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A review for natural polysaccharides with anti-pulmonary fibrosis properties, which may benefit to patients infected by 2019-nCoV

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ARTICLE INFO

Keywords:
Polysaccharides
Pulmonary fibrosis
Mechanisms
Corona virus disease 2019

ABSTRACT

Pulmonary fibrosis (PF) is a lung disease with highly heterogeneous and mortality rate, but its therapeutic options are now still limited. Corona virus disease 2019 (COVID-19) has been characterized by WHO as a pandemic, and the global number of confirmed COVID-19 cases has been more than 8.0 million. It is strongly supported for that PF should be one of the major complications in COVID-19 patients by the evidences of epidemiology, viral immunology and current clinical researches. The anti-PF properties of naturally occurring polysaccharides have attracted increasing attention in last two decades, but is still lack of a comprehensively understanding. In present review, the resources, structural features, anti-PF activities, and underlying mechanisms of these polysaccharides are summarized and analyzed, which was expected to provide a scientific evidence supporting the application of polysaccharides for preventing or treating PF in COVID-19 patients.

1. Introduction

Pulmonary fibrosis (PF) is a worldwide disease, along with progressive and permanent fibrotic “scar” in alveolus and bronchioles of the lung parenchyma (Jacob et al., 2018), which could be induced by genetic susceptibility and various environmental risk factors including virus, bacteria, cigarette smoke, wood dust, stone dust etc. (Martinez et al., 2017; Richeldi, Collard, & Jones, 2017). Interana and pirenidone were approved by the USA Food and Drug Administration for suppressing the progression of pulmonary fibrosis (Raghu et al., 2015). In clinical, glucocorticoid (Ozaki et al., 1982), prednisone (Bickerman, Beck, & Barach, 1955), cyclophosphamide (Miniati & Cerinic, 2007), and other immunosuppressive modulators have also been used for delaying the degree of pulmonary fibrosis and prolonging the survival of patients. However, all of them cannot improve the risks of incidence and mortality of pulmonary fibrosis (Canestaro, Forrester, Raghu, Ho, & Devine, 2016), but cause a large economic burden for patient (Raimundo et al., 2016). Therefore, it is still urgent developing new therapies and drugs for treating pulmonary fibrosis.

The transformation of epithelial into myofibroblasts and the amount of extracellular matrix (ECM) generated by fibroblasts are considered as crucial developmental milestones in pulmonary, wherein a pivotal fibrogenic cytokine TGF-β is aberrantly expressed, which in turn triggers epithelial-mesenchymal transition (EMT) process thereby enhancing ECM deposition mediated by both Smad dependent and independent pathways (Bale, Venkatesh, Sunkoju, & Godugu, 2018; Liu, Lu, Kang, Wang, & Wang, 2017). Furthermore, some other mechanisms have been also found to be imperative in pulmonary fibrosis progression including inflammation, oxidative stress, deregulated ECM and EMT signaling (Liu et al., 2017).

Human coronaviruses were first described in the 1960s for patients with the common cold, which in the past two decades have caused severe acute respiratory syndrome (SARS), Middle East respiratory syndrome (MERS), as well as COVID-19 now. A typical clinical feature...
associated with SARS is pulmonary fibrosis, which resulted in a high mortality and a low quality of life for recovered patients (Venkataraman & Frieman, 2017). Several long-term follow-up studies have demonstrated that more than 20% of SARS survivors exhibited radiographic evidence of lung fibrotic changes (Hui et al., 2005; Xie et al., 2005). Recently, a retrospective analysis on the pulmonary computed tomographic imaging of fifty patients with COVID-19 pneumonia also observed the formation of fibrotic stripes during their rehabilitation period (Xu et al., 2020). It will be a high probability that pulmonary fibrosis is one of the major complications in COVID-19 patients. Therefore, there will be a huge need for effective and safety agents and therapeutic strategies for treating pulmonary fibrosis induced by COVID-19 (Wang et al., 2020).

With the broad spectrum of bioactivities and highly safety, polysaccharides have attracted more and more attention (Muhama, Zulkifli, Selvakumaran, & Lazim, 2019; Shi, 2016). In addition, several natural polysaccharides, such as polysaccharides-K (PSK) and polysaccharides-peptide (PSP) from Coriolus versicolor (Kidd, 2000), and fucoidan (Wang, Geng, Yue, & Zhang, 2019), have been applied in clinical for treating diseases and improving health. In fact, many studies have been conducted to reveal the anti-pulmonary fibrosis activities and underlying mechanisms of naturally occurring polysaccharides from medicinal plants, seaweed, and edible fungi. However, to the best of our knowledge, a comprehensively understanding of these anti-pulmonary fibrosis natural polysaccharides is still limited.

Thus, in present review, the available information about these natural polysaccharides with anti-pulmonary fibrosis activities were collected by searching in the related topics of “pulmonary fibrosis” and “polysaccharides” in the databases of Scopus (http://www.scopus.com) and China National Knowledge Infrastructure (https://www.cnki.net). In addition, the resources, structural features, physiological activities, as well as underlying mechanisms of these polysaccharides were systematically summarized and analyzed, which was expected to provide a scientific evidence for the application of polysaccharides for preventing or treating pulmonary fibrosis in COVID-19 patients.

2. Mechanisms underlying the anti-pulmonary fibrosis activities of natural occurring polysaccharides

2.1. Transforming growth factor β1 (TGF-β1)-Smad2/3 axis

TGF-β1 overproduction has been recognized as the most relevant element related to the progress of pulmonary fibrosis, which plays a crucial role in the induction of EMT, and stimulates differentiation, proliferation and migration of immature fibroblasts, as well as induces phenotypic conversion of fibroblasts into myofibroblasts (Walton, Johnson, & Harrison, 2017). Smad2 and Smad3 are the main transcription factors for TGF-β1 signals, which are phosphorylated and translocated into the nucleus upon ligand binding (Fig. 1).

DOP is a neutral heteropolysaccharides isolated from Dendroboon officinale, which consisted of Man and Glic (in a mole ration of 5.9:1) with an average molecular weight (Mw) at about 1.78 × 10^5 Da, and had a partial structure of O-acetylated glucamannan with β-configuration in pyranose sugar forms (He et al., 2016). It has been demonstrated that DOP could significantly attenuate bleomycin (BLM)-induced up-regulation of TGF-β1 expression and Smad2/3 phosphorylation in the rat lung tissues, and further suppress the transformation of rat type II alveolar epithelial cells into myofibroblasts (Chen et al., 2018) (Fig. 1).

Ginsan, a polysaccharide isolated from the roots of Panax ginseng with an average Mw of 1.5 × 10^6 Da, composed mainly of Glic and Gal (over 90 %, w/w), and 5–8 % Man and Ara (Ahn et al., 2011; Lee et al., 1997). In mouse lung fibroblasts (NIH/3T3 cells) or human lung fibroblasts (IMR-90 and WI-38 cells), ginsan could significantly reduce the phosphorylation of Smad2 and Smad3 induced by TGF-β via inhibiting induction of R-Smads but not inhibitory Smads (Smad6 or Smad7). In addition, ginsan significantly reduced the phosphorylation of ERK and Akt induced by TGF-β, but did not affect the phosphorylation of either JNK or p38, which indicated that ginsan downregulated both Smad-dependent and independent signaling pathways induced by TGF-β. Furthermore, ginsan could obviously reduce the increases in protein expression of TGF-β receptors (TβRI and TβRII), and block the decrease in protein expression of TβRIII, a known co-receptor of TβRII (Ahn et al., 2011) (Fig. 1).

ERK signaling is another important mechanism implicated in the process of TGF-β1-induced EMT during the pulmonary fibrosis. Wang, Zhang et al. (2019) had demonstrated that a sulfated low-molecular weight fucoidan isolated from brown seaweed (LMWF) can ameliorate TGF-β1-induced EMT both in vivo and in vitro models of pulmonary fibrosis by downregulating ERK signaling, which significantly inhibited the over expression of TGF-β1 and p-ERK1/2 in mice induced by BLM, as well as suppressed the over expression of p-ERK1/2 in TGF-β1-induced A549 cells (Fig. 1).

The high binding affinity for TGF-β1 have revealed to be closely related to the anti-pulmonary fibrosis activities of MS80, a marine-derived sulfated oligosaccharide (1→4 α-D-glucose) isolated from seaweed with the average Mw of 8 × 10^3 Da, which via competitively inhibiting the heparin/HS-TGF-β1 interaction, can arrest TGF-β1-induced human embryonic pulmonary fibroblast (HEPF) cell proliferation, collagen deposition and matrix metalloprotease activity, as well as deactivate both the ERK and p38 signaling pathways (Jiang & Guan, 2009).

2.2. DANCR/AUF-1/FOXO3 regulatory axis

The differentiation antagonizing non-protein coding RNA (DANCR) is a newly identified long noncoding RNA (IncRNA) with pivotal roles in cell proliferation, migration, invasion, and stem cell differentiation, which was shown to promote EMT progression and invasion capability of malignant cells (Yang, Sun, Gao, Meng, & Yang, 2018). DANCR co-operated with an adaptor protein AU-binding factor 1 (AUF1) has been demonstrated to regulate EMT and fibrogenesis by upregulating FOXO3 protein levels without influencing its mRNA expression. The root of Angelica sinensis is a well-known traditional Chinese medicine, which has been used for thousands of years to prevent and treat various diseases. The polysaccharides have been proved as one of the major effective ingredients in A. sinensis, and more than 30 polysaccharides have been identified from A. sinensis, most of which were hetero-polysaccharides (Jin, Zhao, Huang, Xu, & Shang, 2012). Treatment with...
total polysaccharide of *Angelica sinensis* (ASP) exhibited significant downregulatory effects on the expression of DANCR, which in turn represses AUFI-mediated FOXO3 translation to suppress the EMT and pulmonary fibrosis both in *in vitro* and *in vivo* (Qian, Cai, Qian, Wang, & Zhang, 2020) (Fig. 2).

2.3. Antioxidant ability

The pathological progresses of pulmonary fibrosis are complex, but oxidative stress injury plays an important role (Cameli et al., 2020). FYGL-1 is a neutral heteropolysaccharide isolated from *Ganoderma lucidum*, consisted of Gal, Rha and Glc (in a mole ration of 1.00:1.15:3.22) with an average Mw of $7.8 \times 10^6$ Da, and the backbone structure consisted mainly of 1,2-linked-β-LRha, 1,3,6-linked-α-D-Galp, 1,2,6-linked-α-D-Glc and 1-linked-α-D-Glc (Pan et al., 2012). Treatment with FYGL-1 (at 100 and 300 mg/kg for 28 days) led to a markedly reducing in the pulmonary index, inflammatory cell infiltration and collagen deposition in rats induced by BLM, which was associated with increased levels of glutathione, glutathione peroxidase, catalase and superoxide dismutase and decreased contents of malondialdehyde and hydroxyproline in the lung tissues (Chen et al., 2016).

Fig. 2. Total polysaccharides from *A. sinensis* (ASP) suppress the pulmonary fibrosis via DANCR/AUF-1/FOXO3 regulatory axis.

2.4. Reducing the recruitment of macrophages and neutrophil in the lung tissues

Pulmonary macrophages express several fibrotic mediators and play important roles in lung injury healing and fibrosis, which are currently classified into two phenotypes, classically activated macrophages (M1) with secretion of Th1-related cytokines (including TNF-α, IL-1β, and IL-6) and alternatively activated macrophages (M2) with release Th2-related cytokines such as IL-10 and IL-13. However, the migration and invasion ability of both M1 and M2 macrophages are promoted by monocyte chemotactic protein 1 (MCP-1), a chemokine mainly expresses in alveolar macrophages and participates in the chemotaxis of cells. Zhou et al. (2020) found that the total polysaccharides extracted from *Ophiocordyceps lanpingensis* (POL) could suppress the increasing level of MCP-1 expression during the process of BLM-induced pulmonary fibrosis, and consequently reduce the increased amount of M1 and M2 macrophages, as well as the expression levels of relative cytokines. Finally, the generation of myofibroblasts was alleviated based on the reduced expression of α-smooth muscle actin (α-SMA) protein (Fig. 3). According to Yu et al. (2018) studies, the increased expression of TIMP-1, CXCL1, MCP-1, MIP-2, and interleukin-1 receptor antagonist (IL-1RA) were strongly associated with radiotherapy-induced lung fibrosis through the induction of M2 macrophages and neutrophils. However, the administration of fucoidan isolated from *Sargassum hemiphyllum* (FSH) in irradiated mice significantly attenuated these cytokines expression in the collected pleural fluid and reduced pleural fluid-induced collagen expression in fibroblasts, which were correlated with the reduction of neutrophil and macrophage infiltration in lung tissues (Fig. 3).

Fig. 3. Natural polysaccharides suppressed pulmonary fibrosis via reducing the recruitment of macrophages and neutrophils in the lung tissues.
| Name/source | Structural characterization | Experimental model | Effects | Reference |
|-------------|-----------------------------|--------------------|--------|-----------|
| ASP/Asplenium sinensis | Mw: 5.1 – 740 kDa; Containing Glc, Gal, Ara, Man, Fuc, Xyl; Linkage: backbone →4)-α-D-Glcβ-(1→ with α-1,6-Galβ, β-1,4-Araf residues at a branching position of O-6; backbone →3)-α-D-Glcβ-(1→backbone→4)-α-D-Glcβ-(1→2)-α-D-Rhapβ-(1→ with β-1,6- and β-1,4-Galp and α-1,5-Araf at a branching position of O-4; backbone→4)α-D-Glc→(1→6)α-D-Glcβ-(1→4)-α-D-Glcβ-(1→4)-α-D-Glcβ-(1→. | In vivo: BLM-induced Sprague-Dawley rats; In vitro: TGF-β1 induced alveolar type II epithelial (RLE-6TN) cells. | In vivo, ASP administration suppressed the increasing of collagen deposition accompanying with restoring collagen-1 protein levels by inhibiting the DANC/RAF1/FOXO3 pathway in BLM-induced PF model rats. In vitro, ASP restored intercellular junction and spindle-like structure with downregulation of growth rate, migratory ability and α-SMA expression and upregulation of E-cadherin in RLE6TN cells with TGF-β1 treatment. | Jin et al. (2012), Luo et al. (2017), Qian et al. (2020) |
| RAP/Astragalus radix | Mw: 5.6 – 7600 kDa; Containing Glc, Rha, Ara, Xyl, Man, GaL, GaL; Linkage: backbone 1,2-Rha, α-1,4-Gkpa, β-1,3,6-Galp, α-T-Arap with 1,2,4-Rhap backbone→4)-α-D-Glcβ-(1→6)-α-D-Glcβ-(1→4)-α-D-Glcβ-(1→4)-α-D-Glcβ-(1→. | In vivo: BLM-induced Wistar rats; In vivo: Astragalus polysaccharides administration inhibited inflammation of pulmonary alveoli and upregulated the serum level of IFN-γ with downregulated the serum levels of IL-4 and TNF-α in BLM-induced PF model rats. | In vivo, administration of PBS improved pulmonary histomorphology and histopathology with inhibiting collagen deposition, decreased pulmonary index and hydroxyproline contents in BLM-induced PF model rats. | Li et al. (2011), Yin et al. (2012) |
| PBS/B. striata | Mw: 401.3 kDa; Containing Man and Glc. | In vivo: BLM-induced adult male Sprague Dawley rats; | In vivo, After DOP administration in BLM-induced PF model rats, the pulmonary index, hydroxyproline expression and serum TGFβ1 concentrations were lower, with the improvement of histopathology, morphology and the inhibition of neutrophil-dominant inflammation, expression on mRNA and protein of TGFβ1, Smad2, Smad3 and α-SMA protein expression. | Guo et al. (2016) |
| DOP/D. officinale | Mw: 178 kDa; Containing Man and Glc (5.9:1); Linkage: acetylated glucomannan with β-α configuration in pyranose. | In vivo: BLM-induced adult male Sprague Dawley rats; In vitro: TGF-β1 induced rat type II alveolar epithelial cells. | In vitro, DOP decreased the expression of α-SMA, Smad 2/3 protein, pSmad 2/3 protein and the synthesis of collagen 1 and fibronectin within increasing the expression of E-cadherin. | Chen et al. (2018), He et al. (2016) |
| BHP-1/L. davidi var. unicolor | Mw: 1.91 × 10^7 Da; consisted of Glc and Man in a relative molar ration of 5.9: 2.0; Backbone: mainly contained α-1,4-linked α-Glc and β-1,4-linked β-Manp; the branches were probably linked at the O-2 and or O-3 of the Man and Glc residues, with Tα-o-Gkp as a terminal structure. | In vivo: BLM-induced SPF Kunming mice. | Combined with bone marrow mesenchymal stem cells transplantation, BHP-1 decreased the pulmonary index and improved the pulmonary histopathology and collagen deposition with downregulating the protein expression of TNF-α and NF-κB in BLM-induced PF model mice. | Luo et al. (2013), Hui et al. (2019), Hu et al. (2019) |
| TPOB/O. basilicum | Containing Glc, Xyl, GaL; Linkage: backbone β-α-Gkp with α-β-Xylp; β-α-Galp-(1→2)-α-β-Xylp at a branching position of O-6. | In vitro: TGF-β1 induced human A549 cells. | In vitro, TPOB changed less in morphology and reduced hydroxyproline contents with upregulating the expression levels of E-cadherin and downregulating the expression levels of Vimentin, α-SMA and COL1 in human A549 cells with TGF-β1 treatment. | Hoffman et al. (2005), Yan et al. (2017) |
| Ginsan/P. ginseng | Mw: 3.5 – 160 kDa; Containing Glc, Gal, Man, Ara; Linkage: backbone →5)-α-D-Arap(1,3)-α-D-Galp at a branching position of O-6. | In vivo: BLM-induced male CSLRI/6 mice; In vitro; TGF-β1 induced NIH3T3 cells and IMR-90 and WI-38 cells. | In vivo, Ginsan changed nothing in morphology and attenuated TGF-β expression in lung tissue with downregulating the levels of collagen and α-SMA in BLM-induced PF model mice. In vivo, Ginsan suppressed the expression of α-SMA, FN, procollagen type 1 and downregulated both Smad-dependent and -independent signaling pathways with modulates TGF-β receptor levels (downregulating the TβRI and TβRII protein activity). | Ahn et al. (2011), Sun (2011) |
SMA and downregulation of E-cadherin expression in RLE-6TN cells. The similar inhibitory effect of ASP was also observed in TGF-β1-induced HEPF cell assay, which showed that ASP (at 25 μg/mL) significantly reduced the elevated content of hydroxyproline and the upregulated protein expressions of α-SMA and CTGF (Luo et al., 2017).

Chen et al. (2018) have demonstrated that DOP (a neutral heteropolysaccharides isolated from D. officinale) treatment significantly ameliorated indices for both pulmonary inflammation and fibrosis in a BLM-induced pulmonary fibrosis model in rats, which can significantly lower BLM-induced elevations of pulmonary index, total cell numbers and differential neutrophil counts in bronchoalveolar lavage fluid (BALF), as well as attenuate the average scores of BLM-induced histopathological changes, and decrease the BLM-induced increase in hydroxyproline content in lung tissue. A polysaccharide isolated from rhizome of Bletilla striata (PBS), with an average Mw of 4.01 × 10^5 Da and mainly consisted of Man and Glc, also exhibited the similar anti-pulmonary fibrosis activities in BLM-induced SD rat model, which obviously improved the BLM-induced histopathological changes of lung tissues and significantly lowered the pulmonary index and hydroxyproline content (Guo et al., 2016). In addition, the PBS did not cause any side-effect in vivo with oral administration of 4000 mg/kg/day (He et al., 2017).

RAP was a water-soluble polysaccharide purified from Radix astrogalli and composed of Rha, Ara, Glc, Gal, and GaIα in a mole ratio of 0.03: 1.00: 0.27: 0.36:0.30 with an average Mw at 1.3 × 10^5 Da. The backbone of RAP consisted of 1,2,4-linked Rha, 1,4-linked Glcα, 1,4-linked Galα, β-1,3,6-linked Galp with branched at O-2 of the 1,2,4-linked Rha, and O-3 of 1,3,6-linked Galp, while the side chains mainly consisted of α-1,3-Araf and α-1,5-linked Araf with O-3 as branching points and trace Glc and Gal (Yin et al., 2012). The potential inhibitory effects of RAP against BLM-induced pulmonary fibrosis had been demonstrated in a Wistar rats model, which significantly inhibited the alveolar inflammation, downregulated the serum levels of interleukin-4 (IL-4) and tumor necrosis factor-α (TNF-α), but upregulated the serum level of interferon gamma (IFN-γ) (Li et al., 2011).

A total polysaccharides (TPRH) was isolated and purified from Radix hedyari, mainly composed of Ara, Man, Gal and Glc in a mole ratio of 0.42: 0.53: 0.34: 1.00, with an average Mw of 1.9 × 10^5 Da, and the purity of which was 94.3 % (Lei, Zhao, Wang, Yao, & Ding, 2008). Treatment with TPRH (200 mg/kg), combined with prednisone (3 mg/kg), exhibited a significant inhibitory effect against alveolar inflammation in BLM-induced Wistar rat pulmonary fibrosis, and further alleviated fibrosis degree, histopathology changes, as well as the expression levels of collagen and TGF-β1 with reducing the contents of hyaluronic acid and laminin in lung tissues of rats exposed to BLM (Lei et al., 2008). Meanwhile, TPRH demonstrated to possess the similar ameliorate effects on pulmonary histopathological morphology, inflammation of pulmonary alveoli and proliferation and deposition of collagen fibrils with reducing the contents of hyaluronic acid and laminin in lung tissue of BLM-induced pulmonary fibrosis model rats (Su et al., 2016).

In both BLM-induced C57BL/6 mice and TGF-β1-induced murine and human normal lung fibroblasts models, ginsan (a polysaccharide isolated from the roots of P. ginseng) exhibited the significant inhibitory effects against pulmonary fibrosis, which changed nothing in morpology, attenuated TGF-β expression, and reduced the levels of collagen and α-SMA in lung tissues, as well as suppressed the expression of α-SMA, fibroconnectin (FN) and collagen type 1 (Ahn et al., 2011). In addition, administration of ginsan (6 g/day) showed no significant adverse effect in a 14-week randomized, placebo-controlled, double-blind clinical trial (Cho, Son, & Kim, 2014).

BHP-1, a polysaccharide isolated from the bulbs of Lilium davidii var. unicolor, was mainly consisted of Glc and Man in a mole ration of 5.9: 2.0, with the average Mw of 1.93 × 10^5 Da. The backbone of BHP-1 mainly contained α-1,4-linked Glcα and β-1,4-linked Mannp, and the branches were probably linked at the O-2 and O-3 of the Man and Glc.
| Source/Name | Experimental model | Source of extraction | Effect of combination of FSH, BHP-1 or BMSCs transplantation | Other effects |
|-------------|--------------------|---------------------|--------------------------------------------------------------|--------------|
| L. japonica | In vivo, BLM-induced pathogen-free Kunming mice | Mw: 0.8kDa; containing rich L-fucose and sulfated ester groups with some D-xylose, D-galactose, D-mannose, glucuronic acid and a mixture of fatty acid methyl esters. | Reduced lung histopathology and hydroxyproline content, alleviating the expression levels of E-cadherin and downregulating the expression levels of vimentin, α-SMA, type I collagen and fibronectin-EDA (Fn-EDA) (Fan et al., 2017). RSA, an acidic heteropolysaccharide isolated from *Rhodiola sachalinensis*, was mainly consisted of Ara, Rha, Xyl, Glc, Gal, and GaL in a mole ratio of 1.00: 3.23: 0.26: 0.34: 0.84: 10.24, which showed a potential suppression of pulmonary fibrosis in TGF-β1-induced human A549 cells, with improving morphological change and reduction in hydroxyproline contents, upregulating the expression levels of E-cadherin and downregulating the expression levels of Vimentin, α-SMA, COL-1 and fibronectin-EDA (Fn-EDA) (Han et al., 2002; Li, Gao, Zhao, & Hong, 2016). | In addition, the crude polysaccharides from three different complex prescriptions of Traditional Chinese Medicine (Fei Kang Ling, Gua Lou Xie Bai decoction, and Yu Ping Feng) also exhibited potential anti-pulmonary fibrosis activities in different in vitro and in vivo assays (Gao, 2016; Jiang, 2008; Xu et al., 2014). |
| MS80 | In vivo, BLM-induced male Balb/c mice, lung fibrotic histopathology and lung hydroxyproline content was significantly improved. | MS80 is a marine-derived sulfated oligosaccharide (1→4 α-D-glucopyranosyl-(1→3)-L-fucopyranosyl-(1→6)-D-glucopyranosyl, with sulfated residues and some D-xylose, D-galactose, D-mannose, glucuronic acid and a mixture of fatty acid methyl esters. | In vitro, MS80 inhibited the combining capacity of TGF-β1 with heparin examined by surface plasmon resonance and the proliferation, collagen deposition and matrix metalloproteinase activity of HEPF cells with TGF-β1 or BLM. | 3.2. Polysaccharides from algae |
| 3.2. Polysaccharides from algae |
| MS80 is a marine-derived sulfated oligosaccharide (1→4 α-D-glucopyranosyl-(1→3)-L-fucopyranosyl-(1→6)-D-glucopyranosyl, with sulfated residues and some D-xylose, D-galactose, D-mannose, glucuronic acid and a mixture of fatty acid methyl esters. | In vitro, MS80 significantly inhibited the proliferation, collagen deposition and matrix metalloproteinase activity of HEPF cells with TGF-β1 or BLM. | | |
| LMWF, another sulfated polysaccharide extracted from brown seaweed, has also demonstrated to possess a significant inhibition of BLM-induced pulmonary fibrosis in C57BL/6 mice, as evidenced by improved lung histopathology and hydroxyproline content, attenuated the expression levels of TGF-β1 in BALF and lung tissue, and the lung EMT phenotype including the expression trends of E-cadherin, α-SMA and fibronectin (Wang et al., 2019) (Table 2). Furthermore, LMWF displayed no mutagenicity by either the bacterial reverse mutation or chromosomal aberration assays in vitro at 5000 μg/mL, as well as no toxicological indications in vivo by repeated oral administration of LMWF (2000 mg/kg/day) for 28 days (Hwang, Yan, Lin, Li, & Lin, 2016). In addition, LMWF could significantly inhibited the morphologic alterations and proliferation of A549 cells induced by TGF-β1, and attenuated TGF-β1-induced EMT phenotype such as less expression of E-cadherin and over expression of vimentin, α-SMA and fibronectin (Wang et al., 2019). | |
Table 3

| Name | Source | Structural characterization | Experimental model | Effects |
|------|--------|----------------------------|-------------------|---------|
| FYGL-1 | *G. lucidum* | Mw: 7.8×10^5 Da, 25-30% polysaccharide content | In vivo: BLM-induced male mice | increased the pulmonary index, inflammatory cell infiltration and collagen deposition |
| FMP-1 | *M. esculenta* | Mw: 4.7×10^3 Da, 1,4-linked Glcα, 1,6-linked Galβ, 1,3-linked Glcα | In vitro: H2O2-induced human alveolar epithelial cells A549 | inhibited pro-inflammatory and pro-fibrogenic factors, upregulated the expression levels of pro-inflammatory and pro-fibrogenic factors |
| POL | *O. longipes* | Mw: 4.7×10^5 Da, 1,4-linked Glcα, 1,6-linked Galβ | In vivo: BLM-induced male mice | significantly ameliorated the pulmonary index, histopathological changes, and collagen deposition |

3.3. polysaccharides from fungi

Four kinds of natural polysaccharides from fungi were collected in the present review. The resources, structural features, and anti-pulmonary fibrosis activities of these polysaccharides were displayed in Table 3.

Polysaccharides from *Cordyceps* (polysaccharide content > 64 %, average Mw < 2×10^3 Da) exhibited a significant inhibition of pingyangmycin-induced pulmonary fibrosis in mice, which could increase the IL-1RA levels, and reduce the hydroxyproline content and fibrosis area (Hu, Yang, Bai, & Fu, 2019). FYGL-1, a neutral heteropolysaccharide isolated from *G. lucidum*, showed the similar inhibitory effect on BLM-induced pulmonary fibrosis in rats, which could suppress the pulmonary index, inflammatory cell infiltration and collagen deposition, as well as ameliorate the oxidative stress in the lung tissue, such as upregulating the levels of glutathione, glutathione peroxidase, catalase, superoxide dismutase (SOD) and downregulating the levels of malondialdehyde (MDA) (Chen et al., 2016).

POL is a homogeneous polysaccharide isolated from *O. langipes*, primarily composed of Gal, Man and Glc in the mole ration of 5.30:13.38:81.31, with an average Mw of 3.2×10^3 Da (Zhou et al., 2020). POL could improve histopathological changes, collagen deposition and the accumulation of macrophages (via inhibiting the expression levels of NO2, CXCL10/IP10, MARCO and ST2), downregulate the expression levels of pro-inflammatory and pro-fibrogenic factors including TNF-α, IL-1β, IL-6, OSM, IL-10, IL-13, α-SMA, MCP-1 and TGF-β1, as well as inhibit oxidative stress (such as MDA production and SOD level) in BLM-induced pulmonary fibrosis mice (Zhou et al., 2020).

FMP-1 is a heteropolysaccharide from the fruiting bodies of *Morchella esculenta*, which has an average Mw of 4.7×10^3 Da and consists of Man, Gal and Glc in a mole ratio of 1.00:7.84:1.24, with the backbone made up of 1,4-linked Glcα and 1,6-linked Galβ (Cai et al., 2018). At the doses of 0−300 μg/mL, FMP-1 exhibited a potential anti-pulmonary fibrosis activity in H2O2-induced human alveolar epithelial A549 cells, without inhibitory effect on cells’ proliferation, which could attenuate H2O2-induced cytochrome c and Caspase-3 release to prevent cell apoptosis via inhibition of MDA and ROS levels, and enhanced the enzymatic activities of SOD and total antioxidant capacity (Li et al., 2018). In addition, the high degree of branching, low molecular weight and favorable structures (e.g. 1,4-linked Glcα, 1,6-linked Galβ and 1,6-linked Manp) are supposed to play an essential role in excellent antioxidant activities (Cai et al., 2018).

4. Conclusion

In present review, the structural features, physiological activities, and underlying mechanisms of 16 kinds of natural polysaccharides from plant, algae and fungi were systematically summarized and analyzed. The anti-pulmonary fibrosis activities of natural polysaccharides such as ASP, BHP-1, ginsan, TPRH, MS80, LMWF, and FYGL-1, have been demonstrated in different *in vivo* and *in vitro* assays, which can significantly ameliorate the pulmonary index, histopathological changes, and collagen deposition in rats or mice induced by BLM. And the main mechanisms of the anti-pulmonary fibrosis activities of these polysaccharides were targeting the TGF-β/Smad2/3 and DANCER/AUF-1/FOXO3 regulatory axis, and reducing the recruitment of macrophages and neutrophil. Furthermore, the sources of these polysaccharides are most edible materials, and the safety of ginsan, PBS and LMWF have been demonstrated in different *in vivo* and *in vitro* assays. Therefore, these polysaccharides maybe considerate as the safety and effective alternative agents for preventing or treating pulmonary fibrosis in COVID-19 patients.
Compliance with ethical standards
The authors declare that they have no conflict of interests.

Acknowledgements
This work was financed by the National Natural Science Foundation of China (No. 81602991) and GDAS’ Project of Science and Technology Development (No. 2019GDASYL-0103093).

References
Ahn, J. Y., Kim, M. H., Lim, M. J., Park, S., Lee, S. L. O., Yun, Y. S., & Song, J. Y. (2011). The inhibitory effect of Ginsn on TGF-beta mediated fibrotic process. Journal of Cellular Physiology, 228(5), 1241–1247.

Bale, S., Venkatesh, P., Sunkejou, M., & Goduge, C. (2018). An adiopathy: Withafenin A ameliorates in vitro and in vivo pulmonary fibrosis by modulating the interplay of fibrotic, matricellular proteins, and cytokines. Frontiers in Pharmacology, 9, Article 248.

Bickerman, H. A., Beck, G. J., & Barach, A. L. (1955). The use of prednisone (Meticorten) in pulmonary fibrosis and emphysema. Chest, 37, 46–54.

Cho, Y. J., Son, H. J., & Kim, K. S. (2014). A 14-week randomized, placebo-controlled, parallel-group study of Angelica Dahurica polysaccharides on anti-oxidant activity and anti-inflammatory activities. Phytomedicine, 21, 682–691.

Han, L. P., Liang, Z. Y., Zhang, L. P., Han, L. M., Gong, R. C., & Ma, X. H. (2002). Protective role of polysaccharides from the fruits of Schisandra chinensis. Carbohydrate Polymers, 49(3), 459–467.

Hoffman, M., Jia, Z. H., Pena, M. J., Cash, M., Harper, A., Blackburn, A. R., et al. (2018). Idiopathic pulmonary fibrosis. Lancet, 392(10100), 1992–2004.

Hua, P. A., Wang, P. D., Wu, C. W., & Wang, Y. F. (2017). Anti-oxidant activities of a hetero-polysaccharide from the fruits of Rubus idaeus L. Journal of Ethnopharmacology, 195, 325–331.

Kidd, P. M. (2000). The use of mushroom glucans and proteoglycans in cancer treatment. Alternative Medicine Review, 5(1), 4–27.

Lee, Y. S., Chung, I. S., Lee, I. R., Kim, K. H., Hong, W. S., & Yun, Y. S. (1997). Activation of multiple effector pathway of immune system by the antigenic immunomodulator acidic polysaccharide ginsin isolated from Panax ginseng. Anticancer Res., 17(3A), 525–531.

Lei, F. F., Zhao, J. X., Wang, X. X., Yao, B. T., & Ding, K. (2008). An experimental observation of pulmonary fibrosis in rats and its transforming growth factor beta induced by total hedyassin polysaccharide. Journal of Chinese Medicinal Materials, 31(3), 873–877.

Li, J., Zhang, Y., Liu, Y. Q., Li, J. T., Wei, S. C., Su, W., & Nie, L. (2011). Effects of Astragalus polysaccharides on level of cytokines and pathological structure of lung tissue in rat model of pulmonary fibrosis. Lishizhen Medicinae et Medicae Sinica, 22(7), 1684–1685.

Li, W., Cai, Z. N., Mehmood, S., Wang, Y. J., Zhang, Z. H., & Chen, Y. (2018). Polysaccharide FMP-1 from Morchella esculenta attenuates cellular oxidative damage in human alveolar epithelial A549 cells through PI3K/AKT/Nrf2/HCN-1 pathway. International Journal of Biological Macromolecules, 120, 865–875.

Luo, Y. L., Yao, Z. H., Zhao, L., & Hong, B. (2016). Study on rhodiolia polysaccharide against pulmonary fibrosis. Journal of Molecular Science, 32(1), 34–39.

Luo, Y., Li, F., Pan, L. R., & Wang, Y. F. (2017). The antifibrotic effect of puerarin ameliorates bleomycin-induced pulmonary fibrosis in mice by regulating Nrf2/Bach1 equilibrium. BMC Pulmonary Medicine, 17, Article 63.

Luo, Y. A., An, F. Y., Li, N. L., Liu, Y. Q., Li, C. H., Zhang, Y. H., & Chen, J. (2017). Effects of Angelica polysaccharides on the small molecules in expression of α-SMA in CTG in human embryonic lung fibroblast cells induced by TGF-β1. Pharmacology and Clinical Medicine of China, 33(3), 73–78.

Yang, Y. L., Wang, Y. L., Pan, Z. L., Li, N. L., Li, Y., Yan, X., & Zhang, L. (2013). The effects of polygalacturonic acid combined with BMSCs transplantation on expression of TNF-α and NF-kappaB in bleomycin-induced pulmonary fibrosis mice. Journal of Third Military Medical University, 35(5), 431–434.

Martinez, F. J., Collard, H. R., Pardo, A., Raghur, G., Richeldi, L., Selman, M., et al. (2015). Antifibrotic polysaccharide A. Nature Reviews Disease Primers, 1.

Mehmood, S., Yu, C. J., Raghur, G., Richeldi, L., Selman, M., et al. (2014). Idiopathic pulmonary fibrosis. Nature Reviews Disease Primers, 4.

Okatko, T., Nakayama, T., Ishimi, H., Kawano, T., Yasukoa, S., & Tohbara, E. (1982). Glucocorticoid receptors in bronchoalveolar cells from patients with idiopathic pulmonary fibrosis. The American Review of Respiratory Disease, 126(6), 968–971.

Pang, W. D., Wang, Q. Q., Chen, C. H., Teng, B. S., Wang, C. D., Xu, Z. K., et al. (2012). Protective effect of polysaccharide isolated from Ganoderma lucidum fruiting bodies on bleomycin-induced pulmonary fibrosis in rats. International Journal of Biological Macromolecules, 92, 278–281.

Qiao, Y. J., Sun, J. H., & Kim, K. S. (2014). A 14-week randomized, placebo-controlled, double-blind clinic trial to evaluate the efficacy and safety of Lingzhi polysaccharides (Y-75). Journal of Traditional Medicine, 12, Article 283.

Raimundo, K., Chang, E., Broder, M. S., Alexander, K., Zazzali, J., & Swigris, J. J. (2016). Novel signaling mechanisms in idiopathic pulmonary fibrosis via a DANCR/AUF-1/FOXO3 regulatory axis. American Journal of Respiratory and Critical Care Medicine, 192(9), 1147–1162.

Tamura, K., Chang, E., Broder, M. S., Alexander, K., Zazzali, J., & Swigris, J. J. (2016). Clinical and economic burden of idiopathic pulmonary fibrosis: A retrospective cohort study. BMC Pulmonary Medicine, 16.

Richeldi, L., Collard, H. R., & Jones, M. G. (2017). Idiopathic pulmonary fibrosis. Lancet, 389(10082), 1941–1952.

Shi, L. (2016). Bioactivities, isolation and purification methods of polysaccharides from natural products: A review. International Journal of Biological Macromolecules, 92, 36–48.

Sun, Y. K. (2011). Structure and biological activities of the polysaccharides from the leaves, roots and fruits of Panax ginseng CA Meyer: An overview. Carbohydrate Polymers, 85(3), 490–499.

Venkatesh, S. R., & Freiman, M. B. (2017). The role of epithelial growth factor receptor (EGFR) signaling in SARS-coronavirus induced pulmonary fibrosis. Antiviral Research, 143, 142–150.

Wei, L., Johnson, K. E., & Harrison, C. A. (2017). Targeting TGF-beta mediated TGFbeta signaling for the prevention of fibrosis. Frontiers in Pharmacology, 8, 189.

Wang, J., Wang, B. J., Jiang, Y. M., Chen, Y. M., Shen, C. L., & He, W. F. (2020). Advances in the research of mechanism of pulmonary fibrosis induced by Coronavirus Virus Disease 2019 and the corresponding therapeutic measures. Chinese Journal of Burns, 36(1).
Wang, J., Geng, L. H., Yue, Y., & Zhang, Q. B. (2019). Use of fucoidan to treat renal diseases: A review of 15 years of clinic studies. In Zhang (Vol. Ed.), Glycans and glycosaminoglycans as clinical biomarkers and therapeutics, Pt B: Vol. 163, (pp. 95–111).

Wang, L., Zhang, P., Li, X. P., Zhang, Y., Zhan, Q. Y., & Wang, C. (2019). Low-molecular-weight fucoidan attenuates bleomycin-induced pulmonary fibrosis: Possible role in inhibiting TGF-beta 1-induced epithelial-mesenchymal transition through ERK pathway. American Journal of Translational Research, 11(4), 2590–2602.

Xie, L. X., Liu, Y. N., Fan, B. X., Xiao, Y. Y., Tian, Q., Chen, L. G., … Chen, W. J. (2005). Dynamic changes of serum SARS-coronavirus IgG, pulmonary function and radiography in patients recovering from SARS after hospital discharge. Respiratory Research, 6(3–5).

Xu, L., Li, L. C., Zhao, P., Qi, L. W., Li, P., Gao, J., & Fei, G. H. (2014). Total polysaccharide of Yu Ping Feng protects against bleomycin-induced pulmonary fibrosis via inhibiting transforming growth factor-beta 1-mediated type I collagen abnormal deposition in rats. The Journal of Pharmacy and Pharmacology, 66(12), 1786–1795.

Xu, Y. H., Dong, J. H., An, W. M., Lu, X. Y., Yin, X. P., Zhang, J. Z., … Gao, B. L. (2020). Clinical and computed tomographic imaging features of novel coronavirus pneumonia caused by SARS-CoV-2. The Journal of Infection, 80(4), 394–400.

Yan, D. D., Tian, J. Z., Zhang, D., Hou, L., Li, L., Zhang, C. H., … Zhu, Q. J. (2017). Inhibitory effect of Basil polysaccharide on TGF-beta-induced epithelial-mesenchymal transition of AS549 cells. Traditional Chinese Drug Research and Clinical Pharmacology, 28(2), 154–159.

Yang, J. X., Sun, Y., Gao, L., Meng, Q., & Yang, B. Y. (2018). Long non-coding RNA DANCR facilitates glioma malignancy by sponging miR-33a-5p. Neoplasma, 65(5), 790–798.

Yin, J. Y., Chan, B. C. L., Yu, H., Lau, L. Y. K., Han, X. Q., Cheng, S. W., … Han, Q. B. (2012). Separation, structure characterization, conformation and immunomodulating effect of a hyperbranched heteroglycan from Radix astragali. Carbohydrate Polymers, 87(1), 667–675.

Yu, H. H., Ke, E. C., Chang, C. L., Yuan, K. S. P., Wu, A. T. H., Shan, Y. S., & Wu, S. Y. (2018). Fucoidan inhibits radiation-Induced pneumonitis and lung fibrosis by reducing inflammatory cytokine expression in lung tissues. Marine Drugs, 16(10).

Zhan, Y. F., An, X. N., Wang, S., Sun, M. J., & Zhou, H. M. (2020). Basil polysaccharides: A review on extraction, bioactivities and pharmacological applications. Bioorganic & Medicinal Chemistry, 28(1), 115179.

Zheng, C., Li, Y. S., Liu, H. B., Yuan, D., & Lu, B. R. (2001). Sulfoglycolipid from the marine brown alga Sargassum hemiphyllum. Journal of Asian Natural Products Research, 3(2), 117–122.

Zhou, S. B., Zhou, Y. C., Yu, J. J., Du, Y. X., Tan, Y., Ke, Y. M., … Ge, F. (2020). Ophiocordyceps lanpingensis polysaccharides attenuate pulmonary fibrosis in mice. Biomedicine & Pharmacotherapy, 126, Article 110058.