Dr. Yogen Kanthi
Behind the Mask
July 6, 2021

Barr: Good morning. Today is July 6, 2021. My name is Gabrielle Barr, and I am the archivist with the Office of NIH History and Stetten Museum, and today I have the pleasure of speaking with Dr. Yogen Kanthi. Dr. Kanthi is a Lasker Clinical Research Scholar. He is also the Chief of the Vascular Thrombosis and Inflammation Laboratory at the National Heart, Lung, and Blood Institute, and an Assistant Professor at the University of Michigan. Today he is going to speak about some of his COVID research and experiences. Thank you for being with me.

Kanthi: It is great to be here. Thank you, Gabrielle.

Barr: Absolutely. You have done a lot of research, and COVID has not just impacted the lungs but also affected patients in all sorts of ways, and we are going to speak about that during your interview. What percent of those diagnosed with COVID have had subsequent cardiovascular problems that have ensued?

Kanthi: That is a really great question actually, and it is harder to answer than we might expect, but first of all, thank you for the opportunity to participate in this interview. We are in a really spectacular environment at the NIH, where there is a complete mission alignment and hundreds of thousands of people who have been working really hard trying to find both a better understanding of the biology of this disease, and also new and novel treatments for COVID and other diseases. I am grateful to be a part of this spectacular scientific community.

With COVID itself, the initial thought was that this is a lung disease, that it is a disease that affects the alveoli, or the air spaces, itself in the lungs, causes inflammation, and reduces oxygenation. We have learned since then that there is a large vascular component to this disease, and that inflammation in the blood vessels also leads to clotting in those blood vessels, and certainly that then leads to more inflammation. So, there is a big cycle over here. If you look at the numbers themselves, of all the people who get COVID, only a very small fraction of them will actually end up in the hospital with oxygenation problems, or with blood vessel problems, or with problems in their gut, for example, but of the people who are hospitalized, a large number of those patients actually end up with vascular problems. We know that of the people who are hospitalized, about 15 to 25 percent, at different points in time, have ended up needing to get transferred to the Intensive Care Unit. Those patients, certainly, do have vascular problems, but even the other 75 to 85 percent of patients who are not moved to the ICU, but require hospitalization, could have vascular problems. If you look at statistics, reports from across the world suggest that anywhere from around 10 to 60 percent of patients will have a diagnosed blood clot in one of the large blood vessels, and this does not count most of the small blood vessels, so we think that the number of people overall who probably have clots, including the small blood vessels, is much

higher than that. Therefore, a sizable number of patients with this disease end up with blood clot problems.

Barr: When COVID affects some of the other organs like the liver, or kidneys, or another organ, is that due to vascular issues or are there other issues caused by COVID?

Kanthi: This is a tough question to answer but I think a really good and important one. This virus and the consequences of the viral infection cause so many different types of problems that oftentimes there is an overlap of different pathological processes that result in organ dysfunction. One of the large contributors to that is vascular disease so, if there is a liver problem, the liver cells called hepatocytes may be inflamed but, in addition to that, you could have blood clots, for example, in the portal veins that feed into the liver, and then cause other problems to the liver cells themselves. We think it is a combination, and it is going to be hard to separate these processes and say it is only just one part of the process that is causing a problem, when in fact, it is many different areas that come together and result in this culmination that causes organ failure.

Barr: When was the connection made between COVID and cardiovascular issues, and were you surprised given your review of past coronavirus pandemics?

Kanthi: This is an interesting reflection because now, here we are on July 6, 2021, and we first started thinking about this in December 2019. Around February 2020, I was discussing this with a colleague of mine, Jason Knight, who is a rheumatologist at the University of Michigan. We have worked closely together for a number of years. We started to talk about the reports of inflammation that we were seeing. We looked at these preprint reports from Wuhan of the clinical characteristics of patients infected with COVID, coronavirus, or SARS-CoV-2, and saw that there was a lot of inflammation. There was this idea of a cytokine storm per se, and it made us think a little bit about the other possible consequences. There were not any reports early on that there was a blood clotting problem associated with this disease, but if I look back, about 10 years prior to this, when we had the H1N1 epidemic, I was a cardiovascular medicine fellow at the University of Michigan, and I remember being on rounds and in the Surgical Intensive Care Unit, when I started talking to my colleagues there, Dr. Andrea Obi, Dr. Tom Wakefield, Pauline Park. We started thinking a bit about what was going on with the patients they were seeing. Oftentimes, they were seeing people admitted with H1N1, and these patients were developing a large amount of clots in their lungs.

So, they actually started taking those patients, and prophylactically, if they were admitted to the ICU with H1N1 without any diagnosis of a clot, they were putting patients on blood thinners. They found that there were fewer blood clots, and perhaps it translated to an improvement in respiratory function. That laid the context for how to think about COVID 19. I was surprised when we did not, initially, see reports of blood clots, and there are probably reasons for that related to the lower risk of thrombosis and people who are of East Asian descent. Perhaps that was the reason.

Barr: How has COVID differed from your reviews of H1N1 [influenza] and SARS-CoV-1 in terms of cardiovascular issues?

Kanthi: H1N1 and other viral illnesses are certainly new strains and new variants of a virus, but this really turned out to be a completely new virus. We have known that there is no host defense for this; there is no previous exposure, and we ended up rallying all of the tools in our armamentarium to try and help our bodies survive. As a result of that, it is a moment in time, when we rally every single defense and the result of that is potentially catastrophic, but the other side of this is that, in some people, the defense is potentially life-saving so, it is this careful line that we are trying to walk, and all too often, in COVID-19, we end up flipping over to the side where we have disastrous consequences with a lot of organ failure.

Barr: Why do so many COVID patients have neutrophil extracellular traps, NETs? You may want to explain what those are, and what are ways that these NETs cause cardiac and vascular issues like thrombosis?

Kanthi: So, neutrophils are the most abundant circulating immune cell of white blood cell, and we make about 100 billion of these a day. These are cells that are already fully armed in circulation and their job is to be the first responders to any sort of insult, whether it is an infection or an injury of some kind. What we end up seeing here is that there is an evolutionarily conserved mechanism by which neutrophils, when they encounter pathogens like bacteria, for example, will start to detect it. They then start to unravel their DNA. Then they squeeze it out of their side, they spit it out in the form of a sticky web or sticky trap. This web then starts to ensnare things like bacteria and coat it with antibacterial proteins that end up killing the bacteria. This is a great way to try and coral off any infective organisms and prevent the dissemination of them. This is really important so, for instance, if we are walking in the park and we fall and we cut ourselves, and bacteria enters over there you need to be able to stop the bacteria from spreading around, otherwise you would get sepsis. These neutrophil extracellular traps have been found in what we call NETs. They were first described in 2004 or so and have been found since then in a whole host of organisms, and it seems to span the spectrum from very small to very large organisms. We think that this is a mechanism by which mammalian physiology, and actually even other non-mammalian organisms, have been able to try and develop defense mechanisms against pathogens.

This is great if you are walking and you fall and hurt yourself, and it was great in the era of the caveman, but these days, we are not encountering saber-toothed tigers, and we are not scrambling around. So, if this happens in the absence of that trigger, or it happens in an unregulated fashion in the blood vessels, it can be detrimental, and it can start to trigger blood clotting because it forms a scaffold on which other cells can attach, and coagulation starts to occur, and then it can eventually form a clot that can occlude the blood vessel.

There are a number of things that neutrophil extracellular traps do. One of the pathologic consequences is blood clotting or aberrant blood clotting. You need to have a normal, physiologic, "good" blood clot but the "bad" blood clotting is what we worry about. Clinicians have been observing this in a number of other disease states, and in the context of COVID-19, we thought a little bit about whether neutrophils might be involved here. As these first responders, we looked, initially, at the descriptions in those preprints from Wuhan about what the blood counts look like, and if you look carefully there is a profound drop in the lymphocyte count. Normal lymphocytes may be up here and then, in circulation under COVID, the lymphocyte number drops considerably. That is pretty characteristic of some viral infections, but if you look at the total number of white blood cells, that did not change very much, in fact, it went up a little bit. So something is making up that gap. Neutrophils were a leading candidate for that so that is one of the reasons that we started to focus on this mechanism, checking whether neutrophils and neutrophil extracellular traps may be involved in COVID-19, and if they could be contributing to the pathogenesis and some of the complications that we see with this disease.

Barr: Why were neutrophils explored later than other types of immune responses?

Kanthi: This is a good question. To those of us that are more neutrophil centric in the world, it seemed pretty straightforward and clear that we really should investigate it but not everybody feels that way about it depending on their area of expertise and experience. However, lymphocytes seem to be involved because the numbers drop really precipitously and so, that was a very obvious place to look. In addition to that, the T cells, the cytotoxic T cells, are really an important contributor to the inflammatory response, and the immune response to viral infections, and so that was an obvious target to look at. Macrophages do similar things. They gobble a lot of debris and can help clear out bad things in the system. They generate a lot of inflammatory chemicals called cytokines and that seems to be an obvious target [for research] as well. Early on, there was an interest in wanting to see whether we could figure out why there is so much inflammation, and the focus was on lymphocytes and macrophages or monocytes, and we happened to be in the right place, thinking about this at the right time, to really reflect on whether or not neutrophils were drivers of this pathogenesis.

Barr: Will you speak about how you and your team went about studying neutrophil extracellular traps and thrombosis and COVID-19, and what your role was in the study?

Kanthi: This was around the time that I was starting to make the transition from my faculty position at the University of Michigan to the NIH. I maintain that faculty appointment there still, so that I could continue some collaborative work. At this time, we recruited, myself and my colleague Jason Knight, a smart junior faculty rheumatologist named Ray Zuo to try and help us answer some of these questions. Ray, heroically, ran around the hospital trying to collect blood samples that were discarded. The hospital itself was shut down to collecting research samples proactively, or prospectively, and so what we had to do was look for samples that were otherwise going to be thrown away. In fact, when someone got blood drawn, if their chemistries were being checked, for example, and there was some leftover serum

from that, we asked if they would be willing to give that to us, to be able to try and study those samples. This way, we were able to collect samples.

We measured in that circulation that there were very high levels of these neutrophil extracellular traps. These markers in patients who had COVID-19 seemed to go up the sicker they were. So, if you were on a mechanical ventilator, your levels were sky high, and if you were just on a little bit of oxygen and you were on room air, your levels were much lower. We then, also took some of the serum from patients with COVID-19 and we tried to see whether there was something in circulation that could actually trigger NET formation. We took serum from patients with COVID-19, we incubated it with healthy neutrophils, and found that something was circulating in the bloodstream of patients with COVID-19 that could make a healthy neutrophil start to explode and form these NETs. There really did seem to be something—that is, something beyond the virus and the disease itself –some factor in blood that might be triggering this.

We asked ourselves whether NETs could be a good biomarker to figure out which patients might be sick, sicker, or might be more likely to become sick. We measured it, but it was not as clear, and we could not really predict, based on an early blood draw, whether people who had high levels of NETs, or low levels of NETs were more or less likely to end up on respirator with respiratory failure. So, we looked at a different type of biomarker, a protein called calprotectin, or MRP14, that is in circulation, but its detection comes primarily from immune cells and it is really in high levels in neutrophils so we looked to see if you could measure levels of calprotectin in the blood, and whether that might be able to help guide us in deciding which patients were the sickest of the sick.

It was a fascinating result. I will never forget this. We found that if you measure calprotectin on the first or second day of a patient being hospitalized with COVID-19, it could actually predict better than any of the other biomarkers out there whether that patient would end up in respiratory failure on a mechanical ventilator. We thought that this would be a great tool to be able to help us decide which patients were the sickest, and perhaps then as we were in the point of triage at the time, figure out which patients might benefit the most by directing precious resources to try and prevent them from getting sick enough that they end up on a mechanical ventilator, because the mortality rate then was 25 to 30 percent if you were on a ventilator.

That was an interesting approach, but a blood calprotectin level was not an FDA approved test at the time. Since then, there are now two groups, one in the UK, one in the US, that are trying to move a blood calprotectin measurement assay into the clinical sphere, to see whether it is a tool that might help clinicians who are on the front lines figure out whether or not a patient is at high risk for respiratory failure. We are excited and hopeful that it will move forward. There is another group in France that had the same finding around the same time as us so we are glad to see that our work has been validated.

Barr: That is exciting. I hope that it comes through fast, and for the patients as well. Can you, with a few drugs to combat SARS-CoV-2, have therapeutics that tackle NETs, like anticoagulants that been used for COVID patients? At what point are these therapeutics distributed to these patients?

Kanthi: The NIH and the NHLBI, in particular, have directed a lot of resources and effort towards answering these questions. There have been a number of different trials that have been done, and I will start with the one that has been the most important one so far. It is part of the ACTIV-4A trial [Accelerating COVID-19 Therapeutic Interventions and Vaccines]. This is a trial that the NHLBI sponsored and there are several different PIs [Principal Investigators] on it including Dr. Judith Hochman, Dr. Macky Neal, Dr. Jeffrey Berger. This trial is not published yet, but the results have been released in a number of different ways. What the investigators did was try to find out whether anticoagulation with heparin (this is a very common anticoagulant or blood thinner that we use in clinical practice), whether anticoagulation patients with heparin, once they are admitted to the hospital would benefit from them and reduce some of the organ failure that goes along with it.

What they found was that if people were in the hospital, and they would be called moderately ill, which means that they were in the hospital but not requiring ICU care, and they received a therapeutical level of heparin anticoagulant (this is a therapeutic dose [because] it comes in three different doses, the prophylaxis dose, the intermediate, and the high-level therapeutic as though you were trying to treat a blood clot that has already occurred), they had an improvement and a reduction in the amount of days requiring organ support, and also, obviously, survival. We are waiting to see the full data because that is quite exciting. It would be one of the few anticoagulant therapeutics that seems to work. This did not hold true for the patients who are the sickest, those who are in the ICU. We still need to have some sort of intervention for them.

Another trial that I am involved in, the ACTIV-4B trial, which is also sponsored by the NHLBI and run by Dr. Jean Connors, is to test whether or not oral anticoagulants given to patients when they are diagnosed with COVID, but not hospitalized, would make a difference. That trial was just recently closed because there was an unclear benefit, or it was unclear whether there would be a benefit at all in that trial compared with placebo. So that is all work that is still pending and has not been published in terms of the final trial results, we look forward to seeing that.

Other trials have been ongoing, with drugs like aspirin, as well and other oral anticoagulants, and we will see what some of these results show, but it seems to me that not all COVID is the same, that the timing of intervention may be extremely important, but figuring out that precise moment in which to start a particular therapy is going to be important because the disease tends to change very quickly, and quite dramatically, between each of these stages.

Barr: Do you think it is too late for some of these patients by the time they receive these treatments in the hospital? I mean, most people have had COVID for a while by the time they are admitted.

Kanthi: You are asking a really good and insightful question. We know from some of the other studies with antiviral drugs, for example, on COVID, that there is no silver bullet for this. The disease is diffuse enough in terms of the organs that it affects and what it does, what it triggers in the immune response, that we are going to need a multi-pronged approach. There is not going to be one drug that is a panacea

for this. We will need several different drugs, possibly a cocktail of them. In this case, what we can say is that anticoagulation does not seem to benefit patients once they are really sick, and it does not seem to benefit patients if they are not very sick at all, so that in-between spot does seem to be the part that is just hot enough for the drug to actually be able to have an effect.

Barr: You spoke about anticoagulants but what are some other types of therapeutics that are out there to treat patients with vascular issues stemming from COVID? Can you speak about those existing drugs that are being used, as well as, if you know, of any trials of new therapeutics being developed?

Kanthi: This is something that reflects a big needs gap which the NIH, amongst others, is trying to fill. We know that inflammation and coagulation, which is blood clotting, are very closely related to one another. I will go back to my analogy about falling as you walk in the forest. If you fall and you injure yourself and you start bleeding from your skin you have to do three things to survive: one is you have got to stop the bleeding, so you need to have a clot that forms there; you also have to be able to fight off any invading bacteria, so you need an immune response and potentially clotting to try and corral or quarantine those pathogens; and the third thing is you need to have healing of the wound and so and you need it to be sterile.

In all three of those processes, inflammation and coagulation work closely together. If you trigger a whole lot of inflammation, you are likely to get some clotting complications as well, and treatment also needs to be done under the same idea. If you are treating only the coagulation part of it, you are missing a treatment for the inflammation side of it so, we probably need to tackle those two things together. The way to really approach this is to use either multiple different drugs that are very precisely effective, different pathways, or to use, potentially, drugs that have effects on both sides of this process, both the inflammation and the coagulation side. There are many examples of drugs out there that can do that.

One that we focused on was a drug called dipyridamole. This is an old drug that has been around in some form or another since the 1950s or ‘60s. It has been used as an anti-platelet drug**,** and it prevents platelets from becoming sticky and forming clots. It was approved by the FDA back in the ‘80s to try and prevent stroke in patients who have either mechanical heart valves or who have had a stroke before. It is used in combination with aspirin, and it is dirt cheap, we are talking about a dollar a day for the treatment itself. It has been used in over 20,000 patients in clinical trials, it is given by mouth, and has a very good safety profile. Remarkably, as we figured out a couple of years ago, this drug increases intracellular cyclic AMP, and in the process of doing that, it shuts down neutrophil extracellular trap formation. We thought that it would be potentially accessible to patients, and it would be something that could be moved to the clinic very quickly, so we have been interested in doing this in a different disease called antiphospholipid syndrome. We decided to look at this drug itself and see whether we could use it in patients with COVID-19. We did some studies in tissue culture, in which we took serum from patients with COVID, we incubated it with healthy human neutrophils which had been either treated with a placebo or treated with dipyridamole in advance. We found that the cells that were treated with dipyridamole had a 50% more protection against NET formation than the cells that received the placebo. Therefore, we thought that dipyridamole was possibly a drug that might work well in

patients and be accessible not just to people in the U.S., but also, given its low cost, patients in other countries such as India, where my family is from. If you convert its price, it costs one cent a day. We thought that this drug would be quite accessible and scalable to a global audience, and we did end up starting a clinical trial and I will be happy to speak about that.

Barr: Sure, we can speak about your clinical trial but before that, we were also going to speak about your study that looked at thrombosis autoantibodies and serum from patients hospitalized with COVID, and explain some of the short-term and long-term implications of that study.

Kanthi: As I mentioned earlier, there is this disease called antiphospholipid syndrome. It is an interesting disease, an autoimmune condition, that is not understood very well but we know that patients develop auto antibodies, and these antibodies target their own phospholipids, or phospholipid binding proteins. In the past, we had taken some of these IgG fractions, which are antibodies from patients with antiphospholipid syndrome, and injected those treating neutrophils, which tended to cause neutrophil extracellular traps. In animal models, these antibodies can cause clots, and sure enough that marries what we see in the clinical manifestations of antiphospholipid syndrome. Although this autoimmune disease has some overlap with lupus, it is a distinct entity on its own, and patients with antiphospholipid syndrome, develop a lot of different complications that are related to the vascular system; they develop blood clots both in arteries and in veins, and they also develop clots in small blood vessels and in big blood vessels. This is quite unusual since most clotting disorders do not manifest like this. Patients also have things like heart attacks, fibrosis, cognitive dysfunction, and kidney problems. A number of these malfunctions may be related to vascular disease that is underlying it. One of the reasons I became a vascular medicine specialist is because anywhere there is a blood vessel is a fair game organ for me to study, and it is important to me because the vascular system has more real estate than a football field. It is an impressive system in itself, and perturbations in it have significant consequences for patients.

We started looking at autopsy specimens from a colleague and friend of mine, Dr. Sharon Fox from Louisiana. She did an autopsy series in Black patients in New Orleans, and identified clots in both the arteries and in veins in patients with COVID, and also in the small blood vessels and the big blood vessels. In many ways, it was reminiscent of antiphospholipid syndrome, so we decided to measure antiphospholipid antibodies in patients with COVID-19 to see if they might be overexpressed or present here. What we identified was quite surprising in our cohort at the time of 172 patients. There had been a few smaller case studies of five or ten patients that have been described, but in our cohort of 172 hospitalized patients with COVID-19, we found that about 50% of them had an elevation of at least one of these antiphospholipid antibodies, and 30% of them had a really high elevation of these antibodies, much more than just the abnormal number. If we look at the normal population, we think that only about five percent or so of people will have an elevated antibody, so 50%, and the order of magnitude was quite alarming and concerning.

We then took these antibodies, we took whole IgG fractions from patients with COVID-19, and we went in and isolated the antibodies from those patients and found that those antibodies themselves can trigger neutrophils to explode and form NETs. In animal models, these antibodies themselves can start

to also form clots. They can trigger venous thrombosis, in particular, and so in many ways, it was suggested to us that there was something, there was a circulating antibody of some kind, or multiple different kinds, that seemed to be triggering clotting in patients with COVID-19. We think that the antiphospholipid antibodies are just one of probably a repertoire, a generalized response of the body, to produce a lot of antibodies, many of which will be self-directed.

Our colleagues in NIAID, Drs. Steve Holland, Luigi Notarangelo, Mihalis Lionakis, and Jean-Laurent Casanova at Rockefeller, together had found, just a few weeks before our description, antibodies that blocked type 1 interferons. Type 1 interferons are important because this is your physiologic antiviral response in the body, and in the context of COVID, if you suppress the type 1 interferon response you now have suppressed the body's immune response and its ability to fight SARS-CoV-2. We know that there were autoantibodies and our description of this was the first sign that these autoantibodies might, actually, be driving a clotting diathesis in patients with this disease, and perhaps now it makes us think about different ways to approach and target the disease itself.

When we took the healthy neutrophils and treated them with dipyridamole, the drug that I mentioned to you before, and then tried to add these autoantibodies in, the cells were 50% more protected against NET formation, compared with the cells that received a placebo and were treated with the COVID immunoglobulins. Dipyridamole was becoming more and more intriguing for us. Since this discovery of autoantibodies, several other groups have validated our results. They also found that there are pro-thrombotic antibodies, or that there are other autoantibodies that may be driving thrombosis, and different cell types we more recently found, and that these autoantibodies coming from patients who have COVID-19 and have high levels of antiphospholipid antibodies also trigger the blood vessels to themselves, to directly, become inflamed, and so the endothelium itself becomes stickier, it recruits and attaches white blood cells much more, which we know can be a trigger of thrombosis and other vascular inflammation.

There is still a lot to be learned. We are just scratching the surface, and we think it is probably going to have long-term implications for patients with COVID. It also gives us better ideas by which we might be able to approach other critical illnesses, whether it is trauma, or sepsis, or heart attacks, things of this sort.

Barr: Do the people that have a high level of these autoantibodies, have any genetic preconditions for that? I am asking because the NIAID scientists who were studying interferon and discovered that there is a genetic component to those people not being able to combat COVID-19.

Kanthi: It seems that the genetic component that you are referring to were people who have low levels of type 1 interferons to begin with. I do not know if the genetic component itself has described whether or not there were autoantibodies associated with COVID-19, but it is an important question and one that we have been thinking about. We know that these autoimmune diseases outside of the context of COVID-19 can have genetic risks. There is some clustering of families, certain types of HLA, or human leukocyte antigens, for example, that can be seen in patients with different autoimmune diseases, so

there is discussion about this. We are hoping to be able to collect enough data points to be able to better understand it. There are a number of groups who are investigating this area, and I think it is going to be very interesting and exciting. What we are trying to do is see whether we can look at the RNA, not the DNA, to try and better understand what the pathways might be that drive this disease. Understanding the cause of the disease in terms of genetics is important, but once the disease actually starts, we need to be able to understand how to shut it down, or how to treat it, and the transcriptomics, the RNA sequencing, is going to be important because it is a real-time description of what the cell is trying to do, and hopefully, that will open up some pathways that we can then investigate and start to find new targets for.

Barr: Can you speak about the clinical trial that you helped launch when you were at the University of Michigan (you are still at the University of Michigan, except just remotely) to test the drug dipyridamole, a repurposed FDA approved drug in patients with COVID-19?

Kanthi: Yes, I would be delighted to do so. First, I think it is important to thank the NIH and the NHLBI for giving me the space to be able to do these things. They gave me unconventional freedom to be able to try and move some of these initiatives forward. And also, I think it is important to thank the University of Michigan Taubman Medical Research Institute which supported our clinical trial, and also the Frankel Cardiovascular Center there who supported the early work that we were doing there. It is important to acknowledge the people who enabled us to investigate, when Jason and I were thinking about drugs and how we might be able to move this discovery of neutrophil extracellular traps and methods to target it in the disease.

We focused on dipyridamole because we had been studying it before, and we knew it was cheap and accessible. There were other drugs that we had been thinking about. My team had discovered a couple of years ago that interleukin one beta (IL-1β), one of the fundamental pyrogens, or inflammatory cytokines, is intimately involved in clotting and triggering clotting, and that we could reduce clotting in an animal model by using Anakinra, which is a drug that is available for patients with autoinflammatory syndromes and certain types of other diseases. We thought about using that as a drug but we realized that it would be $60,000 dollars a year. That is the cost for each drug and per patient if you were giving it for a full year, and it would not be accessible to people across the world, but rather having something that was known to be safe, oral, easily available, accessible, and inexpensive, would be useful if it worked.

We started to think more about dipyridamole, and we wrote a clinical protocol for this very quickly. We are basic and translational scientists, and although we are clinicians, we focus a lot on animal studies. I had not done clinical trial work before, so I searched and searched for people, called friends to try and get advice from people with more experience in this realm, and thanks to them, we were able to launch our first clinical trial. We had good support but there were a lot of 18, 19-hour days. We came up with the idea in late March and by mid-May we had FDA approval to proceed with the trial. So, in a matter of six to seven weeks, we moved this forward from conception to launch.

We designed this trial in hospitalized patients, since we thought that it is where the biggest need was at the time, and also because we did not know how to run a clinical trial in the outpatient setting. That would be a lot harder than doing it in the inpatient setting, so we designed the trial for hospitalized patients. Initially, it was set up as an 80-patient trial, but the DSMB (Data and Safety Monitoring Board) suggested that we increase the sample size. It was a randomized placebo-controlled trial that was single blinded. We—Jason, myself and Ray Zuo—we enrolled patients on the phones every day, talking to patients and talking to their families, going in to get consent from families and from patients, to be able to enroll them. I am so grateful, not only to our team, but importantly, to our patients who in the throes of the scariest time and illness of their life were willing to participate in clinical trials. It really speaks to their strength and their generosity, for wanting to do something that might be able to help others.

We ended up enrolling 100 patients. The trial is closed now, and the data is being analyzed. Right around this time, steroids were also introduced, as studies came out from the UK suggesting that dexamethasone might be helpful in certain categories of hospitalized patients. In particular, the patients that we intended to enroll were now eligible for steroids, but we had not anticipated that when we wrote the trial. We think that steroids and dipyridamole probably act in a similar direction to suppress inflammation. Our expectation was that perhaps dexamethasone might actually overpower any effect of dipyridamole, and this is still something we are waiting to see in the final results. Our statisticians are looking at the data.

We have built into this a lot of mechanistic studies as well, to try and understand whether biomarkers or biological processes like cell signaling, might be modulated differently by treatment with dipyridamole, and to understand longitudinally what happens to patients when they are in the hospital either receiving placebo or this drug. We think it will give us a good idea of what the natural history of this disease might look like, so we built up a very large biorepository which we are starting to study together with other collaborators. I think this is going to be helpful for not only this disease but for the understanding of other diseases as well. Our discoveries of neutrophil extracellular traps, coagulation markers, and also the autoantibodies have led directly to the design of our trial. We also believe that up to nine other clinical trials have built their foundation on our mechanistic studies amongst others, so these are useful contributions. The teams, both at the NIH and at the University of Michigan have worked around the clock every day from mid-March 2020 to mid-March 2021. They may have taken two to three days off in total. Just an incredible dedication by these teams to really try and band together and understand this disease and try to approach it. The teams should be proud of these scientific contributions for the rest of their careers. It is an incredible mark on the pandemic, and in our approach in response to the pandemic.

Barr: Definitely. Have you been part of other COVID research and initiatives? You mentioned some of the things that you have done.

Kanthi: As a result of this, there have been a lot of other discussions about other drug targets. I have been involved in some agent prioritization committees that are hosted by NIH and NHLBI, things like, CONNECTS for example, to try and understand, look at different drugs and drug targets that may be

proposed, and then, the agents that are submitted for consideration, to try and vet those and understand which ones might have the highest likelihood of success, success not just being what might be the best drug but what are the logistics around getting this drug into people and what is the safety of the drugs for people. Also, can we track the results and outcomes of it in a very effective and efficient manner through these multi-platform trials, for example? I have been involved in a number of initiatives like that.

My team here at the NIH is in the Translational Vascular Medicine Branch, in which Manfred Boehm is our Chief. He and I have been talking for some time about wanting to explore whether or not there may be targets that otherwise affect blood vessels, things like the renin-angiotensin-aldosterone system, in particular. We have been looking at Angiotensin-1, Angiotensin 1-7, and Angiotensin-2. The ACE-2 receptor is important in COVID pathogenesis, but it may also be important in the vascular inflammatory complications.

Doug Rosing in the Clinical Cardiology Branch, Manfred Boehm, the Head of the Cardiovascular Regeneration Branch, who helped recruit me here to the NIH, and myself, talked for a long time about some of these agents, and we were able to advance a few of these into consideration at the CONNECTS agent prioritization committee, and now the ACTIV 4d RAAS multi-platform trial is being launched. The trial includes a couple of agents that we proposed and helped shepherd through, and the committee themselves had looked at them very carefully and then received approval through a number of different layers.

There is another very important trial that has been ongoing and just completed here at the NIH Clinical Center and at INOVA Fairfax in Virginia with a drug called Fostamatinib. This is a spleen tyrosine kinase inhibitor used to treat multiple myeloma, typically, in the in the pre-COVID era, and when our data on NETs came out, Admiral Rick Childs, who is the Clinical Director for NHLBI, talked with me about Fostamatinib for a while. He had started to advance the drug here, to see whether it might be a drug that works and that could be a candidate to stick into a clinical trial for COVID-19. This trial, which has since launched and been completed, was led by Dr. Jeff Strich, who is in the Critical Care Medicine Department, here at the Clinical Center. In this trial, patients were randomized to receive either fostamatinib or placebo in a placebo-controlled trial. The participants were hospitalized either at the Clinical Center, NIH or at INOVA Fairfax. The results of the trial have been released, a little bit, in a press release from the company itself, but we should wait until the clinical trial results have been formally published before discussing them, and I think that Dr. Strich and Dr. Childs would probably be able to tell you a lot more about it. We are helping with many different aspects of the trial from the biorepository and mechanistic studies aspect. We are trying to understand some of the clotting processes, some of the vascular inflammatory processes, understanding how antibodies might be driving some of this disease states, and other thoughts that Fostamatinib might be able to affect it. This is exciting to me because I think that this drug has strong potential, it is well tolerated, again, it is expensive, but if we are looking at a life-or-death situation, then perhaps, cost is less of a concern in the developed world.

Those are other initiatives that are ongoing, and then finally, I participate in a couple of other national committees through things like: The American Society of Hematology, The Society for Vascular Medicine, some additional panels to help advise and provide some guidance on how we should be

thinking about COVID-19 from the inflammatory and the vascular complication perspective, and how do we design new initiatives to be able to develop either funding sources or guidance for clinicians and for patients, and set new research agendas for the future itself of this disease and others. I have been fortunate to be surrounded by a lot of really smart people who teach me more than I teach them.

Barr: That is a great thing. In addition to being a scientist, you are also a clinician so, can you speak a little bit about what it was like for you to treat your patients during this time of COVID, and as well as, if you have worked with any COVID patients?

Kanthi: We have been doing a lot of our work remotely since the pandemic started, and a lot of the work went through some of our clinical trials, for example. We were on the phone with patients almost every day, talking to the physicians and the other clinicians trying to brainstorm on ways that we might be able to improve the care of these patients and in some cases, making suggestions if a patient seems to be not doing very well even if we were blinded to the course of treatment, but trying to think about ways that we might be able to preempt disastrous consequences for a patient, whether a patient might need more or less oxygen, or other different types of treatments whether it is steroids, anticoagulants, or other anti-inflammatory agents, anything that might be able to help that patient.

In addition to that, part of this disease has been that we see families that get infected not just from their loved ones being in the hospital, but oftentimes several family members who are hospitalized and are suffering at the same time, some passing away. Consequently, there seem to be this other ancillary function that physicians are drawn to which is being able to talk to families and help explain and guide them through this process. It is a challenging time. There is so much fear associated with this, there is grief associated with it, and we, as physicians and clinicians, also see this directly. It is a privilege to be to be involved in patient care in this way. I still get calls and emails from my patients giving me updates on how they are doing, and these are patients that I have been taking care of for a number of years now.

There is still a lot to be done. It is a humbling disease, and I think every physician and clinician, whether you are a physician, nurse, or a physician extender, or involved as another health care worker (people who deliver food and clean the rooms), we all see firsthand what the impact of this disease is. A patient may be doing well one day, and then six hours later on may be in respiratory failure, so we all understand the gravity and the profound impact that this disease has on patients, their families, and on the people who are in the hospital taking care of them.

Barr: How did you have to transition? You said that you did a lot of your patient care remotely, which is quite a transition from what you used to when you were able to see them in person. What kind of things did you learn how to do in order to communicate with your patients better, show that you are still empathetic? Sometimes that is hard over a computer or over a phone and doing just sort of examinations remotely that can be a challenge as well.

Kanthi: It has certainly been an adjustment. We went from being in my clinic, which was a vascular medicine clinic, where a lot of the diagnosis is by looking and feeling and touching, to see what the skin feels like, or what someone's pulse might be, or looking at a rash carefully, things that are difficult to do remotely. Being able to try and do this via web portals can be challenging. How do you ask someone who is 80 years old to be able to take their webcam on their computer and use that to show me what their toes look like and what their foot wounds look like? That is very difficult to do.

We certainly, had a lot of struggles, where people were using their iPhones and iPads and instead of looking at their feet, the camera pointed the wrong way, and we were looking at their dog instead. So, there are a lot of struggles that go along with it, but I think the patience here is important. The lesson here is that it is important to be patient and understand that it is just as difficult for our patients who are on the other end of this, who otherwise feel in many ways deprived of the care that they would normally receive in a clinical setting, where their blood pressure is being checked, or I can physically, look at their pills and count up the medicines that they are using, and make sure they are taking the right dose itself. Being able to do all of those things remotely is difficult, and so, patients felt this loss just as much as we as clinicians did. It has been an adjustment, and as long as we both understood that it will take longer to do a clinic visit. The visit is not going to be 45 minutes. It could be multiple 45 minutes, or it could be an hour and a half sometimes, just to try and establish some of those same standards and protocols. As long as we maintained that flexibility, and I have been fortunate that the NIH and the University of Michigan gave me that type of flexibility; we were okay. We managed to make that work.

The empathy part is really difficult. It is hard to communicate tone over email, and it is hard to communicate empathy via web portals, but making sure that I was asking some of those same questions that I always did before, asking patients about their grandchildren, asking younger adults about their other families and their job, and how they are managing to maintain not just their physical but their mental health in the middle of the pandemic, and sharing some of the struggles that we ourselves, and that I myself, as a physician, have gone through in this disease, all of this has helped re-establish some of that empathy.

My personal family has been significantly, affected by the pandemic itself, not just people who have had COVID, and some who have passed away, but also people who have had other diagnoses that were delayed as a result of the pandemic itself. We had three to four people in the family who have had an advanced malignancy that went undiagnosed for too long because of the pandemic and we have lost people in our family. Understanding how my family feels and how I feel helped me understand how patients might be feeling at home themselves. Talking to patients who are either my established patients from my clinical practice, or in talking with patients who are hospitalized, especially those that we were considering for our clinical trial, understanding that they too are in these same scary moments and being able to share with them that this is not a foreign fear to me and my team, and that we are living with the same fear, helped them understand that we treat them like we would treat family. This approach helps re-establish that foundation of empathy that we rely on
and need to be able to take better care of patients. It has been incredibly challenging, but also incredibly powerful and rewarding to be able to provide not only for my family, but also for our patients and their families during the pandemic.

It is a gratifying time in my life, and a landmark and transformative moment in my career. We moved three times. We moved from Ann Arbor, Michigan to Washington DC. We also, then, had to quickly, re-establish a second home in Columbus, to take care of our family in Ohio, Cincinnati, and Columbus who were in part diagnosed with some cancers, and be able to provide full-time care for them while being able to run our clinical trial and keep our translational research going at the same time, trying to establish the research lab here at the NIH. I have been fortunate to be able to bring a talented scientist with me from Michigan, and then start recruiting a postbac in the lab, and so far, we were really just two to three people in our lab trying to move this forward and collaborate with the team at the University of Michigan. It has been an interesting time. We have had three major moves along with this so I am used to living out of a backpack, and just needing a laptop to be able to function, and a good cellphone connection.

Barr: This is one of my last questions. How has your work with COVID-19 made you a better physician and scientist?

Kanthi: Something I reflect on quite a bit. It has made me more humble. I think that I am more willing to think beyond some of the parameters that I explored before, to understand that disease can really manifest in so many different forms and that patients' lives are profoundly affected by this, and when patients do mention an obscure symptom of some kind, I take this seriously, more than I did before. Now I think more carefully about the disease manifestations to be able to have more in and deeper empathy for what patients go through in their lives, and to find ways to be a fierce advocate for patients who have diseases both common and rare, so that we can explore mechanisms of disease and then try to find ways that we can move this forward in the clinic to be able to find new treatments. This is the whole reason I moved to the NIH, to be able to accelerate some of this movement of basic discoveries to patients, and there is no better place to do that than the NIH and the Clinical Center here. This place is going to have a transformative effect on many different diseases; it already has. I am hoping to be able to be a part of more of these, as things move forward. The final point I will mention is that COVID-19 has helped me recognize the willingness of people to cross traditional boundaries and collaborate with one another, to see what we can learn from one another, to be able to try and inform our disease states and band together to tackle important problems in new and interesting ways that have not been done before. This part is the one I am most excited about for the future. I am re-energized and have a lot of hope for what we, as a community, are going to be able to establish and do.

Barr: I wish you and your team all the best and continued success, and of course, continued health. Thank you for everything that you do both caring for patients and in your research.

Kanthi: Thank you, Gabrielle, for this opportunity and for all that you do to archive this important time which we will think about for centuries ahead for the NIH. I appreciate your efforts.