the neurologic functions lost after spinal cord injury.
Methods and Results: 277 patients suffering from spinal cord injury were submitted to an intrathecally treatment with peripheral stem cells. The cells were harvested from the peripheral blood after a treatment with G-CSF and then concentrated to 4～6 ml. 43% of the patients improved; ASIA score shifted from A to B in 88 and from A to C in 32. The best results were achieved in patients treated within one year from the injury.
Conclusions: Since mesenchymal cells increase in the peripheral blood after G-CSF stimulation, a peripheral blood harvest seems easier and cheaper than mesenchymal cell cultivation prior to injection. It seems reasonable treatment for spinal cord injury.

Keywords: Spine injury, Stem cells, Mesenchymal cells

Introduction

Grounded in half a century of research, the study of stem cells is one of the most exciting and rapidly advancing disciplines in biomedicine today. Breakthrough discoveries in both the laboratory and clinic have sharply expanded the use and supply of life-saving stem cells. New treatments include graft-versus-tumour therapy for currently incurable cancers, mesenchymal cells for autoimmune diseases, and tissue repair.

Adult stem cells can either proliferate without differentiating for a long period (a characteristic referred to as long-term self-renewal), or can give rise to mature cell types that have characteristic shapes and specialized functions.

Some adult stem cells have the capability to differentiate into tissues other than the ones from which they originated; this is referred to as plasticity.

Substantial basic and limited clinical research exploring the experimental uses of stem cells for other diseases is underway. Among the primary applications are autoimmune diseases, such as diabetes, rheumatoid arthritis, Crohns disease, graft versus host disease, and systemic lupus erythematosus (1).

A few recent reports indicate that scientists have been able to induce bone marrow or adipose stem cells to differentiate into other types of tissue, such as brain, muscle, and liver cells. In a mouse model indicates that cells from grafts of bone marrow may home to damaged skeletal and cardiac muscle or liver and regenerate those tissues (2, 3).

An old statement in Medicine stressed that the nervous system has no plasticity and that any lesion in central and peripheral nervous system is irreversible. However there are several reports of improvement of vascular brain le-

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sions, Parkinson's disease and Parkinsonism, multiple sclerosis (4, 5) and traumatic spine lesions by means of intrathecal injections of stem cells. These cells were harvested from peripheral blood and bone marrow, selected or unselected from nucleated cells suspensions (6).

Mesenchymal stem cells can be harvested in bone marrow, Wharton jelly of placenta and adipose tissue. However the same cells can be isolated even from the peripheral blood after stimulations with G-CSF (7, 8).

Here we report our experience with intrathecal injections of peripheral blood stem cells in a series of traumatic spine lesions.

Materials and Methods

Patients

277 Iraqi patients, 252 males 25 females with spinal cord injury including partial and complete cervical or dorsal lesions were assessed clinically in the Neurosurgery Department of Medical City complex in Baghdad by trained neurosurgeons after getting patient consensus according to the ethical committee plan which is headed by high authorities in Iraq Health Ministry.

The period of follow up was from January 2009 till January 2011.

The age of the patients ranged from 18 to 65 yrs, mean 34.5.

The range of period from trauma till starting cell therapy is from 6 to 104 months, mean 39 months.

The site of the lesion was dorsal in 208 (75.1%) and cervical in 69 (24.9%).

Procedure

1. G-CSF (Neupogen-Roche) which was given in dose of 5 μg per kilogram body weight daily for 3 days subcutaneously followed by collection of nucleated cells according to the protocol for peripheral hematopoietic stem cells on the fourth day using WBC protocol of blood cell separation.

2. The procedure of peripheral stem cell separation was done by blood cell separator (Cobe Spectra version 7 lrs turbo).

3. A further step was the Buffy coat separation, by means of sedimentation in HES and centrifugation of the supernatant. Then total white blood cell count was done and total and mononuclear cells counted by manual method in the whole product. The cells were resuspended in 4～6 ml of saline.

4. The next procedure was the spinal injection of the cell product into the spinal canal through fifth lumbar vertebrae in sitting position under local anaesthesia by zylocaine within 24 hours of collection.

The procedure was repeated from 1 to 4 times (mean 1, 83), with intervals of 6～8 weeks.

The mean period of follow up was 12 months.

Results

Yield

Mean total mononuclear cell count per product was 5×10^8 (range 1～8).

Outcome

No response, with unchanged ASIA score, in 157 patients.

Clinical improvement was seen in 120 patients after 4 weeks of starting therapy (43.3%).

ASIA score shifted from A to B in 88 and from A to C in 32.

A subgroup (12 patients) whose spine lesion lasted for less than one year had the best outcome: the percentage improvement reached 50%.

Complications: the only complication in 90% of cases was transient backache and meningism. No serious side effects.

Discussion

Cell therapy for spinal cord injury is a new concept as a part of regenerative medicine field. Articles in the available sources are scarce and solid proof of benefit from stem cell therapy is difficult to obtain. The best way to prove the effect would be to track the SC inside the body to prove their participation in repairing the spinal cord lesion, but this is a very complex and difficult procedure. At the present time, we can depend on clinical evidence that demonstrated the positive effect of SC therapy even in advanced cases.

To explain the positive effects of autologous stem cells in spinal cord injury cases we can offer the following explanations:

- First of all even if the blood brain barrier prevents nucleated cells from penetrating into the nervous tissue, mesenchymal cells are able to overcome this barrier. Furthermore the neutrophils contained in the Buffy coat are able to open the barrier by means of a mild local inflammation.

- Chemokines hypothesis (9): local stem cells have a limited proliferative potential leading to slow regeneration, and a trauma starts apoptosis even in cells not

directly affected. However there are many factors that result in sending chemical, hormonal, cytokine or other factors leading to enhance healing and regeneration of the damage by leading to the activation of CNS local stem cells to participate in new cell formation. Furthermore there are probably factors leading to an inhibition of apoptosis.

- Regeneration hypothesis: injections of stem cells could help regeneration through their differentiation into neuronal cells.

Therefore autologous stem cells could be used (derived from patients' own bone marrow, but even from skin and blood). Pre-clinical research shows that effectiveness and safety profile of treatment with autologous bone marrow cells are relatively positive. Virtually no rejection occurs and therefore ethical considerations are limited.

As compared to embryonic stem cells mesenchymal stem cells (adult stem cells) bear no risk of DNA instability and tumour production.

It is beyond doubt that there is lot of research work to be done on the nature of brain damage and natural repair mechanisms of nervous tissue, on the interaction between immune system and stem cells and on various other aspects in this complex neurobiological arena.

However, there is growing, although sometimes inconclusive or casuistic evidence of clinical relevant brain-repair and protective properties of transplanted stem cells. Given the urgency of finding a cure for this widespread, disabling disease, most scientists argued that it is justifiable to arrange a rapid onset of well managed trials.

The proof of concept is available. Now it is time to proceed to the clinic. Therefore, we presented today a concept of a groundbreaking clinical trial, to be executed with more than 200 spinal cord lesions patients in our centre. The goal is primarily to evaluate safety and feasibility of the stem cell treatment, and secondly to investigate the repair and protective effect in the brain.

Potential conflict of interest

The authors have no conflicting financial interest.

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