

Stem cells as therapeutical option for the treatment of bronchopulmonary dysplasia

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Stem cells: present and future

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Abstract

During the past decades clinical results in neonatology have improved dramatically and increased the survival rate of preterm infants significantly. However, the short and long term outcome of these high-risk preterm infants is mainly influenced by respiratory diseases and neurological damages. Despite great advances in perinatal medicine, there is still no satisfactory treatment for bronchopulmonary dysplasia (BPD) and current approaches are only supportive, have strong adverse effects or only show small benefits. Stem cell based therapies as well as other modes of regenerative strategies are applied as standard therapy in childhood predominantly in paediatric oncology. To date, such therapies have successfully been applied to treat immunodeficiency disorders and aplastic anaemia. But regenerative medicine might be an option for the treatment of BPD in preterm infants. According to some first preclinical results stem cell administration appears as a promising tool to improve the clinical outcome in high-risk infants. For severe neonatal diseases, e.g. hypoxic-ischemic encephalopathy (HIE) in term neonates or BPD in preterm infants, a number of animal models have been established. Although these studies showed positive effects of stem cells in animal models of BPD several questions still remain. Further studies with appropriate preclinical neonate models and carefully controlled clinical trials are needed to assess the significance of regenerative therapies. In this review, we summarize recent results of some experimental and clinical studies that used

stem cells to treat BPD associated with impairment of lung development.

Keywords

Preterm infants, bronchopulmonary dysplasia, stem cells, regenerative therapies, lung development.

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Background

With the advances in modern perinatal medicine more preterm infants born with a very low gestational age are offered a chance to live. At the same time, this means that these children are born at a very early developmental stage which is accompanied by complications threatening these children's lives. One of the major complications of preterm birth is bronchopulmonary dysplasia (BPD), characterized by impaired lung development [1]. Despite great advances in perinatal medicine, there is still no satisfactory treatment for BPD and current approaches are only supportive, have strong adverse effects or only show small benefits [1]. In recent years, the use of stem cells in the therapy of BPD has been investigated intensively. Several animal studies showed promising effects of stem or progenitor cells in lung regeneration and were followed by first clinical studies in humans [1, 2]. Still, many questions remain, as in these studies, different cell populations were used, the cells were applied at different times and using different routes. Nevertheless, the treatment with progenitor/stem cells offers a very promising option for new therapies of BPD. In this review, we summarize the results of some experimental and clinical studies that used stem cells to treat BPD associated impairment of lung development.

Experimental and clinical studies

In experimental studies, mice or rats are most often used to simulate the situation in preterm infants.

Rodents are born with lung development being in the saccular stage and with this the lung of newborn rodents resembles the lung of human preterms at 26-28 weeks [1]. Generally, the newborn animals are exposed to hyperoxic conditions (60% to 90% O₂) for 1 to 2 weeks causing structural changes and impairment of further lung development that results in a lung structure that corresponds to the situation in the lung of a preterm infant suffering from BPD [1, 3]. BPD is associated with prenatal insults, for instance chorioamnionitis or growth restriction which may also contribute to the development of the disease besides the postnatal stress of exposing a lung to oxygen when it is not ready to breathe yet. Therefore, some new models include prenatal hits like inflammation or hypoxia to combine more than one factor contributing to the disease and to get closer to the situation in humans [4-6].

Aslam et al. used a mouse model in which newborn mice were exposed to hyperoxia (75%) to induce lung damage. At day P4 the animals were given bone marrow derived mesenchymal stem cells (BMSCs) or BMSC conditioned media (CM). On day P14 lung morphometry, vascular changes associated with lung hypertension and lung cytokine profiles were assessed. Both, stem cells as well as the conditioned media reduced alveolar loss and lung inflammation in the animals. These results suggest that BMSCs act in a paracrine manner via the release of immunomodulatory factors [7].

In the same year, another study by van Haften et al. also showed a positive effect of mesenchymal stem cells (MSCs) in newborn rats. The animals were exposed to hyperoxia (95%) from birth to day P14. BMSCs were applied at day P4 via intratracheal injection. The treatment improved survival of the animals and attenuated alveolar and lung vascular injury [8].

The aim of a study by Ahn et al. in 2011 was to investigate the long-term outcome and safety of MSCs derived from human umbilical cord blood in a rat model. Newborn rats were exposed to hyperoxia (90%) for 14 days after birth. After that they were allowed to recover in room air until being sacrificed at day P70. At day P5 the animals received 5 x 10⁵ human umbilical cord blood derived mesenchymal stem cells (hUCB-MSCs). In the stem cell treated animals impaired alveolar and vascular growth as well as the inflammatory response were attenuated with no long-term adverse effects being seen [9].

In 2011 de Paepe et al. used a model with transgenic mice based on doxycycline-dependent Fas ligand overexpression to induce lung injury.

The animals were given CD34⁺ haematopoietic progenitor cells derived from human cord blood at day P5 via intranasal inoculation. Engraftment, differentiation of the respiratory epithelium, proliferation and cell fusion were studied 8 weeks after the inoculation. The group found that haematopoietic stem cells are capable of reconstituting injured alveolar epithelium [10].

In a model combining prenatal hypoxia (10%) with postnatal hyperoxia (75%), a crude preparation of mononuclear cells derived from umbilical cord blood was able to reduce lung damage. Aim of the study from 2013 was to investigate the capability of fresh cell preparations derived without further manipulation to attenuate lung injury and to improve lung development. Mice were prenatally exposed to hypoxia to induce growth restriction. Besides a reduced birth weight, hypoxia also negatively influences lung development [11]. The prenatal hypoxia was combined with postnatal hyperoxia to further impair lung development [5]. The animals were given mononuclear cells derived from human umbilical cord blood at day P7 and sacrificed at day P14. Stereological analysis showed improved thickness of septa and reduction of inflammation as well as normalization of mRNA expression of mTOR in comparison to untreated animals with lung injury [12].

hUCB-MSCs were used by Chang et al. in 2013 aiming to optimize the timing of stem cell transplantation. They exposed newborn rats to hyperoxia (90%) for 2 weeks and treated the animals at day P3 or P10. They found a time dependent attenuation of hyperoxia-induced lung injury with significant protection in the early but not in the late phase of inflammation [13].

Then, in 2014 a Korean group of neonatologists successfully applied MSCs to 9 human preterm infants with threatening BPD [14]. The children were born with a mean gestational age of 25 weeks. The mean birth weight was 793 g. Thus, the children were characterized as being extremely premature. Intratracheal stem cell transplantation was performed at a mean of 10.4 days after birth. Three patients were given a low dose treatment with 1×10^7 cells/kg and 6 patients were treated with a high dose of 2×10^7 cells/kg. The treatments were well tolerated without any serious adverse effects. With this, the group was able to show that allogeneous transplantation of stem cells is an option for treatment in preterm infants.

Although the aforementioned studies showed positive effects of stem cells in animal models of

BPD several questions still remain. In the studies, different stem cell populations from different sources were used: MSCs as well as mononuclear cells or progenitor cells showed positive effects. The cells had been derived from bone marrow or umbilical cord blood. It still needs to be further elucidated which kind of cells will be best suitable for therapeutic use in human patients. Important aspects are efficacy and long-term safety, but also how easily the cells can be obtained. In this context it needs to be considered that in Europe cells that are more than minimally manipulated are classified as Advanced Therapy Medicinal Products (ATMPs) requiring EU-marketing authorization [15]. This may be a hindrance when timing is of importance and autologous cell populations are supposed to be transplanted to the patient soon after obtaining the cells. Timing is another important factor that needs further examination. As BPD is a disease with early onset and in some cases being influenced by prenatal factors like intrauterine infection or growth restriction, it may be mandatory to begin therapy as soon as possible after birth to prevent lung injury [15]. This consideration is supported by the results of Chang et al. who showed a time dependent effect in mice with better results in case of early therapy compared to administration of the cells at a later time [13].

Further aspects that are not to be neglected are the optimal cell number and the method of application. Besides different cell populations, the studies also used different cell concentrations. Thus, the optimal cell number for therapy still needs to be examined. Furthermore, the question about the application remains. In the studies described before the cells were applied systemically or topically into the airways. First results in the animal models indicate that the direct application into the airways yields better results [16].

In summary, the majority of experimental studies applied stem cells in the first days after birth of the animals, favouring an early start of the therapy. The intratracheal route seems to be more effective than the systemic application of cells [2].

Mechanism of action

Most studies using stem cells in animal models of BPD only showed weak homing of the cells [9]. Thus, the positive effects on lung development and regeneration seen in the studies may not be explained by the stem cells substituting missing cells in the damaged tissue [2]. Another explanation

for the effect of stem cells is the modulation of inflammation via soluble factors [2]. Different studies showed that application of stem cells influenced the concentration of pro-inflammatory cytokines and attenuated the inflammatory response [9, 15]. These results are supported by studies using conditioned media instead of the cells themselves. Conditioned media, containing soluble factors released by the stem cells, are also able to attenuate lung injury in animal studies [7].

Summary

Despite the advances made in perinatal medicine in the last years, the treatment of diseases like BPD is not satisfactory [1]. Therefore, new therapies are needed that are able to not only reduce the damage of the existing lung structure but also to improve further lung development on the long term. Regenerative therapies using the capabilities of stem cells offer promising options in this aspect. Many studies were able to show that stem cells are able to attenuate lung injury in animal studies and support the development of a normal lung structure. In addition, stem cells can also be applied in case of inborn heart cardiac anomalies, cardiomyopathies, pulmonary hypertension, necrotizing enterocolitis, acute respiratory distress syndrome or chronic infections [15]. Although several questions still remain (which cell populations to use, time and route of administration), several studies showed the potential of stem cells to be used in the therapy of neonatal complications.

Declaration of interest

The Authors declare to have no conflict of interest.

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