Received: 24 April 2020 | Revised: 1 June 2020 | Accepted: 3 June 2020

DOI: 10.1002/ijc.33145

IJC International Journal of Cancer

LETTER TO THE EDITOR

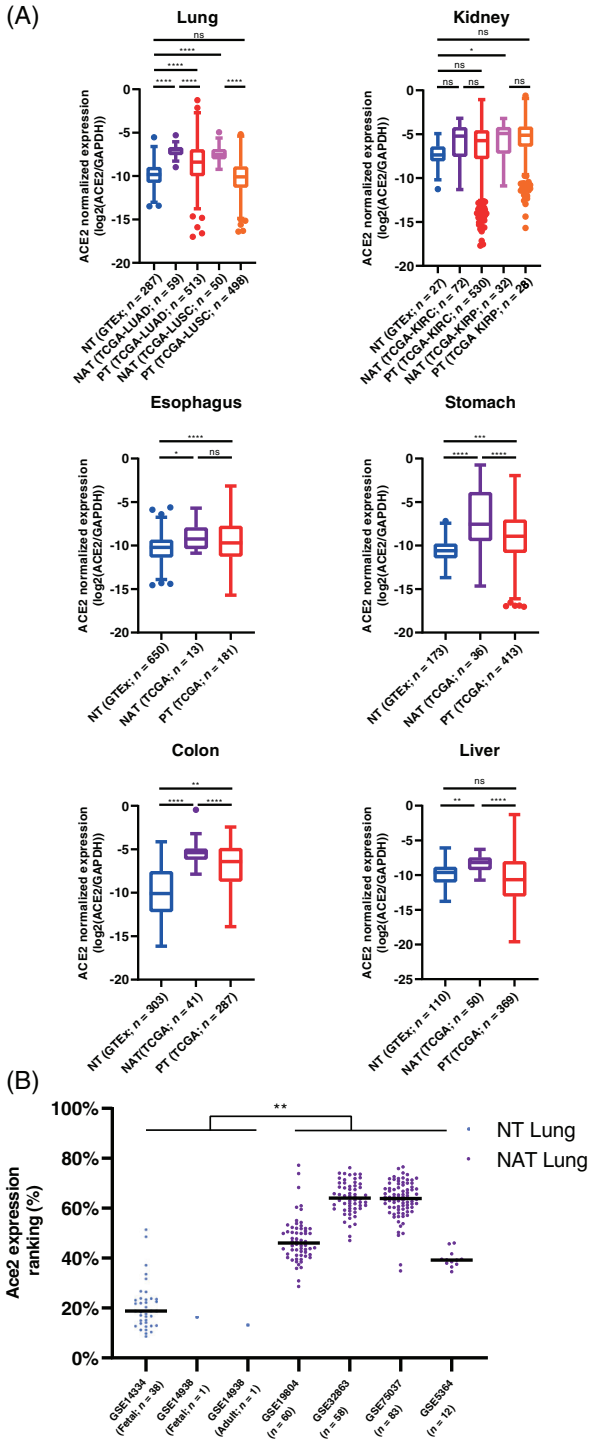
Elevated expression of *ACE2* in tumor-adjacent normal tissues of cancer patients

Dear editor,

The recent outbreak of a novel betacoronavirus known as severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) has raised the concern that cancer patients might be particularly susceptible to infection by this virus.[1-3] Importantly, the guidelines for cancer patients during the COVID-19 pandemic focus on lung cancer patients who are undergoing active chemotherapy or radical radiotherapy, and on patients with blood cancers.[1] Intentional postponing of adjuvant chemotherapy or elective surgery for stable cancer has even been proposed to alleviate the risk.[2] However, it is currently unknown whether patients with other epithelial solid tumors, or cancer patients not currently undergoing chemotherapy, are also more susceptible to COVID-19.

SARS-CoV-2 requires the angiotensin-converting enzyme 2 (ACE2) to enter human cells.[4,5] Moreover, *ACE2* gene expression levels in epithelial tissues corresponded to survival after SARS-CoV infection in transgenic mice[6] and soluble human ACE2 inhibited SARS-CoV-2 infections in engineered human tissues.[7] *ACE2* mRNA levels are particularly high in the human kidney, testis, heart and intestinal tract.[8,9] Albeit not highly expressed in most cells of the normal lungs, *ACE2* expression levels in the airway epithelia increase due to chronic exposure to cigarette smoke,[8,10] which was associated with infection susceptibility.[11] *ACE2* expression levels have also been suggested to underlie the increased susceptibility of patients with hypertension and diabetes to SARS-CoV-2 infection,[12] and to be increased in patients with comorbidities associated with severe COVID-19.[13] Therefore, *ACE2* expression in epithelial tissues, and in particular in the airway epithelia, seem to have considerable effect on COVID-19 morbidity and mortality.

We compared *ACE2* mRNA levels between normal tissues (NT), primary tumors (PT) and normal tissues adjacent to tumors (NAT), using data from The Cancer Genome Atlas (TCGA) and GTEx[14] (Supporting Information). Across multiple tissues, *ACE2* mRNA levels in PT were significantly higher than in NT of the respective tissue (Figures 1A and S1). Surprisingly, *ACE2* expression levels in NAT were also significantly higher than in NT across tissues, and in all cases were



Abbreviations: ACE2, angiotensin-converting enzyme 2; COVID-19, coronavirus disease 2019; FANTOM5, Functional Annotation of the Mammalian Genome; GTEx, genotype-tissue expression; HPA, Human Protein Atlas; NAT, normal tissue adjacent to tumor; NT, normal tissue; PT, primary tumor; SARS-CoV-2, severe acute respiratory syndrome coronavirus 2; TCGA, The Cancer Genome Atlas.

at least as high as in the respective PT (Figures 1A and S1). This result suggests that the NAT of cancer patients would likely be more susceptible to SARS-CoV-2 infection than the corresponding tissues of healthy individuals.

Focusing on the lung due to its relevance in the disease etiology, we next queried the mRNA expression levels of *ACE2* in two additional datasets of normal human tissues, the Human Protein Atlas[15] and FANTOM5.[16] In concordance with the GTEx data, the expression levels of *ACE2* in whole-lung tissues from healthy donors were negligible (median of 0.7pTPM, 1.8pTPM and 2.6 scaled tags per million, in GTEx, HPA and FANTOM5, respectively). Next, we compared the relative expression levels of *ACE2* between healthy and tumor-adjacent lung tissues, using six published gene expression microarray datasets[17-20] (Supporting Information Methods). *ACE2* expression levels in the tumor-adjacent normal lung samples were detected at discernible levels, and were significantly higher than those in the healthy normal lung samples (Figure 1B). This analysis confirmed that the mRNA levels of *ACE2* are elevated in tumor-adjacent lung tissues of lung cancer patients.

This observation raises the possibility that lung cancer patients may have an increased risk to SARS-CoV-2 infection, regardless of chemotherapy-induced immune suppression. Furthermore, patients with other types of cancer, such as renal or gastrointestinal cancers, may also have elevated infection risk. However, to determine whether this is indeed the case, two questions require urgent attention: (a) Is *ACE2* expression level in non-lung epithelia associated with SARS-CoV-2 infection risk?; and (b) Is *ACE2* upregulation limited to the tissue adjacent to the tumor (presumably due to the tumor microenvironment), or are *ACE2* levels systemically elevated in cancer patients? In addition, although *ACE2* mRNA levels are upregulated in NAT and PT compared to NT, we cannot rule out the possibility that ACE2 protein levels are not significantly different due to post-transcriptional regulation mechanisms. Until these questions are resolved, we propose that the discussion of cancer guidelines during the COVID-19 pandemic should expand beyond patients with treatment-induced immune suppression.

FIGURE 1 Elevated expression of *ACE2* in tumor-adjacent normal tissues of cancer patients. A, mRNA levels of *ACE2* (normalized to GAPDH) in normal tissues (NT), Normal tissues adjacent to tumors (NAT) and primary tumors (PT) across eight tumor types representing six distinct tissues. KIRC, Kidney renal clear cell carcinoma; KIRP, kidney renal papillary cell carcinoma; LUAD, lung adenocarcinoma; LUSC, lung squamous cell carcinoma. Bar, median; box, 25th and 75th percentiles; whiskers, 1.5 × IQR of lower and upper quartile. **P* < .05; ***P* < .01; ****P* < .001; *****P* < .0001; One-way ANOVA with Tukey's test. B, Relative mRNA levels of *ACE2* in normal lung tissues (NT) and normal tissues adjacent to tumors (NAT), calculated based on six gene expression microarray datasets. Bar, median. ***P* = .0045; two-tailed Student's *t* test for a comparison between the median values of the datasets in each group [Color figure can be viewed at wileyonlinelibrary.com]

KEYWORDS
ACE2, COVID-19, NAT, SARS-CoV-2

ACKNOWLEDGEMENTS
Research in the Ben-David lab is supported by the Azrieli Foundation, the Richard Eimert Research Fund on Solid Tumors, the Tel-Aviv University Cancer Biology Research Center and the Israel Cancer Association (grant #20200111).

CONFLICT OF INTEREST
We declare no conflict of interest.

DATA AVAILABILITY STATEMENT
The data that support the findings of our study are available in Xena at https://xena.ucsc.edu/, in the Human Protein Atlas at https://www.proteinatlas.org/, and in GEO at https://www.ncbi.nlm.nih.gov/geo/ (accession numbers GSE14334, GSE14938, GSE5364, GSE19804, GSE32863 and GSE75037).

Tom Winkler
Uri Ben-David

Department of Human Molecular Genetics and Biochemistry, School of Medicine, Tel Aviv University, Tel Aviv, Israel

Correspondence
Uri Ben-David, Department of Human Molecular Genetics and Biochemistry, School of Medicine, Tel Aviv University, Tel Aviv, Israel.
Email: ubendavid@tauex.tau.ac.il

ORCID
Uri Ben-David https://orcid.org/0000-0001-7098-2378

REFERENCES

1. Burki TK. Cancer guidelines during the COVID-19 pandemic. *Lancet Oncol*. 2020;21:621-630.
2. Liang W, Guan W, Chen R, et al. Cancer patients in SARS-CoV-2 infection: a nationwide analysis in China. *Lancet Oncol*. 2020;21:335-337.
3. Xia Y, Jin R, Zhao J, Li W, Shen H. Risk of COVID-19 for cancer patients. *Lancet Oncol*. 2020;21:180.
4. Hoffmann M, Kleine-Weber H, Schroeder S, et al. SARS-CoV-2 cell entry depends on ACE2 and TMPRSS2 and is blocked by a clinically proven protease inhibitor. *Cell*. 2020;181:271-280.
5. Zhou P, Yang XL, Wang XG, et al. A pneumonia outbreak associated with a new coronavirus of probable bat origin. *Nature*. 2020;579:270-273.
6. McCray PB, Pewe L, Wohlford-Lenane C, et al. Lethal infection of K18-hACE2 mice infected with severe acute respiratory syndrome coronavirus. *J Virol*. 2007;81:813-821.
7. Monteil V, Kwon H, Prado P, et al. Inhibition of SARS-CoV-2 infections in engineered human tissues using clinical-grade soluble human ACE2. *Cell*. 2020;181:905-913.
8. Smith JC, Sheltzer JM. Cigarette smoke exposure and inflammatory signaling increase the expression of the SARS-CoV-2 receptor ACE2 in the respiratory tract. *Dev Cell*. 2020;53:514-529.e3. https://doi.org/10.1016/j.devcel.2020.05.012.

9. Hikmet F, Méar L, Uhlén M, Lindskog C. The protein expression profile of ACE2 in human tissues. *bioRxiv*. 2020;2020:016048. https://doi.org/10.1101/2020.03.31.016048.
10. Cai H. Sex difference and smoking predisposition in patients with COVID-19. *Lancet Respir Med*. 2020;8:20.
11. Guan W, Ni Z, Hu Y, et al. Clinical characteristics of 2019 novel coronavirus infection in China. *N Engl J Med*. 2020;382:1708-1720.
12. Fang L, Karakiulakis G, Roth M. Are patients with hypertension and diabetes mellitus at increased risk for COVID-19 infection? *Lancet Respir Med*. 2020;8:21.
13. Pinto BGG, Oliveria AER, Singh Y, et al. ACE2 expression is increased in the lungs of patients with comorbidities associated with severe COVID-19. *medRxiv*. 2020;2020: https://doi.org/10.1101/2020.03.21.20040261.
14. Goldman M, Craft B, Hastie M, et al. The UCSC Xena platform for public and private cancer genomics data visualization and interpretation. *bioRxiv*. 2019;2019: https://doi.org/10.1101/326470.
15. Uhlén M, Fagerberg L, Hallström BM, et al. Tissue-based map of the human proteome. *Science*. 2015;347:1260419.
16. Lizio M, Harshbarger J, Shimoji H, et al. Gateways to the FANTOM5 promoter level mammalian expression atlas. *Genome Biol*. 2015;16:22.
17. Yu K, Ganesan K, Tan LK, et al. A precisely regulated gene expression cassette potently modulates metastasis and survival in multiple solid cancers. *PLoS Genet*. 2008;4:e1000129.
18. Lu TP, Tsai MH, Lee JM, et al. Identification of a novel biomarker, SEMA5A, for non-small cell lung carcinoma in nonsmoking women. *Cancer Epidemiol Biomarkers Prev*. 2010;19:2590-2597.
19. Selamat SA, Chung BS, Girard L, et al. Genome-scale analysis of DNA methylation in lung adenocarcinoma and integration with mRNA expression. *Genome Res*. 2012;22:1197-1211.
20. Girard L, Rodriguez-Canales J, Behrens C, et al. An expression signature as an aid to the histologic classification of non-small cell lung cancer. *Clin Cancer Res*. 2016;22:4880-4889.

SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of this article.