Recovery from Severe Neurological Sequelae due to Anti-N-methyl D-aspartate Receptor Encephalitis after Three Infusions of Allogeneic Umbilical Cord-derived Mesenchymal Stem Cells: A First Case Report

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Abstract

Purpose

Anti-N-methyl D-aspartate (NMDA) receptor encephalitis is caused by altered patients' immune reactions. This study reports the first patient with severe neurologic sequelae after NMDA receptor encephalitis treated with allogeneic umbilical cord-derived mesenchymal stem/stromal cells (UC-MSCs) via intrathecal route.

Methods

UC-MSCs were obtained from a healthy donor and expanded under a xeno- and serum-free condition. The patient received three intrathecal UC-MSC infusions at the dose of 10^6 cells/Kg. The outcome was accessed using the German Coma Recovery Scale (CRS), the Gross Motor Function Classification System (GMFCS), the Gross Motor Function Measure–88 (GMFM-88), Manual Ability Classification System (MACS), Modified Ashworth Scale, and the Denver II test.

Results

A 5-year-old girl suffered from a permanent vegetative state with diffuse cerebral atrophy due to NMDA receptor encephalitis despite intensive treatment of immunosuppressive medicaments and IVIG. After three UC-MSC infusions, her cognition and motoric functions improved progressively. In the last visits, she could walk, practice writing, and count numbers. Urinary and bowel functions were completely controlled. Cerebral atrophy reduced on brain MRI.

Conclusions

The outcomes of this patient suggest a potential cell therapy for autoimmune encephalitis and its neurological consequences.

Introduction

Anti-NMDA receptor encephalitis is a brain inflammation caused by autoimmune antibodies targeting the NMDA receptor on the surface of neurons.¹ Treatment of autoimmune encephalitis (AE), including anti-NMDA receptor encephalitis, include corticosteroids and intravenous immunoglobulins, and/or plasma exchange followed by other immunosuppressants in refractory cases.² Although the response rate of patients to those treatments is generally high, 50% of patients still have cognitive and behavioural problems, 33% have ongoing seizures, and about 2% of patients suffer from severe disability. For patients with anti-NMDA receptor encephalitis, only 29% of cases achieve full recover.³

Recently, stem cell therapy has emerged as a new therapeutic option for many diseases. Bone marrow-derived mononuclear cells and mesenchymal stromal/stem cells (MSCs) infusion were safe and might improve the clinical outcomes in patients with various neurological conditions such as cerebral palsy, persistent vegetative state after drowning, autism, stroke, and brain trauma.^{4–8} MSC administration was also applied for AE. Bone marrow-derived MSCs were able to inhibit pathogenic T- and B-cells, prevent demyelination, increase axonal density, and protect oligodendrocytes from apoptosis in mouse models of autoimmune encephalomyelitis. As a result, the disease severity was reduced and survival rate was increased in treated mice.^{9,10} In humans, autologous adipose-derived mesenchymal stem cell infusion improved daily activities in all of six treated patients with autoimmune refractory epilepsy and decreased epilepsy frequency in three cases.¹¹

In this manuscript, we report the first case of a study describing a child who recovered from severe neurologic sequelae due to anti-NMDA receptor after three infusions of allogeneic UC-MSCs.

Methods

Preparation of cell therapy study

UC-MSCs were processed under xeno-free and serum-free conditions as described in detail previously.¹² Cryopreserved UC-MSCs at passage 3 were thawed and cultured up to passage 6. The cells were prepared in 10 mL of NaCl 0.9% (Braun, USA) at a dose of 1×10^6 cells/Kg patient body weight for the infusion. Release criteria of cell therapy products were: (1) free of bacteria, fungi, and mycoplasma, (2) viability $\geq 90\%$, (3) UC-MSC markers fulfilled the minimum criteria for MSC according to ISCT 2006,¹³ (4) product was within endotoxin limits for intrathecal route of application, i.e. 0.2 EU/Kg/h.

Intervention

In accordance with the severe condition of the patient, the parents were explained in detail the potential risks and benefits of the cell therapy and intrathecally infusion route. Upon obtaining their written informed consent and approval from the Hospital's Board of Directors, UC-MSC therapy was applied. The cells were infused intrathecally through the space between the fourth and fifth lumbar vertebrae using an 18-gauge needle within 30 minutes under general anaesthesia as described previously.⁶

Outcome measurements

Outcome was assessed using the German Coma Recovery Scale (CRS),¹⁴ the Gross Motor Function Classification System (GMFCS),¹⁵ the Gross Motor Function Measure – 88 (GMFM-88),¹⁶ the Manual Ability Classification System (MACS – link <https://www.macs.nu/>),¹⁷ Modified Ashworth Scale for muscle spasticity,¹⁸ and the Denver II test.¹⁹

Patient Report

Medical history before cell therapy

The child developed normally during the first 58 months until suffering from convulsions, weakness of the left arm and foot on August 10th, 2018. On EEG, slow delta waves with a frequency of 2-3 Hz were observed in the right hemisphere of the brain. The patient was diagnosed with AE based on positive anti-NMDA receptor in cerebral spinal fluid on August 22nd, 2018. Treatment was initiated with Solumedrol 20mg/kg/day for five days followed by Prednisolone with 30 mg/day, decreasing by 10 mg every seven days and lasting for 16 days; IVIG 5 g/day for 7 days; Depakine 300 mg/day; and Risperidone 1mg/day. From September 1st, 2018 onwards, the patient became unresponsive so that oral feeding through nasogastric tube was required. Treatment was switched to Cellcept 500 mg per day and Topamax 2 mg/kg/day for 10 days as of September 11th, 2018, without success. Rituximab 240 mg was given resulting in reduced myoclonus, but general muscular spasticity increased. An examination on December 27th, 2018, indicated complete loss of awareness, intense muscle spasticity of the whole body, and intermittent seizures. The patient did not respond to Cellcept 250 mg/day, Tocilizumab 120 mg, Depakine 150 mg/day, and Keppra 500 mg/day. The patient received acupuncture therapy in another hospital for two months without improvement.

Evaluation before cell therapy

The patient was admitted to Vinmec International Hospital on March 26th, 2019 (7 months after the onset of the illness). Her body weight was 17 kg. Her awareness was completely lost with a CRS of 6 points (Table 1). Intermittent seizures were observed. The GMFCS scored at the level V, the GMFM-88 at 23 points, and poor hand function with a MACS at the level V. Increased muscle tone measured using Modified Ashworth Scale was two points for upper and lower limbs (Table 2). Feedings were maintained through the nasal gastric tube. Urinary incontinence and constipation were noted. Personal-social, fine motor, language, and gross motor ability according to the Denver II were impaired (Table 3). Brain MRI revealed diffuse cerebral atrophy in the supratentorial region, dilatation of the third ventricle and bilateral lateral ventricles (Fig. 1A).

Allogeneic UC-MSC infusion and improvements

In accordance with the patient's severe condition, the parents were explained in detail the potential risks and benefits of the cell therapy and intrathecal infusion. Upon obtaining their written informed consent and approval from the Hospital's Board of Directors, UC-MSC therapy was applied.

The first infusion was performed on April 4th, 2019, with 17 million UC-MSCs. The characteristics of the MSC product is presented in the Table S1. No severe adverse events occurred during and after the procedure and the patient was discharged 48 hours after the infusion. Medication was continued with Risperidone 1 mg/day and Keppra 500 mg/day. Daily physical therapy was done at home by the patient's mother.

At re-examination on November 26th, 2019 (7 months 22 days after the first cell infusion), the patient's body weight increased to 19.5 kg. Muscle spasticity and dysphasia were reduced so that the nasogastric tube feeding was discontinued and switched to normal oral feeding. The patient was able to react to external stimuli with a German CRS of 10 points (Table 1). Motor functions showed no significant change, both GMFCS and MACS remained at the level V, and GMFM-88 scored 24 points. Muscle spasticity measured 2 points in the upper extremities and 1 point in the lower extremities (Table 2). Better head and neck control was also observed. The patient was able to turn to the sides. Denver scores remained unchanged (Table 3). Constipation and urinary incontinence persisted.

The second infusion of UC-MSCs was safely carried out on December 6th, 2019, with 19 million UC-MSCs (Table S1). The patient was discharged after two days and then continued to receive Risperidone 1 mg/day, Keppra 500 mg per day, and daily physical therapy at home. The doses of medication were reduced gradually and discontinued one month after the second infusion without the manifestation of epilepsy.

Re-examination on June 9th, 2020 (14 months 5 days after the first MSC therapy) showed her improved awareness with a German CRS of 19 points (Table 1). Patient's gross and fine motor skills resulted in better scores in all analysed tests: the GMFCS reduced to the level IV, GMFM-88 increased to 106 points, and MACS improved to the level II. Muscle spasticity was reduced from 2 points to 0 point for the upper limbs and remained 1 point for the lower limbs (Table 2). The patient was able to sit and use her hands to pick up foods and other objects. Denver II scores also showed improvement in all areas including personal-social, gross motor, fine motor-adaptive, and language (Table 3). However, urinary incontinence and constipation remained unimproved.

The third administration of 22 million UC-MSCs was performed without side effects on June 10th, 2020 (Table S2), followed by daily physical therapy at home after discharge.

Re-examination four months later (18 months after the first infusion) revealed that patient's awareness further improved with a German CRS of 21 points (Table 1). Examination of motor function indicated GMFCS at the level I, GMFM-88 of 255 points. MACS was at the level I and muscle tone reverse to normal with the modified Ashworth scale at score 0 (Table 2). She could walk normally, eat independently, and practice writing. Significant improvements were also observed at Denver II tests (Table 3). Urinary incontinence and constipation were slightly improved. Brain MRI indicated cerebral atrophy reduced remarkably with mild dilatation in the left lateral ventricle (Fig. 1B).

Three months later (21 months after the first infusion), she could draw, write, and speak some words. Her urinary and fecal function were completely controlled. In the last examination (28 months after the first infusion, 14 months after the third infusion), she could count numbers and prepared for school.

Discussion

For patients with severe psycho-neurological sequelae after AE, there is currently no effective treatment. Our patient received two lines of medication and rehabilitation, but all interventions failed to improve her condition. The patient was in a vegetative state when she was first admitted to Vinmec International hospital and received cell therapy as a last resort. The patient's cognition and motor skills recovered progressively following UC-MSC infusions. After the second administration, she was able to sit up, and after the third infusion, the patient can walk normally without any assistance. In addition, hand skills progressed significantly. At the last check-up, the child could do all daily activities and practice writing and drawing. Language bounced back, the epilepsy

disappeared, and no medications were further required. Improvements could also be observed on cerebral MRI, in which cerebral atrophy reduced remarkably after three UC-MSC infusions.

To the best of our knowledge, this is the first patient with severe neurological sequelae following anti-NMDA receptor encephalitis whose motor functions and cognitive behaviours recovered after intrathecal infusions of UC-MSCs. We chose UC-MSCs because they have several advantages over other sources. These cells are easy to harvest and expanded in culture.²⁰ Moreover, they are an ideal choice for allogenic use due to their low immunogenicity and strong immunomodulatory potency.^{21,22} UC-MSCs also secrete numbers of growth factors to stimulate angiogenesis and neurogenesis.²³

Conclusion

Our case report suggests that UC-MSC cell therapy may ameliorate severe neurological sequelae due to anti-NMDA receptor encephalitis. A study with a larger sample size should be performed to evaluate the efficacy of UC-MSCs for AE as well as severe neurological sequelae due to AE.

Declarations

Ethics approval and consent to participate

The study was approved by the Vinmec International Hospital's Board of Directors and the Research Institute for Child Health, Vietnam National Children's Hospital, Ethics Committee (No. VNCH-RICH-2019-40). An informed consent was obtained by the patient's parents before the cell therapy.

Consent for publication

Informed consent for publication was signed by the patient's parents.

Availability of data and materials

The authors confirm that the data supporting the findings of this study are available within the article and its supplementary materials or are available on request from the corresponding author.

Competing interests

L.N.T., V.T.H., H.L.T., P.A.T.N., D.M.H., D.V.N. are employed by the not-for-profit Vinmec Healthcare System. M.H. M.H. declared an advisory role for Regenerative Medicine at Vinmec Healthcare System.

Disclosure

The patient was not required to pay for any part of the cell therapy, including fees for umbilical cord collection, cell culture, cell processing, cell infusions, related medications, and single room stay and care during the treatment.

Funding

Not applicable.

Authors' contributions

L.N.T.: conception and study design, provision of study materials, performing clinical assessments and follow-ups of the patient, data analysis, manuscript writing and revision, and final approval of manuscript. V.T.H.: provision of stem cell processing and characterization, data analysis, manuscript writing and revision, and final approval of manuscript. H.L.T.: performing clinical assessments and follow-ups of the patient, data analysis, manuscript writing and revision, and final approval of manuscript. P.A.T.N.: performing UC-MSC infusion, patient care during and after the intervention, and final approval of manuscript. D.M.H.: provision of stem cell processing and characterization and manuscript revision, and final approval of manuscript. D.V.N., H.C.V., V.N.T.B: collection of data, data analysis, and final approval of manuscript. M.H.: data analysis, manuscript writing and revision, and final approval of manuscript.

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References

1. Dalmau J, Lancaster E, Martinez-Hernandez E, Rosenfeld MR, Balice-Gordon R. Clinical experience and laboratory investigations in patients with anti-NMDAR encephalitis. *Lancet Neurol*. 2011;10(1):63–74. doi:10.1016/S1474-4422(10)70253-2.
2. Stingl C, Cardinale K, Van Mater H. An Update on the Treatment of Pediatric Autoimmune Encephalitis. *Curr Treat Options Rheumatol*. 2018;4(1):14–28. doi:10.1007/s40674-018-0089-z.
3. Hachohen Y, Wright S, Waters P, et al. Paediatric autoimmune encephalopathies: clinical features, laboratory investigations and outcomes in patients with or without antibodies to known central nervous system autoantigens. *J Neurol Neurosurg Psychiatry*. 2013;84(7):748–55. doi:10.1136/jnnp-2012-303807.
4. Sharma A, Sane H, Gokulchandran N, et al. A Clinical Study of Autologous Bone Marrow Mononuclear Cells for Cerebral Palsy Patients: A New Frontier. *Stem Cells Int*. 2015;2015:1–11. doi:10.1155/2015/905874.
5. Liem NT, Chinh VD, Phuong DTM, et al. Outcomes of Bone Marrow-Derived Mononuclear Cell Transplantation for Patients in Persistent Vegetative State After Drowning: Report of Five Cases. *Front Pediatr*. 2020;8:564. doi:10.3389/fped.2020.00564.
6. Nguyen Thanh L, Nguyen H-P, Ngo MD, et al. Outcomes of bone marrow mononuclear cell transplantation combined with interventional education for autism spectrum disorder: Bone Marrow Mononuclear Cells & Interventional Education for ASD. *STEM CELLS Transl Med*. Published online September 9, 2020. doi:10.1002/sctm.20-0102.
7. Dharmasaroja P. Bone marrow-derived mesenchymal stem cells for the treatment of ischemic stroke. *J Clin Neurosci*. 2009;16(1):12–20. doi:10.1016/j.jocn.2008.05.006.
8. Wang S, Cheng H, Dai G, et al. Umbilical cord mesenchymal stem cell transplantation significantly improves neurological function in patients with sequelae of traumatic brain injury. *Brain Res*. 2013;1532:76–84. doi:10.1016/j.brainres.2013.08.001.
9. Zhang J, Brodie C, Li Y, et al. Bone marrow stromal cell therapy reduces proNGF and p75 expression in mice with experimental autoimmune encephalomyelitis. *J Neurol Sci*. 2009;279(1-2):30–8. doi:10.1016/j.jns.2008.12.033.
10. Gerdoni E, Gallo B, Casazza S, et al. Mesenchymal stem cells effectively modulate pathogenic immune response in experimental autoimmune encephalomyelitis. *Ann Neurol*. 2007;61(3):219–27. doi:10.1002/ana.21076.
11. Szczepanik E, Mierzewska H, Antczak-Marach D, et al. Intrathecal Infusion of Autologous Adipose-Derived Regenerative Cells in Autoimmune Refractory Epilepsy: Evaluation of Safety and Efficacy. *Stem Cells Int*. 2020;2020:1–16. doi:10.1155/2020/7104243.
12. Hoang VT, Trinh Q-M, Phuong DTM, et al. Standardized xeno- and serum-free culture platform enables large-scale expansion of high-quality mesenchymal stem/stromal cells from perinatal and adult tissue sources. *Cytotherapy* Published online October 2020:1465324920308562. doi:10.1016/j.jcyt.2020.09.004.

13. Dominici M, Le Blanc K, Mueller I, et al. Minimal criteria for defining multipotent mesenchymal stromal cells. The International Society for Cellular Therapy position statement. *Cytotherapy*. 2006;8(4):315–7. doi:10.1080/14653240600855905.
14. von Wild K. Neuroethics with regard to treatment limiting and withdrawal of nutrition and hydration in long lasting irreversible full state apallic syndrome and minimal conscious state. *J Med Life*. 2008;1(4):443–53.
15. Palisano RJ, Rosenbaum P, Bartlett D, Livingston MH. Content validity of the expanded and revised Gross Motor Function Classification System. *Dev Med Child Neurol*. 2008;50(10):744–50. doi:10.1111/j.1469-8749.2008.03089.x.
16. Russell D, Rosenbaum P. Gross motor function measure (GMFM) score sheet (GMFM-88 and GMFM-66 scoring). Published online 2013.
17. Compagnone E, Maniglio J, Camposeo S, et al. Functional classifications for cerebral palsy: Correlations between the gross motor function classification system (GMFCS), the manual ability classification system (MACS) and the communication function classification system (CFCFS). *Res Dev Disabil*. 2014;35(11):2651–7. doi:10.1016/j.ridd.2014.07.005.
18. Bohannon R, Smith M. Modified Ashworth Scale Instructions. Published online 2016. <https://www.sralab.org/sites/default/files/2017-06/Modified%20Ashworth%20Scale%20Instructions.pdf>.
19. Frankenburg WK, Dodds J, Archer P, Shapiro H, Bresnick B. The Denver II: a major revision and restandardization of the Denver Developmental Screening Test. *Pediatrics*. 1992;89(1):91–7.
20. Xie Q, Liu R, Jiang J, et al. What is the impact of human umbilical cord mesenchymal stem cell transplantation on clinical treatment? *Stem Cell Res Ther*. 2020;11(1):519. doi:10.1186/s13287-020-02011-z.
21. Ding D-C, Chou H-L, Chang Y-H, Hung W-T, Liu H-W, Chu T-Y. Characterization of HLA-G and Related Immunosuppressive Effects in Human Umbilical Cord Stroma-Derived Stem Cells. *Cell Transplant*. 2016;25(2):217–28. doi:10.3727/096368915X688182.
22. Mattar P, Bieback K. Comparing the Immunomodulatory Properties of Bone Marrow, Adipose Tissue, and Birth-Associated Tissue Mesenchymal Stromal Cells. *Front Immunol*. 2015;6. doi:10.3389/fimmu.2015.00560.
23. Petrenko Y, Vackova I, Kekulova K, et al. A Comparative Analysis of Multipotent Mesenchymal Stromal Cells derived from Different Sources, with a Focus on Neuroregenerative Potential. *Sci Rep*. 2020;10(1):4290. doi:10.1038/s41598-020-61167-z.

Tables

Tables 1

Changes of German Coma Remission Scale.

	Before the 1st treatment (March 2019)	After the 1st treatment (November 2019)	After the 2nd treatment (June 2020)	After the 3rd treatment (October 2020)
Arousability / attention	2	2	5	5
Motoric response	3	3	6	6
Response to acoustic stimuli	0	1	2	2
Response to visual stimuli	1	1	2	3
Response to tactile stimuli	0	1	2	3
Logomotor (speech motoric) response	0	2	2	2
Total	6	10	19	21

Tables 2

Changes of motor functions.

		Before the 1st treatment (March 2019)	After the 1st treatment (November 2019)	After the 2nd treatment (June 2020)	After the 3rd treatment (October 2020)
GMFCS		V	V	IV	I
GMFM-88	Lying and rolling	17	18	51	51
	Sitting	6	6	48	60
	Crawling and kneeling	0	0	4	42
	Standing	0	0	3	37
	Walking, running, and jumping	0	0	0	65
	Total	23	24	106	255
MACS		V	V	II	I
Modified Ashworth Scale		Score 2 for both upper and lower extremities	Score 2 for the upper and score 1 for the lower extremities	Score 0 for the upper and score 1 for the lower extremities	Score 0 for both upper and lower extremities

Table 3

Changes in Denver II tests

Denver II test	Before the 1st treatment (March 2019)	After the 1st treatment (November 2019)	After the 2nd treatment (June 2020)	After the 3rd treatment (October 2020)
Personal – Social	0.5	0.5	3	10
Fine motor – Adaptive	0.5	0.5	7	15
Language	0.5	0.5	6	6
Gross motor	0.5	0.5	6	24

Figures

Figure 1

Figure 1

A. Brain MRI before the 1st transplantation showed diffuse cerebral atrophy, dilatation of the third and bilateral ventricles. B. Improved brain MRI image with milder dilatation of the lateral ventricles, subarachnoid and sulcus were observed after three UC-MS administrations.

Supplementary Files

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