Journal of Clinical Neuroscience 20 (2013) 310–312

Contents lists available at SciVerse ScienceDirect

Journal of Clinical Neuroscience

journal homepage: www.elsevier.com/locate/jocn

Case Reports

Possible induction of acute disseminated encephalomyelitis (ADEM)-like demyelinating illness by intrathecal mesenchymal stem cell injection

Nirmeen A. Kishk [a], Noha T. Abokrysha [a,*], Hala Gabr [b]

[a] Department of Neurology, Cairo University, Kaser Al-Aini Hospital, Al-Manyal Street, Garden City, Cairo 11562, Egypt
[b] Department of Clinical Pathology, Cairo University, Kaser Al-Aini Hospital, Cairo, Egypt

ARTICLE INFO

Article history:
Received 14 June 2011
Accepted 6 April 2012

Keywords:
Demyelinating disorder
Mesenchymal stem cell
Multiple sclerosis

ABSTRACT

We report a 27-year-old woman with an episode of encephalitis and optic neuritis, followed by autologous bone marrow mesenchymal stem cell transplants and possible induction of acute disseminated encephalomyelitis-like demyelinating illness.

1. Case report

We report a 27-year-old female patient who, in 2006, started to develop an acute onset of flu-like symptoms, including fever. Three days later she developed an acute onset and a stationary course of weakness affecting both upper and lower limbs. She sought immediate medical advice.

Clinical examination revealed a high grade fever (39 °C) but otherwise she had stable vital signs. Neurological examination showed that the patient was conscious, oriented and had no history of convulsions. Her lower limbs had a motor power grade of 0, while the upper limbs showed distal more than proximal weakness (distal grade 3, proximal grade 4). There was also an associated retention of urine, but the cranial nerves were not affected.

Laboratory tests revealed an elevated erythrocyte sedimentation rate (1st hour, 42 mm/hour; 2nd hour, 85 mm/hour). Her haemoglobin concentration was 9.4 g/dL, and total leucocyte count (TLC) was 10,700/μL with a neutrophil count of 12%. She had a random blood glucose test result of 150 mg/dL, and normal liver and kidney function. Her cerebrospinal fluid (CSF) was clear and colourless, with a glucose concentration of 70 mg/dL, protein 10 mg/dL, and cells, 20/μL (mainly lymphocytes).

The MRI showed an elongated area of an abnormal, high T2-weighted signal in the cervical cord parenchyma opposite C6 and C7, with cord oedema (Fig. 1a).

MRI brain and evoked potential (visual and brain stem) were normal. She was diagnosed as having acute transverse myelitis. She was subjected to methylprednisolone treatment (intravenous, 1 g/day) for 7 days and then prednisolone (oral, 60 mg/day) for 1 month; the dose was then tapered gradually over a 2-month period with no improvement. One month after the end of the prednisolone treatment, the patient underwent five sessions of plasma exchange with mild improvement in the motor power of the upper limbs (to grade 3+ distally, and 4+ proximally). However, there was no improvement in sphincter control or of the motor power of the lower limbs.

1.1. Intrathecal injection of autologous bone marrow stromal stem cells

The patient received three successive intrathecal injections of autologous bone marrow stromal stem cells (on a monthly basis). Informed consent was obtained from the patient. Ethical approval was obtained from the ethics committee of the National Cancer Institute, Cairo University.

The posterior iliac spine was sterilized, and local anesthesia (1% xylocaine) was applied to the periosteum and skin of that area. The bone marrow was then aspirated (10–20 mL) under aseptic conditions on preservative-free heparin. Fresh bone marrow was collected and processed for each delivery. Human mesenchymal stem cells (MSC) were isolated from bone marrow aspirates, washed with Dulbecco's low glucose modified Eagle's medium (Hyclone, Logan, UT, USA) containing 1% antibiotic–antimycotic (Gibco, Grand Island, NY, USA), and centrifuged through a density gradient (Ficoll-Plaque 1.077 g/mL, GE Biosciences, Piscataway, NJ, USA) for 30 minutes in a centrifugal field of 9000 m/s to remove lymphocyte and erythrocyte populations. Mononuclear cells were resuspended in complete culture medium (Minimum Essential Media [MEM], Gibco/BRL, Gaithersburg, MD, USA); 2 ml of the patient's own serum, selected for rapid growth of MSC (Atlanta Biologicals, Atlanta, GA, USA); and 100 units/mL of penicillin, 100 μg/mL of streptomycin, and 2 mM L-glutamine (Gibco/BRL). All cells were plated in 15 mL of medium and incubated in T25 flasks (Falcon Plastics, Los Angeles, CA, USA) at 37 °C, with 95% humidity and 5% carbon dioxide, in complete Dulbecco's modified Eagle's medium in concentrations of 500,000 cells/mL. After 24 hours, non-adherent cells were

* Corresponding author. Tel.: +20 966509321228.
E-mail address: nhtaha@yahoo.com (N.T. Abokrysha).

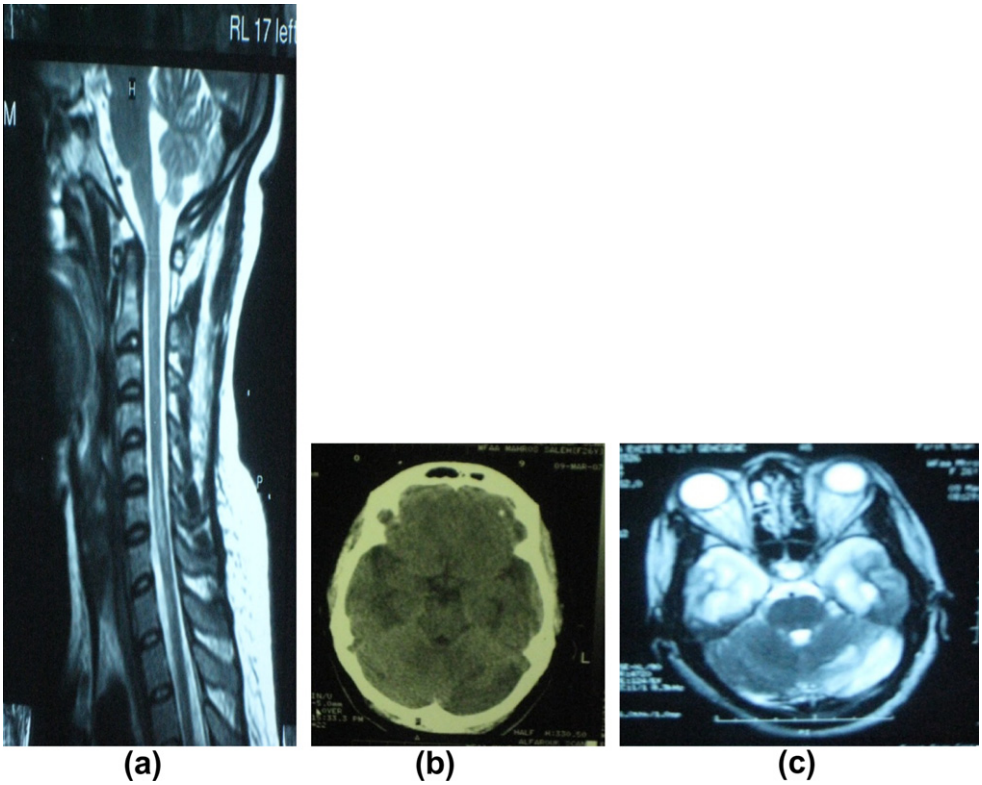

Fig. 1. Imaging studies in a 27-year-old female patient with transverse myelitis; (a) sagittal T2-weighted cervical MRI showing an elongated area of abnormal, high T2-weighted signal in the cervical cord parenchyma opposite C6 and C7 with cord oedema; (b, c) after the third dose of stem cell therapy; (b) axial CT scan showing bilateral temporal and left cerebellar hypodense areas (inflammation), which were (c) confirmed by axial T2-weighted MRI.

discarded, and of the adherent cells, the spindle-shaped cells were morphologically evaluated. The adherent cells were then washed with phosphate-buffered saline (PBS) and harvested by incubation for 4 minutes at 37 °C in 4 mL of 0.25% trypsin/1-methylenediaminetetraacetic acid. After incubation, the trypsin was inactivated by the addition of 5 mL of MEM. The cells were washed twice with PBS. Cell viability was determined using the trypan blue exclusion test. The cell population was characterized by typical fibroblast-like morphology, immunophenotyping (CD34 negative [–ve] and CD44 [+ve]), and ability to differentiate. The cell numbers were counted with a haemocytometer, and flow cytometric enumeration of CD44+ cells was done using a FACS Caliber flow cytometer (Becton Dickinson, Franklin Lake, NJ, USA) with the use of phycoerythrin-conjugated anti-CD44. The MSC were injected intrathecally in the lumbar region at the L3–4 or L4–5 levels. The dose was 5–10×10^6 MSC/kg of mononuclear cells.

The first two sessions showed no side effects from the injections, which were associated with mild improvement of the trunk area. However, at 6 hours after the third injection (March 2007), the patient developed a fever of 38 °C, and a disturbed level of consciousness (localizing to pain).

On examination, the patient had bilateral hyperaemic optic discs, more on the left side. A CT scan showed bilateral temporal and left cerebellar hypodense areas (inflammation), confirmed by MRI (Fig. 1). The TLC was 16,400/μL. The CSF showed a high protein content with few lymphocytes; the glucose concentration was normal. All other laboratory results were normal.

The patient then received methylprednisolone (1 g/day) for 10 days. The patient became fully conscious. The television (TV) pattern of visual evoked potential was carried out for each eye; it showed delayed P100 response latency over the left side with significant interside difference, a picture suggestive of a prechiasmatic demylinating lesion affecting the left visual pathway.

2. Discussion

MSC are multipotent non-haematopoietic progenitors, which have been explored as a promising treatment in tissue regeneration. Both *in vitro* and *in vivo*, the MSC inhibit the function of T, B, natural killer and dendritic cells.[1] This, in part, is the basis for autologous stem cell therapy (SCT) trials in refractory autoimmune disease, but conversely may predispose to the development of novel immune-related complications during reconstitution.[2] Acute optic neuritis and encephalomyelitis in a young person, such as our patient, that developed 6 hours after the third injection of MSC suggests a causal relationship. This may be one of the first reports of a multiple sclerosis (MS)-like disorder following bone marrow transplantation.

Mohyeddin Bonab et al. conducted a pilot study of ten patients with MS.[3] All patients had progressive MS that had not responded to disease modifying agents, including mitoxantrone. Patients were injected intrathecally with culture-expanded MSC. They were followed with monthly neurological assessment and an MRI scan at the end of the first year. During the 13 to 26 months of follow-up (mean: 19 months), the Expanded Disability Status Scale score of five patients increased from 0.5 to 2.5. In the functional system assessment, three patients had deteriorated, six patients showed some degree of improvement and one showed no difference. MRI assessment after 12 months showed no difference in seven patients, an extra plaque in two patients and a decrease in the number of plaques in two patients. [3] In addition, Armstrong et al.[4] reported a 62-year-old man with a relapsing remitting phenotype, characteristic of early adulthood MS. Onset with sequential, but rapidly remitting episodes of optic neuritis, transverse myelitis, and a brainstem syndrome followed autologous SCT for plasma cell leukemia.

3. Conclusion

MSC possess immunosuppressive potential and are proposed as a tool for cell therapy; however, under certain circumstances, they could change to become immunostimulating. It is therefore important to better understand the immune plasticity of MSC.[5]

References

1. Larghero J, Vija L, Lecourt S, et al. Mesenchymal stem cells and immunomodulation: toward new immunosuppressive strategies for the treatment of autoimmune diseases? *Rev Med Interne* 2009;**30**:287–99.
2. Krupica Jr T, Fry TJ, Mackall CL. Autoimmunity during lymphopenia: a two-hit model. *Clin Immunol* 2006;**120**:121–8.
3. Mohyeddin Bonab M, Yazdanbakhsh S, Lotfi J, et al. Does mesenchymal stem cell therapy help multiple sclerosis patients? Report of a pilot study. *Iran J Immunol* 2007;**4**:50–7.
4. Armstrong RJ, Elston JS, Hatton CS, et al. De novo relapsing-remitting multiple sclerosis following autologous stem cell transplantation. *Neurology* 2010;**75**:89–91.
5. Sun B, Zhang X, Wang G, et al. Regulation of suppressing and activating effects of mesenchymal stem cells on the encephalitogenic potential of MBP68-86-specific lymphocytes. *J Neuroimmunol* 2010;**226**:116–25.

doi:http://dx.doi.org/10.1016/j.jocn.2012.04.013

Commentary on "Possible induction of acute disseminated encephalomyelitis (ADEM)-like demyelinating illness by intrathecal mesenchymal stem cell injection"

Helmut Butzkueven *

Department of Medicine, Melbourne Brain Centre at the Royal Melbourne Hospital, University of Melbourne, Royal Parade, Parkville, Victoria 3050, Australia

ARTICLE INFO

Article history:
Received 13 May 2012
Accepted 11 June 2012

Keywords:
Mesenchymal stem cell therapy
Multiple sclerosis
Serious adverse event

ABSTRACT

The case report that is the subject of this Commentary describes a 27-year-old woman, who, 3 months after a devastating low cervical myelitis, underwent intrathecal mesenchymal stem cell (MSC) infusions. Six hours after the third infusion, she became unconscious, febrile and cerebral MRI showed acute bitemporal and left cerebellar lesions, consistent with an acute disseminated encephalomyelitis. It is likely that this is the first reported patient with neuroinflammatory exacerbation after MSC therapy. This case suggests that, in addition to their malignant potential, autologous MSC expanded *in vitro* can exhibit immune-activating properties leading to autoimmune exacerbation.

The report that is the subject of this Commentary in the current edition of the *Journal of Clinical Neuroscience* describes a 27-year-old woman, who, 3 months after a devastating low cervical myelitis, remained paraplegic in spite of pulsed methylprednisolone therapy and plasma exchange. The patient then underwent three intrathecal infusions of autologous bone-marrow derived mesenchymal stem cells (MSC), injected without concomitant immunosuppression. Six hours after the third infusion, she became unconscious and febrile and cerebral MRI showed acute bitemporal and left cerebellar lesions, consistent with an acute disseminated encephalomyelitis (ADEM). The patient regained consciousness but the neurological outcome was not reported.[1] This is possibly the first report of a neuroinflammatory exacerbation after MSC therapy, although a 17-year-old patient in the United States was recently reported with devastating ADEM after receiving stem cell therapy of unknown origin or quality in Costa Rica.[2]

A large variety of putative stem cell therapies for multiple sclerosis (MS) have been proposed and tried in single cases or in small series of patients. One of these, the injection of autologous MSC, was found in animal models of MS to have therapeutic utility. Researchers originally believed these cells could differentiate into, and thereby replace, neurons or myelinating oligodenrocytes, but the mechanism of action was ultimately found to be immunomodulatory.[3,4] Autologous MSC, administered intravenously or intrathecally, have potential utility in the treatment of MS, and several small, open-label studies in groups of seven to 15 patients with progressive MS, respectively, have been reported.[5–8] These studies have reported no serious adverse events, but clinical outcomes have been mixed. Disability was reported to have improved in one, stabilised in four, and worsened in five patients after a mean follow-up of 19 months.[5] In contrast, two studies reported either a 0.8 point mean improvement in the Expanded Disability Status Scale (EDSS) score at 12 months[6] or EDSS improvement in five of seven patients at 6 months.[7] The final study, in 11 patients with secondary progressive MS with visual impairment, reported a small improvement in visual acuity, contrast sensitivity and visual evoked potential latency at 6 months.[8] While these studies pave the way for future randomised controlled trials, serious adverse events are likely. The main concern has been the malignant potential of MSC, as these cells are expanded *in vitro* using mitogens and positive selection of malignant clones is an expected phenomenon.[9,10] However, the following case report in this journal raises another serious concern – the potential for immunomodulation in the wrong direction.

References

1. Kishk NA, Abokrysha NT, Gabr H. Possible induction of acute disseminated encephalomyelitis (ADEM)-like demyelinating illness by intrathecal mesenchymal stem cell injection. *J Clin Neurosci* 2013;**20**:310–2.
2. Alderazi Y, Coons SW, Chapman K. Catastrophic demyelinating encephalomyelitis after intrathecal and intravenous stem cell transplantation in a patient with multiple sclerosis. *J Child Neurol* 2012;**27**:632–5.
3. Zappia E, Casazza S, Pedemonte E, et al. Mesenchymal stem cells ameliorate experimental autoimmune encephalomyelitis inducing T-cell anergy. *Blood* 2005;**106**:1755–61.
4. Grigoriadis N, Lourbopoulos A, Lagoudaki R, et al. Variable behavior and complications of autologous bone marrow mesenchymal stem cells transplanted in experimental autoimmune encephalomyelitis. *Exp Neurol* 2011;**230**:78–89.

* Tel.: +61 3 8344 1832.
E-mail address: butz@unimelb.edu.au