

# Chapter 2

## New Strategy of Clinical Studies for Premature Babies with Ischemic Brain Damage

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**Abstract** In Japan, we started autologous cord blood therapy for newborns with HIE in 2014 (described in Chap. 1). Another research group started autologous cord blood therapy for patients with cerebral palsy in 2017. However, cerebral palsy is induced in nearly twice as many premature babies with IVH and PVL as that by term newborns with HIE (Touyama et al. *Brain Dev* 38:792–799, 2016; Koterazawa et al. *No to Hattatsu* 48:14–19, 2016; Glinianaia et al. *Dev Med Child Neurol* 59:864–870, 2017). Therefore, we are now promoting a new clinical study protocol of cell therapy for premature newborns with PVL or IVH.

**Keywords** Cell therapy · Periventricular leukomalacia · PVL · Intraventricular hemorrhage · IVH

### 2.1 Introduction

Clinical studies of regenerative medicine such as various types of stem cells, embryonic stem (ES) cells, and/or iPS cells have increased rapidly, and the number of clinical studies had exceeded 500 worldwide in 2016 [1]. However, hyperbole, distortion, and overselling of these regenerative medicines have also been reported in some journals [2]. Among these different strategies of cell therapies, the intravenous administration of UCB stem cell therapy could be the safest and most feasible because UCB has been used for hematopoietic stem cell transplantation in patients

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with hematological diseases over several decades. Stem cells, obtained from umbilical cord blood, normally discarded after birth, are emerging as a safe and potentially effective therapy. Therefore, cell therapy using UCB has been expanded for novel applications. Recent reports have stated that the most common indication for UCB therapy is neurological diseases, including studies of cerebral palsy [3]. Other indications included diabetes mellitus, cardiac and vascular diseases, and hepatic diseases. Thirty-one studies administered total nucleated cells, mononuclear cells, or CD34+ cells, while 20 studies used cord blood-derived mesenchymal stromal cells. Eleven out of 46 studies described that cellular products used autologous products. They reported that 4/4 showed clinical benefit in cerebral palsy [4]. Furthermore, autologous UCB therapy has fewer ethical issues than allogenic UCB because autologous UCB has no possibility of rejection and no necessity of immune suppressive therapy to prevent rejection or GVHD. Therefore, autologous umbilical cord blood stem cell therapy could be the most feasible therapy for premature newborns with IVH or PVL. We are now challenging new project to investigate the feasibility and safety of autologous cord blood cell therapy for premature babies with IVH or PVL.

2.2 New Idea of Clinical Study Protocol

Major inclusion criteria included premature newborns who were born at 24–33 weeks old. The collection of baby cord blood cells is performed immediately after birth. The required volume of collected cord blood is over 15 mL. Separation is performed in cell processing center within 36 h of birth and returned to the hospital. The separated cord blood cells are administered intravenously within 36–72 h after birth (Fig. 2.1).

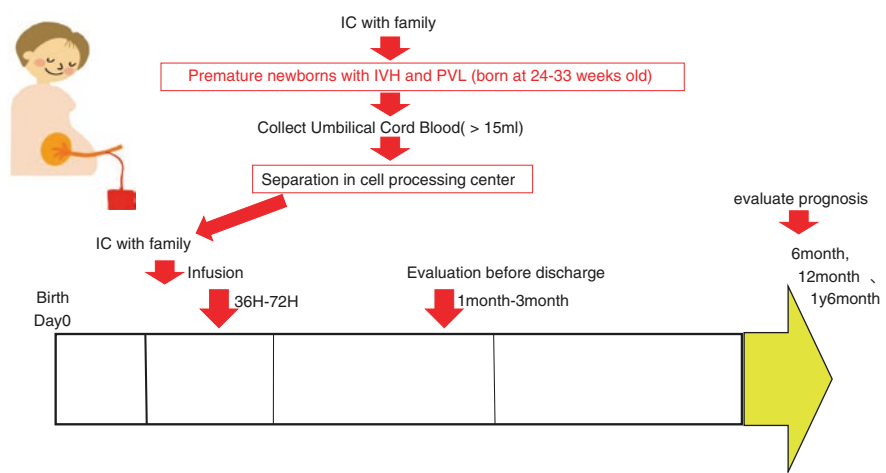


Fig. 2.1 New idea of cell therapy for premature newborns with IVH and PVL

## 2.3 Discussion

### 2.3.1 *Therapeutic Effect of UCB in a Hematopoietic Disease*

In 1982, Nakahata and Ogawa reported that umbilical cord blood contains hemopoietic colony-forming cells with extensive capability to generate mono- and multipotential hemopoietic progenitors [5]. Since then it has been confirmed that UCB also contains various rich stem cells such as hematopoietic stem cells, endothelial progenitor cells, and mesenchymal stromal cells.

CD34 surface antigen has been widely used as a marker of hematopoietic stem cells and endothelial progenitor cells. UCB contains about 0.3–2% CD34+ cells, while peripheral blood of an adult contains < 0.01% CD34+ cells [6–8].

Firstly, the therapeutic effects of UCB have been shown in hematological diseases, such as leukemia, Fanconi's anemia, and aplastic anemia, replacing the hematopoietic stem cells over the past few decades [9–14].

### 2.3.2 *Therapeutic Effect of UCB in Various Intractable Diseases*

In recent years, UCB has been identified as a source of endothelial stem/progenitor cells and has an effect on various intractable diseases including cerebral palsy, diabetes mellitus, cardiac and vascular diseases, and hepatic diseases. Various types of stem cells are possible sources of cell therapy for clinical applications especially for neurological diseases [15–17].

In 2004, Kurtzberg reported that allogeneic UCB has been used for patients with inherited metabolic disorders and neurodegenerative diseases, i.e., Hurler's syndrome and Krabbe's disease, with the aim of delivering the deficient enzyme through the stem cells. Seventeen of the 20 children were alive at the median point of 905 days after transplantation (event-free survival rate, 85%). Transplantation improved neurocognitive performance and decreased the somatic features of Hurler's syndrome. Cord blood from unrelated donors appears to be an excellent source of stem cells for transplantation in patients with Hurler's syndrome. Eleven asymptomatic newborns (age range, 12–44 days) and 14 symptomatic infants (age range, 142–352 days) with infantile Krabbe's disease underwent transplantation of UCB from unrelated donors after myeloablative chemotherapy. Transplantation of UCB from unrelated donors in newborns with infantile Krabbe's disease favorably altered the natural history of the disease. However, transplantation in babies after symptoms had developed did not result in substantive neurologic improvement [18, 19].

Also in 2004, Taguchi reported that CD 34+ cells are effective for brain damage after stroke and started phase 1–1/2a clinical studies for patients with stroke for the first time. Phase 1/2a study of CD 34+ cells therapy for 12 patients with stroke showed

favorable neurologic recovery and improvement in cerebral blood flow and metabolism with no serious adverse events. In comparison with historical controls, patients receiving cell therapy had significantly better neurologic outcomes. The results indicated that intravenous transplantation of autologous bone marrow mononuclear cells is safe and feasible. Positive results and trends favoring neurologic recovery and improvement in cerebral blood flow and metabolism by cell therapy underscore the relevance of larger-scale, randomized controlled trials using this approach [20, 21].

### ***2.3.3 Therapeutic Effect of UCB in Cerebral Palsy***

In this chapter, we focus on the potential therapeutic effects of cell therapies especially UCB therapy for newborns with ischemic disease which has progressed dramatically over the past few decades. In 2006 Meier et al. reported the effectiveness of intraperitoneal infusion of UCB cells in rats with neonatal hypoxia [22]. Kurtzberg is conducting a phase 2 study for cerebral palsy by using autologous cord blood cells at Duke University in the USA. On the other hand, Cox et al. reported a feasibility study showing that autologous bone marrow mononuclear cells were logistically feasible and safe to prescribe intravenously for children suffering from head trauma within 48 h of incident in 2011 [23]. Wang and Sharma also started a clinical study using autologous bone marrow mononuclear cells for cerebral palsy patients in 2013 and 2015 [24, 25].

It was also reported that concomitant administration of allogeneic UCB and recombinant human erythropoietin may boost the efficacy of UCB, as it has neurotrophic effects. Thus, allogeneic UCB treatment might ameliorate motor and cognitive dysfunction in children with CP undergoing active rehabilitation, accompanied by structural and metabolic changes in the brain [26]. Subarachnoid placement of stem cells was performed for 180 cases with diplegia and quadriplegia after trauma in India. This was effective in 32% of patients with no short- and long-term adverse effects. In the long-term follow-up, functional indices improved in 57 (31.67%) patients, including 54 patients with traumatic paraplegia/quadruplegia, 2 with cerebral palsy, and 1 with viral encephalitis [27]. Recently, Mancias-Guerra started an open-label phase 1 trial to investigate the safety and tolerability of intrathecal delivery of autologous bone marrow nucleated cells in children with cerebral palsy [28].

### ***2.3.4 Possibility of Therapeutic Effect of UCB in Premature Newborns with PVL and IVH***

Rizk reported that the most common indication for UCB therapy was neurological diseases (25 studies), including studies of cerebral palsy (12 studies). Other indications included diabetes mellitus (nine studies), cardiac and vascular diseases (seven studies), and hepatic diseases (four studies). Most studies administered total

nucleated cells, mononuclear cells, or CD34+ cells (31 studies), while 20 studies used cord blood-derived mesenchymal stem cells. Forty-six studies described cellular products obtained from allogeneic sources, while 11 studies used autologous products. They identified three indications where multiple prospective controlled studies have been published (4/4 studies reported clinical benefit in cerebral palsy, 1/3 studies reported benefit for cirrhosis, and 1/3 studies reported biochemical response in type 1 diabetes) [4]. Autologous UCB therapy has fewer ethical issues than allogeneic UCB because autologous UCB has no possibility of rejection and no necessity of immune suppressive therapy to prevent rejection or GVHD. Autologous UCB stem cell therapy could be the most feasible therapy for premature newborns. Recent experimental and clinical reports have indicated that cord blood stem cell therapy might provide protective effect on hypoxic-ischemic brain damage by the process shown below:

1. Immunomodulation/anti-inflammatory action [29–31]
2. Reduction of apoptosis and oxidative stress [32–34]
3. Enhancement of regenerative process by secretion of various cytokines [35–38]
4. Enhancement of regenerative process by angiogenesis to better circulation of the brain [39–46]
5. Enhancement of regenerative process by neurogenesis [47–54]
6. However, clinical studies of autologous UCB stem cell therapy for premature newborns with IVH or PVL have not been published. We need to challenge the new clinical study to investigate feasibility and safety of autologous UCB cell therapy for premature babies with IVH or PVL.

## 2.4 Conclusion

Autologous UCB stem cell therapy might be the most feasible therapy for premature newborns who were born at 24–33 weeks old. Further clarification is required on the feasibility and efficacy of cell therapy for premature newborn babies with IVH or PVL.

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