Support comes from AstraZeneca, a biopharmaceutical business that is pushing the boundaries of science to deliver new cancer medicines. More information at astrazeneca-us.com. Welcome to Yale Cancer Answers with doctors Anees Chagpar and Steven Gore. I am Bruce Barber. Yale Cancer Answers is our way of providing you with the most up-to-date information on cancer care by welcoming oncologists and specialists who are on the forefront of the battle to fight cancer. This week it is a conversation about new developments in pancreatic cancer research with Dr. Mandar Muzumdar. Dr. Muzumdar is an Assistant Professor of Genetics and Medical Oncology at Yale School of Medicine, and Dr. Chagpar is an Associate Professor in the Department of Surgery at the Yale School of Medicine and the Assistant Director for Global Oncology at Yale Comprehensive Cancer Center. Chagpar So, maybe we can start off by talking a little bit about pancreatic cancer. What is it, how common is it, who gets it and why? Muzumdar So, pancreatic cancer is a cancer that originates in the pancreas. It accounts for about 3% of all cases of cancer in the United States. So, that leads to about a little bit more than 50,000 new cases in the US and more than 300,000 cases worldwide. An important part of pancreatic cancer is that it tends to be very aggressive at diagnosis and more than 80% of patients are diagnosed at a stage where they cannot undergo surgery for the disease. And so that leads to a very high rate of death. So, even though it is only 3% of all cancers in the United States, it accounts for about 7% of all deaths from cancer in the United States. Another important part of pancreatic cancer is that unlike the other two major leading causes of cancer deaths in the United States, both lung and colorectal cancer that have been decreasing in incidence and death rates due to smoking cessation programs and screening, pancreatic cancer is still on the rise. It has slowly and steadily been rising over the last decade. Chagpar So, why is that. I mean, when we think about cancers increasing in incidence, usually that is because either we are increasing the risk factors causing that cancer or we are getting better at picking it up and doing screening. So what is the case in pancreatic cancer? Muzumdar So, in pancreatic cancer, we have not been as fortunate in terms of screening. It is often diagnosed at a very late stage, often times because the symptoms are very vague, sometimes just a little bit of abdominal discomfort and just a little bit of feeling full very early, and so we do not think that we are catching new cases earlier. What we think is happening potentially is that the risk factors that contribute to pancreatic cancer are becoming more prevalent. One of the important risk factors for pancreatic cancer is actually obesity, and we know that obesity is on the rise over the last several decades and that is concordantly risen with the incidence of pancreatic cancer. Chagpar And so, I mean, but obesity causes or is a risk factor for so many cancers right? It is a risk factor for breast cancer and a variety of other cancers and certainly a risk factor for other bad diseases – heart attacks, strokes, etc. So, how is it obesity actually impacts pancreatic cancer? Do we know? 00:03:23 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Muzumdar So, what we have learned from epidemiologic studies is that obesity is associated with an increased risk of developing pancreatic cancer, and it is also associated with an increased risk

of having more aggressive or metastatic disease at diagnosis. So, it increases the risk of both developing it but also the degree and severity of the disease. And we do not understand the mechanism behind how that works. One of the things we have been very interested in trying to study is to study that particular aspect in the laboratory and some of my research in pancreatic cancer actually focuses on trying to develop better models to understand how obesity contributes to pancreatic cancer development and how it might contribute to its growth and aggressiveness. And we have actually been using mouse models that closely recapitulate, actually closely mimic the human disease in terms of how the cancer looks under a microscope, how it behaves in terms how it metastasizes and responds to therapy, and then integrating different modalities and try and make these mice obese. And what we have been starting to find in these mice is that if you make them obese and give them a genetic predisposition for pancreatic cancer, they get faster and more aggressive disease, similar to what is seen in humans. And it turns out, if we can make these mice lose weight aggressively very early on, we can actually get rid of that more aggressive disease and slow it down. And so we think that we have a model now in which we can study the mechanisms behind how obesity contributes to pancreatic cancer in hopes of hopefully identifying new targets for therapy and prevention in the disease. Chagpar So, it sounds like there is an interaction there between obesity and this genetic predisposition. So, if you had fat mice and they did not have a genetic predisposition, do they get pancreatic cancer more than skinny mice without a genetic predisposition? Muzumdar So, one of the things we know is one of the big risk factors which we cannot change for pancreatic cancer is age. Just having enough time for some mutations, what we think, to accumulate, are mice's lifespan. The average lifespan of the mice we use in the laboratory is only 2 years, and so it is fairly short. So, the mice that are obese do not develop we think those mutations within that time span of their life. So, we give them a little bit of push. We use genetic engineering to try and initiate a specific mutation we know is very common in pancreatic cancer and then combine it with an obesity model in the mice as well. Chagpar So, that is really interesting. So, in people though, they have got age… At what age do people get pancreatic cancer on average? Muzumdar Yeah, so the median age of pancreatic cancer is in the late 60s in people. We know that the incidence goes up with age. So it really age that seems to be an important risk factor for pancreatic cancer and we think at least in some part is due to the potential for the development of mutations that can help initiate or start up the disease. 00:06:15 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Chagpar So, the reason I asked that of course is because some people they were obese as kids, some people are obese as adults and the binding that you had where if you get mice to lose weight, then you can potentially can prevent them from getting pancreatic cancer or at least not get pancreatic cancer as quickly as they otherwise would with this genetic predisposition. So my question is, I was always a fat kid, so even though as an adult I have kind of lost weight, is that enough or if I gained weight again and lost weight right before my 60th birthday, would that prevent me

from getting pancreatic cancer. I mean, is there a time continuum here that we need to look at? Muzumdar Now, that is a really great question, which we do not have a great understanding from human epidemiologic studies. The nice thing about our mice is we can manipulate when they lose weight during the course of how tumors develop. And so that is one of the major things we are trying to study is if we can delay when they lose weight, would that affect ability for tumors to grow and indeed once tumors are fully established in these mice and we make them lose weight, they actually do not have any difference in the outcome from pancreatic cancer. But we are trying to narrow down that window and find when is it really impacting the disease, and then we have to think about how that translates to human populations in terms of time. We do not have a great understanding just yet. Our hope is that if we use the mice to identify mechanisms that are important, we can then start evaluating those mechanisms in human specimens and samples and patients, and then we can take that data and identify perhaps there is a time window where we can intervene with this particular disease. Chagpar Especially on the prevention side, I mean what about if you took mice and they were fat for a year out of their 2-year lifespan versus mice who were obese for 6 months out of that 2-year lifespan or a year and a half out of that 2-year lifespan, and all had this genetic predisposition at birth, does it make a difference, how long they were obese for? Muzumdar Yes, so that is exactly what we are trying to test at this moment is if we can induce the weight loss at different time points earlier or later before they develop the full advanced tumors, can we actually have an impact on the disease and what is that time course. Chagpar And then, the question is after they already have a tumor, does losing weight at that point make the cancer less aggressive? I mean is their outcome better if they lose weight? 00:08:54 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Muzumdar So, one of the things we see a lot in pancreatic cancer patients is that they often come to us with one of the prevailing symptoms being a significant amount of weight loss, and so that is going to be confounding how we view it, because weight loss in that case may be actually be associated with the severity of their disease. One of the things we have noticed in the mice is if we make them lose weight once they are already starting to lose a little bit of weight from their disease, or at a time point where we know that they have developed quite a bit of disease, we do not really have an impact on outcome, and so we think a late weight loss once the disease is established is likely not to have a significant effect on the outcome, but perhaps early weight loss as a preventative or now we have been thinking about in a different way, can we intercept how cancers progress from early to late disease, could that be a way of either preventing or intercepting the disease before it manifests into a very advanced tumor that we often times have a very hard time treating. Chagpar The other question is, what about the people who like me are kind of yo-yo people? You know, we are obese and then we are not obese and then we are obese again and then we are not obese. How does that impact tumor development? Muzumdar That is a great question. One of the things we can really do easily in mice is we can make them lose

weight and stay lean, which is sometimes very hard in humans. We have all faced that same similar situation. Chagpar We just got through the holidays, we all have been there. Muzumdar That's right. And so, we are trying to do is trying to model that as well in the mice to see if there is a temporal correlation if we can go up and down in terms of weight and can we see different effects in terms of is it still driving tumors, how long do they have to keep the weight off and things like that. So, those are all very important questions that we feel now have a model to actually address. Chagpar So, this is all fascinating, but I guess my other question is, there are so many other factors that potentially are etiologic or causative agents for pancreatic cancer, and I wonder how obesity plays into that. For example, does alcohol increase your risk of pancreatic cancer and for anybody who was drinking over the holidays, my bet is that you put on a couple of pounds. So, how does that work? 00:11:05 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Muzumdar Sure, there is a lot of risk factors that have been associated with the pancreatic cancer. Some of which are stronger than others. Alcohol, for example, has shown in some studies to be associated with increased risk of pancreatic cancers, and others again in epidemiologic studies, not showing that association. There is a clear association between chronic inflammation in the pancreas and the development of pancreatic cancer, and we know that chronic alcoholics in some cases can develop chronic inflammation in the pancreas, what is also known as chronic pancreatitis and that is a well established fact. So, it may be that alcohol acts to promote pancreatic cancer development via its effects on the pancreas in creating inflammation. There are other risk factors that have been associated as well; smoking is another big one and fortunately there have now been improvements in smoking cessation programs that really decrease that risk compared to the obesity role, and there have been variable associations with blood type, with race, there is a slight predisposition for males over females, many of these are clearly not modifiable, and so we have been really focusing on trying to identify whether modifiable factors such as obesity can actually alter the pathogenesis or the development in the disease, and we think at least now we have a model to study that. Chagpar So, how does that work exactly. I mean, you alluded to the fact that you are research is trying to identify this and identify biomarkers, etc., and I want to unpack that a little bit more. Is obesity causing inflammation kind of like a chronic pancreatitis or is it acting via a different mechanism to cause pancreatic cancer? Muzumdar That's a great question. And so, we look at the actual pancreas of these mice as they develop tumors. And what we do see is increased evidence of inflammation in these particular mice, and we think we know some of the factors that actually specifically contribute to that and some of the factors that are involved in the inflammatory process itself. So, what we are trying to do now is actually block those factors using drugs, collaborating with my colleagues here at Yale who are developing new therapies to try and test whether we can use that instead of weight loss as a way of intervening in the pancreatic cancer development process. And that would potentially give us an alternative mechanism, potentially something that we can actually treat patients with as a

preventative, as a treatment for pancreatic cancer. Chagpar I will tell you a lot of people would rather take a drug to prevent inflammation than trying to lose weight. It is just easier. So what I want to do after we come back from the medical minute is really talk more about pancreatic cancer and obesity and some of the other factors that might play into this. I mean, we have all heard about obesity causing resistance to insulin, where does insulin come from – your pancreas. So, does all of that work together for pancreatic cancer? If you are curious to know the answer to that, I am too… We are going to find out more right after we take a short break for a medical minute. Please stay tuned to learn more information about pancreatic cancer with my guest Dr. Mandar Muzumdar. 00:14:09 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Medical Minute Support comes from AstraZeneca, a science-led biopharmaceutical company dedicated to partnering with leading scientific companies, organizations and the community to improve outcomes for advanced cancer patients. More at astrazeneca-us.com. There are over 13 million cancer survivors in the US and over 100,000 here in Connecticut. Completing treatment for cancer is a very exciting milestone, but cancer and its treatment can be a life-changing experience. Following treatment, the return to normal activities and relationships may be difficult and cancer survivors may face other long-term side effects of cancer including heart problems, osteoporosis, fertility issues and an increased risk of second cancer. Resources for cancer survivors are available at federally designated comprehensive cancer centers such as Yale Cancer Center and at Smilow Cancer Hospital to keep cancer survivors well and focused on healthy living. More information is available at YaleCancerCenter.org. You are listening to WNPR, Connecticut's public media source for news and ideas. Chagpar This is Dr. Anees Chagpar and my guest Dr. Mandar Muzumdar. We are here talking about research advances for pancreatic cancer. Now right before the break, I was curious Mandar about this whole concept of obesity and insulin-related growth factor, I mean, we all hear about these things and insulin resistance, does that have anything to do with pancreatic cancer or is that just unrelated? Muzumdar So, one of the things we know is that obese individuals often times demonstrate insulin resistance and you can actually tease some of these different factors out in epidemiologic studies by looking at certain blood markers of insulin resistance for example. What we do see is that there is an association of increased insulin resistance or diabetes with the development of pancreatic cancer and it seems to be potentially contributing with how obesity contributes to pancreatic cancer development. We think that some of the factors like you have alluded to potentially insulin like growth factor or insulin itself could actually be a driver of pancreatic cancer development, but we do not have specific evidence if that is true in the disease, and again, we are using our models to try unravel that particular aspect of obesity and insulin resistance. Chagpar Interesting. So, the other thing that I wanted to ask you about is, you said that you can genetically predispose these mice to get cancer. Well, if you can genetically predispose them to get cancer, then you know of genes that can be turned on that make cancer. So, do we know what those genes are, do they exist in humans, and if so, can we turn them off?? 00:16:59

into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Muzumdar That's a great question. So, one of the things that we have really learned a lot about cancer in general over the last 10-20 years is that cancer in many facets is really genetic disease. It is really due to mutations and specific genes in our own normal cells that now make them inappropriately active or inappropriately inactive. And so, we call those oncogenes or tumor suppressor genes. In the pancreatic cancer, there are 4 hallmark genes that are mutated. The predominant mutation in an oncogene, something that gets activated, is in pancreatic cancer is K-RAS and it is actually mutated more than 90% of cases of pancreatic cancer. And so, it is one of the most common mutations found in a particular cancer type if you look at all cancers, and in fact, K-RAS is actually the most frequently mutated oncogene of all cancers, about 20-25%. But if we can understand the biology about how K-RAS contributes to pancreatic cancer development and how it is maintained, we think we might get some further insight in terms of how K-RAS actually contributes to many different cancer types. So, what we do in the mice is actually engineer them to have K-RAS mutations that are specifically expressed in the pancreas, and interestingly enough, these mice get pancreatic cancer that looks and behaves like human pancreatic cancer. And that was really an important finding because it really suggested that there was a causality between the presence of these particular mutations at high frequency in the disease and their ability to actually start up tumors and generate tumors. Now, what we have been interested now is since it is such an important and prevalent mutation in this disease, is how does it actually make tumors tumors and how does it potentially maintain tumors. It is really the major gene that is mutated that we think is potentially active. So, if we could inactivate it, would it be a great target to develop therapies for. The unfortunate thing is K-RAS has been one of the holy grails of proteins that we really wanted to block. We have know about RAS mutations for more than 30 years, but we have never been able to actually develop really effective inhibitors. There is a renewed effort from the National Cancer Institute and many private investment firms to try and actually develop new K-RAS inhibitors using improved technology. So, we have taken an approach to try and understand whether K-RAS is really important for maintaining pancreatic cancer cells using genetic tools that we have developed. And so, we can actually use advances in genome editing to completely ablate K-RAS and pancreatic cancer cells. Now, unfortunately when we do that, we actually see that about 50% of pancreatic cancer cells actually survive in the complete absence of any K-RAS being made. So, suggest that resistance is likely to develop even if we add K-RAS inhibitors in at least about half of the tumors. The nice thing is we can now study how those cells resist K-RAS mutations and what maintains their growth once K-RAS is gone. And it turns out that most of them resist in the same mechanism and so we have another inhibitor that we can add on to develop combination therapeutics. The other thing that this now allows us to do is really study K-RAS biology in great detail because now we have cells that have K-RAS and those do not that came from the cells that had K-RAS. So, we can see what is different about the cells, what

is so different about their behavior, what is different about the genes that are expressed, what differences are there in the proteins that are actually present, and can we now harness those differences to understand new pathways that K-RAS is directing that we can actually target and treat with. It does not mean we have to inhibit K-RAS, which is challenging and perhaps we can... 00:20:29 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Muzumdar ... directly inhibit something that K-RAS has been regulated or particular state that K-RAS is generated in the cancer cells, and we think that is a particularly attractive view because most all of our other cells besides pancreatic cancer should have normal K-RAS. So, if we can identify particular features that are generated by mutant form of K-RAS in the cell that now we can target, we would get a drug that is specific for the cancer cells while saving and salvaging all the normal cells in our body, hopefully decreasing the number of side effects that we see. Chagpar So, K-RAS is a gene that is in all the cells of your body, and when it is mutated, it increases your risk of developing cancer. But you have been able to use genome editing at least in mice to create a mutation. So, my question is, even before people get cancer, is there a way to look to see if they have a mutation that maybe they were born with and turn it off? Muzumdar Yes. So, there are certain forms of pancreatic cancer, about 10% that run in families, and we know at least a subset of those particular genes that are associated with them, and they seem to be associated with particular genes that are involved in repairing the DNA, so making sure there is no mistakes that lead to mutation. K-RAS is an interesting mutation because it is not something that you are born with, but develops in sporadic cells in your body at some point later during time. And it turns out that not all cells that have K-RAS mutations will actually develop cancer, and we think other things may feed into that; for example, perhaps inflammation from the obesity that is now in mice that have K-RAS mutations drives the development of pancreatic cancer. And so, we can certainly look for mutations that run in families because we think they are actually inherited and so you can look at normal blood cells and look whether those blood cells carry the same mutation. This should be found throughout your body, but certain mutations like K-RAS and the 3 other hallmark mutations that are found in pancreatic cancer are often times mutations that are developed as the cancer grows or develops. One of the other things that we know, is some of these familial types of cancers with particular mutations can actually underlie specific sensitivities, so these cancer cells now no longer are good at repairing their DNA. We know that some of our chemotherapeutics damage DNA in certain ways that perhaps they cannot repair as well. And so, we are starting to take advantage of this genome technology in trying to profile the genetics of the tumors to try and take the arsenal of tools we have already such as certain chemotherapeutics and apply them in specific populations and hopefully improve the outcomes in therapy. Chagpar So, what you are talking about is looking at people who have a K-RAS mutation in say a sporadic pancreatic cancer and you will be able to tell that because on a biopsy, you can look for K-RAS mutations and then because you know that they are not going to re-

spond or they not going to repair their DNA as well, then you can hit them with a chemotherapeutic agent that causes them to knock out their DNA. 00:23:41 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Muzumdar Correct. And so, what we are trying to do more and more now is actually profile not just K-RAS mutations which like I said more than 90% of the tumors will have, but look for other mutations such as DNA repair mutations that may suggest that they are now going to be sensitive to this particular types of chemotherapy, and it actually gets a little bit more sophisticated than that in that it is not just mutation that seems to matter, we can also look at what genes are actually present or expressed by the particular cancer cells, and it turns out that you can look at cancer cells that have mutations, particular DNA repair pathways, and they express certain set of genes and you can look at pancreatic cancers that all for the most part have K-RAS mutations but do not have mutations of specific DNA repair pathways, but they express the same genes as if they did have a mutation in a particular DNA repair, and those cancers may also be a biomarker to predict who is going to respond to these particular types of chemotherapies. Chagpar So, is there, I mean when we think about preventing cancers, I mean it is great that we can say "Oh! You have got a K-RAS mutation or you have got a DNA mismatch repair mutation," you know we know that drug X is going to be better for you or not, but what about really thinking about preventing these cancers, I mean, the idea that obesity may kind of trigger this and interact with K-RAS is interesting, but what if you could look at whether somebody has inflammation in their pancreas has a K-RAS mutation without a tumor and is obese. Is there a way to do that or do you always have to see the mutation in a tumor. Muzumdar Yes. So, we have actually had quite a bit of a renaissance in terms of looking at mutations and other features of organs without actually going and sampling and taking a biopsy of the organ, particularly in the case of pancreas which is actually quite hard to get a good biopsy without causing significant amount of inflammation by itself. We can now look in the blood stream for example for gene mutations such as K-RAS in circulating DNA that is just being spit out by different cells in the body. It turns out a lot of our own cells will do that, and it turns out that cancer cells tend to do a little bit more across cancer types. Now the problem with K-RAS mutation is as I suggested earlier is K-RAS is mutated in many different cancer types, so if you find it in the blood, you do not know which organ it necessarily is coming from. Additionally, you do not know whether they actually have cancer because they may have K-RAS mutations without cancer but they may be the people that you screen a little bit more aggressively. You will look for pancreatic cancer more. The lifetime risk of developing pancreatic cancer is 1.5%, which means 1 in 65 people will develop the disease, and that increases again if you have certain risk factors like family history or obesity and things like that. But we do not always know who to screen, we do not know who is actually at the highest risk and it is possible that some of these blood borne markers as they become better developed and tested may give us some insight in terms of who are the people we need to screen and perhaps who are the people we need to give these chemopreventative agents that we are trying to identify and hopefully

deliver, and then we can be able to then when we do our clinical trials do validate them, identify the people who are most likely to benefit from it. 00:26:56 into MP3: https://ysm-websites-live-prod.azureedge.net/cancer/2018-yca-0121-podcast-muzumdar_325496_5_v1.mp3 Chagpar Yeah. I am sure that there might be people in our audience who are thinking you can look at my blood and you can see circulating DNA and you can see whether that DNA has a K-RAS mutation, and you have just told me that K-RAS mutations predispose to all kinds of cancers which are bad, it does not matter whether it is pancreatic which is really, really bad or breast which is a little bit, but cancer in general is bad, and so, if we are then able to turn that K-RAS off, either through genetic engineering or through lifestyle factors like reducing body weight, maybe that is something we ought to do regardless. I mean, how good is this circulating DNA testing in terms of predicting this and is it something that people should be going to their local family doctor once a year and saying "so how is my circulating DNA?" Muzumdar That's a great question. One of the things that we have developed over the years is better sensitivity to pick up DNA. So, we can identify DNA in the blood at lower and lower levels. The problem is that it is also less specific, so it can come from any different places and it can even come from tissues that are never going to develop pancreatic cancer. For example, you may develop a K-RAS mutation in your pancreas, but it is possible that an immune cell that is circulating around in your pancreas says, "well this is a funny looking cell and I don't think it should be here and I'm just going to kill it before it actually causes any cancer." So, it becomes a little bit of a tricky slope, if you keep screening, you will find things; the problem is you may find things that were never going to cause problems in the first place and then you will go fishing and hunting for things that maybe you should not have in the first place. And this has been the issue with other screening modalities such as prostate-specific antigen or PSA in prostate cancer, and so I think we do not know enough in terms of the specificity. We think we can now become more sensitive in terms of our methodology in identifying the DNA, but we do not have enough knowledge at this point to say this particular identification of this mutation is going to be specific for this disease. Now, maybe that down the road, we are able to get to that or it is not going to just be one biomarker, one DNA mutation but maybe it is a bunch of proteins that we look at the same time. I think as we start to put things together, we hopefully will be able to improve our ability to predict risk, then identify the patients who need to be screened, then hopefully take advantage of some of the chemopreventative type methodologies that we are trying to develop and test in our mice models to then apply to that specific patient population. Dr. Mandar Muzumdar is an Assistant Professor of Genetics and Medical Oncology at Yale School of Medicine. If you have questions, the address is canceranswers@yale.edu and past editions of the program are available in audio and written form at YaleCancerCenter.org. I am Bruce Barber reminding you to tune in each week to learn more about the fight against cancer. You are on WNPR, Connecticut's public media source for news and ideas.