Since January 2020 Elsevier has created a COVID-19 resource centre with free information in English and Mandarin on the novel coronavirus COVID-19. The COVID-19 resource centre is hosted on Elsevier Connect, the company's public news and information website.

Elsevier hereby grants permission to make all its COVID-19-related research that is available on the COVID-19 resource centre - including this research content - immediately available in PubMed Central and other publicly funded repositories, such as the WHO COVID database with rights for unrestricted research re-use and analyses in any form or by any means with acknowledgement of the original source. These permissions are granted for free by Elsevier for as long as the COVID-19 resource centre remains active.
Role of inositol to improve surfactant functions and reduce IL-6 levels: A potential adjuvant strategy for SARS-CoV-2 pneumonia?

Antonio Simone Laganàa,b,⁎, Vittorio Unferb,c, Simone Garzon1, Mariano Bizzarrib,c,d

a Department of Obstetrics and Gynecology, “Filippo Del Ponte” Hospital, University of Insubria, Varese, Italy
b The Experts Group on Inositol in Basic and Clinical Research (EGOI), Italy
b Systems Biology Group Lab, “La Sapienza” University, Rome, Italy
d Department of Experimental Medicine, “La Sapienza” University, Rome, Italy

ABSTRACT

To date, the spread of SARS-CoV-2 infection is increasing worldwide and represents a primary healthcare emergency. Although the infection can be asymptomatic, several cases develop severe pneumonia and acute respiratory distress syndrome (ARDS) characterized by high levels of pro-inflammatory cytokines, primarily interleukin (IL)-6. Based on available data, the severity of ARDS and serum levels of IL-6 are key determinants for the prognosis.

In this scenario, available in vitro and in vivo data suggested that myo-inositol is able to increase the synthesis and function of the surfactant phosphatidylinositol, acting on the phosphoinositide 3-kinase (PI3K)-regulated signaling, with amelioration of both immune system and oxygenation at the bronchoalveolar level. In addition, myo-inositol has been found able to decrease the levels of IL-6 in several experimental settings, due to an effect on the inositol-requiring enzyme 1 (IRE1)-X-box-binding protein 1 (XBP1) and on the signal transducer and activator of transcription 3 (STAT3) pathways. In this scenario, treatment with myo-inositol may be able to reduce IL-6 dependent inflammatory response and improve oxygenation in patients with severe ARDS by SARS-CoV-2. In addition, the action of myo-inositol on IRE1 endonuclease activity may also inhibit the replication of SARS-CoV-2, as was reported for the respiratory syncytial virus. Since the available data are extremely limited, if this potential therapeutic approach will be considered valid in the clinical practice, the necessary future investigations should aim to identify the best dose, administration route (oral, intravenous and/or aerosol nebulization), and cluster(s) of patients which may get beneficial effects from this treatment.

ARTICLE INFO

Keywords:
SARS-CoV-2
Novel coronavirus
Pneumonia
Acute respiratory distress syndrome
IL-6
Inflammation
Myo-inositol
Inositol

https://doi.org/10.1016/j.mehy.2020.110262

Received 9 August 2020; Received in revised form 30 August 2020; Accepted 5 September 2020

⁎ Corresponding author at: Department of Obstetrics and Gynecology, “Filippo Del Ponte” Hospital, University of Insubria, Piazza Biroldi 1, 21100 Varese, Italy.
E-mail address: antoniosimone.lagana@uninsubria.it (A.S. Laganà).
1 www.inositolgroup.com.

This is an open access article under the CC BY-NC-ND license (http://creativecommons.org/licenses/by-nc-nd/4.0/).

© 2020 Elsevier Ltd. All rights reserved.
SP-A helps to regulate surfactant secretion and uptake; SP-B and SP-C facilitate adsorption and spreading of phospholipids on the liquid film lining of the alveoli. SP-D may play a role in surfactant reuptake and recycling. Additionally, SPs also regulate inflammatory responses and interact with the adaptive immune response. As a consequence, surfactant degradation or inactivation may contribute to enhanced susceptibility to lung inflammation and infection [6].

The proteins and lipids that form the surfactant have both hydrophilic and hydrophobic regions, which have a specific organization in the surfactant film to mediate the air–water interface at the alveolar surface. The hydrophilic head groups of phospholipids and the hydrophobic regions of proteins interact with water, and the hydrophobic tails and regions are facing towards the air. With this organization, the surfactant film reduces the surface tension, avoiding alveolar and lung collapse at the end of expiration, then preventing atelectasis, but also increases pulmonary compliance by recruiting collapsed airways.

Indeed, surface tension acts as “collapsing” force, which increase with the reduction of the radius of the alveoli. According to Laplace’s law, the gas pressure (P) needed to keep equilibrium between the collapsing force of surface tension (γ) and the expanding force of gas in an alveolus of radius r is expressed as follows, keeping constant the temperature and the volume of the whole system: P = 2γ/r. The normal surface tension for water at the air–liquid interface is 70 mN/m; conversely, in the lungs, it is lower due to the surfactant (25 mN/m). Nevertheless, we should consider that the surface tension decreases as temperature increases, because cohesive forces decrease with an increase of molecular thermal activity, and so it may be influenced by pathological conditions such as fever.

Moreover SPs, making variable weak bonds with lipid components by holding them longer when the interface is compressed, allow to modulate the surface tension based on the alveolar diameter. As a consequence, the surface tension varies according to the volume of air in the lungs, with the surface tension usually lower than at equilibrium during ventilation. At the end of the expiration, compressed surfactant phospholipid molecules decrease the surface tension to very low, near-zero levels. This protects lungs from atelectasis at low volumes but, at the same time, prevents tissue damage at high volumes [7,8]. PS greatly reducing surface tension, reduces the pressure difference needed to allow the lung to inflate, increases compliance, and allows the lungs to inflate much more easily, thereby reducing the work of breathing. In addition to maintain airway patency, the airway surfactant is expected to facilitate particle transport, decrease shear forces, and improve host resistance [9].

In this context, myo-inositol an myo-inositol-1-phosphate interact with SP-D. Specifically, the inositol ring binds the lectin calcium in the usual C-lectin mode, with two vicinal equatorial ring oxygens coordinating the calcium ion [10] and, in this way, contribute to the stability of surfactant pool sizes. Moreover, it orchestrates the interconversion between ultrastructural aggregate forms of the surfactant [11]. In addition, the correct molecular conformation of surfactant, due to the abovementioned complexes, allows robust aggregation and opsonization when binding microbial surfaces, stimulating a direct induction of microbial permeability and growth arrest [12].

As previously summarized [13–16], pathways initiated by either G protein-linked receptors or receptors linked directly or indirectly to tyrosine kinases phospholipase C (PLC) [17], determine the hydrolyses of phosphatidylinositol (4,5)-bisphosphate to form 1,4,5-triphosphate (InsP3) and 1,2-diacylglycerol. In addition, PLC can be activated by lipid products of phosphoinositide 3-kinases (PI3Ks) [18]. Robust data suggested that SP-A induces Ca2+ mobilization by activating these pathways involving PLC [19]. In this view, the activation of PI3K(s) has paramount importance as an upstream signaling molecule involved in SP-A up-regulation of PS and contributes to surfactant homeostasis at the alveolar level.

The hypothesis

Treatment with myo-inositol may be able to reduce IL-6 dependent inflammatory response and improve oxygenation in patients with severe ARDS by SARS-CoV-2. In addition, the action of myo-inositol on inositol-requiring enzyme 1 (IRE-1) endonuclease activity may also inhibit the replication of SARS-CoV-2.

Evaluation of the hypothesis

Evidence for inositol treatment in neonatal acute respiratory distress syndrome

Abnormalities in PS composition and/or reduced surfactant synthesis have been described namely in respiratory distress syndrome [20] (RDS, formerly known as hyaline membrane disease) in the infants, as well as in many similar illnesses [21]. Indeed, the absence (as well as the inadequacy) of PS in the liquid film lining of alveoli causes an increase in surface tension and alveolar collapse [22]. If not treated, such atelectasis causes an increased work of breathing, intrapulmonary shunting, ventilation-perfusion mismatch, hypoxia, and eventual respiratory failure. Surfactant deficiency is mainly due to immaturity in fetal lung and can be predicted by analysis of PS in amniotic fluid, even if current available tests suffer for a lack of sensitivity (sensitivity ranging from 30 to 70%, whereas specificity is significantly high, ranging from 95 to 99%) [23,24]. Ontogeny of PS is tightly coupled to the length of the pregnancy, reaching a critical phase in the latest months, for the PS maturation occurs very close to term [25]. The risk of RDS decreases with increasing gestational age: 60% of babies born at fewer than 28 weeks’ gestation, 30% of babies born between 28 and 34 weeks’ gestation, and fewer than 5% of babies born after 34 weeks’ gestation develop RDS [26]. Acute respiratory distress syndrome (ARDS) has also been observed in adults, associated with different diseases [27]. ARDS usually requires mechanical ventilation and is characterized by an inflammatory condition of the airway epithelium, which involves apoptosis, recruitments of immune cells from the circulation, and protein extravasation of blood proteins into the airways [28]. In this scenario, surfactant replacement therapy has been found beneficial in the early phase of neonatal ARDS (nARDs), due to prematurity, congenital pneumonia, sepsis, meconium aspiration syndrome, bile aspiration, or pulmonary hemorrhage [29].

First attempts in RDS treatment have been made with glucocorticoids and other anti-inflammatory drugs [30]. These preliminary trials demonstrated that such compounds may, to some extent, protect pre-mature infants from RDS, but only if they are given before birth [31,32]. With the advent of therapies for RDS, including antenatal steroids and surfactant replacement therapy, mortality from RDS has decreased from nearly 100% to less than 10% in recent years [33]. Yet, many foetuses do not respond to glucocorticoids, particularly foetuses from multiple pregnancies [34]. Additionally, glucocorticoid-based treatment is still controversial given that glucocorticoid inhibits cell growth and tissue differentiation, thus eventually impairing alveolar maturation [35]. Natural and synthetic surfactant preparations exist, and both are effective in the treatment and prevention of RDS. Natural surfactants are derived from animal lungs (bovine or porcine) and contain phospholipids with SP-B and SP-C, first-generation synthetic surfactants contain only phospholipids without proteins [36]. A Cochrane meta-analysis comparing natural surfactant to first-generation synthetic surfactant confirmed that greater early improvement in the requirement for ventilator support, fewer pneumothoraces, and fewer deaths associated with animal derived surfactant extract treatment. There is also a marginal decrease in the risk of bronchopulmonary dysplasia when using natural surfactant [37]. Although natural surfactants appear to be associated with higher rates of intraventricular haemorrhage, grade 3 and 4 intraventricular hemorrhage rates are not increased. The conclusion of this meta-analysis is that natural...
surfactants are the more desirable choice over the first-generation synthetic surfactants, which is likely due to the inclusion of the SPs in the natural surfactant. Exogenous natural surfactant given at birth of shortly after birth, expands the lung and ameliorates RDS, with a decrease in both neonatal mortality (by 45–55%) and pulmonary air lack syndromes (pneumothorax and interstitial emphysema). Composition of exogenous surfactant is of critical importance. Indeed, presence of SP-B and SP-C proteins is mandatory for surfactant activity, and their presence allows to attain better clinical results (and lower neonatal mortality), than that obtained with synthetic, protein-free surfactant [38]. Nevertheless, surfactant replacement therapy is severely hampered in the case of inflammation of the airway epithelium, which causes accelerated surfactant metabolism [28]. Failure of surfactant replacement therapy is associated with prolonged mechanical ventilation and this leads to secondary damage to the neonatal lung by induction fibrosis and collapse of alveolar-capillary in the distal areas of the lungs [39].

In human infants with RDS, a premature drop in serum inositol levels predicts a more severe course [40]. Inositol supplementation increases the saturated phosphatidylcholine/phosphatidylglycerol ratio in the surfactant of newborns and produces a rise in serum inositol concentration. In humans, free inositol levels in sera from preterm neonates are 2–20 times higher than the levels in maternal or adult sera [41,42]. Consistently, human milk has a high concentration of inositol, with preterm milk being the richest source, and studies in newborns suggest an endogenous synthesis of inositol during fetal life. Infants who are breast fed have higher serum inositol levels compared to those that are not at 1–2 weeks of life [43].

Robust evidence from animal models already suggested that inositol addition in in vitro condition increases the synthesized amount of surfactant phosphatidylinositol by fetal type II cells [44]. Addition of inositol supplement [45] to the feeds of glucocorticoid-treated pregnant rabbits is able to increase surfactant levels in alveolar lavage from fetuses [46]. In this model, myo-inositol decreased betamethasone-induced inhibition of lung growth and potentiated the hormone-induced increase in alveolar space saturated phosphatidylcholine; furthermore, when lung explants from 26-day-old fetuses were grown in the presence of dexamethasone and thyroxine, the addition of myo-inositol switched the acidic surfactant phospholipid from phosphatidyglycerol to phosphatidylinositol and further increased the incorporation of surfactant-associated saturated phosphatidylcholine; finally, myo-inositol increased the incorporation of dihydro-nicotinamide adenine dinucleotide phosphate and acetate into the fatty acid moiety of surfactant phosphatidylcholine. Consistently, myo-inositol-dependent alterations in lipid synthesis may be induced in isolated type II pneumocytes by manipulation of the myo-inositol concentration in the culture medium or, as found in the Sprague-Dawley rats, by a diet supplemented with inositol every 6 h starting 48 h after birth correlated with a significant increase in phosphatidylcholine and phosphatidylinositol and a concomitant significant decrease in sphingomyelin and phosphatidylserine in the tracheal aspirate [53], confirming that surfactant synthesis and secretion are affected by inositol levels as in the animal models. Another study demonstrated that, compared to placebo-treated population, the intravenous administration of 80 mg/kilogram/day of inositol to premature infants with nARDS, starting 4 to 12 h after birth and subsequently every 12 h for 5 days, was associated with reduced severity and mortality due to respiratory failure, increased survival rate without bronchopulmonary dysplasia, and reduced rate of retinopathy [54].

The safety of intravenous administration of inositol was confirmed also in a population of extremely preterm infants. In a recent multicenter study, Phelps et al. [55] included 76 infants who were born between 236/7 and 296/7 gestational weeks, who were ≥ 600 g birth weight, who had no major congenital anomalies, and who had received no human milk or formula feedings since birth. The infants were allocated between 12 h and 6 days of age to receive a single low (60 mg/kg) or high (120 mg/kg) dose of 5% myo-inositol intravenously over 20 min in a 1:1 randomization with placebo delivered in one of two volumes to maintain masking (5% glucose). According to the data analysis, heart rate, blood pressure, and respiration did not differ between placebo and inositol infants at either dose, nor significant differences in adverse events occurred between the 3 groups. From the pharmacokinetic point of view, the central volume of distribution was 0.5115 L/kg, the clearance was 0.0679 L/kg/h, the endogenous production was 2.67 mg/kg/h, and the half-life was 5.22 h when modeled without the covariates. In addition, during the first 12 h renal inositol excretion quadrupled in the 120 mg/kg group, returning to near baseline after 48 h without any significant diuretic side-effect.

More recently, the same authors performed another multicenter trial [56], enrolling 125 infants with similar characteristics of the previous study [55]. The infants were randomized to receive 5% solution of myo-inositol intravenously at neutral pH, at the doses of 10, 40, or 80 mg/kg/day, divided q12h given over 20 min, or placebo (5% glucose for intravenous infusion and dispensed at the equivalent various volumes to maintain masking). This further large trial found that inositol, even at doses up to 80 mg/kg/day for 7–10 weeks, is well tolerated and does not increase adverse events. At 80 mg/kg/day, the pharmacokinetic analysis showed that mean serum levels reached 140 mg/L, declined after 2 weeks, converging in all groups by 6 weeks, with a mean volume of distribution 0.657 L/kg, clearance 0.058 L/kg/h, and half-life 7.90 h. Interestingly, in the inositol groups there were fewer adverse events and co-morbidities compared with the placebo group.

Potential role of inositol to counteract viral infection of the low pulmonary tract

The endoplasmic reticulum (ER) stress response is known to play a role in diverse biological processes including apoptosis, inflammation, and metabolism [57]. In addition, cellular apoptosis has been shown to be mediated by the ER stress response versus several viruses, including the severe acute respiratory syndrome coronavirus (SARS-CoV) [58]. In detail, the ER stress response depends on three main cascade pathways activated by three proteins, whose upstream signals are mediated by ER-resident transmembrane proteins: activating transcription factor 6 (ATF6), protein kinase R-like ER kinase (PERK), and IRE-1 [59]. According to recent pieces of evidence, IRE1 endonuclease activity has been demonstrated involved in the inhibition of the replication of respiratory syncytial virus [60]. This mechanism may be due, at least in part, to the destabilization of phospholipid membrane homeostasis. Indeed, ER-resident transmembrane proteins such as the protein translocon subunit Shb1, which form part of the unfolded protein
response program, are prematurely degraded by membrane stiffening at the ER [61]. Nevertheless, another study shows that influenza A viral infection activates IRE-1, and inhibiting that pathway inhibits influenza virus replication [62]. In this scenario, IRE-1 appears to have diametrically opposed effects for RSV vs influenza, and so it would be difficult to predict how it might interact with SARS-CoV-2.

Inositol’s action on the lung cancer microenvironment and IL-6 levels

Robust epidemiological data correlated cigarette smoking to both high-grade preinvasive bronchial lesions (i.e., moderate dysplasia, severe dysplasia, and carcinoma in situ) and invasive lung cancer [63]. In particular, smoking increases lung cancer risk 5- to 10-fold with a clear dose-response relationship; in addition, exposure to environmental tobacco smoke among non-smokers increases lung cancer risk by about 20% [64]. Interestingly, a diet supplemented with inositol, combined with aerosolized budesonide, has been found to inhibit the process of lung tumorigenesis in a dose-dependent fashion in a mouse model exposed to the carcinogen benzo(a)-pyrene or 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone [65]. In details, according to a robust investigation in this animal model, budesonide at 10 µg/kg body weight administered by aerosol for 20 s three times a week, plus 0.3% myo-inositol added to the diet, reduced the pulmonary tumor formation by 60%; similarly, budesonide at 25 µg/kg body weight plus 0.3% myo-inositol added to the diet reduced it by 79% [66]. In another study, diet containing both myo-inositol and dexamethasone resulted in an additive effect on the inhibition by 40% of pulmonary adenoma induced by benzo[a]pyrene in female mice [67], suggesting a potential therapeutic role of the combined treatment even after the pre-cancerous lesion formation and initiation.

The efficacy and safety of oral myo-inositol for the chemoprevention of lung cancer was also tested and confirmed in a cohort of smokers with persistent bronchial dysplasia, despite treatment with inhaled budesonide [68]. In this study, after a treatment with 18 gr/die of oral of myo-inositol mixed with juice or water divided into two doses daily, the regression of pre-existing dysplastic lesions at repeat auto-fluorescence bronchoscopy and biopsy was significantly increased. More recently, the same group investigated the effects of oral myo-inositol, 9 gr once/day for two weeks and then twice/day for 6 months, on bronchial dysplasia rate, Ki-67 labeling index, blood and bronchoalveolar lavage fluid levels of pro-inflammatory, oxidant/anti-oxidant biomarkers, and airway epithelial gene-expression signature for PI3K activity [69]. Confirming the previous findings [68], the complete response rate was higher in the group treated with oral myo-inositol...
compared to placebo; in addition, the population who showed complete response to this treatment, gene-expression signature reflective of PI3K activation within the cytologically-normal bronchial airway epithelium was significantly decreased; finally, and most importantly, oral treatment with myo-inositol has been found to significantly reduce interleukin (IL)-6 levels in bronchoalveolar lavage fluid.

One year later, the preliminary findings in a selected population with bronchial dysplasia were further investigated at a molecular level in the mouse model. In this study [70], mice with oncogenic Kras expressed in the airway epithelium (CcspCre/+; KrasLSL-G12D/+), which are known form lung premalignant lesions in a stereotypical fashion over the ten weeks following weaning, were raised on diets compounded with myo-inositol. As expected, mice treated with myo-inositol diet showed a significant decrease in the number, size, and stage of lesions as compared to those raised on control diets. Interestingly, also in this mouse model, there was a significant decrease in IL-6 levels at both proteomic and cytokine analyses.

We take the opportunity to summarize the available pieces of evidence suggesting that the strong reduction of IL-6 after treatment with myo-inositol could depend on two different mechanisms that may occur simultaneously, as we recently pointed out [71]. On the one hand, the change in signal transducer and activator of transcription 3 (STAT3) phosphorylation could reduce macrophage recruitment and their phenotype switching, with consequent reduction of IL-6-producing cell population [72]. On the other hand, the fine-regulated cross-talk between IL and 6-mediated pathways and surfactant-derived phosphatidylglycerol subtraction may account for the return to the homeostasis in the bronchoalveolar microenvironment [73–75].

These elements might have a role of paramount importance even for the adjuvant treatment of several conditions which show increased bronchoalveolar levels of IL-6, such as SARS-CoV-2 pneumonia [76,77]. Indeed, the infections in the upper and lower respiratory tract by novel coronavirus-19 in humans causes a mild or highly acute respiratory syndrome, with consequent release of pro-inflammatory cytokines (the so-called “cytokine storm”), including IL-1β and IL-6 [78], lymphopenia (in CD4 + and CD8 + T cells) and decreased interferon (IFN)-γ expression in CD4 + T cells [79]. In addition, recent findings from the Chinese population who underwent SARS-CoV-2 infection confirmed that increased expression of IL-6 in serum is expected to predict the severity of SARS-CoV-2 pneumonia and the prognosis of patients: higher are the IL-6 serum levels, worse is the prognosis among sub-clusters of infected patients [80,81]. This element appears very important, considering that the action of the inositol on the IRE1-X-box-binding protein 1 (XBP1) pathway is able to inhibit the IL-6 production and, in this way, reduce the prof-inflammatory cascade by activation of the IL-6-STAT3 signaling [82–84].

Conclusions and future perspective

To date, the spread of SARS-CoV-2 infection is increasing worldwide and represents a primary healthcare emergency. Although the infection can be asymptomatic, several cases develop severe pneumonia and ARDS characterized by high levels of pro-inflammatory cytokines, primarily IL-6. Based on the available data, the severity of ARDS and serum levels of IL-6 are key determinants for the prognosis of the patients.

Available robust in vitro and in vivo data suggested that myo-inositol is able to increase the synthesized amount of surfactant phosphatidylinositol and its functions, ameliorating both immune defensive system and oxygenation at the bronchoalveolar level. In addition, myo-inositol has been found able to decrease the levels of IL-6 in several experimental settings, due to a robust effect on the IRE1-XBP1 and STAT3 pathways. In this scenario, treatment with myo-inositol may be able to reduce IL-6 dependent inflammatory response and improve oxygenation in patients with severe ARDS by SARS-CoV-2. In addition, the action of myo-inositol on IRE1 endonuclease activity may also inhibit the replication of SARS-CoV-2, as was found for the respiratory syncytial virus.

Since the available data are extremely limited, if this potential therapeutic approach will be considered valid in the clinical practice, the necessary future investigations should aim to identify the best dose, administration route (oral, intravenous and/or aerosol nebulization), and cluster(s) of patients which can benefit from this treatment.

Declaration of Competing Interest

Vittori Unfer is employee at Lo.Li. Pharma s.r.l. (Rome, Italy). All the other authors have no proprietary, financial, professional or other personal interest of any nature in any product, service or company.

References

[1] Sunde M, Pham CLL, Kwan AH. Molecular Characteristics and Biological Functions of Surface-Active and Surfactant Proteins. Annu Rev Biochem 2017;86:585–608.
[2] Mindt BC, Fritz JH, Duerer CU. Group 2 innate lymphoid cells in pulmonary immunity and tissue homeostasis. Front Immunol 2018;9:840.
[3] Rodriguez RJ. Management of respiratory distress syndrome: An update. Respiratory Care. Daedalus Enterprises Inc.; 2003. p. 279–86.
[4] Ryndikiewicz MJ, Wu H, Calaressa TR, Nikolaidis NM, Head JR, Seaton BA, et al. Differential ligand binding specificities of the pulmonary collectins are determined by the conformational freedom of a surface loop. Biochemistry 2017;56:6905–105.
[5] Kishore U, Greenough TJ, Waters P, Shrive AK, Ghai R, Kamran MF, et al. Surfactant proteins SP-A and SP-D: Structure, function and receptors. Mol Immunol 2006;43:1293–315.
[6] Wright JR. Host defense functions of pulmonary surfactant. In: Biology of the Neonate. Biol Neonate 2004; p. 326–32.
[7] Schürch S, Bachofen H, Postmeyer F. Surface activity in situ, in vivo, and in the captive bubble surfacometer. Comput Biochem Physiol A: Mol Integr Physiol 2001;129:195–207.
[8] Fujioka H, Halpern D, Gaver DP. A model of surfactant-induced surface tension effects on the parenchymal tethering of pulmonary airways. J Biomech 2013;46:319–28.
[9] Crouch EC. Collectins and pulmonary host defense. Am J Respir Cell Mol Biol 1998;19:177–201.
[10] Crouch E, McDonald B, Smith K, Roberts M, Moyle T, Seaton B, et al. Critical role of Arg/lys243 in the species-dependent recognition of phosphatidylinositol by pulmonary surfactant protein D. Biochemistry 2007;46:5165–9.
[11] Ikegami M, Grant S, Korthagen T, Scheule RK, Whitsett JA. Surfactant protein-D regulates the postnatal maturation of pulmonary surfactant lipid pool sizes. J Appl Physiol 2009;106:1545–52.
[12] Sawada K, Ariki S, Kojima T, Saito A, Yamaaze M, Nishitani C, et al. Pulmonary collectins protect macrophages against pore-forming activity of Legionella pneumophila and suppress its intracellular growth. J Biol Chem 2010;285:8434–43.
[13] Laganà AS, Garzon S, Casarin J, Franchi M, Ghezzi F. Inositol in Polycystic Ovary Syndrome: Restoring Fertility through a Pathophysiology-Based Approach. Trends Endocrinol Metab 2018;29:766–80.
[14] Facchetti F, Appetecchia M, Argenzio C, Bevilacqua A, Rezerra Espinola MS, Bizzarri M, et al. Experts’ opinion on Inositols in treating polycystic ovary syndrome and non-insulin dependent diabetes mellitus: a further help for human reproduction and beyond. Expert Opin Drug Metab Toxicol 2020;16:255–74.
[15] Garzon S, Laganà AS, Monasta G. Risk of reduced intestinal absorption of Myo-inositol caused by D-chiro-inositol or by glucose transporter inhibitors. Expert Opin Drug Metab Toxicol 2019;15:697–703.
[16] Laganà AS, Rossetti P, Sapia F, Chiofalo B, Buscema M, Valenti G, et al. Evidence-Based and Patient-Oriented Inositol Treatment in Polycystic Ovary Syndrome: Changing the Perspective of the Disease. Int J Endocrinol Metab 2017;15:e43695.
[17] Bill CA, Vines CM. Phospholipase C. Advances in experimental medicine and biology. New York LLC: Springer; 2020. p. 215–42.
[18] Zhao K, Li G, Yao Y, Zhou Y, Li Z, Guo-Q, et al. Activation of phospholipase C-γ1 and translocation of phosphatidylinositol 3,4,5-trisphosphate 3-phosphatase contribute to GL-V9-induced apoptosis in human gastric cancer cells. Exp Cell Res 2017;356:8–19.
[19] Beharka AA, Crowther JE, McCormack FX, Denning GM, Lees J, Tibesar E, et al. Surfactant Protein A Activates a Phosphatidylinositol 3-Kinase/Calcium Signal Transduction Pathway in Human Macrophages: Participation in the Up-Regulation of Mannose Receptor Activity. J Immunol 2005;175:2227–36.
[20] Rubarth LB, Quinn J. Respiratory Development and Respiratory Distress Syndrome. Neonatal Netw. 2015;34:231–8.
[21] Clements JA, Avery ME. Lung surfactant and neonatal respiratory distress syndrome. Am J Respir Crit Care Med 1998;157:S59–66.
[22] Beharka AA, Crowther JE, McCormack FX, Denning GM, Lees J, Tibesar E, et al. Surfactant Protein A Activates a Phosphatidylinositol 3-Kinase/Calcium Signal Transduction Pathway in Human Macrophages: Participation in the Up-Regulation of Mannose Receptor Activity. J Immunol 2005;175:2227–36.
[23] Gluck L, Kulovich MV, Boer R, Brenner PH, Anderson GG, Spellacy WN. Diagnosis of the respiratory distress syndrome by amniocentesis. Am J Obstet Gynecol 1971;109:440–5.
[24] Hallman M, Teramo K. Measurement of the lecithin/phosphatidylglycerol ratio and
phosphatidylglycerol in amniotic fluid: an accurate method for the assessment of fetal lung maturity. Br J Obstet Gynaecol 1981;88:806–13.

[57] Hotamisligil GS. Endoplasmic Reticulum Stress and the Inflammatory Basis of Metabolic Disease. Cell 2010;140:900–17.

[58] Ye Z, Wong CK, Li P, Xie Y. A SARS-CoV protein, ORF-6, induces caspase-3 mediated, ER stress and JNK-dependent apoptosis. BBA 2008;1780:1383–7.

[59] Kennedy D, Samali A. ER stress: causes for studying ER stress and UPR in human cells. Methods Mol Biol 2015;1296:239–53.

[60] Hassan L, Gaines KS, Hotel DJ, Wisby RM, Miller SE, Powers LS, et al. Inositol-requiring enzyme 1 inhibits respiratory syncytial virus replication. J Biol Chem 2014;289:27573–60.

[61] Shyu Jr P, Ng BSH, Ho N, Chaw R, Seah YL, Marvalmid C, et al. Membrane phospholipid alteration causes chronic ER stress through early degradation of homeostatic ER-resident proteins. Sci Rep 2019;9:6377.

[62] Hassan LH, Zhang MS, Powers LS, Shaw JD, Baumgartner J, Rutkowski DT, et al. Influenza A viral replication is blocked by inhibition of the inositol-requiring enzyme 1 (IRE1) stress pathway. J Biol Chem 2012;287:6479–89.

[63] Moro-Sibillà D, Jeannart M, Lantuejoul S, Arbib F, Lavrerve MH, Brambilla E, et al. Cigarette smoking, preservable bronchial lesions, and autoantibodies against bronchopulmonary syncytial bronchoeptosis. Chest 2002;122:1902–8.

[64] Schwartz AG, Cote ML. Epidemiology of lung cancer. Adv Exp Med Biol 2014;803:21–41.

[65] Hecht SS, Kenney PM, Wang M, Udupa B. Dose-response study of Myo-inositol as an inhibitor of lung tumorigenesis induced in A/J mice by benzo[a]pyrene and 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone. Cancer Lett 2001;167:1–6.

[66] Wattenberg LW. Chemoprevention of pulmonary carcinogenesis by brief exposures to aerosolized budenolide or beclomethasone dipropionate and by the combination of aerosolized budenolide and dietary myo-inositol. Carcinogenesis 2000;21:179–82.

[67] Ehrenstein RD, Wattenberg LW. Studies of chemopreventive effects of Myo-inositol on benzo(a)pyrene-induced neoplasia of the lung and forestomach of female a/j mice. Carcinogenesis 1993;14:1975–7.

[68] Lam S, McWilliams A, LeRiche J, MacAulay C, Wattenberg L, Szabo E. A phase I study of Myo-inositol in patients with lung chemoprevention. Cancer Epidemiol Biomarkers Prev 2006;15:1526–31.

[69] Lam S, Mandrekar SJ, Gesthalter Y, Allen Z, Odile DE, et al. A Randomized Phase Ib Trial of Myo-inositol in Smokers with Bronchial Dysplasia. Cancer Prev Res 2016;9:506–14.

[70] Unver N, Delgado D, Zeleke K, Cuminan A, Tang X, Caetano MS, et al. Reduced IL-6 levels and tumor-associated phospho-STAT3 are associated with reduced tumor development in a mouse model of lung cancer chemoprevention with Myo-inositol. Int J Cancer 2018;142:1405–14.

[71] Bizzarri M, Lagana AS, Aragona D, Unfer V. Inositol and pulmonary function. Could Myo-inositol treatment downregulate inflammation and cytokine release syndrome in SARS-CoV-2? Eur Respir Rev 2020;29:451–2.

[72] Lam S, Mandrekar SJ, Ho Y-C. SARS-CoV-2: A Storm is Raging. J Clin Invest. 2020;130:3426–32.

[73] Conti P, Ronconzi G, Caraffa A, Ballarin E, et al. Induction of Pro-Inflammatory Cytokines (IL-1b and IL-6) and Lung Inflammation by Coronavirus-19 (COVID-19 or SARS-CoV-2): Anti-Inflammatory Strategies. J Biol Regul Homeost Agents 2020. https://doi.org/10.23812/JORT-EST-000199.

[74] Pedersen SF, Ho Y-C. SARS-CoV-2: A Storm is Raging. J Clin Invest. 2020;130:2203–9.

[75] Chen G, Wu D, Guo C, Yao H, Huang D, Wang H, et al. Clinical and immunologic features in severe and moderate Coronavirus Disease 2019. 2020;130:2620–9.

[76] Chen J, Jin H, Hu J, Wang Y, Ma Z, Zhang J. Endoplasmic reticulum stress operates in silica nanoparticles-induced macrophage apoptosis via activation of CHOP-mediated apoptotic signaling pathway. Int J Mol Sci 2019;20:5848.

[77] Fang P, Xiang L, Huang S, Jin L, Zhou G, Zhang L, et al. IRE1α-XBP1 signaling pathway regulates IL-6 expression and promotes progression of hepatocellular carcinoma. Oncol Lett. 2018;16:4729–36.

[78] Choi S, Snider JM, Olakkengil N, Lambert JM, Anderson AK, Ross-Evans JS, et al. IRE1α-XBP1 signaling pathway regulates IL-6 expression and promotes progression of hepatocellular carcinoma. Oncol Lett. 2018;16:4729–36.

[79] Suzuki Y, Sugimoto J, Cukrowska KE, Tseng C-T-K. Severe Acute Respiratory Syndrome (SARS) Coronavirus-Induced Lung Epithelial Cytokines Exacerbate SARS Pathogenesis by Modulating Intrinsic Functions of Monocyte-Derived Macrophages and Dendritic Cells. J Virol 2007;81:3039–48.

[80] Konti P, Ronconzi G, Caraffa A, Gallego CE, Ross R, Fradis I, et al. Induction of Pro-Inflammatory Cytokines (IL-1b and IL-6) and Lung Inflammation by Coronavirus-19 (COVID-19 or SARS-CoV-2): Anti-Inflammatory Strategies. J Biol Regul Homeost Agents 2020. https://doi.org/10.23812/JORT-EST-000199.

[81] Pedersen SF, Ho Y-C. SARS-CoV-2: A Storm is Raging. J Clin Invest. 2020;130:2203–9.

[82] Chen G, Wu D, Guo C, Yao H, Huang D, Wang H, et al. Clinical and immunologic features in severe and moderate Coronavirus Disease 2019. 2020;130:2620–9.

[83] Chen J, Jin H, Hu J, Wang Y, Ma Z, Zhang J. Endoplasmic reticulum stress operates in silica nanoparticles-induced macrophage apoptosis via activation of CHOP-mediated apoptotic signaling pathway. Int J Mol Sci 2019;20:5848.

[84] Feng P, Xiang L, Huang S, Jin L, Zhou G, Zhang L, et al. IRE1α-XBP1 signaling pathway regulates IL-6 expression and promotes progression of hepatocellular carcinoma. Oncol Lett. 2018;16:4729–36.

[85] Choi S, Snider JM, Olakkengil N, Lambert JM, Anderson AK, Ross-Evans JS, et al. IRE1α-XBP1 signaling pathway regulates IL-6 expression and promotes progression of hepatocellular carcinoma. Oncol Lett. 2018;16:4729–36.