



Speakers:

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Speaker: ... from the University of Arizona. She's in a cab. She should be here any minute. She is from the University of Arizona Cancer Center. She's a Board of Directors member with the Foundation and she is also the head of our Nurse Leadership Board. So, we are waiting on her any minute now. As an added bonus, immediately following this program will be a program on iron overload. So, I hope you can make it for that also.

So without further ado, we'd like to get the program started and if you please join me in welcoming our first guest speaker that would be great. Thank you.

(Applause)

**Enkhtsetseg Purev, MD:** Good morning, everyone. So, I'll start talking and explaining to you what MDS is and... can you hear me? Hello. Is it better? Alright. So when I start talking about what MDS is I like to start with a survey which was done back in 2013 and presented at ASH which kind of looks at the perspective of the patient and the doctors how do they view MDS and as you can see, first of all, more patients responded than the doctors, but patients view MDS and less than 10 percent look at MDS as a leukemia or a cancer. However, when you ask the doctor more than half of them look at MDS as a leukemia or a cancer. The patients a very minority of them only 29 percent thought that MDS is a curable disease. In contrast the MDs look at it as a curable disease more than half of them and this is just to give you an overview of the patients who responded – a median age of 66, more female than male responded and almost half of them were in an active treatment.

So, to explain you what MDS is I will start talking about the role of the bone marrow what it is. So, bone has... in the middle of the bone there is a bone marrow and if you look under the section and it was stained with ferrous dye, you can see this trabecular bone which is the hard part of the bone and in the middle of it there's many, many cell fleet. So, the stem cells live in between of the cells and they give a rise to all the blood cells. So, they give a rise to the red blood cells which carries oxygen, they give rise to white blood cells which fight infection and the platelets which stops the bleeding.

Just to give you a brief overview of hematopoiesis how the blood cells are formed in the bone marrow it starts with the multiple hematopoietic stem cells and with the various cytokines or signals from other cells they differentiate into the common myeloid and common lymphoid. As you can see it's quite a complex process with the cross talk amongst the cells with multiple cytokines and I highlighted the cytokines which is used in the clinic nowadays. It's cloned. So for example, TPO or Eltrombopag helps production of the megakaryocytes, EPO or Darbepoetin you probably know gives helps production of the red blood cells and GCSF or Neupogen in clinic helps production of the white blood cells.

So, what does myelodysplasia mean in general? So, it's from the three words. The myelo in Greek means bone marrow and dysplasia itself like the meaning of this word is abnormal appearing cells. So if you look under the microscope they don't look... they just simply don't look normal and syndrome means just a group or signs of the disease which occurs together. So myelodysplasia is basically a group of the signs and symptoms of the bone marrow so that the cells look abnormal under the microscope.

So this is how the normal bone marrow looks. So, this is from the healthy donors and this is the section of the marrow and you can look at... it's not very high magnification. So, here's your trabecular bone which is your hard part of the bone and this is in the middle of the bone. So, this person probably like 50 years old man and all the cells look normal. If you look under the microscope here's like precursors of the red blood cells. Here's the precursors of the platelets and here's the precursors of the white blood cells. In a patient who has MDS so this is for example the course of red blood cells and it is abnormal. So, as you can see it takes a lot of experience and many years of looking on the microscope to be able to diagnose. So, we heavily rely on our pathologists. So, this is, for example, normal precursor red blood cells just has one nucleus in the middle, very kind of clean borders, but here you see, for example, like three nucleus kind of overlapping each other. Here you see the precursor of the platelet which has one giant nucleus which shouldn't look like that if you look on the healthy one. This is precursor of the platelets. You can see several nucleus overlapping each other and this precursor of the platelet is first of all it's very giant nucleus and it's just one. This is, I think it's peripheral blood. It's a precursor red blood cells. We call it snowmen so that two nucleus kind of sitting on each other.

So how many patients are there with MDS? So, here you see the incidents rate on the (inaudible 6:20) axis and X axis showing the age of the patient. You can easily see that this disease is disease of older people. We do see sometime patients with MDS which is less than 40, but you can see the majority of them have (inaudible 6:37) the age of 60. So, in red they're showing you the overall incidents, in blue male and yellow showing the female. So, there is a little bit more male predominance in this disease, but the message of this is the disease of the older people.

So, a lot of patients will come with MDS. The first question they ask why did I develop this disease? What did I do wrong? Did I eat something which I'm not supposed to, but in reality most of the patients the causes are not known. Advanced age is, obviously, the risk factor because the older you get more divisions your DNA goes through and more mistakes it makes. There's a little bit of predominance in male. Prior exposure to chemotherapy and radiation is one of the risk. There's association of the congenital disease like (inaudible 7:32) anemia, (inaudible 7:34) disease, aplastic anemia. There are some familial MDSs with certain mutations. When we look for them we usually advise for the family members to be tested as well and then environmental toxins can be also be associated. The defect in DNA repair and that tumor suppressive gene mutations.

So, there are in recent years with advances of science we now have capacity to sequence the DNA very quickly and not very expensively. So, with the whole (inaudible 8:11) sequencing we sequenced a lot of... nowadays we sequence patients and these groups sequence the healthy donor volunteers

and then we found that 10 percent of subject above age 65 have certain mutations, but they do not have any sign of disease and under the microscope they don't have any dysplasia either and this is called CHIP which is Clonal Hematopoiesis of Indeterminate Prognosis and it happens in the older patients and only one percent in the patients less than 65 and these are the most frequent mutations occurring in the CHIP TET2 and (inaudible 8:53) 3B1 and they found that the overall progression is 0.5 to 1 percent per year per (inaudible) to the MDS.

So, this is diagram just showing how a single gene mutations can link to the disease. So for example, you look here there's all the stem cells. None of them have the mutations and with the age if you develop some mutation you can make a CHIP. So with CHIP, you have certain mutations and your blood cells are formed from this particular clone. However, you don't have any problems clinically or under the microscope your marrow looks good. If you continue accumulating more and more mutations, you can further progress to the MDS and then additional more mutations later on it can be progressed to the leukemia.

So, if you look at the pathogenesis of MDS, so all these changes can lead to the Myelodysplastic Syndrome. The chromosomal changes, the gene mutations abnormally new system because your new system would be able to recognize abnormal clone and eradicate it. Abnormal regulation of DNA synthesis and also hostile environment in the bone marrow they all can lead to the formation of the disease.

So, majority of the patients who are diagnosed with MDS initially come without any symptoms. They go to see their primary care doctors. They do get the blood work done and then they look at the blood work CBC and they see that there is some sort cytopenia in one lineage or all three. So however, a fair number of patients come with the signs and symptoms. If you have anemia, patients come with the fatigue, latitude, they have shortness of breath, some of them have chest pain. If patient have a low white blood cell count they can have frequent infections mostly like sinusitis, bronchitis and it takes them longer to recover from cold than other family members. If they have low platelet count they can come with the bleeding or bruising.

So, when we diagnose patients with MDS we do a lot of tests. Physical exam, we look at the peripheral blood and look at the reticulocytes count which is basically baby red cells and by this we can tell how functional the bone marrow is and then we do another additional (inaudible 11:40) tests and that's why when you come to clinic we take 50 tubes of blood so we're able to do all this testing to rule out other causes of the cytopenia and then bone marrow biopsy and aspirate is essential to diagnose the MDS because we look at the bone marrow morphology, cellularity, how they look under the microscope and we also calculate the percentage of blood. We also look at the chromosomes and this can be done only from the bone marrow and then we look at the various iron stain and the reticulin stains as well.

So in order for us to diagnose and give diagnosis of MDS, there's a World Health Organization criteria which has to have either more than 10 percent dysplasia, 5 to 19 percent blasts, characteristic cytogenetic abnormality meaning your chromosome has to have certain changes and then we have to

have evidence of the clonality and we can either do it by flow (? 12:49) or by FISH. In using these we can establish the diagnosis and then subtype and prognosis.

So unfortunately, some of you are probably familiar with this procedure when we do the bone marrow biopsy it goes to the cortical bone and then will go to the bone marrow and we take aspirate and the core meaning the piece of the bone and we'll look at it under the microscope. So, this is the core which is actual bone part. We look at how much cells are there and it helps us to diagnose and aspirate helps us to look at the morphology of the cells and helps pathologists to look if there's any dysplasia.

So, diagnosis of MDS, it's quite complex as you can understand. It has to meet many criteria and we have to exclude other causes. So, we look at the cytopenia like we're looking at your just regular CBC. We look at the molecule biology if there's any mutations. We look at the morphology of the cells and we exclude other causes and the flow cytometry recently been merged as a very important diagnostic tools for us and the reason for that is that any cells who they have different markers on their surface. So for example when the blast cells which is your immature baby cells make mature to let's say, mature neutrophils, they express different markers and different proteins on their surface and we can detect them using flow cytometry and it's a very useful tool and as you can see at the different stage, different markers expressed in a different level. Like CD45 expressed throughout. CD13 for example is kind of dim on the blast level then it got very strong on promyelocyte then it also disappears on the myelocyte page and then it reappears in the neutrophil. So, using these markers we can tell exactly which stage of the depreciation those cells are and also we also can look at the co-expression of different markers and determine whether it's normal cells or abnormal. For example if we have CD45, but we don't have CD10, it's probably a normal blast. If we have expression of CD45 and then we have expression of CD10, then we can suspect that these cells are probably something abnormal going on.

And I know this is a very complex slide, but I just want to give you message how our pathologist look at the flow. This is blast to myeloid maturation and they just basically have a lot of experience and they look at the different stain and they kind of mentally draw this error. For example if the stain was CD33 and it show ADR the (inaudible 15:53) should be like this and if the arrow goes like this there's something abnormal with these cells.

So, more so than plus and minus, more important is the pattern recognition. For example if you look just plus and minus you can call this person as a human being. Yeah, it has legs, it has arm, it has mouth, it has eyes. However, you know that there's legs don't go well with the arms. There's decrease in the eyes. There's increase in the nails. So, you know that it's not a human being and this is kind of depicting how the pathologist diagnose MDS by the flow.

So, there are many other causes of cytopenia that can mimic MDS. That's why we draw so many blood tubes to check for other things. So, nutritional deficiencies like B12, folate, copper can mimic and on the microscope they can look exactly like MDS. So, when we see patients with cytopenia we always check there institutional deficit. Congenital conditions can also look like or mimic MDS. (Inaudible 17:06) can cause them. Other hematological conditions like aplastic anemia and PNH

often times hard to... especially moderate aplastic anemia looks very much like MDS. Autoimmune disease also might mimic various infectious disease, HIV, hepatitis, so we always check for those and some chronic disease can lead to changes in the iron kind of balance and other solid organ maladies.

So, for minimal diagnostic criteria for MDS, you have to have a constant cytopenia meaning your hemoglobin red blood cells or platelets or white blood cells have to be constantly low and we have to exclude other causes. Then MDS related criteria you have to have dysplasia, as I mentioned earlier, at least 10 percent of the cells or you have to have blasts or typical chromosomal abnormalities for MDS and then you have to have this coo-criteria. If you don't have this abnormal (inaudible 18:49), clonality by molecule testing with like looking at the various mutations and nobody does this anymore. It used to be done before when we looked at the colony formation.

So, here is just depicting the relationship to other disease. So, here's the MDS overlaps with many other diseases. So, it all progresses to the MDS... I'm sorry, AML. However, there is many diseases which is overlap. MDS with the MPN. There's a hypoplastic MDS and fair amount of the patients who have classic MDS also have MNH clone. There's LGL and 5Q is kind of separate (inaudible 18:59).

So, here is I just want to show you the concise history of the MDS how it was diagnosed. So, before 1982 we used to diagnose MDS by just clinical course. We would call it preleukemic stage and then it's refractory anemia. In 1982, the French American British classification came out and that was based on the marrow histology, how the marrow looks. Then IPSS score came out based on the molecular and clinical syndrome and a few years ago they revised this IPSS and then they made it a little bit more specific and I'll go a little bit more in detail.

So, this is French American British classification when they just based it on the clinical picture and cytopenias. So, they call it... divided them into refractory anemia, refractory anemia with the ring sideroblasts, refractory anemia with the excessive blasts. This is excessive blasts in transformation and CMML. So, kind of these two diseases were grouped as a low risk and the RAEB1 and RAEB2 was high risk and this showing the median survival of this patient without any treatment in mass. So, the low risk patient, obviously, the lived longer without any treatment and high risk they either progressed to AML or died from other causes.

And here the WHO also classified this and they see also put the number of blasts and this is just an overview how there are many different classification which we use to classify MDS, but I think the most commonly used nowadays is IPSS score revised and the reason we use this scoring system more often because it's more... It takes into consideration many factors. So, the marrow blast is very important. Karyotype and how it basically changes in your chromosomes. Hemoglobin level, platelets and the neutrophil. So, based on all this we give the score value and then we classify as low, very low, Intermediate 1, 2 and high risks.

So, here is the overview of the chromosomal changes which could appear and happen with MDS and based on what type of the chromosomal changes, we divide them into very good, good, intermediate,

poor and very poor. So, I mean, there's a little bit some debate going on based on mutational study, but this is what was in general used for us and using this we applied it into the previous table and we determine the risk of the patient.

So, why is it is important? Because if you look at this here's the very low, low, intermediate, high and very high. So, there is a very big difference between the median survival of this patient and this is survival without any treatment. So, if the patient is very low risk the median survival is 8.8 years which is very good. The low risk, 5.3, and in contrast the very high patients' median survival is only like 10, 9 – 10 months which is a dramatic difference between very low and high. That's why this scoring system is very important and this showing the total time to progression to AML and, again, here, you see the very high risk is only 0.7. So, like seven to eight months until they progress to AML.

Here is just the graph picture showing the difference between overall survival of this patient and one important thing it's only valid for the adult patient. There's a flaw with this scoring system and it's for (inaudible 23:11) diagnosis and the only treated with supportive care.

So, those numbers are there for us to guide us. However, always ask what does low risk mean, what does high risk mean and what does the age depend on that and what is the worsening blood count. So, the message I wanted to put here sometime we do get caught up in a number of blasts and what is it 10, percent, is it five percent, but we always look more at the trend more so than the actual number. So, let's say the diagnosis you have 10 percent blasts and that actual number is important, but more so than that we look at the trend. Is it going up very quickly or is it going down or staying stable? So, it's the overall dynamic picture is important, too.

This study, I think, was very important for me because it looked at the transfusion dependency. So, instead of putting all these numbers, this group just looked at the prognosis based on how many transfusions patients need. So if you look at this, the shaded one is the not transfusion dependent, this one never needed and the open one is transfusion dependent. So, this is a graph depending on the transfusion. So, the low and Intermediate 1, they only 22 percent were transfusion dependent and majority of them 43 and just occasional. It's not dependent which is kind of fits into this prognostic factor. So, if you are transfusion dependent, you most likely are going to be high risk and if you look at the high risk majority of the patients were transfusion dependent. So, just the simple kind of factors do influence in the prognosis.

Just a couple slides on the mutations because this is becoming more and more important nowadays. So, every patient who come to us will do the mutational study because there are a lot of mutations associated with MDS and here is the most common mutations. Here is the paper published in 2013. There, as you can see, there's a lot of mutations and the graphing out here the majority of the mutations which are common in MDS.

So, I think I'm running out of time. So, just to put the whole Myelodysplastic Syndrome putting the picture together, it's a heterogeneous disease. Some patients come with anemia, some patients come with the thrombocytopenia, but it's all like one spectrum of the disease. It's a clonal stem cell

disorder. It result in (inaudible 26:08) hematopoiesis. There is a lot of dysplastic features in the bone marrow and the results clinically in cytopenia. There is accumulation of blasts. They tend to progress to the AML. It's a disease of older patients and also we also look for the abnormal chromosomes. So, MDS is basically syndrome of the many different kind of symptoms and it's a disease of the bone marrow which results in these dysplastic features.

So, I think that's that and I'm happy to answer questions.

(Applause)

So if there is no questions, we'll move on.

**Daniel A. Pollyea, MD:** Okay. Do we need a break or should we keep going? Keep going. Can you guys hear me okay like this? Okay. Great.

Thank you, Enkh. That was great. Thanks, everybody, for coming today. This is just a great organization, the MDS Foundation that we're now fortunate enough to work with. Several years ago we were University of Colorado was recognized as a Center of Excellence by the MDS Foundation and we're very proud of that. We've worked very hard over the last several years to build a comprehensive MDS program here. You heard from Enkh. You're going to hear from Brett, one of the lead researchers at our institution on MDS in the laboratory effort. You're going to hear from John Gutman in a little bit about how he's leading stem cell transplant efforts in MDS and we're just really thrilled to be able to meet you guys and talk to you and try to move forward together with all the implications of this disease.

So, I'm going to talk... I've been asked to talk a little bit about treatments for MDS and in the course of this please feel free to interrupt me if you have questions. I'll, of course, have time for questions at the end as well, but if you have something you just think you're going to forget or want to talk about or talk in more detail about as we're going please just interrupt me and we'll get to your question and, again, thank you guys so much for coming out on a Saturday, giving up time away from your families and all of your other activities. We get that you are super, super motivated to learn more about this and we want to be able to help you in every way that we can with that mission of yours because we think that's really important.

So, some of what I'm going to say is a bit of an overlap with what Dr. Perv talked about and Brett and I were just talking there's probably some overlap between what I'm going to say and what he's going to say, but I think that's okay because some of these overlap things are really important themes and so probably it doesn't hurt to go through some of these things a few extra times.

So, what I want to talk about a little bit is the definition of MDS. Enkh did a great job on this, but in the context of how this interacts with or how this influences our treatment decisions I think is important to review as well as the categorization, how we risk stratify this disease, because all of that really informs the way we manage patients in the clinic with treatments and then at the end I left this very open ended, but I want to talk about the exciting things in the future that I think are coming and

I think this will be a good transition to what Brett his working on in the lab. We work with our lab colleagues for there's a lot of back and forth and the really, really cool interesting things that people like Brett identify we try to get into the clinic as quickly as possible. So, some of what I'm going to say may seem a bit discouraging and that has been the nature of treating MDS for a long time, but I am very excited about what's just over the horizon in MDS and we have real reason to be very, very enthusiastic and excited about what's coming just around the corner. So, I'm going to talk about that at the end, but the balance of the talk until we get there is mostly, frankly, a history lesson, but that's still sort of what constrains what we're doing, but don't get too down about it.

Okay. So, what is... where does MDS live in the spectrum of other myeloid malignancies? So, that's what MDS is. It's a myeloid cancer. There's a myeloid compartment of your immune system that is a normal functioning part of your blood development system that Enkh talked about and you could have cancers in that compartment of your blood, hematopoietic or cell development system and if you have an acute leukemia or an acute myeloid malignancy, that's AML, and then there are chronic myeloid malignancies or cancers. Myelodysplastic Syndromes are the biggest group member there. You also have these conditions called myeloproliferative neoplasms, which we're not going to talk about today, but Myelodysplastic Syndromes just like these other myeloproliferative disorders can evolve into an acute myeloid leukemia. So, acute myeloid leukemia can be like the final common pathway for a lot of patients with any myeloid cancer and that includes Myelodysplastic Syndrome and one of the big themes in terms of how we think about treating patients with MDS is when you're first diagnosed how likely are you to be someone who moves along this spectrum to AML? If you have a high likelihood of that happening then we take a more aggressive approach. If you have a lower likelihood of that happening, there are some people with MDS whose MDS just sort of simmers and smolders along for many, many months or even years and we like to know that, too. If you're not likely to evolve to AML that informs our treatment.

So, as Enkh told you, Myelodysplastic Syndrome, this is a clonal or cancerous bone marrow condition or neoplasm is a cancer. It's characterized by ineffective hematopoiesis. So, she showed you the blood development system isn't working well. It isn't working properly and this results in bone marrow failure. The bone marrow doesn't work. You can't make all the normal cells that should encompass your blood and you have all kinds of symptoms and consequences of that. Under a microscope, a pathologist will tell you there's evidence of dysplasia or immaturity. The cells don't look mature. They don't... aren't able to mature to their full potential and, of course, there's this tendency to progress to AML.

This slide here from the Leukemia and Lymphoma Society, they put this together and I think it's really useful. These bubbles each represent a blood cancer and the size of the bubble is reflective of how common the blood cancer is. This axis here tells you the five year relative survival rate of each of these conditions and this axis here tells you the age at diagnosis of each patient. So, a lot of information on this slide. So, you can see that, for instance, Hodgkin Lymphoma is quite curable. Non-Hodgkin Lymphoma is quite common and there's a big difference between AML if you're younger versus... or sorry, if you're younger versus older with respect to outcomes, five year survival. MDS sits here. We're not good at treating MDS as you can see by this. It's a disease mostly of older patients and it's really second only to AML in terms of how poorly patients can do. So, the



WHO as Enkh explained is the main sort of characteristic or diagnostic system that our pathologists use to classify and subclassify MDS. So, MDS when we talk about it it's plural, Myelodysplastic Syndromes and this is why it's plural because there's all these different subtypes and it's not too important today to go through all of these, but just know that we and the pathologists have a lot of different criteria to determine which subtype of MDS a patient is when they're diagnosed and subtypes can go... fall into low grade and high grade. And so that's sort of rough cut point as to how likely patients are to evolve to acute leukemia and how unlikely that is to happen based on where you fall in which of those subtypes.

So, from a prognostic standpoint as Enkh explained, the main tool to use is this IPSS score which has been updated in 2012 to the Revised IPSS score. Just know that there are other prognostic tools that people use and they have some strengths and weaknesses, but for the most part we're using the IPSS score. This is valid at the time of diagnosis. So, if you walk into a doctor's office for a second opinion three years into your diagnosis after having been on Azacitidine or something like that this isn't a valid tool anymore. So, this is really only informative at the time of diagnosis.

And so this is one area that we're going to go back over a little bit even though Enkh introduced it because it is so important in our treatment decision making. What are the features that go into calculating an IPSS score? The cytogenetic risk. So, when we do a bone marrow biopsy, we look at the chromosomes of your disease cells because we've learned over the decades in dealing with MDS that very frequently people have abnormalities in the chromosomes. So, what are chromosomes? Chromosomes you can think of like the houses where all the genes of your cells live so that the genes you have all these thousands of genes that do all different things and give the cell all these different signals. They have to live somewhere and they live on the chromosomes and so on a certain region of each chromosome you can find each of these genes. Over the years, we've learned that there are certain structural abnormalities that can occur in these chromosomes. So in the disease cells, sometimes there's chromosomes that are missing. Sometimes there are extra copies of the chromosomes. Sometimes there are switches that happen between two chromosomes. They trade places. Those are called translocations. All of those can have some prognostic importance in risk assessing a person with MDS and that's very, very important to understand and it's crucial to understand at the time of diagnosis. So, that's when this needs to be done because after that point it's questionable how prognostic those chromosome abnormalities may be.

We then look at how many blasts or very immature cells are in the bone marrow. So, we have a very arbitrary system where we say if you're less than 20 percent blasts, we call you MDS, but the second you hit 20 percent or higher then we say you have AML. Obviously, this is a very human criteria. The disease doesn't care. It's not different when it crosses that imaginary threshold at 20 percent, but we say you have AML at that point, but clearly if you have 18 percent blasts even though we call you MDS, you have the same biology as someone with 21 percent blasts, but we need a cut point to make that distinction and it's 20 percent has been chosen, but the more blasts you have those are the really immature cells, the closer you are to AML, the worse the prognosis and so that's why the number of blasts plays into this prognostic scoring system and then how anemic are you? How low are your platelets, how low are your neutrophils? All of those play into this prognostic system.

And the reason this is important is these graphs that we look at all the time are called survival curves and so the slope of the curve gives you a sense of how rapidly a patient gets to this end point. So, this endpoint here is overall survival, how long are people living. So, in the bright red curve here, people aren't living very long for the most part. The average here is about a year or less, but you look at the average of this one, this light blue curve, and it's more like 10 years is the average. So, it's a huge difference in survival for one disease. I mean, that's a huge variation. Same disease, but some people live only a year. Some people live 10 years and so you can make this prediction with some degree of accuracy based on this IPSS scoring system. That's why it's so important that we classify patients.

Now, this is the IPSS is built on these features that we could recognize historically, but now we know as Enkh introduced there are all these genes that get mutated in people with MDS. So, we didn't appreciate this until just a few years ago. We knew that it must be happening, but we didn't have the sophistication to know what genes to look for, which ones were commonly mutated and what all that meant and this has changed very quickly just the last three – four years where we now know which genes are commonly mutated in in MDS and therefore by understanding those which genes are mutated we can have a real appreciation for the differences in this disease between people. So, one person may have a disease, MDS, with three or four genes mutated. Someone else may have MDS, but not have those genes mutated and have a different three or four genes mutated and those differences are really important. They're important for prognostic purposes and they're also important for making treatment decisions and as what Brett's going to touch on in a little bit those are some of the angles, some of the advantages that we are trying to use to be able more effectively target this disease in more patients by understanding the unique features of everyone's disease maybe we can have a particular targeted therapy for that particular patient and so that's where this is going. So, this slide sort of gets at some of the complexity of the disease. So, each one of these bubbles represents a different gene. They're group in according to different gene families, target kinase pathways, splicing factors. You don't need to know all of that, but the size of each bubble is represented of how common these gene mutations are. So, some of them are quite common. Enkh touched on TET2, DMN3TA, SF3B1. Those are pretty common. Some of them are really rare, UTAF2, NPM. So, these are the things that are helping us understand our large scale how this disease works like what's under the hood of this disease and then the next logical step because we're only in our infancy of this, but the next logical step is okay. How do we use this information to have better treatment? So, that's where this is all headed.

So, clearly, the future this is also going to impact prognostication. So, if you have a whole region of a chromosome that's missing in a patient, we might assume that that's bad or we found that's associated with a worse prognosis but we can get even more specific, hopefully, in the future where a person who doesn't have any gene... chromosomal abnormalities which is about 50 percent of people with MDS. If they have particular gene mutations then we can be much more subtle about assigning risk for development of AML or lack of response to conventional chemotherapy, etc., etc. So, this is really going to help us in the very near future prognosticate better.

So, let's with that preamble how do we actually manage MDS? First, let me tell you what happens to patients with MDS and this is based on large numbers, so it's impossible to apply this to an individual patient. We use this information like large scale to sort of think about this disease in big

general terms, but trying to put you, one patient, on this scale, that's not how you show to try to do this, but at the time diagnosis if you follow the patients, about 50 percent of patients are going to die from some complication of having bone marrow failure. So, an infectious complication, a bleeding complication, something like that. Thirty percent of patients will progress to acute leukemia and then 20 percent of patients don't have any sort of cause of death related to their MDS. So, they're going to die of what people die from – heart attacks, stroke, diabetes, getting hit by a bus, all of those things. In 20 percent they're not going to have any cause of death related to their MDS and just to take a pause here, this is why it's so important to know if you're the doctor treating the patient and you're seeing someone for the first time are they going to be in this 20 percent where the best thing I can do is just leave you alone and not do anything because this is not going to be a problem limiting your life or do we need to be real aggressive to try to prevent progression to AML. So I think, again, this is why it's so important. This is the same disease, but the outcomes are so different you have to really take time and carefully consider where a patient is on this spectrum.

So, what do we use for patients with MDS? So, what's FDA approved. So, we have hypomethylating agents. You may hear doctors or other people refer to these sometimes as DNA methyltransferase inhibitors or epigenetic drugs. All that means the same thing. There are two of these – Azacitidine whose brand name is Vidaza approved in 2004 in the United States and Decitabine or Dacogen which was approved in 2006 in the United States. There's an immunomodulatory drug we're going to talk about. Lenalidomide also known as Revlimid then there are iron chelators, which we'll talk about a little bit which are used for supportive care. We also use a lot of things off label in MDS. So, this is it. This is all we have for all the people with MDS. This is all the FDA has given us and so there are some drugs we use off label. We use a growth factor support. So, sometimes we use Neupogen, red blood cell growth factor support, Epogen or Darbepoetin. Now, there are these platelet growth factors which some of us are using off label in some circumstances. Immunosuppressive therapies. You remember that slide Enkh showed you? There's some overlap between MDS and aplastic anemia and the treatments for aplastic anemia is immunosuppressive therapies and in so in those rare cases where there's MDS overlap with aplastic anemia we'll use immunosuppressive therapy. I wasn't planning to get too much into this unless people had questions and then, of course, chemotherapy is sometimes appropriate, more intensive chemotherapy or stem cell transplant which Dr. Gutman is going to talk about.

So, first let's talk about this subtype of MDS. It's pretty uncommon, but it's important to understand. It's called deletion 5Q MDS. These are patients with low risk features, so no increase in blasts. They have a very severe anemia at the time of diagnosis, but unlike most MDS patients who have anemia and low platelets and a low white blood cell count for reasons that aren't quite understood these patients often have a normal to even elevated platelet count at diagnosis. They have one chromosomal abnormality, deletion 5Q. Now, that used to be how we classified it, but in April the WHO met again and they allowed one additional chromosomal abnormality to be present as long as it's not an abnormality involving chromosome 7. Don't worry about these details, but this is the definition of deletion 5Q MDS. The reason it's so important to know and understand this specific subtype is because the FDA has approved a treatment, Revlimid or Lenalidomide, for patients with this entity and it is a dramatic... it has dramatic results in people with deletion 5Q MDS. If you have that subtype, this is a great treatment. It can result in deep responses and it's fairly well tolerated and

those responses can last a long time. They're not cures. This isn't a cure, but these are very effective treatments for that subtype. Now, we have come to understand that not all people with deletion 5Q MDS respond great to the Lenalidomide. Most do, but some don't and now with some of this gene sequencing techniques we can have an explanation for some of those patients and not to get too into the weeds here, but if you have a p53 mutation which is actually quite common in people with deletion 5Q MDS you may not have as good a response rate to the Revlimid or the Lenalidomide as you otherwise would have and so that's something important that we've only recently appreciated, but very important to know the status of P53 in a patient with deletion 5Q MDS. What about using Lenalidomide in people without deletion 5Q MDS? Well, it doesn't work that great. So, we've had a couple studies now come out on this and the consensus is 25 percent. About 25 percent of people without deletion 5Q MDS will be expected to respond. So, it's an option, but not necessarily a great one.

Okay. Azacitidine or Vidaza. This is FDA approved for people with higher risk MDS based on... mostly based on this study or this was a post approval study that solidified the advantage here and this is another survival curve. So, here in green are the patients who got Azacitidine. Again, higher risk patients got Azacitidine and then here in red are the patients who did not get Azacitidine. They got either just supportive care like transfusions. Some of them got chemotherapy. Some of them got low dose Cytarabine, but anything but Azacitidine and you can see based on this curve the average survival for patients on the Azacitidine was about two years which was statistically better than patients without Azacitidine which is about 15 months. So based on this, Azacitidine was approved by the FDA for use in higher risk MDS patients and this is why we often use it.

What about Decitabine or Dacogen? So, there's an interesting story here. We often think of these two drugs as like Coke and Pepsi. I mean, the Azacitidine, Decitabine, they kind of do the same thing, they're kind of cousins and I think that's for the most part true, but even though Decitabine is FDA approved, it doesn't have quite the robust data that Azacitidine does for higher risk MDS patients. So, there was a preliminary study that showed a survival benefit for using Decitabine in higher risk MDA patients, but that was like what we call a subset analysis. So, they did a big study and then they picked out the high risk patients. The big study didn't show a survival benefit, but when they picked out the high risk patients all of a sudden that did show a survival benefit. Now, you may think that sounds fine, but statisticians don't like that. They don't think that that's a fair assessment and so they were asked to repeat the study with just high risk patients and when they did that there was no longer a statistically significant difference in survival between patients who got Decitabine and patients who didn't, who got supportive care. So despite that, it's still FDA approved because there is a lot of use of Decitabine, but on paper a lot of us MDS doctors will not... we'll prefer Azacitidine over Decitabine for this reason because Azacitidine is the one where there's a proven survival benefit. Decitabine, it's a little more murky. In low risk disease patients which we're going to get to in a minute, I think there's evidence for either one is fine, Azacitidine, Decitabine fine. High risk patients little bit of a preference for Azacitidine, but other doctors may have other interpretations of this.

**Q1:** (inaudible 52:39)

**Daniel A. Pollyea, MD:** Thank you. That's a great question. So, Azacitidine can be given subcu under the skin or as an IV infusion. Both of those methods are equivalent. So, there's no advantage of one over the other. Some people prefer the IV, some people prefer the subcu. Decitabine, only IV. Now, there is an oral pill, a form of Azacitidine that's in development right now. It's in the clinical trials sort of world and there's various news on its progression at all the meetings and how it's looking. So, but that may be something that will be available at some point.

So, I already talked about which hypomethylator to use and why. Okay.

So, what can we expect? So, this is an FDA approved therapy. What can we expect? So, there's a survival benefit as I showed you nine months and so with the use of Azacitidine. A hypomethylator can delay progression to AML which is a very good endpoint, a time point. Unfortunately, the response rates aren't very great with hypomethylators. So, what I tell people is at best we can expect about a 20 percent rate of a really clinically significant or meaningful response rate. We're not curing anybody with this approach and we'll come back if a person does respond and that's the only treatment they're getting it, it will at some point come back. There's about 30 to 60 percent of patient will have maybe not like a remission where you don't see evidence of the disease anymore in the bone marrow, but they need fewer transfusions and they feel better. So, that's good and that's what this shows here. Patient reported outcomes or quality of life outcomes improve with hypomethylators and but so what do we do when you use a hypomethylator and then it stops working? This is a problem. This is the problem. So, people don't do well in that scenario. Usually, the progression comes pretty fast once you stop responding to one of the hypomethylators. You can switch to the other hypomethylator. So, go from Azacitidine to Decitabine or the other direction. That usually doesn't work real well because they are so similar and once the disease sort of figures out how to get around one drug then it's going to sort of not have a problem with the other in most cases.

So, what do we do in that situation? I mean, it's our preference any MDS or leukemia doctor that you see in that scenario they're going to recommend a clinical trial and because the outcomes just aren't very good with what the FDA has given us and so a clinical trial should be the first consideration with the thinking, I guess, that something even if it's unproven must be better than what we have in our arsenal and that's one way to think about it but, again, another way to think about it is some of these things that are coming for MDS are really exciting and so this is a way to get access to those things before they're going to be FDA approved.

So, let me just kind of review. This is the final slide then we can take questions. A summary of how we approach MDS at our institution. So, everybody gets supportive care. That means transfusions, that means antibiotics, either prophylactic antibiotics or when they get sick, treating an infection. We didn't really talk about iron chelation. This can be something if someone's interested we can talk about more in the question period or come up to me, but iron chelation is a process to remove excess iron from a person's blood. Excess iron can get there mostly in MDS patients through blood transfusions. So, one transfusion, one unit of transfused blood gives a person about a year's supply of iron and your body is sort of built to retain iron and not excrete it because it's something that wasn't commonly available in our ancestors as cavemen. Getting access to like meat was hard and that's mostly where we get iron and so we're built we've evolved to not to try to retain all of our iron. So,

in the modern day when people are eating lots of meat and they get, in our case, potentially multiple blood transfusions that can be a problem because you don't have a good way to clear your iron and it can deposit into the liver, into the heart and in some case cause some real problems. Now, we used to chelate. Chelation is a process where you pull out the iron from the body and that... there's a couple ways to do that. Most of those ways have a lot of side effects. For the most part it's not an issue. So even if you do have... most people are going to have high iron levels because they're getting transfusions, but it's not bothering them. So, in almost all cases we don't chelate patients and I think I've been in this at the University of Colorado for five years treated lots and lots of MDS patients and I can think of one or two patients that I've ever recommended chelation for. So, we almost never actually need to do it, but it's something that some people still do a lot of and people have a lot of questions about. But then when you talk about lower risk MDS, this is based on that IPSS score and other different scoring systems. If you just have anemia and a deletion 5Q then use Lenalidomide. That's deletion 5Q MDS. If you have anemia, no deletion 5Q and a low erythropoietin level you may be a good candidate for an erythroid stimulating agent like Darbepoetin or Epoetin and maybe even some Neupogen or DCSF there. If you don't have deletion 5Q and you have a high erythropoietin level which is in most cases what patients have. It's pretty uncommon to have a low erythropoietin level. That's when we would recommend a hypomethylator, Azacitidine, Decitabine. If you have anemia and one other cytopenia, one other low cell count then we just recommend a hypomethylator and then in high risk patients the first question to ask and John's going to tell you why is are they a transplant candidate because MDS could be curable in those scenarios and so if that's the case try to get to a transplant. That's really the goal because you can go from whatever prognostic scoring system you want to talk about to potentially curative situation. If they're not a transplant candidate, hypomethylator and my preference is Azacitidine for the reasons that I told you.

Okay. The last few minutes. Some of the promising investigational therapies that I think are coming around the bend. So, using CD33 directed antibodies is really looks good. This has been a successful strategy in clinical trials for AML. We've now applied to high risk MDS. This study has just gotten underway. First cohort or two of patients, really looks promising. I'm excited about what's going to happen there. Venetoclax is sort of the same story. This is an inhibitor of something called BCL2, high success rate in AML and based on that we're launching a study in MDS. That should be coming starting in 2017, very excited about that. Immunotherapies. Everyone's excited about checkpoint inhibitors. A lot of this is going on in solid tumors. You heard some of these dramatic stories. President Carter, other people have had these dramatic responses to some of these checkpoint inhibitors. Could they work in MDS? We're going to see. Those trials are ongoing. Potentially very exciting. We're working a lot on directing therapies towards something called CD123 which is a marker of MDS stem cells. Brett may have some discussion about that and that's another really exciting potential future area.

So with that, that's all I had, but we have a couple minutes and very happy to take questions. Yes, please.

**Q2:** About your fifth or sixth slide you had the first set of survival graphs. Are those among treated patients or untreated patients?

**Daniel A. Pollyea, MD:** Those were untreated patients, the newly diagnosed untreated patients.

**Q2:** And to show... and so later on you had a couple that showed some success with treatments comparing treatments but not of that major... that original set of the population. Why is that? The first brief also showed untreated survival rates were, I think, the same numbers that you had.

**Daniel A. Pollyea, MD:** I'm not sure I understood... The slide at the beginning showed survival by prognostic group and that's in untreated patients and then that Azacitidine study showed that survival for Azacitidine was better than not getting Azacitidine. Say your question again. I'm sorry.

**Q2:** I guess I'm wondering the only survival charts that we saw that were among treated patients were later in the briefing and I believe it was with the one... was it Decitabine, I believe it was, drug, but not survival rates of a broader population of the patients with other treatments.

**Daniel A. Pollyea, MD:** Yeah. So, other treatments besides Azacitidine and Decitabine the only really other treatment we have is the Lenalidomide for the deletion 5Q patients. I didn't show you survival curves there, but they look really good compared to the deletion 5Q patient got Lenalidomide versus not got it, much better, much improved there. The survival curve for Decitabine in the targeted study where they did the high risk patient population and looked at Decitabine, there wasn't actually survival advantage to the other arm where patients didn't get Decitabine and so for that reason there's a controversy as to whether Decitabine should be our first line treatment.

**Q2:** Thank you. And one other, will the briefings be posted on the web somewhere?

**Daniel A. Pollyea, MD:** The slides?

**Q2:** Will the slides be available to us?

**Daniel A. Pollyea, MD:** That's a good question for these guys. I'm happy to share them.

**Speaker:** We'll put it on.

**Daniel A. Pollyea, MD:** Perfect.

**Speaker:** (inaudible 1:03:15)

**Daniel A. Pollyea, MD:** Good.

**Q2:** Thank you.

**Daniel A. Pollyea, MD:** Yeah. Yes, sir?

**Q3:** Are periodic bone marrow aspirates, aspirate processes, the only way to really determine the progression of blasts or what does...?

**Daniel A. Pollyea, MD:** It's a great question. So, we try to infer as much as we can from the bloodwork to get a sense of how a person's responding to a treatment, but it's like reading the tea leaves. The gold standard is the bone marrow, but yeah, I mean if a person goes on Azacitidine and after two cycles their platelets normalize and their red blood cell counts close to normal, without doing a bone marrow we can't say for sure, but we know the bone marrow is going to look better. So, we can read the tea leaves a little bit with the peripheral blood work, but the only way to really know what's happening is...

**Q3:** Are there any markers that in your monthly CBCs that would suggest (inaudible)

**Daniel A. Pollyea, MD:** So, great question. So, yeah. I mean, we key in the platelets, those and the red blood cells. The problem there is that these treatments like Vidaza or Decitabine they're toxic to the normal blood cells, too. So, it can be hard to tease out is a person responding and you just can't see it because the toxicity of the drug is limiting or is it are they not responding. I mean, that's always the sort of question.

**Q3:** And what if you're not on anything?

**Daniel A. Pollyea, MD:** If you're not getting any treatment then, yeah, using... need for transfusions and degree of cytopenia at the interval visits that's why we keep bringing you in every couple months or whatever we do to try and get those trends.

**Q3:** Thank you.

**Daniel A. Pollyea, MD:** Yes, ma'am.

**Q4:** So my name is (Attendee) and I think I have a question I can stump you with.

**Daniel A. Pollyea, MD:** It's not going to be that hard.

**Q4:** (inaudible 1:05:01)

**Daniel A. Pollyea, MD:** Say again?

**Q4:** (inaudible 1:05:03) my body makes good red blood cells and then it...

**Daniel A. Pollyea, MD:** Say the first part again?

**Q4:** Immediately I (inaudible 1:05:11) the test. So, I make good red blood cells and (inaudible 1:05:19).

**Daniel A. Pollyea, MD:** Okay.



**Q4:** So maybe I could talk to you later.

**Daniel A. Pollyea, MD:** That'd be great. Let's do that.

**Q4:** They don't like me. (Inaudible 1:05:31)

**Daniel A. Pollyea, MD:** So, you're talking about an autoimmune hemolytic anemia.

**Q4:** Yeah and I just thought...

**Daniel A. Pollyea, MD:** Yeah. Let's talk about that. Great. Thank you. Yes, ma'am.

**Q5:** If you start out as a low risk MDS patient is there a protocol that they would follow that they would naturally go to first blood transfusion then maybe Vidaza?

**Daniel A. Pollyea, MD:** Sure.

**Q5:** Or can they skip... Can they jump the transfusion go straight to Vidaza?

**Daniel A. Pollyea, MD:** So before in a low risk setting and I should have clarified this a little bit better. A low risk patient not needing transfusions can just be observed and there's really no benefit to introducing therapy in setting. In fact, that will likely make things worse because you'll take a person who's not needing transfusions yet and you'll make them need transfusions because of the treatment itself. So, a low risk patient not needing transfusions, you just watch them. Now as soon as they start to need transfusions that's when we usually introduce treatment with a hypomethylator to help minimize the number of transfusions or maybe even make them independent of transfusions again.

**Q5:** So, would there ever be a case when you don't have transfusions at all?

**Daniel A. Pollyea, MD:** Sure. So, some people they have MDS, they have mildly low blood counts, but they never need a transfusion and they go their whole life like that. They can go decades eventually.

**Q5:** But then what if a doctor suggested you jump and go start having chemotherapy?

**Daniel A. Pollyea, MD:** I would say I want a second opinion.

**Q5:** Okay.

**Q6:** I'm the patient.

**Q5:** This is my sister and that's what they did with her. So, I'm kind of...

**Daniel A. Pollyea, MD:** That's not the approach that I think most MDS doctors will take. Unlike like lung cancer where oh, you have it, you need to do something about it, that's not the case for this. If just the presence of the disease does not automatically warrant treatment. The presence of the disease plus some sort of symptom or sign or problem is what warrants treatment because like what I showed you 20 percent of people with MDS live their whole natural life and never have a problem.

**Q5:** Well, would they see something maybe in her blood or bone marrow aspirate that are seeing we need to start Vidaza?

**Daniel A. Pollyea, MD:** There's some scenarios like that that you could consider, but for the most part if you just pay attention to the patient in the specific scenario, but yeah.

**Q6:** Well, my red blood cell count was 7.8 for quite a while for like three or four months, so that's why they decided to start. That's kind of low.

**Daniel A. Pollyea, MD:** That's pretty low. Yeah.

**Q6:** And I've gotten up to a 10.4.

**Daniel A. Pollyea, MD:** With treatment?

**Q6:** With treatment.

**Daniel A. Pollyea, MD:** Great. Good for you.

**Q6:** With four different sessions.

**Q5:** We were just confused why they didn't try transfusions versus straight...

**Daniel A. Pollyea, MD:** Well, once you need transfusions, I mean, you could do that...

**Q5:** Once you start them...

**Daniel A. Pollyea, MD:** So, that's a good question. There's a gray zone where okay, this patient only needs one transfusion every other month then I'd rather do that than take chemotherapy personally. So, I think that's a gray zone that is acceptable.

**Q6:** Then would a bone marrow transplant be advisable after chemo if your blood cell count is going up?

**Daniel A. Pollyea, MD:** After chemo if your blood cell count's going up that means your bone marrow looks better or you responded. From the outset you had high risk disease features that from

the very beginning someone said, look, we got to get you to transplant then that's the time to do it exactly when you start...

**Q6:** That would be the time to do it?

**Daniel A. Pollyea, MD:** Yeah.

**Q6:** Is there other factors that points to a good decision in that way?

**Daniel A. Pollyea, MD:** Yeah, there would be a lot of other factors. Yes. Yeah.

**Q5:** And they'll probably address more of that with the bone marrow transplant.

**Daniel A. Pollyea, MD:** Yes. I think I'm eating in his time. He's going to yell.

**Q7:** (inaudible 1:09:09) about treatment. You've all been focused on a single drug. What about combination of Procrit and Neupogen and Vidaza?

**Daniel A. Pollyea, MD:** So, we don't usually use Neupogen with Vidaza for a variety of reasons. When you have active disease that would warrant the use of Vidaza, theoretically Neupogen could stimulate the disease cells to evolve to acute leukemia. So, we try to stay away from using Neupogen in more high risk settings. Combinations of other active drugs that's a very, very good question and so combining Azacitidine with some of those novel therapies that I put up there, the CD33 inhibitor, the BCL2 inhibitor that's all ongoing in clinical trials.

I've officially eaten into the next person. I'm sorry. Thank you, guys.

(Applause)

**Brett Stevens, PhD:** I'll get started as long as nobody else had any questions for Dan. Just like to thank the MDS Foundation, again, for putting this on and hopefully you as patients will walk away with this with a good outlook on what we're doing in terms of MDS and MDS research. So as Dan alluded to, I'm on the research side, the lab side, of our leukemia services group and so I'm going to talk a little bit about what we do with MDS patient bone marrow and some of the future therapies that Dan alluded to. So, at the beginning there'll be a little bit of overlap to what both Enkh and Dan talked about, but I'm going to put a little bit of a different spin and stress on it.

So, Myelodysplastic Syndrome. Again, you've seen this curve now three times where you split the disease into multiple risk stratifications and sort of for a lot of reasons which I think you appreciate because of the survival, we concentrate solely on the very high and high risk disease on the research side because this is the disease that has the fewest options in terms of treatment and also carries with it the greatest progression to AML and so I balance my research between acute myelogenous leukemia and MDS and try to apply a lot of what we learned from AML back to the MDS disease

and so, again, high risk MDS has a significantly elevated risk progression to AML leading to the poor survival sometimes less than a year.

And so this is something else and I'll flip on its side. This is essentially what's happening in normal blood and bone marrow as we walk around and so this is a lineage tree where you start with the stem cell that gives rise to multiple progenitor cells and then ultimately the mature cells that carry out function in our blood and bone marrow and so the reason I put this up is because this is in the normal system happened in a very coordinated fashion. We know a lot about each one of these cells, what makes them different from other cells and sort of what makes them grow and survive. What's happening in MDS is that you have some sort of DNA damage or mutation as Dan alluded to in one of those 50 or so genes that you saw, cytogenetic abnormalities. You have some sort of mutational event that leads to what we think is an MDS stem cell and so when I say MDS stem cell, this is actually a subpopulation of the disease. It's a very small population of the total MDS space and that cell has the ability to give rise to the blast cells which is what when you walk into the clinic you hear you have X percentage of blast cells. We predominately study the cell that gives the rise or leads to the growth of these blast cells and so sort of the other path that we look at in the disease and on the research side is this progression from a normal person through low risk to high risk to de novo AML and then relapse refractory AML and so on this progression these cells become increasingly different and diverse and also they become increasingly more difficult to eradicate and so, again, we're primarily concerned with this high risk MDS patient population and preventing this progression to frank leukemia.

And so the questions that we ask of the research side is what makes these MDS stem cells different from a normal stem cell and what makes them different from an AML stem cell and ultimately can we exploit the differences for therapeutic benefit and so we know a great deal about normal stem cells and we know increasingly more about AML and the leukemic stem cells and frankly, we know very, very little about MDS stem cells. It's a fairly understudied area at this point, but we're sort of learning more every year and so we more or less ask the question of is a primitive MDS stem cell acquiring properties of AML stem cells and the consequence of that being progression to frank leukemia and ultimately how can we target those cells and so essentially the question we ask and that our group asks is what makes an MDS stem cell different from a normal cell and then sort of the sub-questions off of that is what characteristics define stem cell. Are there different clearly activities that are hijacked by an MDS stem cell and then ultimately what potentially therapeutic targets can we use to prevent disease progression and sort of how we study that. One of the major tools that we use Enkh alluded to briefly is taking bone marrow cell preps and looking at their characteristics via flow cytometry and so sort of the workflow for that as some of you may know is a bone marrow aspiration. We take that bone marrow aspiration. We then take it. It looks like whole blood and we split it up into each one of those compartments so that initial diagram I showed with all the different mature cells, you've got your red cells, your white cells and your plasma. We then focus our attention here in the middle on these white cells because that's where the blast cells and stem cells are. We then apply antibodies to these cells and these antibodies essentially recognize the characteristics of the cell. So, think of characteristics like eye color or hair color or height. We essentially look at all the different characteristics of the cell via what we call antibodies and so we add those antibodies to the cell. Some of them bind, some of them don't and then whether they are

present or absent is what we study and so we then take those cells, we pass them through a laser. This laser based on the chemical properties of these antibodies excites the cells or does not. We collect that excitation or that signal and then we use software to plot its presence or absence and so Enkh very briefly put a plot up, but I'm just going to sort of give you a brief explanation of what these plots actually show. So, you have an axis here and an axis here and the way that we look at these plots is as you move out on the axis to the right or up is increasing signal or presence of that particular characteristics. So, you can think of it as presence of, for instance, blue eyes and blond hair and then anything over here would mean that you would have blue eyes and blond hair and so we're able to now do this with multiple different characteristics and start to better understand the disease and so at the very heart of this is what separates an MDS stem cell from a normal stem cell and does it have any overlap with leukemic stem cells or acute myelogenous leukemia stem cells and so here's an example of a cell and you can see there's a bunch of different shapes attached to it. Each one of these is a unique characteristic to that cell and so we ask the question of what makes these any different from a normal stem cell and one of the things that comes out is in high risk disease specifically contrary to a risk and normal disease is a protein called CD123. So, you can imagine, for instance, that it's all these green shapes here attached to the cell. Is CD123 a gene and a protein attached to the cells and so, again, you have your close cytometry plots where you can see that the pattern differs between low risk and high risk and the presence of this in the red box here signifies the presence of CD123 and so we've characterized these CD123 positive cells extensively in the lab and it's led to a couple different discoveries and, again, I want to reiterate that what we're looking at is preventing now this progression to AML because these cells we know also express CD123 while these do not and so that leads us to believe that it's integral to the progression of the disease and so we have our CD123 now as a handle, so we can pick it out. We can put it into a box now, for instance. We can say these are all the cells with CD123. These are all the cells with blue eyes, for instance, and then again we now have the ability because we have a hemo to target these cells.

So, we can specifically ask these are the cells that cause progression to AML can we kill them and how can we kill them and so we're doing that with sort of three methods at this point and I'm going to give you the first two which are probably the closest to the clinic and then I'll finish off with a third method which is a little bit further from the clinic and a little bit more preliminary and so two of the most positive and promising targets, I think, in MDS at this point and this is work that I do in collaboration with Dan is the BCL2 inhibitor which he alluded to, Venetoclax, and I'll show you on the next slide sort of what it's doing specifically to these MDS stem cells and the second being protein translation inhibitor almost attacks it which at this point is an approved therapy in CML for patients that don't necessarily respond to other drugs and so we're applying these either approved or clinical trial agents in AML now to MDS to see whether or not we can effectively target the cells that lead to progression of the disease and so here's some work that we've done here in Colorado as well as at other sites throughout the country showing what Venetoclax is specifically doing in AML patients and so I'll just draw your attention down here. These numbers in brackets are the response rate. So, and so as Dan alluded to Venetoclax in AML has an amazing response rate in the upfront patient setting where you're seeing a response rates upwards of 80 percent in the preliminary studies. Omacetaxine which is the second therapy that I alluded to the response rates and I apologize for the size of this. The response rates are not nearly as good as the Venetoclax, but to some degree we're still seeing in various studies response rates of 30 to 50 percent which is actually it's strong in AML

and so if we back this back to MDS and apply this to MDS patient samples what are we doing to the CD123, the MDS stem cells that, again, caused progression of the disease to frank leukemia.

So, the blue bars are those cells that cause progression and the MDS stem cells and this is just a measure of how alive the cells are, again, via flow cytometry that technique that I talked about earlier and so as you can see another cell population within this MDS bone marrow sample under the addition of Venetoclax shows a very little response. Contrary to that in the blue bars where you see these are half or as much as 90 percent less cells. So, this drug is specifically killing these cells. Omacetaxine, the second drug I alluded to, again, we see significant death of those MDS stem cells, again, and it's fairly specific to these cells suggesting that we may not be killing the other cells in the marrow and the blood and so while all that happens in a plastic dish it's not very relevant to a true patient model and so the way that we sort of get around that outside of our clinical practice is we use mice. These mice have the specific immune system that allows us to take human cells and grow them in the mice and so, again, what you end up with is the ability to look at the mouse cells which are seen here on the right side of this plot and the human cells which are seen up here. So, we can now start to ask specific questions. If we give this mouse a drug what happens to these human cells? Can we specifically kill them because we know that these human cells represent as close as we can without going to clinical trials human disease and so when we do that with those agents we actually see quite striking results and so this is just, again, a flow plot and this is just an exercise in pattern recognition just to show you guys that when we take out a patient bone marrow and look at sort of its characteristics and then we inject it into a mouse, the pattern is very similar. So, we are actually fairly close to recapitulating what has happened in MDS patient and so now when we start to give these animals these drugs that being, again, Venetoclax, Azacitidine and drugs that you've heard about multiple times already this morning, Omacetaxine, which was our protein translation inhibitor and then to answer the question earlier combinations of drugs and so sort of what you see here is at least these bars represent the number of human cells in the mouse. You can see that Venetoclax alone actually in this particular set of mice has very little effect. Azacitidine also not terribly surprising because this is a high risk disease has very little affect. Omacetaxine as a single agent has a fairly exquisite toxicity, but where we start to see the greatest effect is now when we start to combine these agent. So, the protein translation inhibitor in Venetoclax or the protein translation inhibitor in standard of care that being Azacitidine. So, this is in the total human cells. This is now when we start to focus on those MDS stem cells. Again, where we see almost complete wipe out specifically of those MDS stem cells suggesting now that this disease or that these animals would not progress or have the ability to progress to frank leukemia and so sort of those are the closest the clinic as Dan alluded to. The Venetoclax therapy should hopefully be in clinic next year. We're also working on an investigator initiated trial to get that up and going with (inaudible 1:26:41) agent.

So, the third one which is a little bit further from the clinic, but still takes advantage of the specific characteristics of the MDS stem cells is sort of immunotherapy and so, again, a couple years ago *Science* magazine which is sort of one of the world leading journals in scientific publications gave cancer immunotherapy the Breakthrough of the Year and so we're already applying this to AML. We'd like to take a step back now again and look at MDS and see whether or not we specifically target these MDS stem cells and, again, we do that by the presence of the characteristic of CD123 and so the way that we're going to do that is through something that many of you have heard. It's

made the cover of *Time* magazine. It's frequently on CNN for its initial results, but that being chimeric antigen receptor T cells and so essentially what we do is we pull out a patient's T cells. We then modify those T cells to allow them to specifically recognize the cancer cells to seek out the cancer cells and essentially carry out the immune modulation and specifically kill these tumor cells and this is work that's underway in other leukemias as well as AML and so here's an example of our collaborators' results in AML where they take a mouse and so the color this is just showing disease in the mouse, AML in the mouse, and so the brighter the color the more disease you have and so you can see that a month into injecting them with just a buffered saline there's no response. The tumor has progressed. It's gone from blue to red. Here is a control experiment, again, where you see no effect, but when we now apply these CD123 specific CAR T cells we can essentially ablate the mouse of all AML and so this is work that's led to a phase one trial that's currently underway at City of Hope with our collaborator and then ultimately we'd like to bring this MDS and that's because of some of the preliminary results we've seen, again, in a dish and so this graph here represents two different bone marrows where upon mock treatment which is similar to this we see no shift to the left. We see no disappearance of those MDS stem cells. We're under stimulation with the CD123 CAR T cell. We now see this shift to the left side of the screen here showing that we're actually effective with kill these cells.

And so I'd just to conclude just summarize sort of what I briefly talked about here and then I'll take some questions. So, I think what I hope you understand is that MDS patient bone marrow samples we can take these bone marrows that some of you have given and consented for our tissue banking purposes. We're able to study those and understand the differences between a normal stem cell and leukemia and MDS. CD123 appears to specifically mark these MDS stem cells and then ultimately we can target these both with clinically available agents that I showed you as well as sort of on the horizon some of these immunotherapies and so this is very much a lab thing to do. We acknowledge all the people that have helped with this. This is not just all of my work. So, Craig leads our division and I work under the direction of him. Dan and I work closely together. This is Dan in action. Enkh who spoke first, she and I work closely together on the immunotherapy and then, of course, I have to thank the patients for their generous donation of their tissue so that we can actually study this disease and so I'll take questions and then also threw my E-mail up here if there's something that you want to address via E-mail question later by all means.

(Applause)

**Jonathan Gutman, MD:** Alright. Good morning, everybody. My name is John Gutman. I'm an associate professor over at the University of Colorado, work with all the folks who have been speaking with you already this morning and I direct our allogeneic stem cell transplant program, our bone marrow transplant program. For reasons that are a little unclear to me, I thought I might have about 10 more minutes than I think I actually allotted. So, we'll try to move through this pretty quickly, but I'm certainly going to be available for questions and I can have a propensity to start talking and not stop. So, I'd want to try and avoid subjecting you to that because I know lunch is coming up, but I also will note I'm about five minutes late getting going.

But let's take some time to talk about stem cell transplant for MDS. I think every... people like to hear about this because it offers the great (inaudible 1:33:28) it is the only strategy that I think we think in this day and age offers the potential cure for this disease, but unfortunately it's also a strategy that is fraught with a lot of challenges, a lot of risks and it's a very logistically challenging strategy and so finding the patients for whom it is right is often as much a philosophical question as it is anything else and so I want to kind of walk through those issues a bit. Spend the first portion of our time talking about what a transplant is because a transplant is really kind of a big hammer that we can use to hit lots of different diseases and MDS is one of those diseases, but the way that we think the transplant works has a lot more to do with the transplant itself than it does the specific disease we're treating and then we can spend the remaining portion of our time looking at MDS itself and how we think about the specific issue and the context of MDS and, again, hopefully some time for questions and I'll certainly stick around a bit and be available to talk more.

Everybody, I think, has probably drawn up this slide, hematopoiesis. If you go off to medical school you're subjected to this and it becomes engrained in your system, but I like to put it up because anytime I want to talk to a patient about a transplant, I think that understanding a little bit about how our normal blood works is very important to establish context for how we think the transplant works. So, in our normal blood system as I think you are probably all fairly well aware, we have three main types of cells. We have what are red blood cells which carry oxygen, give us our energy, we have platelets so that if we get cut we can clot and not bleed to death and then we got a whole bunch of different kinds of cells that in the aggregate we call our white blood cells. They're all these different kinds of cells and when we put the white blood cells together they constitute our immune system and in the normal healthy blood system these mature cells are dying off and they're being replaced. The way that they're being replaced is that living in our bone marrow are these things that we call hematopoietic stem cells. These hematopoietic stem cells are like a fountain of youth for our blood system. They sit in our bone marrow and throughout our lives they don't change, but they have the capacity to continuously pop off cells that go through these divisional maturations to grow up and become mature white blood cells, mature red blood cells, mature platelets. So, when everything's working right as others have said, you get normal numbers of these cells and they're doing off. A single goes to the stem cell pop off to replace it. Something can go wrong virtually anywhere in this process and depending on the cell that acquires abnormalities and the abnormalities that that cell requires leads to the development of blood disorders and blood cancers. Myelodysplastic Syndrome is characterized by abnormalities in the immature cells growing up to become white blood cells that we call myeloid lineage cells, but what's critical in, I think, forming a context for understanding the stem cell transplant is that this fountain of youth is being driven by the stem cells that sit in our bone marrow.

So, what are we talking about when we talk about an allogeneic stem cell transplant? Allogeneic, what we mean when we're talking about allogeneic is that somebody else is going to be the donor for the transplant in contrast with something called an autologous transplant where we collect one's own stem cells, bash them with chemotherapy, put their own stem cells back in. It's a strategy that's not really viable for Myelodysplastic Syndrome. I'm happy to talk about it. We use it for other diseases, but allogeneic transplant, what we're talking, about is taking a donor's hematopoietic stem cells and putting them into a patient with the intent of those donor hematopoietic stem cells reconstituting and



becoming a new blood and immune system for the patient and also in the case of Myelodysplastic Syndrome and anytime we do an allogeneic transplant for someone who has a blood cancer or blood disease that is characterized by a bad cell that's growing because I know we can debate whether we call Myelodysplastic Syndrome cancer or not, but we rely on those donor cells to help to destroy... eliminate any residual disease that's left in the patient. We call the donor cells ability to do that a graph versus leukemia effect or a graph versus tumor effect and that is the key to the curative potential of the allogeneic transplant.

Now when we do an allogeneic transplant, we... there are lots of permeations of the details but the basic concept of what we do is that for a period of about a week prior to the infusion of the donor's stem cells the patient will undergo what we call conditioning and conditioning is some combination of chemotherapy and/or radiation therapy that's intended to knock down the patient's immune system sufficiently so that the donor cells will be able to take and it's also intended to help to kill off residual disease that might be left in the patient to make the transplant more successful. The transplant itself is actually really just a blood transfusion or a marrow transfusion. It is not a fancy dramatic surgery and when I read in the paper about a great bone marrow surgeon performing the procedure, I'm always... I pretend that I'm a great surgeon, but I don't think you'd want me cutting into you at all but it is what I call one step for man and a giant leap for mankind phenomenon, but the event itself is small, but the consequences of the infusion are very significant and, again, lots of permeations of the details but the general notion is that for a period of about six to eight months after the transplant, we're going to have a patient on some form of medicine to knock down the immune system which we try to taper off over time to prevent significant problems from developing after the transplant and just to speak to the logistical requirements here which are, obviously, very important for any patient is thinking about this and I didn't spend a lot of time in this talk. For a period of about three months after the transplant because of all the complex things that are going on we need to have you here local in Denver, so patients who we're treating from Fort Collins, from Wyoming, from New Mexico have to move to Denver and be with us for about three months after the transplant, but it's a very complex process.

So again, how does this transplant work? I've already sort of suggested in the last slide, but the original premise behind these allogeneic transplants was the thought was that maybe we could drop a nuclear bomb on patients and we could destroy any residual disease that was left in their system, but as a consequence of doing that we would wipe out all of their stem cells so they would die of no blood system and the thought was let's get somebody else's system cells in there to regenerate the blood system and that was our original thought, but we came to understand fairly quickly as we started doing these transplants that when someone else is the donor far more goes on and, in fact, those donor cells can act like a drug and they can mediate this graph versus tumor effect and the graph versus tumor effect is what we think really cures patients in the case of the allogeneic transplant. So, the realization that this graph versus tumor effect exists led to a revolutionary thinking about how we might do these transplants because in the early days when we thought that all we were doing was killing everything off and there was no graph versus tumor effect, we had to use these very intensive bombs of chemotherapy and/or radiation to wipe the patient clean to prepare them for the transplant. The problem is is those very intensive conditioning regimens are very toxic and people over the age of 30 to 40 years old we'd just kill them if we try and do it and plenty of younger

patients we would kill in the process. So, we realize the graft versus tumor effect exists. It made us think well, maybe we can do things a little bit differently here. Now, if I were to take a donor cells and just stick them into a patient at this point with no chemotherapy, no radiation before doing it, those donor cells would be rejected, but we learned that if we give just a little bit of therapy, just some gentle small doses of radiation or chemotherapy and we have the right donor that can be sufficient to stun the immune system of the patient adequately to get the donor cells to go in there and start growing and then they can start growing, they can take over and they can do their graft versus tumor effect. When we do a transplant like that we call it a nonmyeloablative or reduced intensity transplant and the development of these nonmyeloablative or reduced intensity transplants has been a huge boon to our field because it allows us to think about doing transplants on patients well up into their 70s now who are otherwise healthy and lots of patients who have Myelodysplastic Syndrome tend to be patients in their 60s and 70s.

So, what we've ended up doing is developing these spectrum of these conditioning regimens of varying degrees of intensity. As I've described to you might say well, why would you ever not do a nonmyeloablative or reduced intensity transplant. It turns out that there's a bit of a tradeoff as there is in all things that we deal with in transplant and the gross tradeoff is that this graft versus tumor effect is imperfect. We don't really understand how it works. We're trying to better harness it, trying better to understand it, but we know sometimes it works, sometimes it doesn't. We know that it is more likely to work when the disease is under the best possible control it can be going into the transplant and so what turns out to be the tradeoff is that the harder we hit you with the conditioning regimen before the transplant the more intense the chemo, the radiation is that we give you, the more we will reduce any burden of disease that's left in your system the less the graft versus tumor effect will have to do and on the margins the less chance that the disease will come back after the transplant, but that comes with the tradeoff of hitting you harder with a more toxic combination of chemo and radiation that might be lethal or certainly cause significant problems. So, the gross tradeoff is the harder we hit you, the less chance the disease is going to come back, but the more chance we might kill you in the process. The gentler we hit you vice versa the more chance on the margin the disease might come back, but the better off we are in terms of risks of causing you acute harm.

So, what we've ended up doing is developing a spectrum of these things of varying degrees of intensity and what we try to do for any given patient is we try to pick for them based on their age, based on the disease, the details of the disease in terms of where it stands at the time that we're moving into the transplant based on their medical issues. We and try and pick somewhere on the spectrum the best balance balances those risks and depending on the details of any given patient we come up with different spots on this spectrum. The second major issue around the transplant in addition to how hard are we going to hit you with the transplant is who do we use as the donor for the transplant. We learned very quickly when we started doing these transplants you can't just use anybody as a donor for one of these transplants and you've got a set of genes in your body that code for the way that your immune system function and for a donor to be appropriate we need to match you as closely as we can at the combination of genes that you have. You get one set of each of these genes from each of your parents, so the way the statistics work if you have a sibling from the same parents there's a 25 percent chance that sibling inherited the same combination of genes that you did

and in 2016 I would say that virtually all transplant centers would say if you have a perfectly matched sibling who is able to be a donor and the bar for that is not terribly high, but we could... I'm happy to talk about that with anyone who needs or who's interested in talking about it more, but if you have a perfectly matched sibling who we consider to be acceptable for a donor that's usually our first choice for donor, but 70 percent of people I meet don't have a perfectly matched donor and for that group of people there are several different alternatives. There's a pool of 25 million people in the world who said they might be willing to be donors and we can go and we can look in that pool for a perfectly matched unrelated donor. Umbilical cord blood which is literally the blood in the umbilical cord blood is very rich in these blood forming stem cells and has been frozen down at public banks around the world and we have an inventory of about 600,000 cord blood units we can potentially use as a donor source for transplant. I put in all this junk. I actually should have taken it out because it gets into the details of how we do the matching, probably a little beyond the scope of things here, but also there's been a lot of investigation for decades that is now starting to bear some fruit, we think, in using what we call a half matched or haplo identical donor. So, a parent, a child, a sister or brother that got half the genes that you got, all these different potential alternative options for donor selection, again, I think I'd be happy to talk in more detail about on an individual level to get into a huge detail is probably a little beyond the scope of our time, but the upside is that we need to find an appropriate donor for patients, but with these options we can find a donor for virtually 100 percent of patients in this day and age. The University of Colorado, we have a particular preference and (inaudible 1:46:55) and we're thinking about using cord blood as the donor source as we can, again, talk about, but there's also this very exciting emerging data around haploid identical transplants and I think that the algorithm for donor selection broadly across the world of transplant doctors is going to continue to be evolving in the next probably decade or so such that we may well be seeing more and more use of donors that historically haven't been the primary source of donor for transplant.

So, these transplants are the best chance of cure for this disease. Why are we not willy nilly doing them on every patient walking down the street? There are five major complications that occur with these transplants in addition to the complex logistical considerations. Again, I can touch on them briefly. I'm happy to hash out with you in more detail, but 1) we can put the donor cells in and they might not take, 2) you could get sick, you could die from the complications of the chemotherapy/radiation we give you to prepare you for the transplant for the drugs that you need to be on in the aftermath of the transplant, 3) which is the single most significant problem with these transplants is that there exists the flip side of that graph versus tumor effect, its bad side where we put the donor cells in and they see the normal body as foreign and they start to attack the normal (inaudible 1:48:16) which we call graph versus host disease. It comes in two flavors, an acute and a chronic form, but the spectrum of what it can do ranges from nothing to potentially being an awful lethal process where your GI track can slough off, your skin can slough off, your liver can shut down, you can lose flexibility in your joints, your skin can tighten, you can develop significant lung problems, wasting syndrome. It is definitely the thing that holds transplant back most significantly. Again, lots of details could be discussed, but it is a very significant issue that we're working very hard to try and sort out. The fourth major risk of a problem that is something that can sort of be caused by the transplant is that you get this new immune system, you're very vulnerable to infection if you get graph versus host disease, we have to knock down the immune system further to manage it and so infection can be a major issue after the transplant and so all things considered depending on

the details of any given patient, we would expect that somewhere between 10 to potentially even up to 40 percent of patients might die in the first year or two after the transplant from some complication related to the transplant and not just die of the... and this number I always say is a little bit... I take it with a little bit of grain of salt because we're talking about death here. The more... perhaps even more significant issue to transplanters and to me for sure is that there are a significant portion of patients who may be cured of their disease and don't die of a complication of the transplant, but they may have long term problems as a result of graft versus host disease that contributes significantly to the quality of their life even if they've been cured of their disease. So, these are very important issues that we have to think about and consider and then on a statistical basis the single most common reason that these transplants fail is that even though we do them to cure of the disease it is not a guarantee of cure and the disease itself can still come back after the transplant. The risk of disease coming back after the transplant is most significantly affected by how well controlled we have the disease going into the transplant. So, it's very important we try to control the disease as well as we can going into the transplant.

With that as background for how we think about transplants, let's just spend a few minutes here talking about Myelodysplastic Syndrome in the context of transplant. As I think I've already alluded to and suggested, perhaps the most challenging issue, I think, for a transplant doctor and for a patient considering a transplant and in the discussion of transplant is trying to find the timing that's right for doing the transplant because as a general rule the healthier you are, the better off you are and the less likely you are to have a problem with the transplant, but the less likely you are also to want to subject yourself to the risks of the transplant and so there have been a number of analyses that have tried to think about where the timing is best to think about doing the transplant and as I imagine, I wasn't here for Enkh's talk, but she may have mentioned and Dan may have mentioned we have a lot of different tools for scoring the prognosis of Myelodysplastic Syndrome and perhaps the most commonly used is the IPSS, International Prognostic Scoring System, which has been more recently replaced by the Revised International Prognostic Scoring System which adds in a few elements, but basically by looking at a few fairly simple clinical variables – how many blasts do we see in a patient's bone marrow, how significant are their blood count abnormalities and what cytogenetic abnormalities do we see associated with their Myelodysplastic Syndrome, cytogenetics we are looking at the chromosomes which is where the DNA is packaged in every cell and in Myelodysplastic Syndrome there are often abnormalities in the chromosomes, but putting those factors together we can come up with a score and we can stratify patients into a risk category and based on their stratification in that risk category we can very grossly prognosticate about how likely they are to be alive in a year and two years and three years if they don't do anything, if they try Azacitidine or if we move onto a transplant and what these scoring systems have helped us to understand is that we think that if you have Myelodysplastic Syndrome and it's not causing you a whole lot of trouble, you don't have a lot of transfusion needs, your blasts aren't elevated, you don't have bad chromosomes, the risks of the transplant outweigh the benefit at that point, but as we advance and the disease starts to become more challenging and the prognosis tends to worsen then we start to think that the risks of the transplant really do start to make sense and that's what these slides are showing in various forms. As a general rule though I think we would say that people who have Intermediate 2 or higher risk in this scoring system, we really get serious about thinking about transplant. The Revised International Prognostic Scoring System just adds in a few more details. The

principles are exactly the same, but as we move into higher risk patient population as the disease evolves thinking about moving in the transplant is really where we really get serious about it.

The other major thing that we have to think about independent of the disease itself and what the disease is doing is who is the patient, how healthy is the patient and how do we think about the patient in the context of this decision because, again, many patients with Myelodysplastic Syndrome are older patients and in our world of leukemia and Myelodysplastic Syndrome you don't have to be that old for us to start calling you a little bit older. As you get into 60s and 70s we start think of you as being a little bit older in terms of your ability to tolerate the therapies that we have to subject people to when we think about more aggressive treatment, but I think that it's very fair to say that in this day and age with younger patients, 50 and under or so we're able to think about more intensive conditioning regimens and a little bit less risk of relapse, but patients well up into their 70s, again, we transplant now with good success if we can bring together all the other pieces as best as we can. In addition to age and I would say that physiologic age is more important to us than chronologic age, so I've seen some 20 year olds that we wouldn't want to touch with anything intense and I've seen 75 – 80 year olds that we think are healthy as horses and able to take some serious intensive therapy. We've also developed a scoring system that we call the comorbidity index that allows us to look at all your different organ systems basically and based on any abnormalities within them assign a score, then we can add that score up and it can help to give us a little bit of a quantitative predictive sense of how likely you are to do well with the transplant in terms of toxicities and help us think a little bit about what the best way to do the transplant is.

So when we look at outcomes associated with transplant for Myelodysplastic Syndrome there have been a number of studies that have tried to assess who is most likely to do well with the transplant and a recent paper that looked at a variety of factors – what was the IPSS score, what were the chromosomes, what was the comorbidity index, how old was the patient, how well controlled was their disease going into the transplant we could come up with a scoring system that gives us a predictive sense of how likely patients are to do in the aftermath of the transplant, but patients who go into transplant with Myelodysplastic Syndrome but have very good prognostic features on all of this other stuff, they're likely to do very well. This is 10 years out, 70 percent of patients still alive. Whereas if we take you to transplant you got all these terrible features likely to do poorly in spite of the transplant and so working at figuring out how to optimize all this stuff is stuff that we're continuously doing, but I think that the best available data would suggest that there's definitely value to be gained from doing a transplant in the right patient in terms of, again, the chance of curing patients and in terms of overall outcome. There have been a couple of studies published that have compared patients who had a perfectly matched sibling donor and therefore were eligible for transplant to patients who did not have a sibling donor and therefore did not receive transplant and what these studies have fairly consistently shown is that the transplant patients there's a much higher likelihood years after treatment that they're going to be alive and they're hopefully cured of their disease. Both of these are the transplanted patients and then these are patients who did not have transplants and there's a lot of upfront toxicity associated with the transplant as well as the disease process itself, but if you get through that early period and you're cured of the disease then we do definitely believe that transplant holds that creative potential and ideally gets you back to a place

where you are living your life wholly and completely ideally off of all drugs although it's only a portion of patients who really do meet that criteria.

And then just one last slide. I think you've heard already from a number of speakers about sort of where we're going with the future of Myelodysplastic Syndrome, but in general in cancer and particularly in Myelodysplastic Syndrome we're getting far more sophisticated about understanding the individual abnormalities that are driving an individual's cancer and we've identified that there are about 13 genes that seem to be aberrant in the majority of patients with Myelodysplastic Syndrome and different patients who have different combinations of abnormalities of those genes and we are beginning to understand the prognostic significance of those individual genes and their abnormalities and that allows us to think a little bit about outcomes associated with different particular abnormalities and perhaps more importantly and more excitingly hopefully gives us targets that we can begin to think about as we think about better and better improved therapeutics down the line.

But hopefully I didn't go over too far and that's a brief introduction to what's a very complex topic and I think hard to kind of summarize in 20 minutes, but hopefully of some value. Happy to take a question or two.

**Q8:** (Inaudible 1:58:47)

**Jonathan Gutman, MD:** So, it's a bit of a complex and evolving question, but as a general rule I would say that we look at five critical genes one of which that you inherit from each of your parents and so really it's 10 because you get one pair from each and generally speaking I would say to be called a perfect match we would be looking for a 10 out of 10 match at what we call high resolution sequencing, but that being said when we use umbilical cord blood as a donor source for transplant or we use a haplo identical donor we are very willing to accept the mismatches because we know that those mismatches don't necessarily correlate with worsening outcomes. This standard for perfect matching goes back to an era before cord blood and haplo identical transplant were really available options.

Yeah?

**Q9:** (inaudible 1:59:46) Robin Roberts' (inaudible)

**Jonathan Gutman, MD:** I saw "Good Morning America" the other day and she looked pretty good, but I think that she's doing well by all counts. I don't know the details of what's going on with her. She had a form of Myelodysplastic Syndrome called secondary Myelodysplastic Syndrome where she had had prior treatment for a breast cancer and the chemotherapy for that prior treatment we think caused her to develop the Myelodysplastic Syndrome and we know that that is historically a group of patients who do quite poorly and for whom we always are thinking about transplanting.

**Q9:** (inaudible 2:00:26)

**Jonathan Gutman, MD:** I don't know what the combination of therapy exactly that they gave her before her... her conditioning regimen was. She had her transplant at Sloan Kettering and they take a particular approach to transplant where they take out a subset of cells called T cells and the details I'm not sure, but she appears to me to be doing well, but you know as well as I do.

**Q10:** Can you (inaudible 2:00:54) donor (inaudible) sometimes even if they're a really good match (inaudible)

**Jonathan Gutman, MD:** That's another question that's debated and around which there's not hugely compelling data in one direction or another, but it is absolutely true that as we are able to do these transplants for older and older patients we are also then dealing with siblings who are older and older as potential donors and so the process of donating for the transplants the way that most of these kinds of transplants are done is that we would do is called a peripheral blood stem cell transplant and what that means is the donor has to take shots for about five days of a medicine called DCSF or Neupogen. It stimulates the stem cells to come out into their blood and then we hook them up to a machine, the blood runs through the machine and we pull the stem cells off. It's a very safe procedure. It's done all over the world all the time on volunteers who don't know the person they're donating for. So, it's a very safe procedure, but the question as you get older whether there might be any issues around the stem cells that you as a donor is one that has been considered and whether a perfectly matched 65 – 70 year old sibling is a better donor than a perfectly matched 20 year old unrelated donor is debated. I think that the aggregate data at this point would say that with a... that siblings as long as they're able to donate sufficiently are still probably the first choice even into their 70s and so. We would use a donor in their 70s probably over an unrelated donor provided that there were no health flags or anything like that in the donor and the data on the question is sort of neutral.

**Speaker:** I think that we're ready for lunch, but I do want to thank Doctors Pollyea, Dr. Gutman and Dr. Stevens. They donated their time today to be with us. We'll have time for more questions after lunch, but I just wanted to also mention. We were with Robin Roberts at an NPN MDS Women's Conference maybe about two weeks ago and she thought it was the radiation that caused the MDS. She's doing remarkably well three years post-transplant just celebrated her third birthday this past September and fortunately Sally Ann, her sister, was almost a perfect match for her, her sibling. So, if we could break for lunch right now we'll have an opportunity. Sandy Curtain is here. She'll begin a program in about a half hour, but if you'd like to go help yourselves to lunch and bring it on in in about a half hour Sandy will start her program. We'll have an opportunity to ask more questions then.