The CypA-netics of Ventilator-induced Lung Injury

For patients with acute respiratory distress syndrome (ARDS), mechanical ventilation is often an obligatory life-saving intervention. Mechanical ventilation itself may, however, evoke ventilator-induced lung injury (VILI) (1). In spite of lung-protective ventilation strategies with, for example, low VT having been implemented into clinical practice (2), ventilated areas of ARDS lungs may still encounter injurious transparenchymal forces because of a marked reduction in aerated lung size (“baby lung”). The absence of a definite safety threshold for VILI therefore necessitates further efforts to minimize VILI even more so as the requirement for mechanical power to ensure adequate ventilation increases the sicker the patient is. To solve this obvious dilemma, personalized ventilation and novel therapeutic strategies based on point-of-care monitoring of the mechanical forces acting on the lung tissue and better insight into the mechanotransduction pathways that convert these forces into injurious cellular responses are required. To this end, a body of work has identified various inflammatory and barrier-disruptive mediators as potential biomarkers and therapeutic targets in VILI. Yet, this knowledge has so far not translated into improved patient care or novel treatment approaches.

In this issue of the Journal, Koh and colleagues (pp. 421–430) report findings from animal experiments and patient sample analyses that suggest secreted extracellular CypA (cyclophilin A) as a biomarker and mediator of VILI (3). Originally, Handschumacher and colleagues had identified CypA as a ubiquitously expressed cytosolic protein that intracellularly binds cyclosporin A, thereby mediating its immunosuppressive activity (4). Subsequently, CypA was shown to also serve as an extracellular signaling molecule that can be secreted by endothelial and epithelial cells, monocytes, or macrophages in response to, for example, oxidative stress or LPS and then acts as a proinflammatory cytokine in acute and chronic inflammatory diseases, including rheumatoid arthritis, coronary artery disease, or sepsis (5). CypA is considered to exert its proinflammatory effects by activation of the transmembrane protein CD147, a member of the immunoglobulin superfamily expressed by many cell types, including epithelial cells, endothelial cells, and leukocytes. Of late, CypA was also identified as an endogenous ligand for another immunoglobulin superfamily receptor, TREM-2 (triggering receptor expressed on myeloid cells-2), to which it binds with even higher affinity to elicit both pro- and antiinflammatory responses (6). Yet, despite abundant evidence for CypA’s involvement in inflammatory processes, its role in acute lung injury and specifically VILI has so far not been addressed.

In their present study, Koh and colleagues show CypA levels to be 5- to 6-fold elevated in the BAL fluid of patients with ARDS as compared with healthy volunteers and similarly in mice ventilated with excessive VT of 35–40 ml/kg body weight as compared with mice undergoing lung-protective ventilation. In overventilated mice, flow cytometric analyses detected a concomitant decrease in intracellular CypA in alveolar epithelial cells but not in alveolar macrophages, whereas cyclic stretch of primary human alveolar epithelial cells in vitro resulted in CypA secretion into the supernatant. In vivo, CypA blockade by MM-284, a nonimmunosuppressive cyclosporin A derivative that inhibits CypA extracellular signaling, improved survival and classic parameters of lung injury in overventilated mice, including lung function and oxygenation, and reduced alveolocapillary barrier dysfunction and epithelial injury. Ex vivo stimulation with recombinant CypA induced inflammatory responses in human monocyte-derived macrophages, including IL-6 secretion, yet not in primary alveolar epithelial cells. These data thus suggest a scenario in which overventilation causes CypA secretion from stretched alveolar epithelial cells, which in turn activates alveolar macrophages, triggering proinflammatory responses that will ultimately drive alveolocapillary barrier failure and impaired lung function and oxygenation (Figure 1). Although this concept is coherent, the exact cellular sources of CypA in VILI, its auto- or paracrine target cells, and the individual receptors mediating these effects (e.g., CD147 vs. TREM2) remain to be validated in vivo by cell-specific conditional knockout models and/or single-cell transcriptomic analyses.

May CypA hence present a promising therapeutic target to reduce lung injury and improve survival in patients with ARDS? The following aspects should be considered. First, although MM-284 attenuated lung injury in overventilated lungs of naïve mice, evidence that CypA blockade similarly reduces VILI in lungs preinjured by, for example, pneumonia or sepsis is presently lacking. The plethora of inflammatory pathways triggered in such critical inflammatory conditions may simply outweigh the benefits of CypA blockade in VILI. On the other hand, therapeutic effects of CypA blockade in ARDS may not be restricted to VILI but may also target inflammatory pathways of ARDS and its underlying diseases. Although this might point toward a broader therapeutic potential of CypA blockade, it also raises the question of the perfect timing for this intervention. In their preclinical study, Koh and
colleagues tested the prophylactic administration before overventilation, which may be translated into treatment start right before intubation in a patient. Unless CypA blockade also has therapeutic potential to reverse rather than prevent ongoing inflammation, however, this approach may prove too late if CypA already contributes critically to the inflammatory condition that is the cause for intubation. Analogously, it remains to be shown whether increased CypA levels in the BALF of patients with ARDS are specific for mechanical ventilation or equally present in patients with nonventilated sepsis or pneumonia, which would preclude the use of CypA as a specific biomarker for VILI.

Second, recent work by Calfee and colleagues identified specific subphenotypes of ARDS, which display major differences not only in terms of presentation but also in response to therapy, stressing the need for personalized therapy (7, 8). As such, a potential treatment response to CypA blockade may be restricted to specific endotypes. Based on CypA’s role as a proinflammatory mediator, it is tempting to speculate that CypA blockade may be particularly effective in patients with an inflammatory phenotype, yet this notion remains to be tested.

Third, therapeutic CypA blockade should probably be commenced as early as possible in the course of the disease. To identify appropriate patients promptly, CypA would ideally be used as a theragnostic or, in other words, as both a biomarker and therapeutic target. As CypA was increased in BALF but not plasma, theragnostic CypA quantification would require BALF sampling, which is typically not feasible in spontaneously breathing patients acutely deteriorating toward the need for intubation. Thus, the earliest time to obtain BALF is commonly after intubation, which would require point-of-care CypA testing capabilities to ensure a timely start of targeted therapy in case of increased CypA levels.

Although experimental research published in this issue of the Journal has thus laid an important foundation for CypA targeting as a putative new therapeutic strategy in ARDS, considerable translational hurdles remain, which necessitate in-depth follow-up analyses to better understand CypA’s mode of action, the exact cell types and signaling pathways involved, and further translational experimental studies to evaluate its diagnostic and therapeutic potential in ARDS and VILI, respectively.

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Figure 1. Overventilation (OV) causes CypA secretion from stretched alveolar epithelial cells, which in turn activates alveolar macrophages, triggering proinflammatory responses that ultimately drive alveolocapillary barrier failure and impaired lung function and oxygenation. Illustration by Patricia Ferrer Beals.
Lymphangioleiomyomatosis (LAM) is a rare, multiorgan disease, affecting primarily women during the childbearing years (1). Patients with LAM have proliferative, smooth-muscle–like cells within the lung and lymphatics, leading to both airway and lymphatic obstruction (2). An inherited form of LAM occurs in patients with tuberous sclerosis complex (TSC), with prevalence estimated at 26–49% of women with TSC and increased incidence with aging (3). Sporadic LAM occurs more rarely with an incidence estimated at 3.5 per 1 million females in the United States (4). From a pulmonary standpoint, women with this disease can present incidentally or, more often, with symptoms including dyspnea, cough, chylous effusions, or pneumothoraces (1).

Based on the high incidence of LAM in patients with TSC, studies historically focused on the role of the mammalian target of rapamycin (mTOR) signaling pathway. Loss of TSC gene function constitutively activates the mTOR signaling pathway, leading to cellular proliferation and survival in numerous disease states including LAM (5). mTOR activation is blocked by sirolimus, leading to a series of in vitro and preclinical in vivo studies in LAM that ultimately laid the framework for the landmark MILES (Multicenter International LAM Efficacy and Safety of Sirolimus) trial (6). This double-blind, placebo-controlled study of 89 women with LAM found that mTOR inhibition with sirolimus stabilized the decline in FEV1 over a 1-year study (6). A study of 89 women with LAM found that mTOR inhibition with sirolimus stabilized the decline in FEV1 over a 1-year study (6). Although cessation of sirolimus in the 12-month follow up was associated with a resumed decline in lung function, the MILES trial was nevertheless transformative for the care of women with this rare disease, with efficacy and minimal side effects described out to 4 years of observational therapy (7, 8).

There is recurrence and/or growth of tumors noted upon cessation of therapy (6), and studies have proposed mechanisms by which TSC2-null cells develop resistance to mTOR inhibition over time (13). Therefore, despite the putative role of TSC2 in LAM, groups have wisely begun to investigate TSC2-independent pathways in disease pathogenesis (14-16) to increase the potential pipeline of therapies for this rare disease.

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