[MUSIC PLAYING]

FRANK SCIURBA: So today we're going to talk about precision medicine in COPD, and I'm going to-- we're going to go bedside to bench here. And I'm going to talk about the bedside and translational clinical practice. These are my disclosures. So we're going to discuss the variation in COPD, both physiologic and biologic, and the really the unmet need with guideline-based therapy that really requires us to start thinking in a more precision direction.

So these are some of the most recent definitions of COPD, which are very different from when I trained that said it's an irreversible disease in smokers. It's common. We should pay attention to it. It's preventable. I'm not going to talk about smoking cessation. It's always the elephant in the room. And so acknowledge, I understand that. But we have to treat these as people and human beings, despite our bad habits, and try and make their life as good as possible.

It is treatable. So while you're learning about your statins and SGL2 modulators, please pay attention to COPD because we can optimize and improve these folks' quality of life. It has airflow obstruction and limitation as its primary defining characteristic. But there are many mechanistic components and variation that go into that, from alveolar loss of recoil to increasing airway resistance. And this variation in heterogeneity likely lies in very different cellular molecular cascades.

So it's an umbrella diagnosis for really many conditions. One of our investigators in the field described it as hundreds of orphan diseases. And really that's the focus of precision medicine, recognizing that our naive classifications require much more detailed understanding and redefinition of the disease. Emphysema chronic bronchitis, refractory asthma, some forms of bronchiectasis are often classified under the title of COPD.

The symptom presentation is often very variable. But dyspnea is common, particularly with exertion, cough, sputum, and chest tightness, and they may not even occur together. And so paying attention to that variation is important.

We talked about the variation in COPD in the 1950s and '60s when the gentleman on the left, represented by Frank Netter, the emphysema is pink, hypermetabolic individual, versus the chronic bronchitiic edematous hypoxemic individual on the right. And with evolution of CT scanning, we can see the panel on the left, the CT image has pretty much complete lung destruction with low attenuation. And on the right, preservation of the parenchyma in that individual has a similar degree of airway obstruction, likely related to the airways.

And it really leads one to believe, mechanistically, these have to be very, very different. And we're naively classifying them and treating them the same. And we've done a lot of work on the association of comorbidities with COPD and the different lung features.

At our institution, we discovered a linkage, independent of tobacco smoking, with lung cancer. Osteoporosis, Jessica Bon has taken on and brought to the next level. Vascular disease has been a focus of Divay Chandra, independently linked with emphysema and COPD, likely from a systems biology standpoint, independent of tobacco exposure.

We worked with the folks in Vancouver and brought these locally and did a lot of work internationally on quantitative analysis of airways and emphysema. And you could see, in this panel, where airways are thin and there's very little low attenuation, individuals, despite tobacco exposure, has relatively normal lung function. These folks have very little emphysema but thick airways. These folks-- a lot of low attenuation. These folks here have both and have the worst lung function. So the anatomy begets the physiology.

Another aspect of variation in the disease is the rate of progression. And if you have 100 individuals, often will show the Fletcher Peto curves of the average rate of decline. But that really doesn't tell what's going on with these folks. And what's going on is some individuals stay pretty much stable. They look like non-smokers. Others decline gradually and much more rapidly. Others seem to have a much more sporadic approach, either related to an autoimmune-type reaction or associated with exacerbations, but inevitably a decline. And these variations likely represent unique mechanisms in these individuals.

Another aspect of the disease that is important to follow that may be independent of FEV1 is the rate of low attenuation or emphysema progression. And this is really the dominant area of focus that Dr. Konigshoff works, with and it's the most difficult thing to study as an outcome in COPD.

This is one of our most rapid progressors in our 700 patient score cohort. You can see-- you can barely see, at two years, any change in that emphysema. And at six years, it becomes more apparent. But it really is subtle. This disease eventually is devastating, but it moves glacially. And so to study it as an outcome is difficult, and that's why I'll make an argument for biomarkers.

This shows the distribution of low-attenuation progression. And if we're going to give a very expensive biomarker to these individuals, we certainly don't want to target these individuals who have zero change in low attenuation at two years. Yet these folks here, which are the minority of the individuals, have had pretty dramatic progression. And if we could give a biologic to stop it earlier on, it would make a huge difference in their lives.

The variation in exacerbation rate, while there is some linkage to severity of lung function, can occur very independently as a domain of this disease that warrants treatment. And exacerbations are the number one driver of costs in COPD, accounting for 60% to 70% of the actual costs.

The GOLD criteria are probably the most common guidelines used in COPD-- goldcopd.org if you're interested. And while they acknowledge severity of lung function impairment, which is the traditional metric, FEV1, of severity, in fact, the dominant classification for this disease is based on symptoms and exacerbation. And individuals with two outpatient treated or one hospitalization are considered this GOLD Class E, which is a big focus of treatment. And the majority of studies going through the FDA right now are attending to the unmet needs for exacerbations. Although that still does not eliminate the need, really, for symptoms and lung function decline independent of exacerbations.

As far as predictors of exacerbation-- and these are what that GOLD E class is based on. Based on the UK database, they identified a sentinel year, the frequency of exacerbations, and then followed these individuals for 10 years. And the individuals that had no exacerbation in that sentinel year had less than a 25% chance of having an exacerbation in the next year and really a low risk. One exacerbation is higher, but two or more outpatient exacerbations or one hospitalization create a fairly high risk for an exacerbation in ensuing years. So to some extent, it's obvious, but this actually exceeds the ability of any other biomarker in predicting who's going to be an exacerbator.

So why do we need biomarkers? And we're not very good at it yet, but I'll show you some progress in this area. We talked about the slow rate of decline, the incredible variation in the disease. And it allows some mechanistic-specific potential targeting. So hopefully we can define biomarkers that determine prognosis those folks that are going to decline and also as an early marker of response. If it's going to take three years to see a change in emphysema, we'd want to know that something's happening before then, if we're giving, again, an expensive biologic.

This paper has been cited over 4,500 times-- I was the tissue deliverer to the famous James Hogg in Vancouver-- that really defined-- I mean, we take for granted inflammation is important in COPD now. But, really, it was not thought of. It was more thought of as mechanical matrix destruction by other mechanisms.

But this paper really is increasingly informative, and it shows the proportion of individual airways that have various acute and chronic inflammatory cells identified. And one thing that was somewhat not even discussed in the paper was about a third of the patients had actually eosinophils in the airway, that, with recent development in therapeutics, becomes an area of interest and probably the most targetable cell with current therapeutics as they're evolving.

Melanie thinks this figure is very simple, too simple. And so-- and it is. There's obviously hundreds of molecular pathways that can affect the inflammatory response, and they all occur in different patients with COPD with different degrees and patterns. And so in a simplified way, the TH2 pathways, IL-5, IL-413, eosinophils, IGE-related pathways, and even within the statement of TH2, there's significant variation in complexity. More thought to have been related to asthma, but increasingly being identified as a contributor to progression in COPD.

More traditionally in COPD, we've thought of TH1 and now TH3 pathways that result in neutrophil attraction and involvement in the lung. And so it's likely that individuals who are dominant in one way or another would require different therapeutic interventions if we're targeting the inflammatory response in this disease.

As far as biomarkers, there's only one biomarker that the FDA has approved. And this was at the behest of John Walsh in the COPD Foundation that really pushed this through after a ton of work by our group in trying to identify biomarkers. And fibrinogen, which is really an acute phase reactant likely linked to inflammation, is associated with exacerbation frequency and, as a threshold of 350 nanograms per deciliter, also associated with survival with nearly double the mortality rate at three years with high fibrinogen versus low fibrinogen.

We've done some work with a lot of biomarkers, and there's really nothing that independently shows significant predictive value. We found some with mild statistical significance. One of the markers-- and Yingze Zhang has done most of this work with us. We found PTX 3, which is a matrix-associated peptide that's one of the most differentially expressed markers, in COPD, linked modestly strong with the degree of emphysema low attenuation and is a mild predictor of survival. And so its use in defining therapeutic targets and response would need to be evolved further.

Blood eosinophils-- and we're talking about eosinophils in the CBCs that you order on your patients regularly, are available in the charts historically on almost all of your patients-- winds up being currently the most usable biomarker that we have available. This slide shows what's been reproduced in multiple studies, that it is a predictor of exacerbation risk and, in this case, likelihood of readmission to the hospital over the prior year, those with high eosinophils versus low eosinophils. And the threshold that they used in this paper was 2%. And we'll be showing you more about the use of eosinophils in current therapy.

Biomarkers may be obtained at baseline in individuals when stable. Dr. [INAUDIBLE] looked at subtyping inflammatory response with exacerbations. And it winds up individuals have tended to have similar inflammatory response patterns or exacerbation phenotypes on repeated exacerbations. And there's a bacterial viral and a TH2-type response and the pauci-inflammatory response with some overlap. But some individuals will spike their eosinophils regularly when they have exacerbations. And that may, in fact, be a targetable inflammatory response.

So in designing therapies for COPD, attending to the different outcomes and the different domains of this disease may influence how we design the trials and what we're looking at as outcome measures. Declining lung function, traditionally used, is not the only parameter. Emphysema progression may be independent of decline in lung function.

Symptoms, quality of life-- we've got our first PRO as a primary outcome in a trial that's ongoing right now, but FDA has, in general, not accepted surveys as outcome measures and symptoms. Activity improvement-- reduction in exacerbation is an acknowledged and important outcome measure that the FDA recognizes. Survival-- obviously, the holy grail that we work toward. And just reduction in costs is also an important feature.

These are the drugs-- mainly three classes of drugs-- beta agonists, anticholinergic, and inhaled corticosteroids-- that have been the basis for the last 30 years. And currently, approved drugs don't go far beyond that. They've gotten longer acting and a bit better.

Currently in COPD, the guidelines are LAMA LABA, long acting muscarinic antagonist and beta agonists, in everybody with COPD and airflow obstruction. That came into the GOLD guidelines this year. No patient with COPD becomes completely asymptomatic. And so maximal bronchodilator therapy is the baseline. That's not precision. The addition of an inhaled corticosteroid, on the other hand, has now gone a bit into the precision realm. And we'll talk a bit more about that.

Triple therapy with inhaled corticosteroids or-- this is a summary in our *JAMA* review article of all the Cochrane analyses of long-acting agents in COPD. And this basically describes that patients with LAMA LABA do better than those with LAMA or LABA alone and really justifies using that. If I can tell you one thing to do now is every one of your patients should be on one of the LAMA LABA preparations.

This is looking at triple versus either the LAMA LABA or the LABA ICS, Advair, which has been used forever, is no longer indicated in COPD because it does not have a LAMA antimuscarinic in it. But while I show in an overall group that the triple is better than any of the doubles, in fact, there's more to that.

Despite all these therapies, independent of the arm, you're left with an unmet need that approximately 50% of patients still have exacerbations. So even though there's statistical significance of these preparations over certainly placebo, over triples, over doubles, it still leaves really a significant unmet need.

And this brings in the first biomarker that is considered in COPD and within the guidelines. And again, that's just your baseline eosinophil count. And if you look at the impact study, which I just showed you, and you look at the individuals with low eosinophils, less than 150 versus those greater than 150, you see a difference in response. This is the group that had just LAMA LABA without the ICS, and they have a significantly higher exacerbation rate than either of the ICS containing arms if the eosinophil count is high. Whereas there's no difference in response by adding the ICS if there's a low eosinophil count.

And so the guidelines really do recommend considering eosinophils in whether we put patients on ICS. And why bother even thinking about it? Because there's a 2% to 3% incidence of pneumonia per year due to inhaled corticosteroids in individual. So there's a downside to treating individuals who don't deserve it. This is a meta-analysis looking at formoterol ICS versus formoterol alone and looking at the only differences occur with higher eosinophil levels in these experiments.

So what is precision therapy? So this is the traditional way we look at intervention in COPD. That's looking at the whole group, looking at the mean change, and saying it's statistically significant or not. In precision therapy, we look at the separate dots, and we try and isolate the green group, who actually does worse, the red group, who has a significant response, and then the yellow group, which may be a cost-effective decision, and the white group where there's no change.

These are the exact same dots as the gray dots. It's just that we now have, hopefully, biomarkers to define these groups to pick out the right patients for the right therapy. There is one therapy that has an endotype, a molecular cause, a phenotype, a pattern of emphysema, and a therapy that affects that. And that's alpha-1 antitrypsin deficiency and a deficiency in the inhibitor of neutrophil elastase.

This is a meta-analysis of two studies that shows a significant reduction in increase in low attenuation or emphysema on serial CT scans in individuals treated versus untreated with replacement therapy. And so, unfortunately-- or fortunately-- alpha-1 is the cause of about 2% to 4% of all cases of COPD. And we haven't been very good at picking out other genetic causes that are targetable, and there's a lot of work going on in that.

There's been a lot of mouse studies that have shown abrogation of emphysema. This is a study that we followed up with that showed an elastase model in a rat given all retinoic acid prevented the development of emphysema. But in the clinical trial looking at low attenuation change on the CT scan and other clinical attributes, it was a negative study.

There's other-- telomerase, losartan-- other selective retinoic acid receptor antagonists. All have been studied that showed preliminary data in rat models but did not pan out in human models. So we need to get better at doing this, both the outcome measures and the targets.

This drug was touted as really the first precision medicine therapy in COPD. It's roflumilast, which is a PDE4 inhibitor. Now, roflumilast does result in about a 14% reduction if it's a selective group of frequent exacerbators with low FEV1 but has huge GI side effects and is not a drug that is generally well tolerated. Doesn't improve quality of life. And so, in general, it's really not a breakthrough, even though it is the first new class of drug in this disease.

Azithromycin, based on its anti-inflammatory and antibiotic effects, showed in an overall group of COPD a 27% reduction in exacerbation rate. If you look at the forest plot of subtypes, in fact, only patients with more moderate disease and former smokers responded to this therapy. So it has a precision twist to it. This is not FDA approved for this purpose but is very commonly used in patients who persist in exacerbations.

So how about all these other pathways? What else is going on with that? Well, IL-5 modulation, which is an approved drug in asthma-- mepolizumab. We did a study in COPD without features consistent with asthma with a broad range of eosinophils. There was about a 20% reduction in exacerbation rate in sister studies. The FDA did not feel it was a significant enough effect and did not approve the drug.

Interestingly, if you look at the range of eosinophil counts in this study, as the eosinophil count gets higher in the individual patients, there's a trend to significance toward a response. And in fact, the historical eosinophil count of greater than 300 was the strongest predictor of reduction in exacerbation in this group.

Two months ago, based on lessons learned from the mepolizumab trial, dupilumab, which is an IL-4, IL-13, TH2 modulator, showed very significant results with a 30% reduction in exacerbation rate in a highly targeted group who have more than 300 peripheral eosinophils and frequent exacerbations. So this trial is waiting for its sister trial to be presented to the FDA but likely, based on these results, will be approved in high eosinophil COPD.

This is another product that's in presentation to the FDA-- an inhaled PDE3/PDE4 inhibitor that largely more targets neutrophilic inflammation in the lung, showing the primary outcome of improvement in lung function but also a serendipitous finding of a reduction in exacerbation rate.

How about the TH1, TH3 neutrophil pathways? There's currently classes of drugs-- the alarmins, which are epithelial cytokines that can drive both TH2 and TH1, but finding that largely TH-1-driven pathways here are affected with anti-IL-33. The first phase II trial showed really no overall effect, but really about a 40% reduction in former smokers versus non-smokers. And so the phase III trials, there's three different products out there that are ongoing.

We made an argument that device intervention was actually the closest to precision therapy. We showed, in lung volume reduction surgery, upper lobe dominant disease really was the only responsive therapy to improving survival in lung volume reduction. And we tried to target valve volume reduction. We identified fissure integrity, a lack of collaterals between lobes as a driver of who responds and who doesn't respond.

We showed that endobronchial valves significantly better than placebo in improving lung function, quality of life. This is an emphysema-targeted therapy. We also have a device targeted RFA ablation of the vagus nerve. This trial is completed enrollment and is ongoing. So I'm going to finish up with that and pass it on to Melanie so she can tell you about the basic science component. Of course, we don't do this alone, and Melanie is one of our contributors. Thank you.

[APPLAUSE]

MELANIE KONIGSHOFF: I think we are on. Thank you so much, Allison, for the introduction, Frank, for being such a great partner in this adventure, actually, that we had in terms of really trying to think about, how can we develop novel therapeutics also in the realm of COPD?

So I just would like to directly go in, no conflicts. The learning objectives you also already see. So I think one of the things that Dr. Sciurba mentioned is really that [INAUDIBLE] there is a lot of ways that we really focus on airway inflammation and remodeling. And that has been something that in COPD has been the focus for a long time in terms of therapeutic development, and I just want to point you to this *Lancet* commission article, which is quite a big article, actually, really outlining a lot of different things which we believe is important to eliminate COPD for the future.

So this is a high aim. And one of the things is that next to airway inflammation, next to airway remodeling, we really also have to go back into our focus on precision medicine approaches, such as go looking into emphysema. And if I talk about emphysema, I really wanted to highlight that this is a alveolar tissue destruction and degeneration that we want to understand better so that we can get target for novel, curative, and regenerative therapies.

So this is the slide from Dr. Sciurba. It's not simple at all. And I think what is really good to show this picture about is that we really have learned a lot about inflammation, even though it just came back to the attention in 2004, if you so want, and also that we have important proof of concept, again, like highlighted in the first part of the talk, that we can really bring biologics to the clinic.

So in general, this is very hopeful. And of course, for this shows us that we have feasibility for drug development in COPD. However, there is a lot of unmet needs that we still have to understand specifically in the realm of precision medicine. I put here the NIH definition for precision medicine, again, just as a reminder about genetic, biomarker, phenotypic characteristics that we have to understand.

And Frank already talked with you about biomarker, and I will come back to this, but also the genetic and the phenotypic basis of these diseases we need to understand better. And right now, we're still lacking a lot of this in terms of endotypes and phenotypes. We also still miss a lot of basic understanding, mainly because we also neglected basic research for the longest time. And there is just also a pretty big lack of models that we need to develop, again, to highlight that we can not only focus on animal models but also really know clinically relevant models that we can move forward.

So there is advances in all of these areas, and I would like to highlight all three of these areas that I really feel like have been made a move in the past couple of years and really bring us closer to precision medicine approaches in COPD. So first of these are the genome-wide association studies and CT-based imaging in large multicenter cohorts, which is a team in Pittsburgh, led by Frank, is actually a big part of.

Then there has been a lot of molecular phenotyping and multiomics also in COPD biospecimen, which I will touch briefly on. And then I really want to talk more about clinical relevant model systems for translational studies and how we can use them to hopefully bring us closer to an understanding of the disease but also to develop really novel therapies.

So first let me talk a little bit about the genome-wide association studies and CT-based imaging. And I think the COPDGene group actually has been really one of the groups that has been-- with large NIH funding-- has made a tremendous effort in that. And again, like in Pittsburgh, there have been a huge part of this adventure.

So there are over 10,000 individuals where we have genetic factors, where we also have CT-based imaging, even plasma biomarkers. And for the longest time, you have seen publications with all these Manhattan plots, as you can see over here, with genetic associations. But what is happening now and what I think is a move that we really can see throughout the community is that gene to mechanism movement, which is also highlighted here by the top initiatives.

So we have to move from just collecting these data to really understanding what these genes and these associations really do. And I highlight one paper here from the group from Andrew Wilson, actually, from the CReM in Boston where they now look for some of these specific genes that have been identified in COPD and use IPS cell technology to see if they can better understand what really this gene is doing in terms of COPD pathomechanism.

And that is one of the, I think, very nice first studies showing us that we can really use all these information that we gained over the past couple of years with genetic associations and now have to move this forward into functional studies. There also, of course, have been a lot of advances in molecular phenotyping and multiomics. So I mean, single-cell analysis has been actually, I would say, like one of the things that has really revolutionized many different diseases that we are studying and, of course, also, COPD.

And within COPD, there has really been an explosion in data, actually, over the past couple of 2 to 3 years. One of these studies over here, you can see, really highlighting, actually, that we haven't really known a lot about the cellular composition. But what we have learned from the studies up to now-- and again, we just see the tip of the iceberg right now-- is that there are, A, novel immuno subtypes that we haven't really been considered beforehand and which will definitely be impacting the current treatment strategies. We also have learned that there is a lot of impaired structural cells, such as stem cells that are impaired. And I will talk about this a little bit further.

But there is really this growing map. And one thing that I would like to convey to you is that I feel is-- it really was a little bit of an a-ha effect for everybody-- is that we study most of these single-cell data in end-stage tissue. And a lot of you would probably say, this is burned-out tissue, and yes, for sure, this is really advanced stages of disease. But there has been the observation that there is a lot of plasticity, even in these burned out lung diseases. And I just want to get this as a thought over to you that we do maybe, even in a burned out disease lung, in a very advanced lung disease, we still have possibility to maybe target specific cellular and molecular mechanisms.

So then I come to the model systems, and that's something else, all of this coming together really allowing us to move forward. And I'm highlighting here a review actually done by a colleague of mine in pulmonary, Dr. [INAUDIBLE], who's really working on Lung-on-a-Chip models that you can see on the right. But there has been a huge advance in general in the development of more clinically relevant models over the past couple of years.

Acknowledging that animal studies, as Frank has outlined before, are not really always bring us really moving forward because some of these studies have never been really translated. So there is a need to really work with human tissue, human cells, and develop other systems. And the one thing that I would like to talk more about for you today will probably be the complex tissue models that you can see down here, so precision cut lung slices, to show you how these technologies can be used to hopefully help us moving forward.

That brings me to our general aim within the lab. It's a very big question that we that we're trying to answer is, Can the human aged and disease lung regenerate? which is a bold question. Sometimes it also sounds like a naive question. But, again, I do feel that, with a current field and how we know more about the cellular mechanisms that we do have a starting point which we want to follow up on.

And what I would like to really show you specifically is how we use as an aim to investigate potential regenerative therapies. So I will focus on regenerative therapies in these clinically relevant model systems.

So if you're thinking about regeneration, there is two main areas of how you can approach regeneration. The one is you really look into endogenous repair capacity, and that is what my talk will focus on, really with the question of, How can we harness the endogenous repair capacity of the lung and understanding what's really going wrong in such disease as COPD? So we have to understand the cells, the pathways involved in this.

There is-- and I just want to mention this, also-- a huge effort around the community to foster regeneration exogenously. There has been bioreactors for lungs, for example. People have built new scaffolds. Of course, this is not really close to translation yet, but there are also efforts that hopefully will help us in the future to better address these open questions.

So I want to focus on the first part over here, tell you a little bit more about the stem cells and the [INAUDIBLE] pathways that we believe might be helpful and give you one example as a concept moves forward. So just a quick note to non-pulmonary people in the room and on the line here. As many different organs, the lung is highly complex, and especially the epithelial cell system is highly complex. We have several different subtypes, even on the basis.

And I put this one out here just to show you how many different cells we basically have. And it's, by now, much more that we know of, but it's a simplified way. And the important thing here is really that you look at the arrows, because the arrows actually mean that we do have stem cells that serve as progenitor cells for other cells. And specifically for the emphysema part in COPD, so really at the distal lung, we are interested in the alveolar part that you can see here on the lower left where you can see 82 cells and 81 cells, which are really our prime cells, with 82 cells serving as a stem cell within the lung.

The pathway I would like to talk about is with beta catenin signaling as one example of a regenerative target that has come up over the past couple of years. And the reason why this actually has came up or has been in the focus of a pathway is that we know that wind signaling is absolutely essential for the lung to develop in the first place. So if you need it to generate, it's most likely also that it's something that you maybe need to regenerate the lung.

It also has been very well documented by now that this is a pathway that is very powerful in setting and making the homeostasis of specific lung stem cells. What I need you to follow up on for the next couple of slides is that beta catenin is one of the classical surrogate markers. We know that out of pathology for different cancers, beta catenin really in the nuclei shows us that this pathway is active and that is highly regulated by a GSK3 beta complex here that I will talk a little bit more about in the future. And basically, we have started to look at this pathway really from the basic discovery, and I want to show you some data in COPD that we have over to preclinical models and also finally to human cohort studies.

So the pathway is indeed reduced in COPD and specifically in emphysematous tissue. I show you here human data as well as mouse data to start with, basically showing you that beta catenin as one of the surrogate markers is not activated. It's not in the nuclei in the disease. So really showing you that, also with other models, that this whole pathway is downregulated.

We also have actually really looked into this in different models over the years, and I hope you can see-- and this is also something I'd like to highlight, that over the past 15 years, we really have built on this story together with other collaborators. And also, now, with new methodologies and technologies still could see, if you look here on the right-hand side on single cell results, that we do have a reduction in wind signaling in the disease and in different models.

I also want to highlight just a little bit that there is a big immune cell compartment in there, which is actually affecting this wind break that we see in COPD. And that is something that I feel is very important, especially if you have listened into Frank's talk where we have a lot of anti-inflammatory therapies already available.

So we did the usual thing, and I really want to go quickly, actually, over this-- is we went into the mouse model, as have done people previously and so on. And yes, indeed, if we activate wind and we have used for activation of wind in this case, something called lithium chloride, which is an FDA-approved drug for bipolar disorders, and could show that lithium chloride indeed, via wind signal activation, leads to an attenuation of emphysema in the specific mouse model.

And over the years, we have confirmed this in other models, like the cigarette smoke model, and really try to build, actually, the evidence for wind signaling and wind signal activation as a regenerative target in COPD. However, this is where a lot of studies also in the past have then stopped because we didn't really had other methods available and just looked into the mouse models.

At this point, we actually said, how can we move forward without really knowing if this plays a role in disease without going directly-- oh, this goes automatically.

[VIDEO PLAYBACK]

OK. So this is how we do it. We actually went to a model, which is called Precision Cut Lung Slices-- apologies for the tone here. I didn't know that this was in there-- where we actually take advantage of living human tissue. And look at the right side here on the scheme.

And we use this after-- we filled these lungs with low melting agarose. We can cut it into very thin slices, which you can see in the middle part. So this allows us really to actually develop something-- apologies.

- Yes, the tissues--

[END PLAYBACK]

MELANIE KONIGSHOFF: Let me go to the next slide. This was actually one of my colleagues in the lab, Dr. [INAUDIBLE], who was working on this. So we'll do these models in a way that we get these 300 to 500 tissue micrometer slices. And what I would like to highlight is that, by getting these tissues from human patients, that we really are able to look at individual patient tissue.

So thinking about precision medicine, you can think about, like, yeah, really looking at individual human lung tissue over time. It has been a great tool for drug discovery and validation. I would like to show this a little bit further. But what I really wanted to highlight here is that we can use this, hopefully, for understanding mechanisms of action of drugs, as well as of specific patient responses in terms of biomarkers.

There's also, of course, a couple of limitations to the system that I also wanted to highlight directly at the beginning. We have a piece of lung in culture. We don't have perfusion or ventilation, for example, just to keep this in mind.

But the principle of how we can use such technologies is then that we have these, for example, diseased lungs, as I just mentioned, from COPD patients, for example. And we can then generate these PCLS, which you can see on the right-hand side, and test different drugs and test for different biomarkers. However, the other way I also would like to highlight is that you can also use donor lungs, again, individual donor lungs with a specific exposure, with a specific genetic makeup, and then can also develop diseases and really try to develop early disease models, which we also have been focused on in the past.

I would like to show you some data of the COPD study now that we have done. So here you can see how COPD lung tissue looks ex-vivo when we use it for these studies. And this is actually both that we use tissue from lung explants. So this is really end-stage disease lungs. But we also use tissue from lung resections.

And what we actually-- basically, for the very first time, we are then asking, can wind signaling really be activated ir this tissue? And that question comes back to the burnout idea that we always all have, is can this tissue, after explantation, even react to something like a stimulus from the outside?

So on the left-hand side, you can see that we have proof of concept that indeed, when we take lithium now as another example, again, we can really activate a pathway. And this is just a few small dots at the beginning, but it was a very important proof of concept for us, showing that it's indeed not a burnt out tissue that is not responding to anything, but indeed we can activate the pathway also ex vivo.

And on the right-hand side, you also see data where we can see now that we have a response from these classical type 2 cells, from these classical stem cells in the lung tissue. And the very nice thing that we feel over here is, on the right-hand side, is that the surfactant protein C secretion, which is one of the key features of every type 2 stem cells within the lung, is also measurable in terms of secretion. So we are also able to really look for secreted proteins when we study these lungs.

And the second part that I would like to highlight here is actually then what we discussed at the beginning. If you're thinking about precision medicine now, now you have these results within your culture dish. But what does it actually mean in terms of the patient data that you have to this? And I think the really advantage of one of these technologies really is that we can do correlation with clinical parameters.

In this case, we had available lung function parameters, and I hope, if you can see on this slide, you can see that the response that we see in terms of treatment is correlating with the lung function of specific patients. And you just put your attention to the right one where we have the surfactant protein secretion. It shows you that if you really have a severely impacted patient with low lung function parameters, as you can see over here, you even get a better response. So there you have a response. Whereas patients which have more normal lung function didn't really had a good response to that treatment.

So as an example of showing you how you can really dissect and coming back to this very nice scheme that Frank had is, like, how can we separate between the green and the blue group? And maybe we can do this with these parameters that we have at hand. So basically the disease stage in this model really impacted the drug response.

So overall, I think that precision lung tissues or precision tissues in general are very helpful for moving forward with precision medicine approaches. And it's summarized over here. And I would like to highlight that especially the biomarker part is something that is very active right now in terms of that we use these PCLS and identify the biomarkers that are secreted. So in this case, we can look for biomarkers that do have a tissue cellular response, but then, hopefully, we can also move further to plasma biomarkers and help with therapeutic responses in vivo in the patient.

I'm highlighting a little bit our PCLS cohort here, which we have built over the past three years since I have moved to Pittsburgh. We do have a lot of different setups now where we do have these precision cut lung slices from different patients. And we have them treated with several different wind activators, obviously, and we can now subject these to multi-omic analysis, for example.

And on the other hand, we'll do this proteomic studies that you can see in the lower right, again, with the idea of looking for biomarkers, some which are known in COPD, like [INAUDIBLE], which has been highlighted, but also some others. And the other aspect over here is really important, I feel, is the early disease modeling where we try to model disease aspects in human tissue. That has been a huge discussion in terms, also, of new therapeutic approaches because, again, we don't want to only intervene in a very late stage. But we also really need to understand earlier mechanisms in the disease to hopefully also intervene very early on.

So for the last couple of slides, I want to move back to the human cohort studies, actually, that we're doing and talking a little bit about lithium, also. We have used lithium as a classical wind-activating drug. But as many of you know, lithium is also an FDA-approved drug for bipolar disorder. It actually has been around for many, many, many different years, and we're thinking about, How can we now actually look into human patients that take lithium as a drug, and how does this affect, potentially, COPD or respiratory health?

Just want to have one slide over here that lithium really has been around for the longest time. Some of you might remember that 7 Up had lithium in it as a mood stabilizer and something which is refreshing in the '60s and '70s. But even before that, there is lithia springs around the country, which always have been, actually, highlighted to be a nerve tonic and to be a mood stabilizer and, again, might even be something to consider for COPD. However, you can see there is smoking advice directly below that. So that's just a small remembering that lithium really has been around for the longest time.

So how can we now really study lithium and its impact in humans on respiratory health and COPD? So there has been actually studies that are spearheaded by Divay Chandra in the division. He really is a great epidemiologist and pulmonologist which we are collaborating with who has helped us in getting some data on this.

So we have looked into two major biobanks. Actually, one is a UK biobank, which is a large-scale biobank with over half a million UK participants. It's the population. It's not really the COPD specific. But we really ask the question, can we identify lithium patients actually on lithium therapy in these cohorts, and do we see an effect on various outcomes?

So what I can show you and I want to just highlight a little bit the numbers because that's not easy to identify. There's, even in this very large cohort, only very, very few lithium users. And that's, of course, making a definitive answer, especially in such a large observational cohort, very difficult. However, what we could see is that lithium therapy indeed was associated with higher lung function measurements.

So this was something which we took further on and looked a little bit closer and also looked into different other outcomes, what's also available within the UK biobank. And one of the things that is available, also, there is a likelihood of an MD diagnosis of emphysema where you can also see that people that are on lithium therapy actually have a highly reduced odds ratio for being diagnosed for emphysema.

So this is, again, a huge cohort which has multi-morbid patients and it has multi-different-- and they could have all different kinds of diseases, obviously. So we also went back to COPDGene, which is a cohort which I introduced to you at the beginning where we really have large data on genetic associations and plasma bio levels. And we also there ask the question, can we identify, within these cohorts, also patients that are on lithium therapy?

And indeed, we have. Again, I show you the very small numbers that you can see over here. In the cohort, we have 45 patients on lithium. And the other number actually was no lithium, other mood stabilizers that people have taken. And some of the promising results that we can see over here is that lithium therapy, in these patients, is associated with less radiographic emphysema, so really bringing, actually, our basic discoveries to start out with to something where we feel like we have, at least in observational studies which we need to further understand, have some evidence that this might also be relevant.

So we now comparing, obviously, these data with all the other data that is available within COPD gene that I mentioned to you. So we're looking for SNPs as well as plasma biomarkers. We also look for disease progression, which, as Frank has highlighted, especially for emphysema, is a longstanding-- it takes some time. And so within COPDGene, now there is data with 5 to 10 year follow-up that we are interested in looking at. And we are also interested in looking into the VA million veterans cohort, obviously.

All right, so this was a little bit of an outline, also, of how we can hopefully come to precision medicine going back to clinical cohorts and use the data that is available by clinical phenotyping as well as genetic association studies to move precision medicine forward.

So with this, I want to end and hopefully give us a couple of minutes for discussion. I hope I could show you a little bit that there is novel endotypes on the horizon for COPD, specifically with this in-depth molecular and cellular characterization, which needs to be expanded upon. But, obviously, it gives us very early hope that we can contribute to that phenotype or endotyping, that we need novel approaches in terms of concept where we want to really target COPD and that regenerative therapies really have been on the horizon on that, and people specifically try to understand what of regenerative targets, such as wind signaling, but also others, And how does that then interplay with the inflammation part? which is for sure very dominant in all of our COPD patients.

And then the use of these models that you see down here with, for example, precision cut lung slices that I mentioned to you, but really that these models where we can combine the human background and the variety, of course, in every individual with a specific reductionist treatment, which hopefully will be some of the basis for precision medicine approaches. And with this, I want to thank everybody again. This is a huge teamwork with the whole Pitt team. And especially I will highlight, obviously, our COPD team. And, yeah, happy to take any questions, together with Frank.

[APPLAUSE]