Molecular Interactions of SARS-CoV-2 in Lung Tissue of Patients with Chronic Obstructive Pulmonary Disease

To the Editor:

Patients with chronic obstructive pulmonary disease (COPD) appear to be underrepresented in coronavirus disease (COVID-19) series, but their risk of severe disease and death if infected by severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) is increased (1, 2). To disentangle these apparently contradictory clinical observations, a deeper understanding of the molecular interactions between SARS-CoV-2 and lung cells in patients with COPD is needed (3). Specifically, the association with COVID-19 severity might be mediated by the expression of the ACE2 (angiotensin-converting enzyme 2) receptor, which facilitates viral entry into lung epithelial cells (4), and is upregulated in patients with COPD (5–8) and/or by tobacco smoking, the main environmental risk factor for COPD, which also upregulates lung ACE2 expression (5, 6, 9). Contradictorily, inhaled corticosteroids (ICS), which are frequently used in the clinical management of COPD in patients, downregulate ACE2 expression in patients with asthma (another chronic inflammatory condition of the lungs) (10), so it is possible that ICS could have a similar effect in COPD. Furthermore, there is evidence that some ICS can reduce viral replication *in vitro* (11). However, the relationship of previous use of ICS and COVID-19 in patients with COPD or asthma is unclear (12).

A recent study has demonstrated 332 high-confidence protein–protein interactions (PPIs) between SARS-CoV-2 proteins and human proteins that are involved in the biology of the virus within cells (13). To get a deeper understanding of the molecular interactions between SARS-CoV-2 and lung cells in patients with COPD, we here investigated how the severity of COPD alters the expression of genes involved in this PPI network, as well as the potential effects of cumulative smoking exposure and/or use of ICS on ACE2 expression.

Methods

We explored these questions in the Barcelona Lung Tissue Cohort (LT-BCN), a collection of pulmonary samples obtained at surgery from 70 patients with COPD (Global Initiative for Chronic Obstructive Lung Disease grades 1–4) (14), all of whom had quit smoking at least 1 year earlier. All participants signed their informed consent, and the ethics committee of our institution approved the study (HCB/2014/1127, R121212-126). RNA was extracted from samples preserved by using RNA*later* (ThermoFisher Scientific), and whole-genome transcriptomics were measured by using the Human Genome U219 Array Plate (Affymetrix). The methodology for microarray hybridization has been described previously (14), and data are available in the Gene Expression Omnibus (GEO; identifier GSE69818).

Microarray preprocessing of each data set has been previously described (14). Observations in the LT-BCN were validated by using data from the Lung Tissue Research Consortium (LTRC), which were downloaded from the GEO (identifier GSE47460). We used linear regression (adjustment for sex because the angiotensin-converting enzyme gene is located in the X chromosome) to identify genes associated with forced expiratory volume in 1 second (FEV_1) % predicted. The enrichment was tested by using a hypergeometric test (GeneOverlap package for R [R Foundations for Statistical Computing]). All analyses were performed by using R.

Results

In former smokers with COPD included in LT-BCN, the level of ACE2 expression was negatively correlated with that of the FEV_1% predicted, and this was reproduced in the LTRC (Figures 1A and 1B). In the LT-BCN, neither use of ICS (in 42% of the patients) nor cumulative smoking exposure (pack-years) influenced ACE2 expression significantly (false discovery rate *P* values = 0.687 and 0.999, respectively). These data were not available in the GEO for the LTRC. Finally, in the LTRC data, increased ACE2 expression was also observed in patients with COPD compared with nonsmoker control subjects (Figure 1B); the latter were not included in the LT-BCN.

Next, we assessed how many of the reported SARS-CoV-2 host PPI genes (13) were differentially expressed in relation to the presence of COPD or the severity of airflow limitation. We found that in the LTRC, 18 SARS-CoV-2 host PPI genes were differentially expressed in patients with COPD compared with nonsmoker control subjects (ACE2, AP2A2, ATP1B1, CIT, DCAF7, ERMP1, FAM98A, GDF15, GNB1, IDE, LOX, NPTX1, PUSL1, SIRT5, TAPT1, TMED5, TMPRSS2, USP54, and WFS1). We could not contrast these observations with the LT-BCN because it only includes patients with COPD; in the LT-BCN, 1,249 genes were significantly related to the FEV_1% predicted, 31 of which belong to the SARS-CoV-2 PPI network. Likewise, in the LTRC, 2,993 genes were also significantly related to the FEV_1% predicted, 34 of which also belong to the SARS-CoV-2 PPI network. Finally, four of these identified genes (ACE2, FAM98A, MFGE8, and HMOX1) were differentially expressed in both the LT-BCN and the LTRC, and when we assessed the presence of an overall enrichment of the PPI network in relation with the severity of airflow limitation FEV_1% predicted, we only observed a significant enrichment for the LT-BCN (overlapping *P* value = 1.8×10^{-12}; odds ratio, 4.9).

Discussion

This is the first study that explores how the SARS-CoV-2–human PPI network (13) changes in relation to the presence of COPD, the COPD severity (as determined by the FEV_1% predicted), and the cumulative exposure to smoking and/or use of ICS. Results confirmed those of previous studies showing that lung ACE2 expression is upregulated in COPD (5, 6). We extended these previous observations by showing that ACE2 expression was related to the severity of airflow limitation but was not related to smoking history and/or use of ICS. Of note, very recently, the lack of an association between serum and lung levels of ACE2 has been reported (15). Finally, our study identified three additional SARS-CoV-2 PPI genes (FAM98A, MFGE8, and HMOX1) that were differentially expressed in relation to the severity of airflow limitation in the lung tissue of patients with COPD. Interestingly, two of them (ACE2 and HMOX1) are implicated in the

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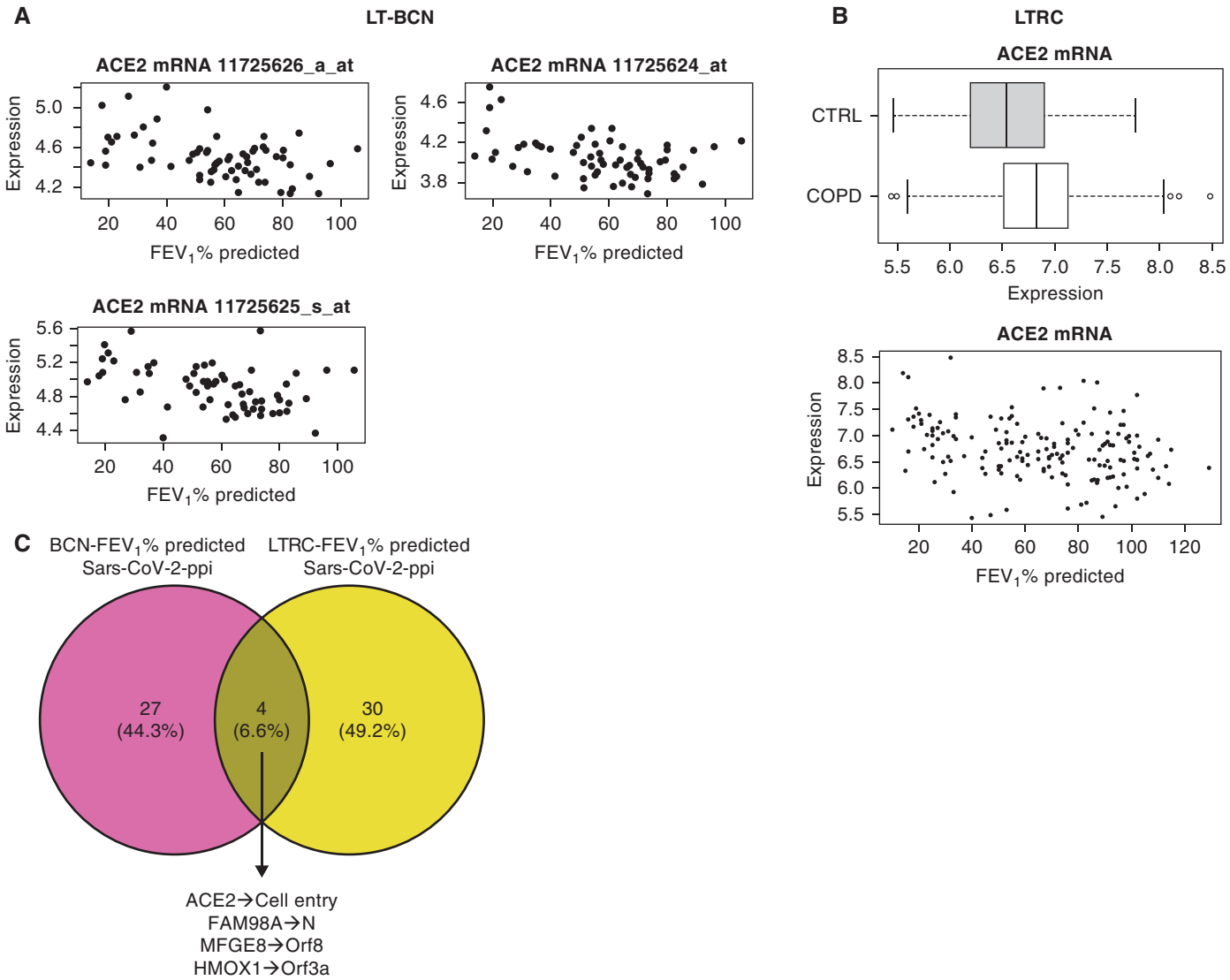

Figure 1. (*A*) Level of expression of the three probes mapping to the ACE2 (angiotensin-converting enzyme 2) gene in relation to the FEV_1% predicted level in the LT-BCN (false discovery rate *P* values for each correlation = 0.015, 0.02, and 0.036, respectively). (*B*) Level of expression of ACE2 in nonsmoking CTRL subjects versus patients with chronic obstructive pulmonary disease (COPD) in the LTRC and correlation of ACE2 expression in patients with COPD with the FEV_1% predicted level. (*C*) Genes differentially expressed in relation to the severity of airflow limitation (FEV_1% predicted) in each data set that is part of the SARS-CoV-2 ppi. BCN = Barcelona; CTRL = control; FEV_1% predicted = predicted forced expiratory volume in 1 second; LT-BCN = BCN Lung Tissue Cohort; LTRC = Lung Tissue Research Consortium; mRNA = messenger RNA; ppi = protein–protein interaction; SARS-CoV-2 = severe acute respiratory syndrome coronavirus 2.

regulation of vasodilation, and aside from this, the HMOX1 gene encodes a heme oxygenase that is an essential enzyme in heme catabolism, which in turn induces ferritin. Ferritin has been identified in many previous studies as a marker of COVID-19 severity. Unfortunately, we lack information on circulating ferritin levels in the patients with COPD studied here.
In any case, altogether, these results contribute to a better understanding of the molecular relationships between COVID-19 and COPD. ■

Author disclosures are available with the text of this letter at www.atsjournals.org.

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Efficacy of Elexacaftor/Tezacaftor/Ivacaftor in Advanced Cystic Fibrosis Lung Disease

To the Editor:

Elexacaftor/tezacaftor/ivacaftor (E/T/I) treats nearly 90% of people with cystic fibrosis (CF) with clinically meaningful results in lung function, weight, and patient-reported outcomes. We wished to further characterize short- and long-term outcomes associated with E/T/I in this population. This retrospective study evaluates clinical outcomes over 1 year of E/T/I administration in patients with forced expiratory volume in 1 second (FEV_1) % predicted <40.

Methods

This is a retrospective cohort study at the adult CF program at Columbia University Irving Medical Center. The study was approved by the Institutional Review Board and via email communication with Vertex Pharmaceuticals, Inc. Patients were included if they had at least one F508del (F) mutation, initiated E/T/I between August and December