REVIEW

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Lung Regeneration Therapy for Chronic Obstructive Pulmonary Disease

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Chronic obstructive pulmonary disease (COPD) is a critical condition with high morbidity and mortality. Although several medications are available, there are no definite treatments. However, recent advances in the understanding of stem and progenitor cells in the lung, and molecular changes during re-alveolization after pneumonectomy, have made it possible to envisage the regeneration of damaged lungs. With this background, numerous studies of stem cells and various stimulatory molecules have been undertaken, to try and regenerate destroyed lungs in animal models of COPD. Both the cell and drug therapies show promising results. However, in contrast to the successes in laboratories, no clinical trials have exhibited satisfactory efficacy, although they were generally safe and tolerable. In this article, we review the previous experimental and clinical trials, and summarize the recent advances in lung regeneration therapy for COPD. Furthermore, we discuss the current limitations and future perspectives of this emerging field.

Keywords: Pulmonary Disease, Chronic Obstructive; Cell- and Tissue-Based Therapy; Drug Therapy; Emphysema; Lung; Regeneration; Regenerative Medicine; Retinoids; Stem Cells

Introduction

Chronic obstructive pulmonary disease (COPD) is a critical condition with high morbidity and mortality not only in South Korea but also all over the world[1]. There are two major pathologic changes in subjects with COPD: emphysema and chronic bronchitis. The former refers to the destruction of alveoli, whereas the latter involves chronic inflammation and subsequent airway remodeling. Several medications with bronchodilating and/or anti-inflammatory effects have been developed and prescribed in clinical practice. However, although many studies have tried to regenerate destroyed alveoli, no therapy has successfully repaired the diseased lungs of patients with COPD. We here review the experimental and clinical trials of lung regeneration therapy, particularly those focusing on COPD, and discuss the current advances, limitations, and future perspectives of this emerging field.

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Endogenous Stem and Progenitor Cells in the Lung

Stem and progenitor cells are characterized by a self-renewal capacity and ability to differentiate into various types of cells[2,3]. Owing to these distinct characteristics, they are expected to play a key role in the regeneration of damaged tissue. Since the discovery of the ability of alveolar epithelial type 2 cells to differentiate into type 1 cells[4], several endogenous stem and progenitor cells in the lung have been excavated and characterized (Table 1)[2,3].

Basal cells, which line the proximal airways, have self-renewal potential and are able to differentiate into club cells, ciliated cells, secretory cells, and even alveolar epithelial type 1 and 2 cells. They can be recognized by such markers as Trp63,

Table 1. Endogenous stem and progenitor cells in the lung

Cell type	Localization	Differentiation	Markers	Reference
Basal cells	Proximal airways	Self, AEC1, AEC2, ciliated, club	Trp63, Krt5, Krt14, Ngfr, Pdpn	Kotton and Morrisey[2], Rock and Konigshoff[3]
Club cells	Proximal and distal airways	Self, basal, ciliated	Scgb1a1, Cyp2f2	Kotton and Morrisey[2], Rock and Konigshoff[3]
BASCs	BADJs	Self, AEC2, club	Scgb1a1, Sftpc	Kotton and Morrisey[2], Rock and Konigshoff[3]
AEC2	Alveoli	Self, AEC1	Sftpc	Kotton and Morrisey[2], Rock and Konigshoff[3], Adamson and Bowden[4]
Distal lung progenitors	Alveoli	Self, AEC1, AEC2, ciliated, club	Itga6, Itgb4	Kotton and Morrisey[2], Rock and Konigshoff[3]

AEC1: alveolar epithelial type 1 cells; AEC2: alveolar epithelial type 2 cells; BASCs: bronchioalveolar stem cells; BADJs: bronchoalveolar duct junctions.

Krt5, Krt14, Ngfr, and Pdpn. Club cells, also known as Clara cells, are the key component of proximal and distal airways and express markers such as Scgb1a1 and Cyp2f2. They are also considered to have the features of progenitor cells since they have self-renewal capacity and the ability to differentiate into ciliated cells, secretory cells, and basal cells. In addition, bronchioalveolar stem cells, characterized by their noticeable localization at bronchoalveolar duct junctions and by the expression of distinct markers such as Scgb1a1 and Sftpc, can self-renew and differentiate into club cells and alveolar epithelial type 2 cells. In the alveolus, besides the well-known alveolar epithelial type 2 cells, alveolar Itga6$^+$ Itgb4$^+$ Sftpc$^-$ cells, also known as distal lung progenitors, are regarded as endogenous pulmonary multipotent cells that have the capacity to differentiate into other cells, including club cells, ciliated cells, and alveolar epithelial type 1 and 2 cells. Although little is understood about the complexities of lung regeneration, there is growing evidence that these endogenous pulmonary multipotent cells are activated by lung injury[2,3] and thus they are currently presumed to play an important role as a cellular reservoir during regenerative processes.

Experimental Backgrounds of Lung Regeneration Therapy

1. Pneumonectomy models

As is well known, salamanders can regenerate whole limbs or tails, even when they lose a considerable part of them, in a phenomenon known as complete replacement[5]. In contrast, it is regarded as difficult to regenerate damaged tissue or organ in mammals, including humans. Moreover, in contrast to the recent advances in endogenous pulmonary stem and progenitor cells[2,3], the lung traditionally has been considered to be a highly quiescent organ with limited reparative capacities. However, since Laros and Westermann[6] reported the compensatory growth of lung and its re-alveolization after pneumonectomy, the paradigm has changed and many investigators have revealed the basic mechanisms and molecular changes occurring during re-alveolization and lung regeneration.

For example, Landesberg et al.[7] reported changes in gene expression after pneumonectomy in a mouse model. After the pneumonectomy, there was upregulation of transcription factors such as early growth response gene-1 (Egr-1), Nurr77, LRG-21, and tristetraprolin. Moreover, these transcription factors were linked to the activation of key signaling pathways and mediators such as mitogen-activated protein kinase, extracellular signal-regulated kinase, c-jun NH_2-terminal kinase, fibroblast growth factor-2 (FGF-2, also known as basic FGF [bFGF]), and platelet-derived growth factor, which are associated with cell growth and differentiation. In addition, Kho et al.[8] revealed that the expression levels of genes related to extracellular matrixes such as collagen and elastin, which are vital to the repair of damaged pulmonary structures, were increased in pneumonectomy-treated mice, suggesting reinforced regenerative processes after a pulmonary insult. In contrast, the genes related to growth inhibition and cell cycle arrest, such as phospholipase A2 group XVI (Pla2g16), were downregulated after pneumonectomy[9]. Accordingly, the lung is currently considered an active organ that has regenerative capacity through various mediators and signaling pathways that promote cell proliferation and differentiation.

2. Cell therapies for lung regeneration in animal COPD models

With the evidence of compensatory growth after pneumonectomy and advances in the understanding of the cellular

and molecular mechanisms of re-alveolization, the lung began to attract considerable attention as an emerging target of regenerative therapy. In addition, the progresses in stem cell technology have made it possible to examine the safety and efficacy of cell therapy, which refers to the administration of stem cells for therapeutic purposes, in animal models of COPD.

At first, embryonic stem cells (ESCs) derived from an early embryonic stage attracted considerable attention by virtue of the extreme multipotency, which means the ability that can be easily differentiated into any types of cells including endogenous pulmonary stem and progenitor cells. For example, Roszell et al.[10] successfully differentiated ESCs into alveolar epithelial type 2 cells, which play an important role in repairing destroyed alveoli. However, several critical setbacks occurred during the experiments with ESCs, namely teratogenicity and immunogenicity. Furthermore, the ethical issues surrounding ESCs made them more difficult to be investigated. In this context, mesenchymal stem cells (MSCs), which can be harvested from adult mesenchymal tissue such as bone marrow (BM-MSCs) or adipose tissue (AD-MSCs), began to be considered as an alternative of ESCs. Moreover, the low immunogenicity of MSCs, which express only low level of human leukocyte antigen-1 (HLA-1) and no HLA-2 and other major costimulatory molecules[11], made them more popular for the investigation.

Due to these advantages, MSCs have been investigated in the context of many animal disease models, including COPD, and have shown promising results in numerous experimental trials. In murine models with papain-, elastase-, or cigarette smoke–induced emphysema, the BM-MSC and AD-MSC successfully regenerated damaged lungs[12-14]. In addition, in our recent study using intravenously infused umbilical cord blood-derived MSCs, we demonstrated both the regenerative capacity and the optimal dose of MSCs (5×10^4 cells) for regenerating destroyed alveoli in mice with emphysema[15].

Despite the experimental successes in animal COPD models, it is still unclear how the administered stem cells treat the damaged lung. Traditionally, the investigators expected that the administered stem cells would engraft on damaged tissue and subsequently differentiate into the required cells, such as endogenous pulmonary progenitor cells. However, contrary to the traditional belief, there is emerging evidence indicating that stem cells act through paracrine and immune-modulatory mechanisms. In our previous study, rats successfully recovered from emphysema not only through BM-MSCs, but also through cell free-conditioned media containing only the soluble mediators[13]. This suggests that certain molecules from BM-MSCs that are dissolved in conditioned media may affect the damaged lung and promote the recovery processes. Moreover, in our recent study, we demonstrated that the intravenously infused MSCs disappeared from the recipients within 24 hours[16], indicating that the mid- and long-term effects of stem cell therapy are probably based on the mechanisms other than direct engraftment. Taken together with the reports that MSCs release a variety of growth factors and immune-modulatory cytokines[17] and that certain mediators such as hepatocyte growth factor (HGF), FGF, epidermal growth factor, vascular endothelial growth factor, and transforming growth factor β are up-regulated during reparative and regenerative processes[12,18], the successes in cell therapy for lung regeneration in animal COPD models seem to be mainly based on the paracrine and immune-modulatory effects of stem cells.

3. Drug therapies to regenerate the damaged lung in animal COPD models

Besides cell therapies, the expanding knowledge on genetic and molecular changes during the repair process has prompted investigators to examine the possible use of certain molecules as key drugs in lung regeneration therapy. Of these molecules, retinoids such as vitamin A and all-*trans* retinoic acid (ATRA) are some of the most widely investigated. Retinoids regulate various gene expressions by interacting with specific nuclear receptors and induce the production of proteins such as elastin, an essential component of lung matrixes. Moreover, they are also famous for their ability to promote alveolar septation and lung development[19]. In this background, Massaro and Massaro[20] examined the efficacy of ATRA in rats with elastase-induced emphysema and successfully demonstrated its regenerating capacity.

In addition to the retinoids, various growth factors involved in cell growth and differentiation are also actively being investigated as candidate drugs for lung regeneration therapy. For instance, HGF, a pleiotropic molecule with proliferative effects on alveolar epithelial type 2 cells and endothelial cells[21], is being actively researched in animal models of COPD. Shigemura et al.[22] showed the ability of HGF to ameliorate pulmonary emphysema using an *in vivo* gene transfection technique in a rat COPD model. In addition, Hegab et al.[23] demonstrated the therapeutic potential of HGF in a mouse model of emphysema via intranasal administration, which is one of the most convenient and safe routes of delivery in animal models.

Besides HGF, bFGF is also being intensively investigated since it plays a critical role in cell division, differentiation, and survival and functions as a potent mitogen. Morino et al.[24] reported that controlled-release FGF-2 microspheres ameliorate pulmonary emphysema in a canine model by improving pulmonary perfusion. In addition, Lee et al.[25] observed the protective effects of FGF-2 in an interferon-γ–induced emphysema mouse model. Moreover, in addition to the emphysema model, the role of FGF-2 in pulmonary epithelial recovery was also demonstrated in a bleomycin-induced pulmonary fibrosis model using FGF-2 knockout mice[26].

Lastly, there is some interesting evidence indicating that

estrogen might be related to lung regeneration. According to a study conducted by Massaro and Massaro[27], the formation of alveoli was diminished in mice that underwent oophorectomy. Interestingly, estrogen replacement was proven to reverse the negative effects of the oophorectomy by inducing alveolar regeneration, indicating the possible use of estrogen as a candidate drug for lung regeneration in female subjects with COPD.

Clinical Trials of Lung Regeneration Therapy for COPD

We have broadly discussed lung regeneration therapy and its promising results in many experimental and preclinical trials. Accordingly, it has emerged as a promising strategy for the definite treatment of chronic progressive and destructive respiratory diseases such as COPD. Furthermore, a recent fascinating study involving the infusion of allogeneic BM-MSCs (Prochymal; Osiris Therapeutics Inc.) into patients with acute myocardial infarction incidentally revealed a positive relationship between the stem cell therapy and increased forced expiratory volume in 1 second (FEV_1) at 6 months from baseline[28]. In this context, there have been several attempts to investigate the safety, feasibility, and efficacy of lung regeneration therapy in patients with COPD.

The latest clinical trials of lung regeneration therapy, as mentioned above when discussing experimental trials, can also be divided into two major categories: (1) cell therapies, which involve the administration of either autologous or allogeneic stem cells to regenerate the diseased lung and (2) drug therapies, which are presumed to induce lung regeneration by stimulating endogenous stem and progenitor cells through various exogenously administered molecules.

Clinical Trials of Cell Therapies in COPD Subjects

According to ClinicalTrials.gov (https://www.clinicaltrials.gov) and the EU Clinical Trials Register (https://www.clinicaltrialsregister.eu), only four early-phase clinical trials of cell therapy in patients with COPD have been completed; seven trials are ongoing and one has an unknown status (Tables 2, 3).

Of these studies, the most fascinating and well-designed one was conducted by Weiss et al.[29] in the United States, which was published in 2013 (NCT00683722). This was a multicenter, double-blind, randomized controlled trial to assess the safety and efficacy of intravenously infused allogeneic BM-MSCs in COPD subjects. Sixty-two patients with moderate-to-severe COPD (Global Initiative for Chronic Obstructive Lung Disease [GOLD] stage II or III) from six hospitals were enrolled and intravenously infused with either commercially prepared alloge-

Table 2. Completed clinical trials of cell-based lung regeneration therapy for COPD

Location	Patients	Cell type	Dose	Frequency	Route	Follow-up	Status	Clinical trial No.	Reference
US	62	Allogeneic BM-MSCs	1×10^8 cells/infusion	Multiple; monthly ×4	IV	24 mo	Completed	NCT00683722	Weiss et al.[29]
Brazil	4	Autologous BMMCs	1×10^8 cells/mL	Single	IV	12 mo; extended to 36 mo	Completed	NCT01110252	Ribeiro-Paes et al.[30], Stessuk et al.[31]
Netherlands	10	Autologous BM-MSCs	NA	Multiple; weekly ×2	IV	2 mo	Completed	NCT01306513	NA
Brazil	10	BM-MSCs	NA	Single	IT	4 mo	Completed	NCT01872624	NA

COPD: chronic obstructive pulmonary disease; BM-MSCs: bone marrow-derived mesenchymal stem cells; IV: intravenous; BMMCs: bone marrow mononuclear cells; NA: not available; IT: intratracheal.

Table 3. Ongoing clinical trials of cell-based lung regeneration therapy for COPD

Location	Patients	Cell type	Dose	Frequency	Route	Follow-up (mo)	Status	Clinical trial No.	Reference
US	200	Autologous AD-MSCs	NA	NA	IV	6	Recruiting	NCT01559051	NA
US	100	Autologous AD-MSCs	NA	NA	NA	6	Recruiting	NCT02041000	NA
US	60	Autologous SVFs	NA	Single	IV	12	Recruiting	NCT02161744	NA
US, Costa Rica, Nicaragua	200	Autologous AD-MSCs	NA	Single	IV	12	Recruiting	NCT02216630	NA
US	75	Autologous SVFs	NA	NA	NA	12	Not yet recruiting	NCT02348060	NA
Brazil	20	Autologous BMMCs and/or AD-MSCs	1×10^8 cells/infusion	Single	IV	12	Recruiting	NCT02412332	NA
Russia	30	Allogeneic BM-MSCs	2×10^8 cells/infusion	Multiple; every 2 mo×6	IV	24	Recruiting	NCT01849159	NA
Iran	12	Autologous BM-MSCs	6×10^7 cells/infusion	Single	IT	12	Unknown	NCT01758055	NA

COPD: chronic obstructive pulmonary disease; AD-MSCs: adipose tissue-derived mesenchymal stem cells; NA: not available; IV: intravenous; SVFs: stromal vascular fractions; BMMCs: bone marrow mononuclear cells; BM-MSCs: bone marrowderived mesenchymal stem cells; IT: intratracheal.

neic BM-MSCs (Prochymal) or vehicle control. The MSC dose was 1.0×10^8 cells per infusion and was administered monthly for 4 months. Subsequently, the subjects were followed for 2 years from the first dose. In this trial, the investigators successfully demonstrated the safety of allogeneic BM-MSCs as no severe or fatal adverse events (AEs) related to the procedure were observed. However, they did not find any significant differences in efficacy-related end points including pulmonary function tests and quality of life indicators. Interestingly, however, in *post-hoc* analysis of subjects with elevated circulating C-reactive protein (CRP) at baseline, the researchers observed an early decrease in the level of CRP in subjects who received MSCs, suggesting possible anti-inflammatory effects of stem cells.

Meanwhile, another interesting pilot study (NCT01110252) was carried out in Brazil[30]. The investigators in that trial enrolled four male patients with advanced COPD whose age ranged from 59 to 76. The participants were intravenously administered autologous bone marrow mononuclear cells, which contain both hematopoietic stem cells and MSCs, to evaluate the safety of the procedure. During 12 months of follow-up, the participants did not experience any AEs related to the drug or procedure except for mild symptoms and signs due to the administration of granulocyte colony stimulating factor, which was subcutaneously injected to stimulate bone marrow before harvesting. In 2013, the Brazilian researchers reported additional follow-up data, up to 3 years, for the same patients[31]. One of the subjects, unfortunately, died of hospital-acquired infection about 12 months after the procedure, but the stem cell therapy was still considered to be safe because the death was not the drug or procedure related and the remaining subjects did not show any serious or fatal AEs.

In addition to these aforementioned trials, two more complete but unpublished studies are noteworthy, one from the Netherlands (NCT01306513) and the other from Brazil (NCT01872624). These studies are compelling due to their unique designs, which combine cell therapy with other procedures. The investigators from the Netherlands enrolled 10 COPD patients with GOLD III stage who were candidates for lung volume reduction surgery. To investigate the safety, feasibility, and efficacy of stem cell therapy with lung volume reduction surgery, the researchers performed the surgery twice in each patient with an interval of several months and administered weekly doses of autologous BM-MSCs twice between the procedures. They compared the number of AEs, indicators of wound healing, and histologic responses between the first (before BM-MSCs infusion) and second (after BM-MSCs infusion) groups of patients. On the other hand, in the study from Brazil, the investigators recruited 10 COPD patients who were undergoing the installation of a one-way endobronchial valve. To assess the safety and efficacy of the procedure with endobronchial administration of BM-MSCs, they divided the

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subjects into two groups: an active comparator group, who received both a one-way endobronchial valve installation and endobronchial MSC infusion, and a placebo comparator group, who received a one-way endobronchial valve installation alone. They compared pulmonary function tests, inflammatory markers, and quality of life indicators between the groups at 4 months from the procedure. The publication of these trials is eagerly awaited because they are expected to provide meaningful evidence that will broaden the use of stem cell therapies by combining with other pulmonary procedures.

In addition to BM-MSCs, COPD therapies using AD-MSCs are also eagerly being investigated. Currently, six clinical trials using AD-MSCs or stromal vascular fractions containing AD-MSCs are ongoing (Table 3). Since it has not been well established yet whether the safety and efficacy of AD-MSCs are comparable to those of BM-MSCs, the clinical trial NCT02412332 being carried out by Brazilian investigators is receiving considerable attention. In this trial, 20 patients with COPD are to be enrolled and randomized into four distinct groups receiving placebo, AD-MSCs, BM-MSCs, or both. By evaluating the number of AEs and indicators of pulmonary functions and radiologic morphologies at 12 months from baseline, the study is expected to reveal interesting results on the effects of cell types on the safety and efficacy of cell therapies using MSCs.

Furthermore, a Russian research group recently added a fascinating new twist to this rapidly expanding field (NCT01849159). They have adopted the concept of preconditioning to potentiate the efficacy of stem cells. To reinforce the effects of MSCs, the investigators plan to culture allogeneic BM-MSCs in hypoxic conditions with 1% oxygen. Although it is a small pilot study that includes only 30 subjects, its findings are expected to give a hint to overcome the disappointing results of lung regeneration therapy in human subjects.

Drug Therapies to Regenerate the Damaged Lung in COPD

In contrast to cell therapies, drug therapies are intended to induce lung regeneration via exogenously administered molecules that are considered to stimulate endogenous stem and progenitor cells, not by the administration of stem cells themselves. Because considerable evidence indicates that the major action mechanism of cell therapy is based on paracrine effects[12,13,16-18], researchers have attempted to regenerate diseased lung with various molecules[20-27]. Of these numerous candidates, to date retinoids are the only one that has been widely investigated in the clinical field (Table 4).

Retinoids are proven to activate genes involved in lung development and to promote alveolar regeneration in animal models[19,20]. In this background, the first pilot study using

Table 4. Completed clinical trials of drug-based lung regeneration therapy for COPD

Location	Patients	Drug type	Dose	Frequency	Route	Follow-up (mo)	Status	Clinical trial No.	Reference
US	20	ATRA	50 mg/m^2/day	4 days/wk, 3 mo	Oral	6	Completed	NA	Mao et al.[32]
US	148	ATRA; 13-cRA	ATRA: 1 or 2 mg/kg/day; 13-cRA: 1 mg/kg/day	ATRA: 4 days/wk, 6 mo; 13-cRA: daily, 6 mo	Oral	18	Completed	NCT00000621	Roth et al.[33]
Multinational*	492	Palovarotene	5 mg/day	Daily, 24 mo	Oral	24	Completed	NCT00413205	Jones et al.[34]
Multinational†	262	Palovarotene	5 mg/day	Daily, 12 mo	Oral	12	Completed	NA	Stolk et al.[35]

*Includes the UK and Switzerland. †Includes Australia, Belgium, Canada, Denmark, the Netherlands, New Zealand, Spain, Sweden, and the UK.
COPD: chronic obstructive pulmonary disease; ATRA: all-*trans* retinoic acid; NA: not available; 13-cRA: 13-cis retinoic acid.

retinoids was carried out in the early 2000s[32]. Twenty patients with severe emphysema were enrolled and randomly assigned into two groups. They were treated with either 3 months of ATRA (Vesanoid; Roche Laboratories Inc.; 50 mg/m^2/day for 4 consecutive days out of every week) or placebo, followed by a 3-month crossover period. In this phase 1 clinical trial, the investigators successfully demonstrated the safety of the drug as no dose-limiting toxicity was observed. However, the physiological and computed tomography (CT) measurements did not change appreciably in response to the therapy. And also, interestingly, the plasma level of ATRA varied considerably among the subjects and decreased significantly over time since it induced its own catabolic enzymes to various degrees in each patient.

In 2006, another clinical trial to assess the feasibility of retinoids for the treatment of emphysema, the FORTE study, was published (NCT00000621)[33]. In that study, 148 patients from five university hospitals were recruited and randomized to receive ATRA at either low dose (1 mg/kg/day for 4 days/wk) or high dose (2 mg/kg/day for 4 days/wk), 13-*cis* retinoic acid (1 mg/kg/day, daily), or placebo for 6 months, followed by a 3-month crossover phase. Subsequently, they were observed for an additional 9 months before the final assessment. In the trial, retinoids were proven to be safe as the drug-related AEs were generally mild. However, no treatment was associated with an overall improvement in pulmonary functions, CT density mask scores, or quality of life indicators at the end of the first 6 months. Interestingly, against general expectations, the diffusing capacity of carbon monoxide (DL_{CO}) was decreased at 6 months in subjects treated with high-dose ATRA. Moreover, it recovered significantly with time after discontinuation of the drug, and this observation was correlated with the plasma level of ATRA. Although it is unclear whether this finding is a measure of lung remodeling or an indicator of drug-related toxicity, it suggested that ATRA might have a physiologic effect on emphysema-damaged lungs.

Between 2011 and 2012, two clinical trials using a γ-selective retinoid agonist (palovarotene) were released[34,35]. In a clinical trial conducted by the TESRA (treatment of emphysema with a selective retinoid agonist) group, the researchers recruited 492 patients with smoking-induced emphysema and randomly assigned them into two distinct groups. Participants were administered either palovarotene (5 mg/kg/day) or placebo and were observed for 2 years. The drug was generally well tolerated but failed to show any significant efficacy in terms of physiological and CT indicators. In *post-hoc* analysis, however, palovarotene showed significant alleviation of declining pulmonary functions such as DL_{CO} and FEV_1 in patients with lower lobe predominant emphysema, indicating a possible disease-modifying effect of the drug in selected patients. In another clinical trial carried out by the REPAIR (retinoid treatment of emphysema in patients on the α_1-antitrypsin international registry) group, the investigators enrolled 262 patients with emphysema caused by α_1-antitrypsin deficiency from 16 centers in 10 countries. Participants were randomly assigned to either palovarotene (5 mg/day) or placebo group and were observed for 12 months. However, similar to the TESRA study, palovarotene failed to show any significant drug-related benefits, although it was generally well tolerated.

Future Perspectives on Lung Regeneration Therapy in COPD

As discussed above, lung regeneration therapy is attracting considerable attention as a novel therapeutic approach for COPD. Considering the achievements of current clinical trials, the administration of stem cells and/or stimulating agents to patients with moderate-to-severe COPD appears to be safe and tolerable[29-35]. Moreover, it may have disease-modifying effects in selected patients. However, despite the advances made in recent decades, we are still far from our goals and there are substantial hurdles to be overcome.

First of all, although it has been investigated in some clinical trials, the safety of lung regeneration therapy remains unclear and needs to be assessed in better-designed trials. It is well known that stem cells have tumorigenicity properties and can cause benign or malignant tumors and, like a double-edged sword, the recent efforts to boost the power of stem cells might amplify the drawbacks as well. Moreover, there are a number of reports linking retinoids to lung cancer in certain individuals, such as active smokers[36], and evidence indicates that patients who have retinoids are frequently associated with dyslipidemia requiring long-term management[37], which can cause and aggravate vascular diseases, especially in combination with smoking. Given that most COPD patients are ex- or current smokers and the possibility of long-term fatal AEs including cancer and vascular diseases, it is vital to carefully re-evaluate the safety profiles of lung regeneration therapy in larger and longer trials.

In addition to the safety issue, the efficacy issue is another important hurdle that should be surpassed. In contrast to animal models that have shown promising results, all of the current clinical trials aimed at regenerating the damaged lung in human subjects, irrespective of the modality, have been unsuccessful in terms of efficacy[29-35]. Since the sources of these discrepancies between the laboratory and clinical results are unclear, it is crucial to assess the possible causes of the recent disappointing results in clinical fields and to identify the possible ways to surmount the barriers.

First, the different pathophysiology and chronicity between human and animal subjects might contribute to the discrepancies in efficacy. In contrast to the animal models, which are usually acquired by acute or subacute exposures to a single offending agent, almost all patients with COPD are chronically exposed to multiple noxious substances, such as cigarette

smoke and air pollution. With the growing evidence of stem cell aging, which is characterized by a diminished functional capacity and increased stem cell senescence, it is currently suspected that endogenous stem cells and pulmonary microenvironments are exhausted due to recurrent and prolonged pulmonary insults in patients with COPD[38]. However, although this hypothesis appears to be plausible, it is supported by only a limited number of experimental studies. Further investigations to provide more direct evidence, such as proving the diminished quantity and quality of stem and progenitor cells in human COPD subjects, are required. Furthermore, studies on how to recover the weakened cellular reservoir in clinical practice are needed to boost the efficacy of lung regeneration therapy.

Second, the issues related to the quality control of administered stem cells and the standardization of the treatment protocol also need to be settled. A huge number of stem cells is required for lung regeneration therapy, but successive *in vitro* passaging frequently cause cytogenetic abnormalities that may result in loss of cellular functions and cell transformations. These factors might be associated with the diminished effects of the administered stem cells and thus the surveillance and correction of these aberrant cells are necessary. Moreover, considering a recent fascinating report showing the different regenerative efficacy of stem cells according to donor age in mice with bleomycin-induced pulmonary fibrosis[39], the importance of stem cell quality control cannot be emphasized enough. Besides the stem cells, other factors potentially influencing the clinical outcome should also be controlled and standardized. Because we are at the first step of this newly expanding field, the current dosing schedule, cell type and origin, and route of delivery are chosen empirically based on clinical trials of other diseases. Furthermore, these uncertainties involve not only cell therapies, but also drug therapies. For example, in previous studies using ATRA, the dosing schedule was determined empirically, although wide intra- and inter-subject variabilities in plasma ATRA levels were observed[32,33]. More studies to determine the optimal settings and protocols for lung regeneration therapy are required.

In addition, candidate molecules that have demonstrated safety and efficacy in animal models should be more readily translated to clinical trials. As discussed above, there are a number of candidates with proven disease-modifying effects in animal models, such as retinoids, FGF-2, HGF, and even estrogen[20-27]. However, only retinoids have been tried in the clinical field[32-35]. Further clinical trials of other emerging candidates are required to diversify our therapeutic options. Moreover, given the possibility that the current unsatisfying results are due to exhausted and damaged stem cells[38], strategies to support the impaired stem cells also need to be developed. For example, one option being investigated involves culturing the stem cells under specific conditions such as hypoxia to potentiate their efficacy (NCT01849159). Hybrid or recombinant cells combined with proven stimulatory molecules such as FGF-2 and HGF should also be investigated. Combinations of candidate molecules might help to enhance the regenerative potency of lung. However, the safety and feasibility of these procedures should be firmly guaranteed in experimental trials before clinical applications.

Conclusion

Given the achievements to date, the goal of regenerating diseased lungs and curing destructive pulmonary diseases such as COPD seems to be within our grasp. Although there are numerous obstacles to be overcome, lung regeneration therapy is expected to be translated safely and effectively from the laboratory to the bedside.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

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