Blood Biomarkers of Emphysema: What Can They Really Tell Us?

Maarten van den Berge[1,2] and Alen Faiz[1,2,3*]

Authors' affiliations-

1. Groningen Research Institute for Asthma and COPD, University Medical Center Groningen, University of Groningen, Groningen, the Netherlands
2. Department of Pathology & Medical Biology, UMCG, Groningen, the Netherlands
3. Respiratory Bioinformatics and Molecular Biology (RBMB) Group, The University of Technology Sydney, Ultimo, NSW, Australia

*Corresponding author: Alen Faiz

Telephone:

E-mail: alen.faiz@uts.edu.au

Address: School of Life Sciences

Building 4, Room 04.07.332

University of Technology Sydney

Thomas St, Ultimo NSW 2007

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Single level transcriptomics and proteomics analyses have been used to unravel the complexity of COPD pathogenesis and treatment response (1-4). Thus far, resected lung tissue or bronchial biopsies and brushes have been considered to be the most relevant samples to investigate. However, these are difficult to obtain, particularly on a larger scale. Therefore, there has been huge interest in less invasive and more easily accessible biomarkers. Here, the manuscript by Suryadevara *et al*(5) takes advantage of available data from the two large COPDGene and ECLIPSE cohorts. The authors show that a multi-omics approach in blood allows for the identification of genes, proteins, gene isoforms and exons relevant to COPD that could not be identified smaller studies using sometimes more relevant tissues.

Another important aspect of the study by Suryadevara *et al* (5) is the focus on emphysema as a continuous trait rather than investigating the heterogeneous group of patients with COPD overall. Thus far, only a few studies have investigated the underlying molecular mechanisms of emphysema using genome-wide data, most of them using single-omics approaches with an assessment of the presence of emphysema rather than analysing the trait as a continuous variable (2, 6). Rathnayake et al, and Morrow et al. showed that transcriptional profiles from bronchial brushes can be used to identify genes associated with levels of emphysema measured by computed tomography-based parametric response mapping (7, 8), with the latter study also investigating blood and macrophages. Interestingly, up to 25% of the genes identified in these studies in bronchial samples were also significantly ($p<0.05$) associated with emphysema and in the same direction in blood as shown by Suryadevara *et al* (5). This is an important observation suggesting that emphysema-associated gene signatures may be shared across the different tissues of the body (figure 1). This is not a new concept as the "united airway field of injury" has been put forward earlier by Spira et al, suggesting shared disease-associated mechanisms across the lower and upper airways, whereas the data

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presented by Suryadevara *et al* now show that this concept may be expanded to blood, which is even more readily accessible. This is not a new concept as the "united airway field of injury" has been put forward earlier by Spira et al (9), suggesting shared disease-associated mechanisms across the lower and upper airways, whereas the data presented by Suryadevara *et al.* (5) now show that this concept may be expanded to blood, which is even more readily accessible.

Another unique aspect of the current study was the investigation into alternative splicing mechanisms. This analysis reveals the importance of gene isoform and dysregulated use of exons contributing to severity of emphysema. Change in function variants produced during translation would not be identified in a traditional gene expression analysis. The investigation of alternative splicing in COPD is not a new concept, however, most studies focus on single genes. One of the best examples is the association of RAGE (AGER) a cell surface receptor which has a splice variant esRAGE, which is associated with the polymorphism rs2070600 one of the top SNP associated with COPD and emphysema (10, 11). Furthermore, SNPs associated with the presence of COPD have been found in the gene FBXO38(12). These SNPs were shown to influence the alternative splicing in this gene, which is thought to increase the susceptibility to COPD. Together, this opens up new avenues for optimal use of RNA-seq data in future studies.

Suryadevara *et al* also investigate whether multi-omics from the blood can be to predict the presence of emphysema (5). Interestingly within their model in COPDgene the best prediction was with the presence of clinical factors, Cellular blood counts and proteomics. Interestingly when transcriptomics was added to the model it decreased the accuracy. It is well established that gene expression levels do not always equate to protein levels. This indicates that protein levels in the blood may be a better predictor than gene expression levels of features of lung disease. This may also be due to these proteins being produced by

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structural cells in the lungs which travel to the blood while the transcriptomics is restricted to blood cells. Unfortunately, in their replication cohort, protein levels were unavailable so the authors were unable to test this theory.

One of the main limitations presented by the authors was the inability to analyse the variability of the immune cell subpopulations and their association of blood counts with emphysema. This is an important consideration as changes in cellular composition is known to greatly influence both transcriptomics and proteomics; therefore, it is difficult to determine whether the emphysema transcriptional signature is due to a change in gene expression or a shift in cellular compositions of rare or common cell types. To overcome common blood cell types, the authors reran their analysis, adjusting for blood cell count data and cellular deconvolution where most analyses remained significant. However, this does not compensate for rarer cell types that may be more abundant during disease. Single cell sequencing studies on PBMC obtained from COPD patients in the future may be able to disentangle this effect.

In conclusion blood transcriptomics and proteomics in large cohorts can provide important insight into complex features of disease. Suryadevara et al. have provide one of the largest and well powered blood-based signature studies to investigate the association of biomarkers with degree of emphysema.

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Figure 1. Transcriptomics associated with emphysema

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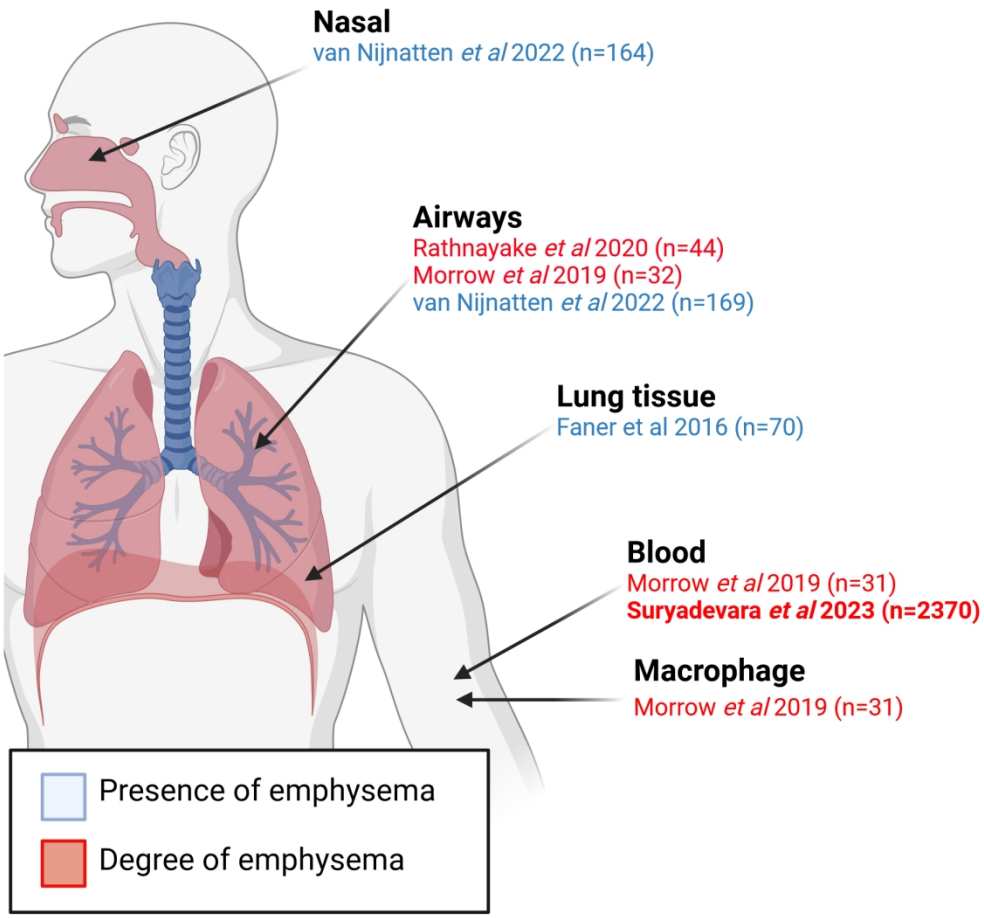

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