Zac Getty:

Good morning everyone. Thank you all for joining us bright and early today for our Genetic Testing Versus Biomarker Testing Webinar. My name is Zac Getty, and I'm the patient education program manager here at Fight CRC. Fight Colorectal Cancer is the leading patient empowerment and advocacy organization in the United States providing balanced and objective information on colon and rectal cancer research, treatment and policy. We are relentless champions of hope focused on funding promising high-impact research endeavors while equipping advocates to influence legislation and policy for the collective good. Before we get started with our webinar today, allow me to go through a couple of housekeeping items. We will have some time at the end of the webinar for general questions, but please, please feel free to use the Q&A panel on the right side of your screen to ask any questions that come up along the way.

You can ask questions during the presentation and we'll be sure to get to them at the end of the webinar. We'll do our best to address every question that we can within the time limits that we have. This webinar and most of our webinars are meant to serve as an opportunity for you to ask your questions, so please don't be shy. We will have a recording of this webinar available on our site within the next few days, and you'll receive that direct link via email as soon as it is available if you have registered for the webinar. We will also provide a transcript of this webinar on our site for those of you that would prefer to read the information discussed today. And also, please feel free to tweet along with us. You can use the hashtag #CRCWebinar.

Please remember to stop by our website at fightcrc.org to check out all of our patient and caregiver resources. This includes Your Guide in the Fight Meetups, which are an online space to meet with other patients and caregivers, which are held three times a month, and touch on a variety of topics, but are also just a great place to find a sense of community, our free Community of Champions app where you can connect with other people in the colorectal cancer space and keep in touch with Fight CRC and know what we're up to. And we also offer an assortment of print and digital educational resources that are free to request and download. We are also now hosting monthly Mega Meetups, which address a specific unique topic each month. Our next Mega Meetup is actually this evening, Wednesday, April 24th, and we'll focus on women's health. You can learn more about that in the Community of Champions app that I mentioned earlier. Just a disclaimer, the information and services provided by Fight Colorectal Cancer are for general informational purposes only. The information and services are not intended to be substituted for professional medical advice, diagnoses or treatment. If you are ill or suspect that you are ill, please see a doctor immediately. In an emergency, call 911 or go to the nearest emergency room. Fight Colorectal Cancer never recommends or endorses any specific physicians, products or treatment for any condition.

Okay, now with that all out of the way, I'd like to briefly introduce our participants today. Joining us today are City of Hope genetic counselor and researcher Heather Hampel, and Dr. Al Benson, Oncologist and Professor of Medicine at Northwestern University. Both of our guests today are actually Fight CRC Medical Advisory Board members as well, and we are so lucky to have them share their time with us today. Heather, Dr. Benson, thank you so much for taking time out of your busy schedule to join us for this early-morning webinar. I'm going to hand the screen off to Heather to give herself a chance to introduce herself a little bit more thoroughly and kick our webinar off. So I'm going to stop sharing. And Heather, it is all yours.

Heather Hampel:

Thanks so much, Zac. I am going to start sharing. And let me know if you can see my screen.

Zac Getty:

I can. It looks good.

Heather Hampel:

Excellent. So as Zac said, I am a cancer genetic counselor. I'm the associate director of the Division of Clinical Cancer Genomics at City of Hope, and a professor in the Department of Medical Oncology and Therapeutic Research. I'm just looking at my [inaudible 00:04:05] on Twitter, which is now called X, my Twitter handle's on there, love to engage with people there if you have any questions or comments about the talk. But I'm going to talk in this first 20 minutes about what we like to call in the business, germline genetic testing. And then Dr. Benson's going to talk about somatic or tumor testing, which is also often referred to as sort of biomarker testing. And we hope by the end you'll have a good feel for the differences between the two and why both are actually important.

So we'll start with the inherited component or the germline component here. Let's see if my slide will advance. There we go. All right. So I'm going to take you guys back to your last high school or college biology class for a little reminder about how we get our genetic material. So this is actually a picture of one cell's chromosomes. And we all have 46 chromosomes, we get 23 from our mom and 23 from our dad, and they come in pairs, and they can put them on charts like this so that we can see them. And what you can't see when you look at this picture of the 46 chromosomes are the genes, because they're just too small, but we think there's about 20,000 genes strung along these 46 chromosomes. And they also come in pairs, because if you've got one gene on, say this chromosome number one from mom, you got a matching gene on the other chromosome number one from your dad.

So you have pairs of genes on your pairs of chromosomes, and each gene codes for one protein that your body needs to grow and develop properly. So I like to say that the mix of these 20,000 genes are the recipe for you. And they determine things like hair color, and eye color, and height, and as we're starting to learn, predispositions to certain diseases. And I always tell people who happen to have cancer running in their family, "Other people may have heart disease running in their family, or diabetes, or other health conditions." We've just come a little further in finding some of the genes that can, when they're not working properly, run in the family and cause an increased risk for cancer. And that allows us to be much more helpful for those families, because if we can identify a family that has one of these hereditary susceptibilities to cancer, we can let people know before they get a cancer that they're at risk so that we can watch them much more carefully with increased surveillance and try and keep them from getting that cancer in the first place, or at the worst case scenario, catch it early when it's treatable so we get a better outcome.

The reason we refer to genetic testing when you're trying to find any changes in the genetic material that you inherit from your mom or your dad as germline genetic testing is because this is the genetics that you got from either the egg or the sperm that you were conceived from, and those cells are known as germ cells. And so that's where this comes from. It has nothing to do with germs or infectious disease, but if you see that word germline, that's what we're referring to. And when we refer to germline genetic testing, we're trying to figure out what gene changes were you born with that you inherited from your mom or your dad that might have given you an increased risk to get cancer.

So I want to spend a minute on this slide because I think this is an important concept for our whole talk today, just to set the stage. So I spend most of my time trying to figure out if people have an inherited cancer susceptibility. So that's what you see on the right side of this slide. And in that case, what happens is somebody is born with one copy of one of these cancer-susceptibility genes from either their mom or their dad, so a germline mutation or mistake. So that gene is not working properly. Luckily, these changes are rare, so typically the other copy from your other parent is working just fine, and that

copy is going to compensate for many years. So we don't typically see a lot of childhood cancers with a lot of our adult-onset, cancer-susceptibility syndromes. These genes are good genes, their job in your body is to protect your cells. Some people describe them like the brakes in the car, as long as they're working, the cells grow nice and slowly like they're supposed to, dividing when they should, and dying when they should.

And so sometimes when I talk to my patients, I say, "Imagine a scenario where cars had brakes on the front tires and the back tires." I know that's not the way it really works, but if you get a car that has the front-tire brakes not working, you could still stop the car, because the second set is working. And so that's how these genes work, they're like the brakes for our cells and our genetic material, making them again, grow slowly like they're supposed to, dividing when they should and dying when they should. So if you're born with the one copy not working, the other one's going to be compensating for many years, but you have millions of cells in your body, and the odds are high that one day over the years in one of those at-risk cells, you might acquire a mistake in the working copy. Now you have a cell with no more protective gene, the brakes come off, it's like you've lost that second set of brakes, and that cell starts to grow too fast, doesn't die when it should, and that cell can become the cancer.

So basically, people who inherit a gene change that increases the risk for cancer are essentially born one step closer to getting a cancer because every cell in their body has the one copy that's not working. And all it takes for a cancer to start to develop then is for them to acquire a change in the working copy at some point during their life. And that's the reason we see all the characteristics we see in a hereditary cancer family, such as earlier ages of diagnosis. If you're born one step closer, it's not going to take as long on average. And so that's why in these hereditary families we see an earlier age of diagnosis. We also typically see multiple generations with cancer. We call this a vertical transmission pattern in the cancer-genetics world.

So if you see in this family over here, the grandma had cancer, her son and daughter had cancer, and then the son's daughter has had cancer. So we have three generations or vertical transmission of cancer. That's more concerning than a few siblings with nobody affected above or below. That vertical pattern gives you the feel that there is a gene change that's running in this family and getting passed down through the generations and causing an increased risk for cancer. The other thing that's much more likely in a hereditary family is for someone to have more than one primary cancer during their lifetime, because you've got this one mistake in every cell of your body, you can get the second mistake one year, have a cancer, successfully treat that, and five, 10 years later this happens again in another cell and we have a whole new primary cancer. Any of us could get one cancer in our lifetime, one in three of us will, but when you start seeing people who get more than one primary cancer during their lifetime, that's a red flag that they might have one of these hereditary susceptibilities.

When I say primary cancer, I'm just trying to differentiate from metastatic sites. So many people may have a colon cancer that spreads to the liver, that's not two primary cancers, that's a colon cancer that's spread. I'm talking about two completely new cancers unrelated to each other. So for example, colon cancer, and then 10 years later an endometrial or uterine cancer. And then clustering of certain cancers. And so many people are well aware that breast and ovarian cancer tend to run in families and can be caused by the BRCA genes when they're not working properly. And as we've come to learn over the years, that also includes prostate and pancreatic cancer, but less people are aware about that connection with colon, uterine, ovarian and stomach cancer.

The most common inherited colon-cancer syndrome that we're going to talk about today is Lynch syndrome, and it can cause all of those cancers. And I think it's much less common for people to be aware that they go together. And so you might say, "Well, my sister had uterine and my dad had colon, what does that have to do with each other?" And you might not put together that that cluster is a

known cluster that can be a sign of a hereditary susceptibility. But again, in this hereditary side, the person was born with a mutation, so it's a germline mutation, that made them at increased risk to get cancer in the first place, and they only get a cancer if they acquire a mutation, which would be a mutation that happens during their lifetime, which we're going to talk about in a minute, those are called somatic mutations, that's when the cancer develops.

And when we do gene testing for these inherited mutations, we can tell somebody if they're born with this mutation, but we can't tell them when they'll get cancer, because that's left up to chance, to when that second mutation will happen. We can't tell them where they'll get cancer. We know the organs that are at risk, but within the same family, one person might get one cancer, one person might get another. We can't tell them how many times they'll get cancer. Someone might get one cancer, someone might get three during their lifetime. And we can't even tell people if they'll get cancer. There are certainly people with these mutations who happen to never get the second mistake and never get a cancer during their lifetime. Obviously that's interesting, maybe we should study them, maybe we can learn from them about things that might reduce risk, or it just might be chance, they might just have gotten lucky and never acquired the second mistake and gotten a cancer. Most of these syndromes don't have a 100% risk for cancer. So even if you have one of these gene changes, you're at increased risk, but the risk is not 100%.

I would say on average, we typically now are believing about 10 to 15% of most cancers fall into this hereditary category. And the only way to find out if you're in this hereditary category is to have germline genetic testing, which typically uses a blood or a saliva sample, because we're trying to figure out what you were born with. We don't need to use your tumor for this, we'll use a sample that's easily available like blood or saliva, because we're looking for what you inherited from your parents, not what's occurred in your tumor. And for that, the rest of the cancers, 85% are sporadic, meaning that they've popped up by chance, maybe environmental exposures, age, age is our greatest risk factor for cancer. And in these scenarios, people are born with all of these cancer susceptibility genes working just fine. They don't have any germline mutations. And if I do genetic testing on their blood or saliva, it will come back negative. But we're all aging and our cells are dividing, and we're acquiring mistakes in our genetic material as we age.

And maybe we get one mistake one year and probably nothing happens and that cell dies, but if it lives long enough and acquires enough mistakes, it can start growing funny, not die when it should and become a cancer. But these are acquired mutations that have happened in our body during our lifetime, we were not born with them and we cannot pass them onto our children. And so the only way to test for these is to test the tumor itself to find out what in the world happened in those tumor cells as they became a cancer, what are the mutations that occurred that are driving this tumor to grow too fast and not die when it should, and are they something we can target with therapy? And so this is very important for your treatment purposes. It is, again, not something you were born with or that you can pass on to your children. So it's not something that we worry about testing your family members for, but this is very, very important for therapeutic purposes. And in families that have sporadic cancers, we usually have later ages of onset, little or no family history of cancer, and single tumors, not multiple tumors.

And so I hope that just gives you a little general background about the difference between what I'll be talking about, which is these hereditary cases over on the right, and what Dr. Benson is going to be talking about, which is testing your tumor, tumor testing, somatic testing, same thing, to look for biomarkers that might give us better ways to treat your cancer. Okay, so in the hereditary scenario, most of the cancer syndromes except one I'm going to talk about today, are inherited in a dominant pattern. So if you remember Punnett squares, also from your last biology class, if you've got a parent

who has a gene change that causes an increased risk for cancer, that's going to be the capital A here, they also have their working copy, the little A, and it's pretty rare that they're going to marry somebody or have children with somebody who also has a gene change. So they're going to likely have working copies, which means when they have kids, it's basically a 50/50 chance you're either going to get the copy that doesn't work or the copy that does. And so if you have a hereditary cancer syndrome running in your family, for the most part, with a couple exceptions, but I'll talk about one today, they're inherited in a dominant pattern. And if your parent has one of these conditions, you have a 50% chance you've inherited it.

If you have inherited it, then you do have that increased risk for cancer that we've just talked about. And unfortunately then you can pass it on to your children, they move to the 50% risk. However, if you don't inherit it, you actually have no increased risk for cancer at all. It's as if you don't have a family history. Your risk is the same as the general public. It doesn't ever go to zero, we all can get a cancer during our lifetime, but you can follow the ACS guidelines for cancer screening in the general population. And even better, if you didn't get it, you can't pass it on. And so your children are not at risk and don't even need to be tested. And this is where the power of germline genetic testing comes in, because again, if we can identify a hereditary cancer syndrome in the family, we can let the children know if they did or didn't inherit it, then they will know if they're at risk or not, they can start that screening, we can hopefully keep them from getting cancer, and then when their kids are old enough, we can figure out if they've inherited it.

And you follow the gene through the family, and we call this cascade testing. You start with the people closest to the person who's tested positive, test them first, only proceed or cascade down to their children if they test positive. If they test negative, you don't need to cascade down to the children because there's no way they can have inherited it. Okay, so with that, as you're thinking about cancer genetics, the first thing we ask is when somebody is saying, "Could my cancer be hereditary?" We ask, at the time of their cancer diagnosis or over their life, if they've had more than 10 pre-cancerous polyps, yes or no. If no, we're going to think about some syndromes that don't cause tons of polyps, the number one one being Lynch syndrome. If yes, we have to start thinking about what kind of polyps have they been, the adenoma type, which are typically pre-cancerous, or the hamartoma type. And there are a number of syndromes that can run in families and cause polyposis.

These are much more rare conditions, the polyposis syndromes, but I will talk about the two most common ones today, which is familial adenomatous polyposis, MUTYH-associated polyposis. But we will start over here on the flow chart for family history where there's cancer without a ton of polyps, and we'll talk about Lynch syndrome. Okay, so Lynch syndrome can be caused by gene changes in one of four genes. And also there's a gene called EPCAM, right in front of MSH22. And if there's a deletion at the end of EPCAM, it causes Lynch syndrome by turning off MSH2. So it's really MSH2 causing it, but because of something upstream. So these are the four genes that can run in families and cause Lynch syndrome, they were all found in the mid-1990s.

And Lynch syndrome, again is the most common inherited condition or cause of colorectal cancer. It's also the most common inherited cause of uterine cancer, or endometrial cancer. And we think about one out of every 279 individuals has Lynch syndrome, which makes it quite common. And if you just map that out, that means there's actually over 1.2 million individuals in the United States who have Lynch syndrome. And we know it causes an increased risk for cancer. The interesting thing though is that these are very preventable cancers. If we start colonoscopy early enough and do it frequently enough, we should be able to hopefully prevent colon cancers, or definitely catch them early when they're treatable and we have good outcomes. With endometrial cancer, it can definitely be caught early, but it also can be prevented with hysterectomy.

And so if we know somebody has this condition, there's actually a lot we can do to help lead to a better outcome. So what's the problem? The problem is that we currently estimate that 95% of people who have Lynch syndrome have no idea that they have it. So we've not done a great job of getting the word out and getting people diagnosed so that they can benefit from all of this increased screening. And so that's why we like to spend a lot of time talking about this and recommending that people who have some family history get genetic testing to see if they have a hereditary susceptibility. Okay, so if you do have Lynch syndrome, you have increased cancer risks, and they actually vary depending on the gene. So MLH1 and MSH2 have much higher cancer risks than MSH6 or PMS2. And here you see those cancer risk by gene on the left side of the screen, and then the general public's risk for that same cancer.

So you may know that the general public has about a 4% lifetime risk for getting colon cancer, but if you have Lynch syndrome, it can be anywhere from 8% with PMS2 up to 61% with the higher-risk MLH1 and MSH2 genes. And so not all Lynch syndrome is created equally. And we actually give different screening recommendations now, depending on which gene is causing the Lynch syndrome. And I'm working with Fight CRC on a project right now to get ICD10 codes for Lynch syndrome, and we actually are asking for five of them, one for each gene to code it separately, including EPCAM, because we think that they're going to be treated differently in the future, and we need to know which type of Lynch syndrome a patient has.

The second most common cancer, endometrial or uterine cancer, the general public has about a 3% lifetime risk for getting uterine cancer. And the risk with Lynch syndrome can range from 13% with PMS2 up to 57% with the higher-risk genes. And increased risk for stomach and ovarian cancer now as well, but also a number of other cancers, hepatobiliary cancer. So we can see cancers of the pancreas or the bile duct, the gallbladder, small bowel, some cancers of the urinary tract, particularly the collecting ducts in the kidneys and the ureters, and unusual skin cancer of the sweat gland, called sebaceous carcinoma, and we have seen glioblastomas, but these are the main four cancers.

So what's the surveillance? The surveillance is obviously going to be increased colonoscopies. So in the general public, we know we now start at 45 and go every 10 years. If you have a parent, sibling or child with colon cancer, you're going to start at 40, or 10 years prior to the earliest diagnosis and go every five years, but if you have Lynch syndrome, if it's one of the higher-risk genes as the cause, you're going to start your colonoscopies at 20 to 25, and you're going to go every one to two years. If you have a gene change in one of the lower-risk Lynch syndrome genes, you actually are going to start at 30 to 35, and they now say you can go every one to three years. So I got to update this slide. But suffice to say, someone with Lynch syndrome could over their lifetime, say they do colonoscopies from 20 to 75, they could have 55 colonoscopies if they were going annually. And in that same time period, somebody in the general population might have three or four, and somebody with some family history who's going every five years, would have perhaps 10.

So a lot more colonoscopies, but obviously the goal here is to catch cancers at the polyp stage. And we know with Lynch syndrome, a polyp can progress to a colon cancer on average in about three years, whereas in the general public that takes on average about 10 years. And so there's this faster polyp-to-cancer sequence, and that's why we have to do our colonoscopies more frequently. For endometrial cancer, we just educate women about the symptoms, which is irregular bleeding or postmenopausal bleeding. A lot of women do consider having a hysterectomy once they're done having children and closer to natural menopause, because you can't get uterine cancer if you don't have your uterus. There is screening, it's an endometrial biopsy every one to two years, beginning at age 30 to 35. It does catch uterine cancers early, but we haven't seen actually a better outcome because they tend to get diagnosed early from symptoms anyway. So while it works, it doesn't really add a lot of net benefits, so a

lot of people don't do the screening, they just watch for symptoms until they're ready to have a hysterectomy.

And while endometrial cancer does tend to get diagnosed early and have a good outcome, ovarian cancer is really almost impossible to screen for. And because there is an increased risk for ovarian cancer with most of these genes, that's where the hysterectomy kills two birds with one stone, because it eliminates the risk for both of these cancers. And that's why many of the women with Lynch syndrome do consider this. PMS2 has gotten a little bit trickier, not a real lot of evidence for elevated ovarian cancer risk, which makes the hysterectomy more of a consideration and not a recommendation. For the upper GI cancers, we now recommend upper GI endoscopy every two to four years starting at age 40. For the urinary tract cancers, especially for MSH2 carriers, especially men with MSH2 carriers, or mutations, or people who have Lynch syndrome with a family history of urinary tract cancers, we recommend an annual urinalysis starting around age 30 to 35.

Pancreatic cancer screening is only recommended if there's been a pancreatic cancer in the family, but if so, starting at age 50, or 10 years prior to the earliest diagnosis in the family, we're going to do an annual MRI or MRCP or an endoscopic ultrasound just to take a look at that pancreas and see what's going on. While there are increased risks for prostate cancer, it's not any earlier, and so we just recommend general population screening. It's controversial whether there's any increased risk for breast cancers, and it's certainly not earlier, so we recommend general population screening. The brain cancer, there's really no effective screening, but if you're getting a physical once a year, they should just do a neurological exam during that physical. In reproductive risk, we do like to let people with Lynch syndrome know that if they feel strongly that they do not want to pass it on, there are some reproductive technologies like pre-implantation genetic testing, which can use in vitro fertilization, diagnose the embryos that have Lynch syndrome, and only transfer the ones that do not.

And so that is a technology that has become increasingly more available. It's still expensive, but has come down quite a bit over the last 30 years. And then we do let people know if they do have Lynch syndrome, and they happen to have children with somebody who has a mutation or gene change in the same gene as them, there is a low chance, but they could both pass the mutation onto their children. And anyone who inherits two mutations in the Lynch syndrome genes, one from their mom and one from their dad, has a much more severe condition called constitutional mismatch repair deficiency, which can cause hematologic malignancies and brain tumors and childhood, then all the same Lynch syndrome cancers, but starting much earlier, in the teenage years. And so that's something that some people who know they have Lynch syndrome, before they have children, will make sure that their partner has testing before they have children, to know if their kids are at risk for that more severe condition, constitutional mismatch repair deficiency.

Okay, moving on briefly to a couple polyposis syndromes, and these are just so much more rare than Lynch syndrome, we're talking less than 1% of colon cancer patients. And they're usually easily diagnosed because you can see all the polyps on the colonoscopy. So someone with classic FAP is going to have over 100 precancerous polyps or adenomas in their colon, and so you just know as soon as you get in with the scope. Now there are a lot of different kinds of polyposis now, so it's still important to do the genetic testing to prove what type of polyposis it is, and so that we know what gene to test the kids for, and the brothers and sisters, to see if they've inherited it, but these are some pictures of what that looks like.

And people with FAP also can get precancerous polyps in their duodenum. They get non-precancerous polyps in their stomach, fundic gland polyps, but they can also get precancerous polyps here. And it's hard to tell when you're looking at this many polyps, which are the ones to be concerned about and which are the ones to not be concerned about. It's important to go to somebody who has a lot of

experience with FAP for your endoscopies, because some people will get stomach removed because of this picture over here, when in fact these are not pre-cancerous polyps and they could have kept their stomach. So just always get a second opinion is a good idea. The risk for colon cancer with classic FAP is actually close to 100% by age 40, and that's because there are just so many precancerous polyps, that if you didn't know this and weren't doing colonoscopies, one of them would become a cancer. If we do know this, actually there's too many of them with classic to just do frequent colonoscopies, and people have to get their colon removed preventively, which is obviously a big surgery, and there's a lot of things to discuss there.

There is a weaker type of FAP called attenuated FAP, where sometimes people don't have to get their colon removed but can get by with very, very frequent colonoscopies, just depends on how many polyps they're making and if they can stay on top of it. FAP can cause other cancers like papillary thyroid cancer, medulloblastoma brain tumors, hepatoblastomas in young children under age five, it's a tumor of the liver, rarely gastric cancer if one of those polyps happens in the stomach. And then a lot of non-cancer conditions, and it is caused by the APC gene. And I just will point out you don't have to have a family history of FAP to get FAP, because 30% of the time this pops up new in either the APC gene you got from your mom or your dad, they didn't have a mutation, it happened new in you. And so you can have no family history and get FAP. And those are the cases that tend to get diagnosed later and based on symptoms, because they had no idea obviously, that they were at risk for this.

And of course we are going to do early-early sigmoidoscopy or colonoscopy starting around age 10, and again, appropriately timed colectomy in the early 20s for people with the classic. Then they're going to do upper GI endoscopy as well with side view to make sure to take a look at that duodenum. And if we know somebody has this as an infant, we'll even do some screening for hepatoblastoma. And then we can do thyroid ultrasound to watch for the thyroid cancers. And lastly, I just want to mention another polyposis condition called MUTYH-associated polyposis. So it turns out MUTYH mutations are very common in the general population. About one in 100 people has a single mutation in MUTYH. That does not cause MUTYH-associated polyposis. This is a recessive cancer genetic condition. So you have to get two mutations, one in the MUTYH gene from your mom, and one in the MUTYH gene from your dad to have MUTYH-associated polyposis. And it's a condition where we can see a small number of adenomas, like 30, all the way up to 100. So it's quite a wide variety of phenotype as we least like to call it. And so if you have people who have what you think is attenuated FAP or that lower polyp count, 30 to 100 polyp count, and when they test negative for APC, about 40% of the time it will be due to MUTYH-associated polyposis.

So I think the biggest deal with this gene is there are so many carriers out there, and the carriers may of course want to have their partner tested before they have children, so they know if their children are at risk for having MUTYH-associated polyposis. For a while we thought there was an increased risk for the carriers who had a single MUTYH mutation, but as increasing evidence is coming out, it's becoming clear that if there's an increased risk, it's very slight for those carriers, and they really are not recommending any increased screening for carriers anymore unless they have a family history of colon cancer, which is basically the same thing we recommend for anybody in the general population. So if you were tested in the last five years and had a MUTYH mutation and they had you doing increased colonoscopy, you may want to check back in because you may not need to do that.

So now that we can do these large multi-gene panel test where we can test all of the colon and polyposis genes in one fell swoop, we've got some nice studies where we look at, say, here's one where they looked at 1,058 colon cancer patients, and they all got a multi-gene panel test for 25 cancer genes, about 10% tested positive. If we do more genes, if we do about an 81-gene panel, we'll see this get closer to 15%. Pretty consistently, three to 4% of colon cancer patients will have Lynch syndrome, so we

talked about that being the most common. The remaining cases, so in this case, if you tested 25 genes, that would be 7%, if you tested 80-some genes, this might be 12%, they're going to have non-Lynch syndrome-gene mutations, including mutations in high penetrance genes like we just talked about, APC, the MUTYH. We also find mutations that may not have been related to the colon cancer, but do increase their risk for other cancers, like BRCA1 and 2, and PALB2. And these are important for the patient to know for their cancer risk, and for their family, even if it didn't have anything to do with their colon cancer risk. And then a large number of them will have mutations in more moderate-penetrance colon cancer genes.

So in the past we would call those carriers of MUTYH into this category, now they're more falling into the low or no increased risk, so they might fall out. There's a mutation in APC that's common in Jewish individuals that does not cause polyposis, it just doubles the risk of colon cancer. That's always common when we test a large number of people, because 10% of Jewish individuals with colon cancer have this mutation. And that's treated very differently than the FAP that I just talked about. And age at diagnosis, family history of colon cancer did not significantly predict who was going to test positive for the non-Lynch syndrome genes, which is interesting. And we're sort of moving to this era where we're starting to consider should we just be offering germline genetic testing to every single patient with colorectal cancer, or any cancer for that matter, because there's a 10 to 15% chance you'll test positive, and we can't always guess who's going to test positive based on the family history.

So take-home messages for you from this germline or genetic testing component of the talk is that the major risk factors for having a hereditary cancer syndrome are earlier ages of diagnosis, more than one cancer in the same patient. If you have three cases total on the same side of the family, or more than 10 colon polyps, those are big risk factors, that you might want to see genetics. And germline genetic testing can be helpful for three reasons. It could affect your cancer treatment. So it can sometimes give your doctors ideas for ways to treat your cancer, and it can help determine if you are at increased risk for getting other additional cancers in the future and need intensive surveillance and prevention to prevent those. And then it also can help you to determine whether any of your family members, so brothers, sisters, sons, daughters, even out to aunts, uncles and cousins, have also inherited that increased risk for cancer, so that they can get intensive surveillance and prevention and hopefully we can keep them from getting cancer in the first place. And I think Dr. Benson will now tell you why somatic or tumor testing or biomarker testing can also be helpful, and that really is primarily for the therapy decision-making. So thank you very much. And I will now stop sharing and turn things over to Dr. Benson.

Zac Getty:

Thank you, Heather. I'm going to share my screen. And Dr. Benson, I'll let you introduce yourself a little more thoroughly than I did earlier and go over biomarker testing.

Dr. Al Benson:

Great, thanks Zac, and thanks Heather, and thanks all of you for joining today. My name is Al Benson, and I'm a GI medical oncologist at Northwestern University in Chicago. Besides my work at the Robert H. Lurie Comprehensive Cancer Center at Northwestern, I work with a variety of national organizations, including the National Cancer Institute Cooperative Groups, in particular the ECOG-ACRIN Cancer Research Group. I do a great deal of work with cancer treatment guidelines, including my association with NCCN, ASCO, and in Europe, ESMO. And also I do work in the area of health policy, and have worked with various patient advocacy groups, including by Fight Colorectal Cancer.

So today we're going to expand upon a topic of biomarker or tumor testing. So these biomarkers can include proteins which are made in response to the cancer or changes known as mutations in the DNA of cancer cells. And these markers are derived from tumor cells or DNA in the bloodstream. So as I'll mention, we can test these biomarkers in blood as well as from the actual tumor. And while these biomarkers may involve changes in the DNA of your cells, getting your biomarkers tested is not the same as having your genetics tested, as Heather has very nicely explained. Genetic testing looks for mutations inherited from your biological parents, as Heather had reviewed, that may predispose you to certain cancers, but we emphasize that the biomarker testing I'm addressing looks specifically at your tumor cells to determine their characteristics.

So who needs biomarker testing? So all colorectal cancer patients need biomarkers tested. However, not all patients will need the same testing performed. The biomarker that your physician will decide to test for will be determined by the type of cancer you have and the stage that you are diagnosed. We emphasize that biomarker testing is now standard of care, and all CRC patients should be tested for their mismatch repair status, known as MSI-H, or deficient mismatch repair. Heather spoke with you about Lynch syndrome, and these MSI-H tumors may be present in Lynch syndrome populations, but they also may be sporadic. It's estimated about 15% of people with colorectal tumors have MSI-H tumors. And so this should be universally tested. And fortunately now many pathologists are routinely reporting the MSI status on the initial pathology report. So if we look at the various stages of colorectal cancer, for stage 1 tumors, for example, really here, the most important biomarker is looking for MSI. And in particular, if it is an MSI tumor, we are concerned that someone may have Lynch syndrome, and so germline testing becomes very important.

For stage 2, again, MSI, there's also a blood marker, CEA, which can be produced by the tumor. It also can be elevated for other reasons such as inflammation, cigarette smoking, endometriosis, but it's something we do look for. Also, there may be a role for looking at what's known as circulating tumor DNA, so we can do blood testing and actually identify fragments of DNA from the tumor. And if there is presence of such after surgery, we're very concerned that an individual is at very high risk of recurrence, and that may drive a treatment decision. For stage 3, it's quite similar, MSI, CEA. And on occasion we might look at ctDNA. I should mention that ctDNA testing is now the subject of many clinical trials because there's much more information we need to gather about this particular technology. For metastatic disease however, there's much more testing that we need to evaluate. And so typically we do what's called next-generation sequencing that actually can test for hundreds of different mutations. However, we are most interested in just a few including RAS mutation, BRAF mutations, HER2 amplification, and NTRK fusions.

So what do your biomarkers mean? Well, they can tell us a great deal about your cancer. They can tell us if you belong to a specific subset of a given cancer. And more and more we view cancers, including colorectal cancer, not as just one entity, but divided into different groups depending upon their biomarker testing, and this can have significant importance in choosing a type of therapy. Many of these can be prognostic, so we get a hint as to how well a person may do over time. Some can predict if your treatment will respond to a given therapy. This has been much more of a challenge, identifying predictive markers as to who will or will not respond to a given therapy.

And this is very much an evolving field. So participation in clinical trials that are inclusive of biomarker testing is really important. And on your left you can see this is an example of a report which may include a whole variety of different mutations or other genomic changes that may not directly apply to you, but these reports will highlight the most important of these. So for example, on this report, it mentions that this individual has wild-type KRAS, meaning there is not a mutation. And that's important to know,

because on the right side here, it lists particular therapies that may be efficacious for the individual with a wild-type RAS tumor.

So what do they mean? Well, there are many different biomarkers that have been studied in colorectal cancer, some of these are more common than others, and not all biomarkers need to be tested in every situation. So as I mentioned, for people who have early-stage colorectal cancer, these biomarkers for the most part will not direct a specific treatment. Although having said that, we are looking at such in clinical trials currently for people, for example, who have BRAF mutations. Many, as I mentioned, will be tested for people with stage 4 metastatic disease, because there, some of these choices of treatment based on the biomarker status are very important in terms of treatment for the individual. And so what we've listed here are some of the more common markers that we test for and can help guide therapy. And if you visit the Fight Colorectal Cancer website, there's more information about these very specific markers.

So how do you know if you've had biomarker testing? Well, you certainly must have a discussion with your healthcare team to determine if biomarker testing has been ordered or will be performed, and when the results will be available. Some testing, such as MSI testing, can be done quite rapidly. And as I mentioned, many many pathologists now are routinely reporting MSI status when they issue the pathology report from either a biopsy or the tumor specimen obtained at the time of surgery, but many of these others will take much longer. This is very complex technology, and it can take two to four weeks before we receive the report. If you know your biomarkers have been tested, but unsure of the results, make sure to discuss this with your clinical team so you understand what has been tested, why it's been tested, and how such testing may affect your care. And this is something even before surgery, you can ask your doctor if biomarker testing will be performed.

There are, as I mentioned, multiple biomarker tests available. And as I mentioned, these can be done on your tumor, which is more typical, or from your blood. So if we do a single biomarker test, this will look at a single gene, multi-gene panel tests look at multiple genes. There's also what's known as whole-exome sequencing, which looks at all the protein coding regions of genes in your cancer's genome. So whole-genome sequencing, such as what's referred to as next-generation sequence or NGS, looks at all the DNA in your cancer. This allows for multiple genes to be tested at the same time. And as a result, as I mentioned, your NGS report may list hundreds of genes that were tested, most of which however, will not be what we call actionable, or in other words, will not affect your treatment or a particular identifiable risk.

There's also what's known as tumor mutation burden that looks at the total number of genetic changes in your cancers. Although, what's known as TMB can be important for some cancers, thus far in colorectal cancer, it has not been a particularly helpful test and we don't routinely recommend that TMB is evaluated. I will say though, for people with MSI tumors, their TMB can be quite high. However, the decision about treatment opportunities related to MSI testing is really on the basis of the result of the MSI test or the DMMR test. Next. So when we talk about testing, I mentioned the tumor biopsy, or actually the entire tumor specimen from the surgery, or what's known as liquid biopsies, which it simply means testing from the blood. And it's a step-by-step process of requesting, obtaining the tissue, sending to the lab for testing. And I will say that growing numbers of centers are doing their own NGS testing, but there are also a number of companies where either the blood or tumor specimen can be sent and then analyzed, which we hope will result in more personalized treatment. And this is what's also referred to as precision medicine, where we're identifying these very specific genomic changes for which there may be a specific treatment.

So a tumor biopsy is a surgical procedure in which a piece of your tumor is removed and then examined by the pathologist to determine among other things the relevant biomarkers in your cancer. These

tumors may be performed, for example, more and more commonly by an interventional radiologist who can use a needle and identify the tumor, for example, under ultrasound, and obtain a piece of that tumor, which is then analyzed. They may also be incisional, where a sample of your tumor is removed, often by a surgeon. So for example, perhaps a liver tumor may or may not be done surgically or by a interventional radiologist. The interventional radiologist may not only do what's called a core or larger sample from a larger needle, but also what we call fine needle aspiration, where a thin needle is inserted into the tumor, cells from the tumor can be acquired and evaluated under the microscope. Now, when a person has tumor testing by the pathologist, these tumors are stored typically for years and years. So we can actually go back to a person's previous tumor sample and do a number of tests, including biomarker testing. And this is a technique that's often done.

So, as I mentioned, the liquid biopsy is from a blood sample, and they primarily look for two things. One, as I described, circulating tumor DNA, which are segments of cell-free DNA released by tumor cells. But there's also the ability to look at actual circulating tumor cells, and these are whole tumor cells that are circulating in the bloodstream. By far the most common tests we do is looking at circulating tumor DNA, and they are different, but they are complementary tests. So the circulating tumor cells may be single or groups of whole but intact cancer cells that are shed by a tumor and actually enter the bloodstream. And now with modern technology, we can actually locate these tumor cells. They can be detected and counted by just simply drawing a blood sample, and they can provide a more complete picture of a tumor's characteristics than we may acquire just from circulating tumor DNA. The circulating tumor cells also can be used to detect biomarkers.

And also, there have been studies that look at circulating tumor cells throughout treatment that serve as a predictive indicator of not only the prognosis, but in some cases whether an individual is responding to treatment. Not all tumors will necessarily shed circulating tumor cells, but they can be used in addition to ctDNA testing perhaps to get a better picture. This is currently an approved test, but as I mentioned, for the most part, when we order what's called a liquid biopsy, we're looking at circulating tumor DNA. So the circulating tumor DNA, as mentioned, is cell-free. So these are just fragments of DNA that's shed by dying or damaged tumor cells and they're released in the bloodstream.

Like circulating tumor cells, ctDNA is just a simple blood draw, and it too may allow us to look at biomarkers. And more and more we are able to identify the critical biomarkers just from looking at ctDNA. Also, ctDNA may have important prognostic information. So for example, if a person has had a successful surgery and there's no obvious tumors seen on CT scans, for example, but there is ctDNA present, that is a worrisome signal that tumor cells remain, and may inform a treatment decision. There's a growing number of companies now that are testing for ctDNA. And ctDNA testing is certainly a work in progress, and we're doing a number of clinical trials to get a better understanding of how best to use this technology to help our patients.

Both of these type of tests can be performed in the doctor's office and sent to a lab. As I mentioned, some centers are doing this within their own laboratories, but others are sent out to commercial companies. After analysis, the clinician will receive a full report. And in addition to providing the biomarker status, we can get additional information such as proof that there may be what's called minimal residual disease, which means that there is presence of tumor at least somewhere, which may be simply circulating tumor cells, which over time may locate in a specific organ and begin to grow. So that concludes my presentation, giving you a quick review of technologies that can give us very critical information about your tumor and inform us as to potential treatment opportunities.

Zac Getty:

Thank you, Dr. Benson. Thank you, Heather, as well. We really appreciate you sharing your time today. I know we are at time. I do have one question, if you're able to answer it, it's directed for Dr. Benson, but I do not want to keep you too long here. We have a question about biomarker testing being standard of care. And the question is, "Out of the roughly 150,000 new CRC cases diagnosed annually, do you have an estimate of the percentage of US patients that get their biomarker testing completed?"

Dr. Al Benson:

We know that it is improving, it is not where we need to be, it does vary across the country. For example, there are some centers where nearly 100% of people are tested, and other places where it is less so. And that's why we are certainly emphasizing in cancer treatment guidelines that are used by clinicians, the importance of biomarker testing. We are urging clinicians to participate in clinical trials which include biomarker testing. And certainly we want individuals to routinely speak with their clinicians about whether biomarker testing has been done and which tests have been ordered. So it is improving, we're not where we need to be. It's very encouraging, as I mentioned, that now more and more pathologists are routinely reporting MSI status, which really needs to be done on every single colorectal cancer patient. So the trend is in the right direction, it's just not where we need to be.

Zac Getty:

Thank you. And then Heather, I do have a question for you as well, if we have time, "What do you do when you have a family member mixed with a Lynch syndrome, what guidance do you give for surveillance if you have a family member known with Lynch?" You're muted.

Heather Hampel:

With Lynch syndrome we talked about, that's the colonoscopy every one to two years starting at 20 to 25 if the mutation's in MLH1 or MSH2 or EPCAM, and every one to three years starting at 30 to 35 if the mutation's in MSH6 or PMS2. I'm seeing here though in that question something about familial type, maybe X. So there are families that look like they have Lynch syndrome, but the tumors don't have microsatellite instability, like Dr. Benson talked about. And we call those familial colorectal cancer type X, because we don't know what's going on, they appear hereditary, but it's not Lynch. So what is it? We're not sure. So until we figure it out, we're calling it type X. And those families, we typically do an intermediate screening starting at around 35 and going every three to five years. So less than Lynch syndrome, but more than a first-degree relative with colon cancer right there in the middle. So typically what's done with those. hopefully that helps with that question.

Zac Getty:

Thank you so much. So we are past time, I do not want to keep either of you much longer. I do appreciate you sharing your time with us today. Real quick, I just want to point out that Fight CRC produces a biomarker brochure that outlines information your biomarker report should contain, and gives more reasoning why biomarkers testing is so important when looking at your treatment options. You can download this brochure for free at fightcrc.org/resources, or request a physical copy and we'll get that in the mail for you. It includes a biomarker passport card, which is a punch-out card that allows you to mark down your known biomarkers, the date of testing, and where your tumor is stored, so you just have that information if you switch doctors or looking for a second opinion or anything like that. We also offer an Understanding Your Genetics brochure, which does not go quite into the depth that Heather went into in her presentation today, but we'll help you understand your genetics and

understand your risk and options, and just a little bit more information about genetic testing in colorectal cancer. Again, you can download that brochure for free or request a copy for free as well.

I do want to take a second to thank both Dr. Benson and Heather for sharing their time with us today. Thank you so, so much for joining us for this early-morning webinar. I would also like to thank the supporter of this webinar, Menarini Silicon Biosystems for supporting our mission in educating patients and caregivers. I like to end each webinar with our mission statement, Fight Colorectal Cancer fights to cure colorectal cancer and serve as relentless champions of hope for all affected by this disease through informed patient support, impactful policy change and breakthrough research endeavors. If you had a question, you didn't get a chance to answer it, please feel free to reach out to me directly, Zac@fightcrc.org, and I will do my best to get you an answer. And keep an eye on your inbox, you will get a link to this recording once we have it edited and up on the website. Again, thank you, Heather, thank you, Dr. Benson, thank you for sharing your time, and thank you for everyone that attended today. And we will see you later. Have a great day.

Heather Hampel:

Thank you.