What is MDS and Standard Treatment Options

Christopher R. Cogle, MD
Preeti Narayan, MD
Christina Cline, RN
Damian Alderman

Audrey Hassan: Good morning, everyone. I want to welcome you to the MDS Foundation Patient and Family Forum today. I want to thank you for choosing to spend your Saturday with us. I'm Audrey Hassan. For those of you that don't me. I know many faces out there and I'm happy to see you coming today, but for those of you that don't know me I'm Audrey Hassan, the patient liaison with the MDS Foundation and Debra Murray, I know many of you know her as well. She's here with me today as well. It's an honor and a pleasure to have today as your guest speakers Drs. Cogle, Narayan, Alderman and Nurse Christina Cline from the University of Florida Health Shands. I want to thank you for volunteering your Saturday today to spend with us.

As a side note, I just wanted to let everyone know as an added bonus immediately following this program will be a program on iron overload, so I hope you can stay for that.

So without further ado, I'd like to get the program started and please join me in welcoming Dr. Cogle.

(Applause)

Christopher R. Cogle, MD: Thank you, Audrey, and thank you for the MDS Foundation for hosting this event. Also I would like to thank the faculty, Christina Cline, Dr. Narayan for presenting and also to Stephie McGrail who's our staff that helped put this on and, of course, you.

We are really excited to present today and to talk with you about MDS. So, the first part of the day is we're going to update you on MDS and what it is and bone marrow transplant and clinical trials and then around lunch we're going to flip the table around literally and we are… what we'd like to do is we'd like to hear from you about what you're interested in finding about MDS. We're going to show you what we know and what we don't know, but we'd like to hear from you and what's important as a patient or as an caregiver helping care of a patient with MDS and so we have about seven questions. We have some citizen scientists who are going to help facilitate that discussion and I will tell you that your answers will shape the way that we do research and so we're really excited about that, that part of our presentation today.

So, let's begin with what is MDS. You know what it is from having to struggle with it, but from the science standpoint if you take apart the name myelo means bone marrow cells or bone

marrow, dysplastic essentially is Greek for funny looking and syndrome means a group of symptoms. So, Myelodysplastic Syndromes really quite simply is funny looking bone marrow that presents clinically in lots of different ways and so if the disease or if your doctor when he or she is explaining what MDS is if you ever feel confused, you didn't really get a good clear answer what it is it's because most physicians and scientists are also somewhat confused about what the disease is because it can present in so many different ways and there is so many causes.

So, what I'd like to is to talk with you about five aspects about MDS and the first aspect is what I call the Picasso premise. So, to appreciate Picasso or any modern art it's quite difficult. When you walk up to a painting you can't just immediately look at the colors and that's it and walk away. What you have to do in your mind's eye to really appreciate modern art is that at the same time that you're looking at a modern art, you also have to know what is normal. So, to appreciate Picasso's "Guitar on Table" this is my guitar on my table and what you're doing simultaneously is know what normal and abnormal is and this is very difficult when it comes to medicine and science. So, here is a picture. So, to appreciate MDS, you first need to know what normal looks like. So, here is a normal bone marrow and what you can see the most obvious thing is a big white circle, actually two big circles, maybe even three, four, actually. So, what those are are actually fat globules because most of our bone marrow is actually fat. In fact, a minority of our bone marrow is cells and so when Dr. Narayan sit down and look at bone marrow we first ask what's the age of the patient because for a bone marrow the cellularity should be roughly 100 minus the age. So, if you're 70 years old and we're looking at a bone marrow only about 30 percent of the bone marrow should be cells and the other 30 percent is usually fat and other stroma or scaffolding inside the bone marrow and what you'll also notice is that a lot of the dark purple nuclei, that's where all the DNA is kept together and condensed, all of those dark purple areas are nice circles. They're roughly nice circles. There's a couple of them that are kind of stretched out a little bit, but most of them are organized and there's a nice even and condensed purpleness to it.

But if you look at MDS what you notice first of all in this cases there's a lot more cells. There's not a lot of fat globule or there's no big fat globules. So, there's more cells and the other thing is look at the purple nuclei or where the DNA are are a little bit more hazy. They're a little bit more drawn out. They're a little bit more opaque, more light's coming through them. There's also some funny looking nuclei and here you can see if I can… there's no pointer there, so you're going to have to take my word for it. Some of these nuclei look like they've got BB shots through them where the white small BB shots on the MDS side. That's abnormal. There's also some misshapen nuclei. So, when we say funny looking to a pathologist they would look on the right side of the MDS and they would say those are some funny looking cells that are in there and that's essentially how to diagnose MDS. We're using very old technology, a light microscope invented in the 1600s to be able to look at "funny looking." So, what's funny looking? It's in the eye of the beholder. So, here's some funny looking cells. So, the myeloid

cells make your white cells, the megacaryocyte cells make your platelets and the erythroid cells make your red blood cells. So, these are three MDS versions of those important bone marrow elements. Now, if you look at the myeloid the reason why I know that's funny looking is look at the nuclei, the dark purple nuclei on that top picture. It's crazy. It's all over the place. There's all these lobules and outpouchings and there's no organization to it and on the megakaryocyte which is the second cell. For that one, for megakaryocytes it should not be one organized purple, dark purple nucleus. That one is supposed to be a little bit crazy, but it's not. So, it's the opposite situation and then finally in the bottom the erythroid, you can see there those cells, those two nucleated cells on the bottom they should be turning pink, filling up with iron and they should be spitting out their nucleus in preparation to become a circulating red cell. So, the fact that these two erythroid cells are still holding onto their nucleus that's abnormal. That's funny looking. So, when the pathologist reads your bone marrow this is the thought process that they're going on. Now, it's difficult and there was a study done a few years ago that asked well if finding something funny is hard then how often does one pathologist at one hospital agree with another pathologist at another hospital and it turns out that there's okay agreement when it comes to the myeloid and megakaryocytes. So, the myeloid, again, making white cells, the megacaryocyte making platelets, but when it came to agreeing on what is funny looking on the erythroid or red cells you can see there's only 75 percent agreement and so that's why you might go to one hospital and be told you have MDS and then you go to another hospital, another doctor and you're told well, I'm not sure this is MDS. So, if that's happened to you this is why because of what funny looking means. Very tough.

So, I had a question about this years ago and thinking well if it's hard to diagnose MDS then how sure are we about how many people in the United States or in Florida have that MDS because if you… so I asked the Cancer Registry which is located down in Miami. It's where the operations are, how do you identify MDS patients and how do you keep track of them? It's essentially a paper process where the patient with MDS has to actually come into a hospital and someone down in the basement probably with no windows is looking through your chart records trying to decide whether or not this is MDS or not which is tough and when you do that it comes out to about three… almost three out of 100,000 people in the United States have MDS using this method. Well, we have a busy clinic upstairs in the seventh floor and Dr. Gordon and his group over at North Florida have a busy group and I heard someone talking about Dr. Cartwright's group in Ochoa. I know they got a busy group and all of us know that there's more than this many patients that have MDS just from our practice. So, I designed a way to use computer records to detect how many cases because it turns out computer files are E-mailed down to University of Miami in a secure and encrypted way that alert them whenever a pathology report says this is MDS or it's got all the features of MDS, but the Cancer Registry is so busy with the paper that they never had time to look at the computer files. So, my group and I designed a computer program that could do this to look at those files and when we did that we found there's actually double the amount of MDS patients if you look and if you look at people who are over

65 we almost… we find that the incidence rate is almost actually triple than what the Cancer Registry is. So, what does that mean? If I had to estimate I'd say that there's about 10,000 people that are diagnosed every year in the United States with MDS. So, 10,000 new people every year. Interestingly, that rate has been relatively constant. So, we don't… and it's been constant in the last decade or two. So, we don't think that this is there's an active and ongoing environmental epidemic where we're all being exposed to something… Splenda or there's nothing like that that we can blame as causing MDS. We think that because of the epidemiology information we think that it's happening within bone marrows and because of aging and not because of some environmental push for this disease and we say that. We're inferring that because of the relatively steady incidence rates in the last decade or two and because we're doing a better job of taking care and managing patients with MDS, people with MDS, we estimate that there's somewhere between 60,000 and 170,000 people in the United States right now with MDS which is a large number of people and that's why we have a symposium like this.

Let's get to the second topic. The second topic is what I call software bugs. So, I mentioned to you that when we look at microscope slides and we look for funny looking cells and so when Dr. Narayan and I look at this we know it's funny looking because those blue dots are iron and they should not… these iron should not be accumulating like a ring around the nucleus. We know that because if a cell was… if the cell had good programming, if the DNA was okay, it would not clump up the iron and instead it would put it inside the heme protein and make hemoglobin. So, in fact these are called ring sideroblasts because of the rings of blue that are around the nucleus and so that's a type of MDS. So, this procedure… So, diagnosing this is with a microscope which, again, was invented in the 1600s and is the cornerstone of the way we take care of patients with MDS, but what we're finding out in the last, I'd say about five years, five to 10 years of MDS is that you can't just stop at the microscope level. You have to dig deeper and when you dig deeper into the cell there are these and you look inside the nucleus, that's the darker part of the cell you see there are what's called chromosomes. I'm going to walk you through this because you must know this to understand MDS. So, you get 23 chromosome… if you look inside your cell you gotten 23 chromosomes from your mom and you got 23 chromosomes from your dad. So, when we look at your bone marrow cells and even your peripheral blood cells what we can do is we can grow those cells in a dish and then we can stain those chromo… your mom and dad's chromosomes. Well, they're really yours, but you're borrowing them for a long time. We're able to stain those chromosomes and then we use a computer program to then sort them to find out which is the mom and which is the dad version and so we can see chromosome one on the upper left, one from the mom, one from the dad. Chromosome two, the same thing and with the aid of a computer what we can then do is ask do the two chromosomes match each other? That's important because what we found in MDS is that they don't always match. So, let's talk about chromosomes real quick because that's where the DNA is. So, each chromosome has about 1,000 to 5,000 genes on it and if you take apart a chromosome it's really like a coiled telephone wire, which there's not many audiences you can

say that to, but we all remember. In about 10 years I won't be able to use that metaphor, but you remember having that one phone in the kitchen and then you would go all over and clean dishes and cook or you'd go to the other room and come back and then it'd be all like super coiled. It turns out that that's exactly what our cells do in condensing our DNA and the genes. So, the chromosomes, they're the big ones that you can see pretty easily, they're essentially a super coil of a string of DNA and that's how your body is able to fit 2 to 4 feet of DNA into a tight space called our cell. So, what we've been able to do is we know how to uncoil all of that where we don't break the DNA and then we now have machines that can read the spelling of the DNA after we uncoil it. When we do that we're finding some very, very interesting things which is that there are gene mutations and chromosome abnormalities that are inside the MDS bone marrow. So, we can see parts of that telephone wire that have been cut, snipped out or that are missing completely and so one of your questions might be well, what's on those snipped out portions that's so important? That is the active area of MDS research today is looking at the snipped out portion. Sometimes there's extra chromosomes and you wonder well, what's on that extra part of the chromosomes that's making this disease. Now, you can see here if you look at the arrow. I'm actually making it easy for you. So, if you were medical students I wouldn't put an arrow. I'd make you tell me which two don't match. It's like that *Highlights* magazine where you get like the polar bar eating a candy cane on one side and then the polar bar on this side, but he's missing like an ear or something. What's missing? So, as you can see is on chromosome five where that arrow is there is a little piece that's missing there. That's that deletion 5Q or that deletion of chromosome five that turns out to respond exquisitely well to the drug Lenalidomide or Revlimid. We'll talk about the clinical response, but what we think is happening is the genes that are missing on that chunk of that chromosome, the drug is able to work on that cell and push that cell to die that it's requiring a gene at the end of that chromosome. So, there's a biologic basis to it and it all begins with looking at the chromosome of your MDS and it is a must ask for your doctors is what are my chromosome results. Must ask that.

Now, for at least half the people with MDS, when we do this test and do the stain and put it under a microscope and we read the 20 cells and half the patients we can't catch it and all the chromosomes look normal and that's because of the fact that we're not looking at enough cells. It's very difficult to do this chromosome staining. It's very laborious. It takes specially trained people and computers, but we haven't perfected it and it's just too laborious. So, half the people it comes out "normal" and so if your doctor says that your MDS cells showed normal chromosomes those are air quotes around normal because we know that it's not normal if you dig deep enough. What we call it is uninformative instead of calling it normal. So, what do you do then in this? Well, we dig deeper and if you remember I said that if you can imagine a super coiled telephone wire what we do is we uncoil the chromosomes and then we read every DNA letter on the chromosomes and when we do that what we have found is that there is about 70 to 100 genes out of 20,000 that are recurrently mutated in MDS. So, you can see here recent study that is listing the most frequent gene mutations in hundreds of patients with MDS and I'm not

going to read each one of you and tell you about each one, but the current state of the art in MDS, the current standard is to do gene sequencing to look at what's changed. So, the second question that you must ask your doctor with MDS are what are the gene mutations in my MDS? You must ask that. It has to be done at a minimum once and we're beginning now do it in a repeated fashion where we do it looking at whether or not the MDS is changing and evolving over time which we know it does and I will show you evidence of that. This gene mutation turns out to be… if you know your gene mutations it can be very helpful in a) cinching the diagnosis. So, we can get away from funny looking and actually get to knowing for sure if this MDS by using genetic data, b) it can tell you how well you're going to respond to different treatments. It can tell you your chances of progressing to acute leukemia and then what Dr. Narayan will talk to you about it can even help you in deciding whether to go to a bone marrow transplant. So, this prognostic information is we're learning a lot about and thirdly this genetic information could also suggest treatment. So, there are some mutations that which I'll talk to you about that suggest you should be treated one way versus another way and fourthly what we're developing is that now that we know the gene misspelling we have a technology now that we can do blood draws and measure the level of MDS just from a blood draw. I am… my group and I are trying to get away from bone marrow biopsies and get more towards blood draws. In fact, we just got a clinical trial or we're going through a research approval with Christina Cline in our research office where we're going to start doing finger stick and saliva collections to see if we can catch the MDS even at that level make it as noninvasive as possible and the way that we're able to do this is because we have we were developing technology here at UF that can find these gene mutations in very small quantities. We're able essentially do a photocopying of the gene because a Xerox machine of once we know the misspelling we can just Xerox copy it in the blood and it becomes apparent and we can see it using a computer or a camera and a computer.

So, it turns out that when we look at patients most patients have at least one gene mutation and the average number of driver gene mutations is about two to four. So, most patients with MDS have two to four driver gene mutations and about 100 to 200 accessory mutations that are in the cell. So, these are really messed up cells genetically and so when we've done correlation studies these are just retrospective studies asking does the gene mutation predict how aggressive the disease is and if it's going to progress to leukemia. It turns out that there are five genes that are particularly bad actors**.** They are here. If patients with MDS have these gene mutations then we're going to watch you more closely and we're going to be more apt to recommend treatment sooner and to then not sit on the disease and just watch it like we do in some patients.

I'll be happy to… if you don't catch something, we'll be happy to provide that after.

So, to tell you where we're at, however, is with this IPSS scoring system which many of you have read about on the web or your doctor has talked to you about it in the room and you can see that it's a scoring system where you get a score for cytogenetic is basically a chromosome.

That's the chromosome (inaudible 27:24). Chromosomes are in there, the percent of myelo blasts that are in the bone marrow, how low your hemoglobin is, how low your neutrophil count is and how low your platelet count is. This is what we're using in the clinic, but I just presented to you a bunch of genetic information which is also prognostic. So, what that has caused and then so there's a scoring system I should mention that you guys know that your score is then predictive of how aggressive your disease is and so where we are with MDS is a collision course on how to assimilate this genetic information with this older more clinical parameters or variables like the marrow blasts and how long your hemoglobin is. So, this is all coming together and this is the big crash and the big excitement, if you will, in MDS right now is how do we bring in this new information and so the old news which you guys know about is that your IPSS risk group does or can predict survival time. Now, remember that's with no treatment. So, as soon as you introduce a treatment these survival curves are hopefully pushed upwards is the intent. So, this is in data and people who decide not to get treatment as far as the survival time here, but when you add in… so if you're a low risk and if you add in a gene mutation that's found that low risk patient is actually behaving like a more intermediate risk patient. So, it's a… so having a gene mutation… having one of those gene mutations that we pointed out there, the TP53, etc. that makes the disease worse no matter what the microscope might tell you and that's what we would guess is that if you know what the bad actors on the gene level it doesn't matter what the microscope says. The biology is it's going to be a more aggressive disease. This gene information is also telling us how well people are going to respond to treatment or not and so, for example, in the beginning I told you that Revlimid or Lenalidomide is a good treatment for many patients that have that deletion of chromosome five and that's true, but still some people don't respond and we never really understood why until we started looking at the gene mutations. When you do it turns out that people with this TP53 gene mutation even though they've got the deletion of that chromosome they don't respond because of this TP53 gene mutation. If this seems complex to you it's because it is and it turns out that a lot of doctors are having trouble not only memorizing this information but also keeping track of it and so it's always been common sense to go to a physician whose seen a lot of MDS and soon you'll be asking them what their genetics credentials are also.

And then just real quickly it turns out that we found a gene mutation that also links to getting a good response to Vidaza or Dacogen and that's a gene mutation called TET2 and if the MDS has that gene mutation it turns out they're going to be more likely to respond to Vidaza or Dacogen. Now, I did not… I'm not presenting to you additional information, but it's more complex than this because if you have a ASXL1 mutation then that will negate the TET2 effect, but if you have an EZH2 mutation that negates the ASXL1 negation and you actually do get a response. So, your doctors are having to remember more than just do you need more refills or a lot of the clinical stuff that has been the primary focus in the past. We're entering into an era of genetic oncology and that's an area of active…. Much active research.

I'm going to jump over this, the transplant, because Dr. Narayan's going to talk about that soon.

So, the current thinking in MDS as I mentioned, we are in a traffic jam. We've got light microscope technology, we've got the clinical medicine we're still using the stethoscopes and but we need to because we need to care for the whole patient. We have many needs. We're in an era where we're looking at gene mutation data and gene sequencing and so whose stuck in the middle? The patient and what we're trying to do is try to make sense all of that.

I'm going to end on number three just for the sake of time and really three, I think we need to know this and to understand about MDS and it's called… this section's called clone wars because when we use this new technology of gene sequencing what we're finding is that are two major things, 1) is that there are many gene mutations inside MDS and in each patient's MDS and 2) it turns out that there are more than one MDS inside each individual patient and over time what we're finding and see here as a depiction is that the clones of MDS give rise to daughter clones, granddaughter clones, great-great-granddaughter clones and on and so by the time a patient with MDS or with a leukemia and possibly any other kind of cancer comes to see a cancer doctor they easily could have several cancers all within their body and they all appear to look like just one MDS because under the light microscope you can't tell. You have to do gene sequencing to fully appreciate the multiple clones of MDS that are in somebody and why does that matter? It matters because here's a case of a patient with let's see, one, two, three, four… four clones in them. We give the chemotherapy. It shrinks down all the clones, but then we stop the chemotherapy and then over time the clones reemerge, but the clones give rise to a great-great-granddaughter red clone that you can see here that is now resistant to the chemotherapy that we just gave. Now, in today's or in yesterday's medicine we would do the bone marrow biopsy and say, unfortunately it looks like the disease has come back and we need to treat it and you would say well, what are you going to treat it with and in yesterday's and even in today's medicine the oncologist would say well, what have you gotten in the past? And you would say something like Vidaza or Dacogen or maybe immune suppression and the doctor would say well, we're not going to give that one again. Let's just give something else. What's next on my recipe book or if we have a clinical trial, we'll get you in a clinical trial. That's today's medicine, but where medicine… where MDs and medicine is going now we are doing gene sequencing on that great-great-granddaughter clone and we are asking where are the new gene mutations. Let's compare that to the beginning at diagnosis and let's find a treatment that this great-great-granddaughter clone will not be able to escape. So, we're going at the gene level to figure out what we can give you. That is today's medicine in a clinical trial that we have here at the University of Florida called I Care, I Care for Cancer Patients, and it will be the future of medicine where it'll… we need more information about the relapsed or the refractory cancer and we do that with gene sequencing.

I'm going to stop there, try to answer some questions and then after questions we'll begin to talk about the standard treatment for MDS. Any questions? Yes, sir?

Q1: Does a transplant get rid of the bad genes that you have?

Christopher R. Cogle, MD: Great question. So, the question was does the transplant get rid of the bad genes? It does in 40 percent of the time and this is what Dr. Narayan will talk about is that the new immune system attacks the MDS cells that have the gene mutations. Now, the new immune system finds those MDS cells by the surface proteins, by the coat that the MDS cells are wearing. We're assuming that the genetically mutant MDS cells are wearing a bright pink coat that the new immune system is going to be able to find. We're assuming that and it does 40 percent of the time. Sometimes the MDS cell even though it's genetically mutant doesn't have a different coat and that could be… that's one of the reasons for the disease persisting despite the transplant. The other and this is really nasty tricky the MDS cells can take off their coat, parts of their coat so that the new immune system can't even see them. So, the MDS cells can evolve and make themselves invisible and that's also been shown in the past, too.

Any other questions? Yes, (Attendee).

Q2: (inaudible 38:31) like this is treatments currently are not disease modifying, but are you saying that this genome this molecular (inaudible 38:42) may be able to see some treatable modifying treatments?

Christopher R. Cogle, MD: So, (Attendee)'s point and question was that that most of the current treatments for MDS are not disease modifying meaning that they might just knock down the disease for a little bit but the disease eventually comes back and that's absolutely correct, (Attendee), and your question then was so if you look at the gene sequencing and then apply drugs to the… and then choose a drug based off of the genetic mutations could that be disease modifying? Yes, it can depending on what drugs are available. So, for example, there is a somewhat frequent gene mutation called IDH or isocitrate dehydrogenase. There's an IDH1 enzyme and there's an IDH2 enzyme and either of them can be mutated in MDS. We see IDH2 mutations in about 10 percent of patients with MDS here at our center. So, if a patient came in and failed the Vidaza and we read their genes and we found the IDH2 mutation we would recommend that that patient received the IDH2 drug. That still doesn't mean we… that we give the drug and we walk away because what can happen though is that MDS cell treated with the IDH2 inhibitor drug could evolve and could develop resistance mechanisms to that drug and then develop a subclone. So, one of the very real possibilities where we're going in oncology in MDS is that we can… it's like Whack a Mole where we can hit the disease and knock it down but still be very vigilant for where it's going to pop back up with gene sequencing and when it pops back up we hit it again with targeted agents and then we still watch and if it pops back up gene

sequencing again targeted therapy, whack it down again and so we're moving… where I see us moving is instead of giving a nuclear bomb blast of drugs and then we'll see you later. Instead I see us having a long term relationship with doctors and patients where we're going to be watching very closely when things crop up, we just treat it and we keep watching you. That's where we're headed.

Q2: Is this molecular target.

Christopher R. Cogle, MD: And (Attendee) is asking is that molecular targeting. That's what that is and it's targeted therapies or molecular gene mutations. There's lots of ways to say it to make it sound more fancy.

Yes, ma'am.

Q3: Are there results showing if there's more male or female MDS patients?

Christopher R. Cogle, MD: So, the question is are the results showing if there's more male or female people that have MDS. Yes, and using the paper method of capturing cases of MDS, the paper method shows that there's more men that are diagnosed with MDS, but when we use the computer method that we're finding that it's that there's more younger women that… Well, we're finding that there's younger women are not being captured and that if there is a male predominance it's not by much and that it may be closer to 50-50. So, is it because younger women don't come into the hospital and get and their case is not captured by the registrars we have chained in the basement with no windows? I don't know. It's speculation, but it doesn't… the old data says it's more men, but I'm doubtful of that.

Yes?

Q5: Earlier when you were talking about (inaudible 43:01) get all these different things (inaudible 43:11). What's the possibility of having one pathologist that is is what I'm saying is Dr. Bennett, for example, actually reads all of them in any particular region or state or whatever and that way there you have it… the information would be more accurate.

Christopher R. Cogle, MD: So, (Attendee)'s question is if there's a problem with variations on how pathologist read bone marrow why can't you imprison one pathologist named Dr. John Bennett to read. That would be hard to do. Audrey and MDS Foundation, you guys have probably a good relationship with Dr. Bennett. Please ask him if he would do that.

Audrey Hassan: (inaudible 44:02) be more than happy.

Christopher R. Cogle, MD: Another way to do this, (Attendee), is to train a computer how to read slides. So, if you have a smart phone and you take a picture of a person you know how the smart phone depending on what kind you have puts like a little rectangle wherever there's a face and if that face moves the rectangle moves with it. The computer is using a computer assisted design to be able… It's been programmed that whenever there's two eyes and a mouth and like a head kind of rectangle or oval or whatever it decides it is told to put a box around it and assume that it's a face. So, it turns out that Dr. Narayan has been working with software engineers for the past many months where we're feeding this computer images of MDS and versus normal bone marrow and the idea is that the computer is learning what are the features about an MDS bone marrow that are different to the normal. Now, we're not pointing it out. We're not showing the ring like the iron, the blue iron. We're not showing the computer that. We're not telling the computer what to notice because we're letting… this is called machine learning. We're letting the computer decide what are the features about MDS that are different than normal and the machine will make up its own rules about what MDS is. So, that is being done. It's a project that we have now. It's very slow going because to train a computer you need… I mean, what have you… how many images?

?: (inaudible 45:58) more than 100. At least 100… you have to (inaudible 46:03 – 46:23)

Christopher R. Cogle, MD: It's been very interesting to teach a computer not… we don't teach it what to look at. We just tell it when it's bad and when it's good. So, it's a very different way of… you can't do that to a medical student. Well, you can tell them they're bad or good, but you have to do it in software terms and you also have to show them the blue dots whereas a computer you don't have to show them. So, that's the idea.

Q5: (inaudible 46:49)

Christopher R. Cogle, MD: Well, yeah. So, maybe Dr. Bennett's off the hook. Any other…? Yes, sir?

Q6: You had talked about a finger stick or saliva test in lieu of a bone marrow biopsy. Is that specific to Shands or proprietary here at this point?

Christopher R. Cogle, MD: So, it is a research study that we will most likely launch in the next couple months depending how quickly Christina Cline and my group can push the research through the approval layers and it is an observational study currently where we're going to follow along with the patients and we'll be doing the finger sticks and saliva… is Dr. Hubosky (sp?) doing that like every two weeks I want to say? I think every two weeks is what the current plan is and we would be following with that technology in a shadow mode while the regular care of the patient is ongoing and then once we get enough patients... I forget what her sample size is,

but once we get to a certain number of patients we'll then analyze all the data and see if we could have been more informative than the actual bone marrow biopsies and other standard testing being done. If it is as good as we think it is and all the preliminary work we've done then the next step would be to then launch a study where we do the finger sticking and make decisions off the finger stick results, but that would be in the future. So, in a couple months we'll be doing the finger stick as an observational study.

Yes, ma'am, (Attendee).

Q7: Okay. This is totally off the wall. I know where I got my MDS from. Can somebody be born with the gene of MDS without having to do chemo and radiation?

Christopher R. Cogle, MD: Great question, (Attendee). Absolutely. There are about nine genes that and there are most likely more, but there are about nine genes that can be mutated at birth and they can be mutated spontaneously during embryonic development and the big ones are RUNX1 and GATA2 that we've seen here. I've had a few patients that present with childhood and adolescent low blood counts and then somewhere in their teens they begin to have more of MDS changes in their blood counts. We do a bone marrow biopsy. We find that it's MDS in a younger person than we would expect like a 12 year old or a 19 year old, much younger than when we would expect the average age of MDS being around 71 and so in those patients we definitely do gene sequencing with special attention to those nine genes and if they have the gene mutation there we'll do I will then do a skin biopsy to see if it's in their skins cells, not just in the bone because if it's the skin cells then it's most likely in every… that gene mutations in every cell of their body not just in their blood system like it is with older onset MDS and if it is which it has been in several patients that we've seen we then recommend bone marrow transplant because chemotherapy is not the cure and so for these younger people a bone marrow transplant and we've had success, several successes by doing that with those patients. There's also the family planning part of this. So in patients that have a RUNX1 gene mutation there's a 50 percent chance that they can if they have children and if they want children that they can pass that gene mutation onto their offspring. So, I have those people that are born with the gene mutations do either sperm banking or if there's an oocyte banking we have them bank for that in case in the future if they want to have children. Did I give you too much information, (Attendee)?

Q7: No, it's I have several girls that have low anemia and I'd like to explain why and nobody can explain why they're (inaudible 51:18) or they have low platelets or sometimes they have too high platelets and they keep going back to the doctors and the doctors keep saying I don't know what to tell you and I (inaudible 51:30) born with the gene (inaudible 51:35).

Christopher R. Cogle, MD: Let's talk during one of the breaks we can help you out. There's lots of reasons to have low red cell counts, a lot of reasons, and number 50 on the list is MDS. So, we would first want to make sure it's not the top 49 before, but yeah, let's talk.

Yes, (Attendee).

Q8: I kind of piggybacking onto her question. If you did inherit the gene could it remain latent though or unproblematic much past childhood?

Christopher R. Cogle, MD: Great question. So, could the gene mutation not cause a clinical problem until much later? Certainly. The nine genes that we know about now we know about them because they presented so early and so reproducibly in young kids and adolescents, but there certainly could be gene mutations that have a low penetrance where they don't really present clinically early on. We just haven't discovered them yet. Whenever I have a patient with MDS, that's usually in their 20s or 30s we usually presenting at that age we do… we look for gene mutations that cause bone marrow failure syndromes like MDS, aplastic anemia, things like that. If a patient presents in their late 40s, 50s and 60s, we still do gene sequencing on their blood elements, but I may not do a familial or inherited work up because late 40s, 50s, 60s is about the time when an age acquired MDS starts to kick in. I don't know if that answered your question.

Yes, (Attendee).

Q9: Four or five years ago you said something like starting research on a willow tree from South Africa. How is that working out?

Christopher R. Cogle, MD: Yeah. It's a great… so the greater question is how is that bush doing in the clinical trial? How is that tree… it's going well. So, actually Christina Cline is the research coordinator of it. We have treated over 30 patients with this drug. I don't… are we talking about… are you talking about the oxy trial? So, I'm going to leave that for Christina to talk about at the end.

We were scheduled to talk about standard treatments and I want to make sure we have enough time for both the treatment talk and bone marrow. How about if I present the three drugs that we're using in MDS, most of you already know about it, but we'll present that quickly, I'll take maybe a five or 10minute break and then we'll bring Dr. Narayan to talk about bone marrow transplant.

So, I split this up into treating low risk patients and treating high risk patients. So, again, these are low risk according to clinical definitions and what… So, the types of treatment for someone with not much of an anemia, no increase in blasts and chromosome abnormalities that are not

high risk or not suggesting aggressive disease is first of all the first question I have is is there a chromosome… is there a deletion of chromosome 5Q? The long arm. You guys saw that with that little arrow. I showed you that there was a piece of that chromosome five missing. You had to squint probably to see it, but the… if there is a deletion in that chromosome, what we think about is starting Lenalidomide first or Revlimid and then after… if Lenalidomide doesn't work we then recommend Azacitidine or Dacogen or a clinical trial. The other question that we ask and think about in low risk patients is we measure the erythropoietin level or EPO level. This EPO is actually a hormone that your kidneys produce. Your kidneys are always sensing the oxygen level in your blood. So, it's more than just your lungs. Your kidneys are also there as the judiciary system to make sure it's the check on the lungs to make sure they're doing their job. The kidneys are also communicating with your bone marrow and if the kidneys sense that more oxygen is needed inside your body, it spits out a hormone called EPO or erythropoietin, that's the long name of it. The EPO is spit out, it goes down to your bone marrow and the bone marrow stem cells then will make more of those erythroid or red cells as a reaction. In some patients who have MDS and low blood counts not much oxygen, not as much oxygen is circulating for some reason the kidney hasn't woken up yet and is not making enough of this hormone. So, quite simply we just give you more of the hormone to wake up the bone marrow, to wake up the normal parts of your bone marrow to pump out more red cells. That's as simple as it is and so there's a rough cutoff if that hormone level of EPO is below 500 there's a good chance you're going to respond to these injections. If your hormone level is above 500, your kidney is doing a pretty good job and adding more of this hormone probably is not going to make a big difference and there's a scoring system that we've developed, which you don't need to know about, but it essence it relates to how much your… the EPO, the serum EPO, S-epo or serum EPO level is in your body. So, the other drug is called Lenalidomide or Revlimid is the other name for it. It's approved for the use of transfusion dependent anemia. That's also approved for multiple myeloma as well. There is a study done several years ago where they gave Lenalidomide every day or every day for three weeks and they look for response and what they found is that about two-thirds of patients were able to get off of transfusions. Now you had to have the chromosome 5 deletion to be in this particular study. So, a two-thirds response rate which is very good, very, very good and they were actually able to get rid of the chromosome abnormality in about half the patients and then there was hemoglobin increases. The time to response was four weeks and the average duration of response was two years. So, that's why we typically give Revlimid for at least two months to see if it works because we want to… if the time to response is about four weeks, we'd like to make sure we have enough time being on drug before giving up on it.

Now, some of you are thinking well, I don't have the chromosome 5 deletion, will it work in my case? That study was done or a small study was done and what they found is if you don't have the chromosome 5 deletion you're still able to get about a quarter of the patients off transfusions and approximately 10 percent of patients were able to get rid of the chromosome abnormality. So, much lower response rate, but there are response rates and that's why in the clinic if we're

backed in a corner and there's no obvious way to treat the doctors we do think about giving Revlimid to someone even though they may not have a chromosome 5 deletion.

Now, with these drugs there are side effects which is what always gives us a pause before we prescribe it. So, thrombocytopenia means low platelet counts. Neutropenia means low neutrophil count and those are the two biggest things that we have to watch out for with this drug. Now, you can also give Vidaza and Dacogen to people that have low risk disease and in fact there was a trial done in private practice setting where they gave three versions, three regimens of the Vidaza, and the first regimen was what's called 5-2-2. So, it's five days on, two days off and then two more days. That's the private practice friendly regimen where you have the weekend to… well in the old days, I guess, the doctors would golf on the weekends, but I think our private practice doctors are working every day these days. It's getting so busy for them. Then there's the 5-2-5 regimen which is five days on, weekend off and then come back for another five days and then it's just the five days just Monday through Friday and this is low risk MDS and what they found interestingly was that if you look at transfusion independence the arm that did the best was actually the five day, the Monday through Friday regimen. We don't know why. Now, this is not for high risk patients which is most patients that you would consider giving Vidaza to. This is for low risk patients and so what I caution doctors out in the community is don't take this five day regimen and treat this way for all your MDS patients. The recommendation is still seven days for the higher risk patient. That's where the data is, but for a low risk patient you would be justified to go to just the five days.

I'm going to skip over this and go to… So, what about high risk patients? So, these are people who have lots of chromosome abnormalities, these are people that have more bone marrow blasts, over five percent in the bone marrow. Here's how doctors think about it. We first ask is this patient a stem cell transplant candidate? That's our first question and the reason is because that tis the only curative treatment that we have today in 2017 and that's why we asked Dr. Narayan to talk about transplant to everyone. If they have a donor then we go as quickly as possible to transplant. Now, quick as possible usually means three or four months. So, in that time we sometimes give Vidaza to get people off of blood transfusions, to see if we can reduce the burden of disease and that's an active area of research right now is whether we should give Vidaza or Dacogen and if so how much before going to transplant. Now, most patients with MDS transplant is not an option. It's way too risky for reasons Dr. Narayan will present or there's no donor and in that case what we recommend is the Vidaza or Dacogen**.** You can sometimes give more higher intensity chemotherapy in the hospital which we do sometimes and also to consider a clinical trial where we're testing out these targeted agents against the genetic mutations.

So the Vidaza was a randomized trial against a conventional regimens like Cytarabine and other agents. This was a randomized trial a few years ago and it was high risk patients that it was

tested in. It was multiple centers and here's the survival curves of people that were on the Vidaza back then. You can see in the orange arm which has got the Vidaza versus the dashed blue arm which the people that got whatever conventional treatment was at the time. There was as survival advantage for people that got the Vidaza and so that's the reason why Vidaza now recommended to many patients with high risk MDS and let me back that up and again the major side effects are the low white count which is neutropenia, the low platelet count which is called thrombocytopenia and anemia as well.

Interestingly when we look at physician behavior when we look at insurance billing only about a third of the patients of MDS actually receive treatment. So, most patients aren't being treated with Vidaza. Now, whether they're being offered Vidaza or Dacogen or Lenalidomide we don't know, but we presented research this past December in California at our annual hematology meeting and we showed that most patients actually don't receive treatment and it could be because of the fear of making the blood counts worse or the patient not being healthy enough to receive the treatment. We hope it's not education of the physician. We hope that if anything it's the other problems.

So, Dacogen or Decitabine is another approved treatment for higher risk patients. There's different ways to give it. Most centers give it as a once daily treatment for five days. What we've noticed with the Dacogen is that after only about two to three months you begin to see the benefit of reduced need for transfusions and separately with Vidaza it can take four to six months. There's been no head to head trial and there may never be comparing Vidaza to Dacogen on how quick they work or how well they work. So, we cannot say that Dacogen is any better than Vidaza or vice versa that Vidaza is better than Dacogen. There's no data to say that and it's really just up to whether you want a five day regimen at this point. Vidaza does have the survival data for it. So, there's that to consider as well. There was a trial of Dacogen looking for survival benefit, but they're not able to show the survival benefit. One of the reasons was that because they stopped the Dacogen after they achieved a response and as many of us know in this room once you stop the Dacogen or Vidaza the disease will come back in a matter of months. That was a major flaw in the design of the trial, but to their credit they made that decision back when they didn't know that if you stopped it the disease would come back. So and I doubt we'll ever know whether Dacogen has a survival benefit if you keeping going with it. We probably will never know and the side effects are pretty much the same with Azacitidine or Vidaza. It's just the low blood counts being the big concern.

I'm going to actually stop there and take some questions, take a small break and we'll get to Dr. Narayan. Yes, (Attendee)?

Q10: Are there any treatments where you use like a cocktail of these kind of drugs?

Christopher R. Cogle, MD: Great question. So, are we using cocktails of these drugs? Yes. So, we've done studies where we've combined Azacitidine with Lenalidomide and when we take all comers there was no greater benefit compared to just doing just the Vidaza in the past. However, when we have done subset analysis looking at genetic mutations in the responders of the cocktail compared to the non-responders there is a gene signature, a gene mutation signature in the cocktail responders that we think we found. So, now it's up to us to design a study that just recruits people with that gene signature and see if they, indeed, get that enhanced benefit. That's where we're at in research. Great question.

Okay. Let's take a five minute break. Let's come back at 11:15. We'll hear about transplant.