Astrid Haase, M.D., Ph.D.

Behind the Mask

October 7, 2022

Barr: Good morning. Today is October 7, 2022. My name is Gabrielle Barr, and I'm an archivist with the Office of NIH History and Stetten Museum. Today I have the pleasure of speaking with Dr. Astrid Haase. Dr. Haase is a Stadtman Tenure-Track Investigator and Acting Section Chief of the RNA Biology Section in the Laboratory of Cellular and Molecular Biology branch at NIDDK [National Institute of Diabetes and Digestive and Kidney Diseases]. Today, she's going to be speaking about her COVID-19 research and experiences. Thank you very much for being with me.

Haase: Thank you very much for the invitation, Gabrielle. I'm really excited to be here. Thanks for doing this. This is going to be a nice resource for people.

Barr: You work a lot with RNA. Can you speak a little bit about what your lab does and how that put you in a position to work on COVID-19?

Haase: Of course, I'm happy to. What is fascinating, and what might come as a surprise, is that throughout evolution we've always lived with viruses. As an RNA biologist, RNA might have been the first molecule that enabled life. What do I mean by this? When you look at the human genome, more than 50% of the human genome looks like fragments of really old viruses. I explain this to new students when they come to the lab as if the human race was not existing anymore and aliens came from space and found our remnants and sequenced our genomes. We would look like a giant, very sophisticated retrovirus because more than half of our genome looks viral. But why are we not sick? Why are we living how we live? We are very stable, aren't we? What is really, really important is that we control these fragments. What happens is that very, very early in development, we locked them all up. We close all of them. We do this by epigenetics, which are modifications that we put on the DNA super early in development that then say they are never allowed to function. This space was also called "dark matter", although I know that physicists suppose it's very different, of course. There's about half of our genomic space that we do not use on a daily basis. In contrast, we don't want this space to ever be active, because it's old sequences from old viruses. By "old" I mean evolutionary time ancient.

Barr: We got over this virus at some point and our bodies think we don't have that anymore?

Haase: That is an absolutely excellent question. That's exactly what happens, and that's exactly what we study. What we know is that we are actually able to accommodate a virus in our genome and get this virus under control. That is exactly the mechanism that my lab studies. We started this in germ line stem cells and germ cells, where all of this happens. If a virus wants to be present in the next generation, they have to enter our germ cells, so our egg and our sperm, because that allows them to be nicely transmitted to the embryo. That's what some retroviruses like to do. Germ cells have this special RNA-based system, so that's what we work on.

It's a small RNA-based system that tries to detect the incoming virus and tries to silence it. We can't really cut it out without doing too much damage and killing the cell. That's really not what animals do anymore. Bacteria can do this. The small RNAs that we work on—they're called PIWI-interacting or piRNAs—really try to detect the new invader. And then they just say, “Wait, there's a virus—okay, you are sitting in our genome now, but we close the door.” It's like in the library—the bookshelf with the really, really valuable books. I have a five-year-old. You don't want to have a five-year-old with chocolate fingers going to the precious books, so this shelf is hopefully locked. There are some other shelves that are open. These things are getting closed for good, already in germ cells, by small RNA-based mechanisms.

That's the point where our interest came in when the pandemic started. We have this long-standing interest in trying to figure out how this small RNA-based immune system is able to recognize incoming viruses and protect us against our resident parasites. An interesting thing is that it’s RNA against RNA. RNA is extremely flexible because it has sequence. We learn this now in middle school—when you have DNA form a double strand, when you have RNA or DNA strands, if they have complementary base pairs, they can actually recognize each other. What is super cool is that short RNAs and the RNAs we work on are only about 25 nucleotides in length—they are really tiny. They can recognize a virus; they just need to contain a complementary sequence. These kinds of mechanisms—that rely on these short guide RNAs to recognize another RNA by complementary base pairing—are called RNA interference pathways. They exist in all eukaryotes that we have looked at, with very similar things in prokaryotes too.

What about COVID? The pandemic started, and I think we were all shocked. Of course, us scientists were perhaps a little bit more shocked than my parents who didn't know much—they thought it was another cold. For us, it was pretty clear that this is something bigger. Over the last years or decades, it has also been clear that something like this was bound to happen. When, for example, SARS-CoV-1 was around, we were just lucky that it got contained before it spread far. With a pandemic this huge, the lab shut down. The most important thing for me at the very beginning was that everyone in the lab was safe. On the personnel side, our lab is very mixed—we have American and international people. Especially for the international ones, these two years have been extremely hard, because some of them have not been able to see their parents. We have students in the lab and a postdoc who have not been able to see their family in the last two years because they just couldn’t do international travel. But we thought perhaps we could help because this is an RNA virus. As dangerous and terrible as it is, in terms of RNA viruses, it’s a pretty sophisticated virus. Coronaviruses are the largest single-stranded RNA viruses, with the size of something that we cannot synthesize in a lab. Nature can always do more than we can do. But we thought that the system we normally study on a daily basis is able to get a virus under control just by making small RNAs that recognize certain parts. Perhaps we can use the same strategies and the same concept to try to develop something that will help us combat COVID-19. A lot of things came together.

Of course, we are not the only ones who think like this. This is a concept that has been harnessed for a long time. It’s really taking off now because we are finally able to do it technically. This has been given the name antisense oligonucleotides, or ASOs. You might know of some ASOs that recently entered the clinic. What people do is use the principle of recognizing an RNA by complementary base pairing, then make a change there that results in degradation by doing something on a molecule. And this works. What did we want to do? We first looked at the virus and thought that since viruses are extremely compact, it must have vulnerabilities. What we

first thought about is that we have to have a very precise project. Otherwise, we can't do experiments. We never worked with antivirus. It's not that easy and has a lot of regulations. It was hard enough to actually keep a lab running during a pandemic, so we didn't want to start anything crazy like that. We also wanted to have a project that we can actually do in the lab. We thought we should look at the viral sequence and look at some vulnerabilities that we can identify. We looked at different stages. Think about it—the virus is coming in, you breathe in, and it's going in your lung epithelium in the cells. There's one crucial step, of course. The virus has to get in, but that's not our part. We look at what happens when the viral RNA is in the cell and is exposed. At the very beginning it needs to make the viral proteins, which means it needs this initial round of translation. This is really not a normal looking RNA. What we looked at is how this initial round of translation starts. Then we can somehow tackle this crucial point. If it can't do this initial round, nothing will happen.

Barr: What does the RNA look like? You said it doesn't look normal at all.

Haase: Viruses don't have a lot of space. They have to be very compact. This virus actually has a lot of RNA structure at the beginning where translation starts. It is very, very compact. It doesn't have a lot of flexible space it can just use. Half of our genome is old viruses. It's kind of a flexible space—a lot of this stuff is junk. Viruses don't have junk. What we immediately saw is where the first ATG [autophagy related] are—where the first translation starts, and it is required to make the first protein—is actually in a very structured region. There is a long stem loop that seems to have a lot of energy. What we were wondering is why this protein has the initial start code and needs to be recognized by the ribosome in such a highly structured and compact region. One of the hypotheses is that this helps translation. For example, there are indications that some viruses use highly structured regions as internal start sites—so internal ribosomal entry sites, or IRES. But this is not the case with coronavirus—it doesn't do an IRES. It seems to do dependent translation. We thought perhaps it is recruiting something, so it could be either positive or negative. That's what we first wanted to test. We had to have something we can move in a testable system because we don't need to work with the entire virus. We just made a construct that contains just it's beginning until the start. We don't make a viral protein; we just make a "sense" of protein. We take something that is green, so it's easy for us to see, and then we just tried it out. We take the beginning of the RNA as it is and see whether it is actually easy for this piece to make a protein. Then we make certain constructs where we make changes—we break the structure, take it apart, and make it very fluff. The ribosome needs to open a structure to access this ATG. We designed a whole set of constructs that tried to make this happen stronger, make it weaker, move the ATG, and see how the normal context of the virus compares to these other constructs. What we saw in this very simple experiment is that having the ATG in the stem loop is not good for translation. It makes it really difficult. If the virus had the option of taking the ATG out of the stem loop, translation would be much, much easier. You might want to know why it has to stand up. The virus doesn't have a lot of space to try to keep the genome very compact. It needs the stem way later in the lifecycle to actually form a proper viral particle—so it needs the stem loop. If you have a lot of space, like if you have a big house, you don't need a folding bed and folding chair or whatever, because you have space to spread stuff out. But think about it like the virus lives in a shoebox and the virus doesn't have space. The virus doesn't have the luxury to, say, put something it needs later in a different room. It has to put everything together. Having seen this told us this is actually something that is not great for the virus. This might be a vulnerability that we could use.

We don't have a solution yet. This is also due to the pandemic. Sometimes it's not easy from basic or fundamental research to actually go and develop a therapeutic. What we want to do is just describe what we see and what we think the implications can be, and then hope that industry is picking that up—or someone who is technically better at doing it. One thing that we are suggesting now, and this is combining it with a mechanism that we usually study, is that having the stem loop is not great for this initial round of translation. What if we forced the stem loop to be even more stable than it already is? You could do this with an antisense oligo. Again, it would be really great to get some more industry help because these things also need to be modified to be applied to sell. What we want to do is have an antisense oligo that now locks the stem loop in. We do the same things as pioneers do in the genome, which is to close this one bookshelf and say "Wait, this is not accessible for you now." The idea we would like to try out, and where we think there's potential for therapy, is if you could design an oligo, you lock in this initial happening. You prevent—or you make it even less efficient for—the virus to do this initial round of making its own proteins. You could really dampen it. What we found and why we think this is possible is that there's a company at the moment that does ASO therapeutics that has a patent and has developed the method to deliver ASOs in aerosol formulation. That is also cool. One idea—and this is really an idea for an application—would be if you could make this little ironic antisense oligo, this ASO, as a therapy. You can give it to people through an inhaler because we know that the virus enters through our respiratory epithelium. We know that so far, ASOs, when they have been used, are relatively stable. We could give people a boost for an inhaler, and they would have this ASO. Of course, we would need to test this ASO to be sure it doesn't do anything else, but that can be done. If the virus comes in, even if the virus enters the cell, it can't do anything because it can't initiative its first round of making proteins.

Barr: Would you have to keep changing it with all the different variants?

Haase: That is a very, very good question. And to be honest, I haven't looked at this yet. The stem loop is so crucial for the virus to assemble virus afterwards that this is not a very mutagenic region. This is not a region that happened to mutate a lot, but for the new variants we would have to look at it again. But that is another cool thing about ASO therapeutics and using RNA in therapy, like with a vaccine—once you actually have the therapy established, you can simply change the sequence. You don't need to develop a completely new product, like now with the updated boosters. You use an RNA, and you are extremely flexible. I have the impression that the best ideas that we get for therapy come from watching what life has already done during evolution. What I find so fascinating is that basic research and the clinic R&D [research and development] are so close together nowadays. I started my training as a medical doctor. I'm not a clinician in the U.S.—I'm completely in the lab now. It's really important to figure out how these basic mechanisms work—just like what we do on a daily basis trying to understand how we close our inner viruses and tell them they can be there, but they're not allowed to do anything. We use these methods to develop therapeutics that are equally cool and can be equally successful.

Barr: You were looking at COVID-19 because of the pandemic, but have you looked at other coronaviruses and if this mechanism could work for them?

Haase: We looked for SARS-CoV-1 and they look similar. If the principle works that you can use an ASO to sterilize viral structures, that could even be used in other regions of the virus. It would be really fun to try this. Everyone does their part—we want to describe what we see and what we think would be cool to try. And then

hope that some cool RNA chemists who actually know how to formulate this and do this in an industry setting will have the capability to try multiple oligos and optimize this. I hope that they will try that.

Barr: Does your lab plan to do any more COVID-19 research?

Haase: We want to finish this. We don't want to give up our old interest in figuring out how our own genome figures things out. Every new insight into what we actually do in early development to control our own inner viruses always spurs a new idea. With most of the basic or fundamental research, we are generating the knowledge that is required to then say, “Wait, there's actually that cool stuff, perhaps we can use this.”

Barr: This is a weird thing to say, because I know you're focused on medicine of the future, but you could probably learn so much about the history of humankind through your research looking at all the different viruses humans have had and have overcome.

Haase: I was super excited when Svante Pääbo was awarded the Nobel Prize in Physiology and Medicine for his amazing work in mitochondrial sequencing and nuclear genome sequencing to actually look at human evolution, from the Neanderthals and the others to the modern human. And we see changes there. There's an entire field called mobile genetic elements or transposons. They were discovered by Barbara McClintock way back when, and she was awarded the Nobel Prize for finding that we have these mobile elements in our genomes. She discovered them first in plants. There's an entire research field looking at our inner viruses and what they tell us about where we come from—and perhaps even where we are going.

Barr: It's so cool—it's really neat to think about. In addition to doing your COVID-19 research, you’re also part of managing a lab. Can you talk a little bit about that experience during a pandemic?

Haase: It was bad. I like working with people, and we have a really nice team here. I really missed seeing people—the pandemic isolated us all a lot. But what I found fascinating is that we are able to adapt and to make the best of it as it happens. Just make the best of it and don’t endanger people. At the beginning, we were mostly at home on Zoom. There was always only one person in the lab. NIH helped us to have the proper PPE and the proper masks. We still wear masks in the lab when we talk to each other. It's not mandatory anymore, but to be honest, if we don't get it [COVID], I prefer that. Now we are all vaccinated and boosted. But I think one point that I want to stress is that I have a five-year-old and one of my postdocs has a five-year-old. These kids were three at the beginning of the pandemic. We were very, very lucky. I have a daughter. We were very lucky that she was still in daycare, and daycare was only closed for three months and then reopened. For some of my colleagues who had kids in school, it was much harder. People had different reasons—they either had kids or they had to take care of a vulnerable family member.

It was not easy, and I'm very proud of my team. They accommodated everything really, really well. We talked to each other, and people were very respectful with each other. It's like a discussion we’re having in society at the moment. If you're a young and healthy person, and you're vaccinated, your risk is very low. But not everyone is a young and healthy person, so we are taking care of others. I wear my mask not only to protect me but also to protect others. Before the vaccines were approved for kids, the masks were protecting our kids, because we

really had no data on how much COVID could harm our kids. It was protecting the elderly and the vulnerable in society. We still should think about how there are some people who don't have a great immune system—people who have to undergo types of chemotherapy, people with autoimmune diseases, and elderly people. For them, it's a luxury to have a good immune system and develop antibodies when you get vaccinated, isn't it? When you and I get vaccinated, I hope that our immune system just kicks in and says, “Oh, thank you for showing me the blueprint. Now I know what the guy looks like, and I can actually protect you when I see him next.” Some people can't do this, so we also want to protect them. We have been very proud of the guys in the lab. They have really been working together.

Barr: How many people do you have in your lab?

Haase: At the moment, because some actually left, we have six.

Barr: You said you had some international folks that worked in your lab. How did you support them in keeping their spirits high during the pandemic?

Haase: That is what the team did. One of my grad students has not seen her family since the start of the pandemic. Now she's renewing her visa and can’t travel either. It has been rough. People talk to each other—they were distancing but saw each other in the lab a little bit. They were very good at helping each other out. My lab was up immediately, and the guys organized that themselves. We could get tested. The NIH was very good [about that] and had a testing facility. At the beginning, we just went testing every week. If you caught the virus, you go home and isolate. That worked pretty well. We tried to stay in contact as much as we could, and we tried to discuss what was going on. The COVID project that I told you about was driven by that student. It was important for her to have the impression that you can actually do something. One of the things that really makes you depressed is when you have the impression that you are faced with a humongous challenge like a pandemic, and you can’t do anything. That’s why we said, “Okay, perhaps it's a small contribution, but every small contribution counts.” She was the one who was really driving the project and started it.

Barr: Is there anything else that you'd like to say about your COVID-19 experiences both professionally and personally?

Haase: So many things. I'm really just happy that we are all here. I know a lot of people have lost people or had more terrible experiences. We were very fortunate. I also have to say, I'm very fortunate for my family because it's really hard when you have to do this alone—taking care of a kid. After the three months that daycare was closed, our daughter told us she really wanted to play with other kids. She's an only child, and while we make an effort, and she recognizes our effort, it's getting a little boring. We were really fortunate that the NIH daycare reopened—they were very good. It will be interesting to watch this generation grow up because these kids are used to wearing masks. Our kid doesn't mind wearing masks. Our kid knows to wash her hands and to have a distance without being antisocial. She's a very social kid. They learn to take care of each other, which is a nice thing. They have a big hero, of course, at the NIH. Since it's NIH daycare, they talked about Dr. Anthony Fauci a lot. And when you ask her what she wants to be when she grows up, she tells us she wants to be a “girl Dr. Fauci.” And I applaud this. She's a little young, but otherwise, we might want to write him a letter and tell him

perhaps he should consider her for the next director of NIAID [National Institute of Allergy and Infectious Disease]. She's a little young, but it's a good inspiration.

Barr: Definitely. Did you worry at all about her development with it being during the pandemic and with everyone wearing masks? Some parents were really worried about their kids not socializing with other kids as much or the masks not allowing for the same kinds of speaking they normally would.

Haase: There are always a lot of things to worry about. We tried to prioritize. There are so many examples of viruses having really bad long-term affects—not only acute but also long-term effects. Now we are talking about long COVID—at the beginning, this was not a thing. I was worried for her to get the virus before she was able to get protection. As an RNA biologist, it was exciting for us to watch how fast the vaccines were developed. Some people say this came out of the blue. Nothing comes out of the blue. This was already in development for so long, but for cancer vaccines. It was just at the right point that we could actually tap into this cool new technology. Once we realized it was possible, we were all hopeful that the kids could get vaccinated soon. I was more worried about that. Of course, we spend time with her at home. My husband is a senior investigator at the NIH in a different Institute. She is used to daycare, but I think she knows how much we love her, and all our free time belongs to her. We read books to her, and we talk to her. These kids learn to read each other's eyes. I have an impression they read each other even if they wear masks. It's different—young kids make the best out of it.

Barr: They adapted. It's a different kind of language.

Haase: Exactly. They're much more adaptable than we are when we are adults. To give you an example, when the pandemic hit, our kid was just turning three, and finally had to learn to go to the potty by herself at daycare. That is a huge change for a kid. You don't have your pacifier anymore, you need to learn to go to the potty, you learn to put on your shoes. For her, putting on a mask was just one more thing. There were so many things to learn.

Barr: It's a different mentality than as an adult, obviously.

Haase: As an adult, it's different, because we have something that we are used to. But think about it—kids change all the time. They learn so much in those first years. For her it might have been just the right age that having a mask was just one more thing. She also had to learn to hold the spoon properly and eat properly at the table. There are so many things that were new. The mask was just one, and from what I've watched with her group, these kids had no problem with this. They're healthy and strong. They met on Sunday mornings and played soccer outside together. At the beginning before they were vaccinated, they all played with masks. Kids have no problem running around wearing the masks or being on the playground.

Barr: It's inspiring as adults to learn from kids.

Haase: You're completely right. Yes. Kids tell you what we can do, actually, and how resilient we are. Kids overcome so much. Look at a baby when they learn how to hold their head or learn how to turn around. We watched when our daughter was a baby, and the kids learn to flip and learn to sit. There's a lot of trial and error.

They fall over constantly but they never give up. We can learn a lot from them—that we are stronger than we sometimes think we are.

Barr: Definitely. Thank you so much for all your work and for sharing your experiences.

Haase: Thank you very much for doing this.