Transcript Details
This is a transcript of an educational program. Details about the program and additional media formats for the program are accessible by visiting: https://reachmd.com/programs/heart-matters/exploring-novel-tools-for-pah-measuring-severity-with-a-new-blood-test/13963/

ReachMD
www.reachmd.com
info@reachmd.com
(866) 423-7849

Exploring Novel Tools for PAH: Measuring Severity with a New Blood Test

Dr. Sorrentino:
Pulmonary arterial hypertension is a rare but devastating condition causing high pressures in the lungs, difficulty breathing, chest pain, and fatigue and can quickly become fatal. A novel blood test has recently been described that can be used to evaluate disease severity, and hopefully, improve survivability in patients with pulmonary arterial hypertension, or PAH.

Welcome to *Heart Matters* on ReachMD. I'm Dr. Matthew Sorrentino. And joining me today are the two co-authors of the study published in *Circulation* coming from the National Institutes of Health, Dr. Michael Solomon and Dr. Sean Agbor-Enoh. Dr. Solomon is the head of the cardiology section for Critical Care Medicine and Co-Director of the Translational Pulmonary Arterial Hypertension Program at the NIH. And Dr. Sean Agbor-Enoh is the chief of the National Heart, Lung, and Blood Institute's Laboratory of Applied Precision Omics.

Dr. Solomon and Dr. Agbor-Enoh, welcome to our program today.

Dr. Solomon:
Thank you. It's a pleasure to be here, Dr. Sorrentino, and I appreciate the invitation.

Dr. Agbor-Enoh:
Thank you very much.

Dr. Sorrentino:
So, Dr. Solomon, let me start by asking you a question. Can you give us a little bit of background on how we currently diagnose and test for patients that we suspect of having pulmonary arterial hypertension?

Dr. Solomon:
Sure. I'd be glad to. So first, I want to start off and just say that there is currently no blood test that can specifically diagnose pulmonary arterial hypertension. So the main diagnostic algorithm for pulmonary hypertension usually follows a stepwise approach. It starts with suspicion by physicians due to their patients' symptoms.

Symptoms and physical exam findings should be followed up with an echocardiogram. Echocardiographic signs of pulmonary hypertension might include a dilated right atrium, right ventricle, or inferior vena cava. You could see hypertrophied right ventricle, a flattened intraventricular septum leading to what we call a D-shaped left ventricle, a decreased tricuspid annular plane systolic excursion, suggesting that RV function is diminished. You could see increased systolic peak tricuspid regurgitation velocity by continuous wave Doppler, which would translate into an elevated estimated systolic pulmonary artery pressure.

But finally, the final step in confirmation is a right heart catheterization. Pulmonary hypertension would be defined by a mean pulmonary artery pressure greater than 20 millimeters of mercury at rest. The definition of pulmonary arterial hypertension also implies a pulmonary vascular resistance of greater than two Wood units and a pulmonary artery occlusion pressure of less than or equal to 15 millimeters of mercury. This is precapillary pulmonary hypertension.

And while pulmonary arterial hypertension is pulmonary hypertension, not all pulmonary hypertension is pulmonary arterial hypertension. So for instance, one might want to do lung imaging, both noncontrast computed tomography and contrast-enhanced computed tomography pulmonary angiography, to rule out, respectively, World Health Organization Group 3, which is pulmonary hypertension associated with lung diseases or hypoxia, and Group 4, which is pulmonary hypertension associated with chronic thromboembolism. And then with Group 2 we've kind of already taken care of at the right heart catheterization. That would be due to left heart disease. But we found that this is purely precapillary. If the pulmonary artery occlusion pressure is less than 15 millimeters of mercury with heart disease, you'd expect it to be greater than 15 millimeters of mercury, which would be a post capillary pulmonary hypertension, and there would be other signs on echocardiography.

Other things you could look at are chest X-rays and an EKG, but normal chest X-rays and a normal EKG do not rule out the diagnosis.

Dr. Sorrentino:
So, Dr. Agbor-Enoh, as you just heard from Dr. Solomon, it could be a pretty elaborate way of diagnosing somebody with PAH. So how does this new blood test help to evaluate patients with PAH? And I guess the real question is, can we not do all this testing if your blood test can give us the answer?

Dr. Agbor-Enoh:
So that's an interesting question. The diagnosis of PAH involves multiple steps and multiple different diagnostic algorithms, some of which are quite invasive, like right heart catheterization, which are quite invasive procedures. So what we're suggesting is a blood test, but not for the diagnosis of pulmonary arterial hypertension however, but more for an evaluation of these patients.

Now, the goal of such a blood test would be to ask a few questions. Question number one−how severe is this patient's pulmonary hypertension? Because treatment and management plans are directed based on a patient's disease severity. Question number two—in addition to the pulmonary artery, what other tissues or organs are affected in these patients with this disease? And then question number three—what is some form of a prediction of poor outcome in these patients?

For example, in this study we look at something we call transplant-free survival, which is a chance that this patient would be alive and free from transplantation. So we looked at whether cell-free DNA could identify that. And something else, which we did not evaluate in this study that's also quite important to patients, is how well they respond to treatment. So those are some of the ways that some of these tests can be used in evaluating patients with pulmonary hypertension.

Dr. Sorrentino:
So this really sounds exciting that we may have a tool that can answer some of those questions. Dr. Solomon, the question I have about this test then is, how did it work out in clinical studies? Does this test correlate with symptom severity? Does it give us some idea of prognosis from the cohort of patients you've looked at?

Dr. Solomon:
Sure. So let me just preface that first by saying, the interesting thing about plasma cell-free DNA is that it's being constantly shed and constantly cleared, so it really does give you a picture of what the patient is at that time that you're doing the test. And so we looked at plasma cell-free DNA, and we looked at it in two pulmonary hypertension cohorts. One was from Allegheny General Hospital, and the other was from Tufts Medical Center, and then we had our own group of controls. We also collected data to calculate what is known as a REVEAL score. REVEAL is the registry to evaluate early and long-term pulmonary arterial hypertension disease management, and that score consists of 14 variables, some of which are invasive. Patients were then divided into REVEAL groups by risk. Based on their score, they were either low, medium, or high. And then total cell-free DNA concentrations were compared among the control, as well as, these various pulmonary arterial hypertension risk groups. We also took a sample subset from one of our cohorts, and we went ahead and performed bisulfite sequencing followed by a deconvolution algorithm, which is essentially a methylation analysis. And what that allowed us to do was look at cell-specific and cell-free DNA patterns to have a good idea of where that cell-free DNA was coming from.

And so when we did all this, I think we found a couple of interesting things. One was that circulating cell-free DNA is elevated in patients with pulmonary arterial hypertension compared to healthy controls, and in two independent pulmonary arterial hypertension cohorts, we found that cell-free DNA concentrations increased with severity of disease and predicted transplant-free survival in the

larger of our cohorts. We didn't have enough outcome data in the smaller cohort to do it in the smaller cohort. Third, the methylation patterns that we determined for the cell-free DNA's origin were biologically plausible sites for PAH, pulmonary arterial hypertension, and that included erythrocyte progenitor cells, myeloid lineage inflammatory cells, vascular endothelium, and cardiac myocytes.

So what we had essentially, was circulating cell-free DNA that was elevated in pulmonary arterial hypertension patients, it correlated with disease severity, and it predicted worse survival. And the results of our methylation analysis on cell-free DNA suggested that it was essentially a biologically plausible marker because it was consistent with prevailing paradigms of the disease.

Dr. Sorrentino:
For those just joining us, you're listening to *Heart Matters* on ReachMD. I'm Dr. Matthew Sorrentino, and I'm speaking with Drs. Michael Solomon and Sean Agbor-Enoh about a novel blood test circulating cell-free DNA that evaluations patients with pulmonary arterial hypertension.

Turning back to you, Dr. Agbor-Enoh, this test really sounds exciting. What are the next steps? How do we get it available and know that it's really going to help our patients?

Dr. Agbor-Enoh:
I think as next steps, I would recommend three steps. Step one is validation. This is shown by our group. It is true that we did not show these findings in one cohort from a different hospital, Allegheny. We also validated that in a different cohort of patients, collected in a very different center, and by different investigators in Tufts. That's true that we did that. It would be great if another group would do the analysis, pull the cohort together, and validate this. So that's step one. I think some degree of validation would be relevant.

Step two, in my mind, is the assay itself. In the assay itself, we extract cell-free DNA, and we use something that we call, a whole genome sequencing approach, which means we sequence all the DNA in order to generate this information with the sequence, one sample at somewhere around 170 to 250 million rates of cell-free DNA. That's pretty deep, and deep equals expensive, so we need to tailor the test to become a test that is quick, that is cheaper, and that can be used. And there are multiple approaches to do that using approaches like PCR, digital droplet PCR, and some other methods which one can use, so there are already technologies that can speed that up.

Step three is going to be applying the test itself to patients, and then seeing how this test performs in a prospective cohort of patients for which the test is being used.

Dr. Sorrentino:
Dr. Solomon, the future of this test, do you think it will actually supplant or replace some of the testing we do now? Do you think its power, from what you've seen so far, will give us a better idea of prognosis and survivability that maybe we can avoid right heart caths and some of the other things we have to do now?

Dr. Solomon:
Sure, that's a great question. Let me take a step back and just say right now we have 14 FDA-approved therapies for pulmonary arterial hypertension, and yet survival remains poor, so it's about 57 to 60 percent maybe at five years. And the key here, is in part I think, that this poor survival is related to the delay between symptom onset and diagnosis.

In one study the average was about 31 months between symptom onset and diagnosis. That's a long time. And one would argue we might be getting our therapy started late, and to start these therapies earlier could be another way of making a difference. And to do that we need to understand the pulmonary hypertension pathogenesis better, and we need improved testing and improved diagnostic testing, and I think that's why this test is so powerful because it does two things. One, it's a single blood test, and it's shown again, to be elevated in pulmonary arterial hypertension and to the associated disease severity. But it also is giving us clues into which tissues are involved, which helps us understand the pathophysiology because we can look at where the cell-free DNA such as the elevated portion of the cell-free DNA are coming from. I think there's other work to be done, some more research, and some more studies, but it's got great potential.

Dr. Sorrentino:
This certainly sounds exciting and like a promising advancement, and I can't wait to see if we can use a test like that in these very difficult to treat and very sick patients. I want to thank my guests, Drs. Michael Solomon and Sean Agbor-Enoh for sharing their insights on this. It was wonderful speaking with you today. Thanks for joining us.

Dr. Agbor-Enoh:
Thank you very much.

Dr. Solomon:
Thank you.

Dr. Sorrentino:
For ReachMD, I'm Dr. Matthew Sorrentino. To access this and other episodes in our series, visit ReachMD.com/HeartMatters where you can Be Part of the Knowledge. Thanks for listening.