Pam Barrett:

SLIDE #1 Hello, everyone. Thank you for joining us for this Us TOO University Patient Education Web and RN Teleconference on Understanding Diagnostic Testing for Prostate Cancer Patients. My name is Pam Barrett, and I am the Director of Development with Us TOO International. I would like to recognize and give special thanks to our sponsor tonight, (Verdex) for making this event possible.

SLIDE #2 In reviewing the agenda for today, the core presentation will last approximately 40 minutes. Then we will have about 15 minutes for questions from the audience afterwards.

Today's presentation will cover the tests and tools used along the entire prostate cancer journey, such as to confirm an initial prostate cancer diagnosis or monitor the PSA post-treatment or while on hormone therapy or if one becomes hormone refractory or castrate resistant.

SLIDE #3 Now I am very pleased to introduce our speaker for today's presentation, Dr. Manish Bhandari. Dr. Bhandari is a practicing oncologist in Cincinnati, Ohio, and focuses on genital urinary cancers, including systemic therapy for prostate cancer. He did his medical school training in Boston at Harvard Medical School at oncology training at the University of Michigan in Ann Arbor. And now for our feature presentation, thank you for joining us. Dr. Bhandari, take it away.

Dr. Bhandari: Thank you, Pam. Good evening, everybody.

SLIDE #4 It is my pleasure to spend this hour with you talking about a very important topic for both patients and the care providers about the use of diagnostic testing in the prostate cancer journey. I understand a lot of you, as this cartoon displays its…I've been diagnosed myself on the Internet and am here only for a second opinion. But on a more serious stream, I am very happy to run through this pack of slides and important data concepts and welcome questions that I will try to answer at the end of this presentation.

SLIDE #5 What we want to talk about today is various types of testing from biopsies to blood markers to scans and other tests that get utilized, which help navigate this journey for patients and their physicians and help us manage the disease better.

SLIDE #6 The tests can be utilized at initial diagnosis to monitor the success of the treatment that is given or active observation, active surveillance as it is often called. And more importantly, along the journey to understand how far and confirm that the disease is in remission or if it's not.

SLIDE #7 The tests actually fall into these main categories. The tests for diagnosis—initial diagnosis could be fluid measurements such as PSA, which is the most commonly utilized; a physical exam is a very important part of the testing, in which as I will talk about briefly the direct, digital rectal exam plays an important role; tissue sampling to make a diagnosis to understand grading and prognosis based on the pathology, for example is used; as well as imaging and various modalities to help answer specific questions about extent of disease or response to therapy.

SLIDE #8

Here for example at the time of initial diagnosis, we see that often a rectal exam is performed because the prostate sits very adjacent to the rectum, as shown on the left side, where the finger has gone into the rectum and the physician is feeling a nodule. But as many of you know, unfortunately the entire prostate cannot be felt through the rectal exam. So while this is a very useful test, unfortunately half the nodules that are cancer cannot be felt by the DRE. And DRE hence is useful but not complete assessment or doesn't make the diagnosis all the time. For example, it shows when a lesion may be small, like T-1, often it is within the capsule, within the prostate, and does not create an indentation of the rectum and cannot be palpated. But equally importantly when it is locally advanced like in T-3 or T-4, that can be very helpful to make a diagnosis.

SLIDE #9

PSA is one of the most heavily utilized, controversial and debatable tests in all of prostate cancer, as many of you know. It is actually a glycogen protein, glycoprotein only secreted by prostate tissue. No other tissue in the body except the prostate makes PSA. And the problem is that not just cancer but normal prostate also makes PSA. Usually levels of PSA between 4 and 10 only have about 20 percent chance of being cancer; however, if they are above 10, it is a two-thirds chance that that PSA excess is coming from the cancer of a prostatic origin. Equally importantly is the fact that 25 percent of men with cancer may not have an elevated PSA. And the extreme, very rare but very aggressive tumors may not make any PSA. So while PSA is helpful, it is not the only test. It is a helpful test, but not the only test, as many of our participants this evening are aware about.

As many of you know, PSA is actually bound to protein in the blood. However, some of it is free floating, and free floating PSA or the free PSA is also measurable. Most men have a low fraction of free PSA. However, when that fraction becomes high, it is less likely associated with cancer; but when it gets very low, it is more likely associated with cancer. A free PSA test may spare an unnecessary biopsy, but this really has to be taken in the context of a lot of other parameters, including the rate of rise of PSA. So it is really best discussed with your physician or urologist about the utility of free PSA and whether that should be used in making a cancer diagnosis and proceeding to biopsy. Also importantly, I know that there is a lot of discussion about PSA. And while it is the same test, a PSA that is tested initially for making a diagnosis for primary screening has very different issues and utility parameters than when it is utilized after patients have had therapy to monitor the disease. And it has very different utility to understand if people are responding to therapy if the PSA is elevated. So even though you might here controversy in the press that a PSA is a useless test, you have to really put that in the context. Very often the controversy is about the utility of PSA for initial screening. And the same test but applied later for disease monitoring has very different parameters and implications and can be a very useful test. So please bear that in mind.

SLIDE #10

I am not coming on to another very useful score or testing modality, which is the Gleason score. As many of you know, a biopsy is obtained. Several tissue cores may be obtained from different sextants or parts of the prostate gland. And a Gleason score is calculated based on the initial …development of the score by Dr. Gleason and is a very fundamental, useful score because it has implications on initial what to

do. Should we just simply monitor for more (indolents) a low Gleason scores. Gleason usually runs from 2 to 10. Or a high Gleason score has implications on the overall life expectancy of the patient. Gleason is determined by the sum of two different numbers. The first number indicates the aggressiveness of the type of cancer cell that's in the most dominant or most numerous part of the tissue. And the second number indicates the second most numerous part of the cancer. Those two numbers are added to give a Gleason sum or a Gleason score. And hence, the Gleason that you see is the summation of two different numbers.

SLIDE #11

A Gleason score from 2 to 4 denotes low, aggressive tumor that can often be actively surveyed or followed. A Gleason score of 5 to 6 is a mildly aggressive cancer. Gleason 7 is more moderately aggressive. And 8, 9, and 10s are felt to be very aggressive. Also note that different histologies can be combined to give you a summation. So 3 plus 4 may be a Gleason 7, and that is not the same as 4 plus 3. Usually 3 plus 4 Gleason 7 will be a little more indulent disease than 4 plus 3 because the first number denotes the majority, the most predominant histology in the biopsy.

SLIDE #12

Initially at the time of diagnosis, staging is also undertaken, and often we hear the term Stage I, Stage II, Stage III. For prostate cancer, what does it mean? Stage I is usually cancer that is only confined to the prostate gland and is usually small enough. Like I showed you in a previous slide, it cannot be palpated with direct digital rectal exam. Stage II is when it is only far in the prostate but big enough to palpate. Stage III is when it is not just confined to the prostate but yet has not metastacized outside of the pelvic area, such as may have spread to the nearby seminal vesicles. And Stage IV usually denotes that it has gotten outside the gland and has metastacized or spread to distant tissue, which is bone metastasis.

These next few slides are more conceptual slides to make us understand better what is the life cycle over the entire course of prostate cancer so we understand the terminology better. Very important to understand is that not every patient will go through all these stages. As a matter of fact, a lot of patients will initially get the initial treatment and be cured and never have to face the further aspects of this disease. But if we were to take the presumption that the entire biology…if we look at the entire life cycle, then what we see is that initially when we are diagnosed either because of a nodule palpated under rectal exam or elevated PSA, initially patients undergo testing and get local initial therapy, whether it be prostatectomy or external beam radiation or radiation beads, as may be relevant and decided by the patient and their physician. And that's called initial local therapy for local treatment.

SLIDE #13

If that treatment unfortunately does not life long control the disease, very often what we find nowadays after a variable interval of time is the PSA starts to rise. And that is often called biochemical progression or PSA relapse or biochemical only disease, which is when the PSA starts to show a trend of rise. Very problematic in this area is that not all PSA rise is absolutely diagnostic of cancer recurrence because sometimes, for example, if there is some viable amounts of prostate tissue left, you could have some PSA rise because of normal prostatic tissue growth. And this really should be discussed with your physician and various

parameters, including what was the Gleason initially if you went for surgery and if all the margins were negative, how long after initial treatment did we obtain nadir, and what was the rate and how long after initial therapy was the PSA rising? All of these parameters, including at that point….what the rate is a term that we will talk about later again, PSA doubling time. All of those get utilized to really see if there is evidence of relapse or biochemical relapse by PSA criteria. And as I mentioned earlier, the PSA—the same test that is used initially for screening has very different scientific parameters and validity when it is used for initial screening as opposed to when it is used for surveillance to see if the cancer has come back.

SLIDE #14

But just to make the concepts correct, I introduce this slide that shows tumor burden. And then you see after the initial drop that the PSA went down, the tumor burden went with initial therapy. When the PSA starts rising but there is no other detectable sign of cancer such as through bone scans, CAT scans, whatever radiographic modality we use—when the PSA is the only marker of disease coming back, that is called biochemical progression of PSA relapse.

SLIDE #15

Then we introduce the term that is called hormone response. Very often if the PSA starts to rise, testosterone or the male hormone is very important and usual growth promoter for prostate cancer. And if therapy such as androgen deprivation or androgen suppression is introduced, such as with a (GNRH) injection, you will see the tumor burden decrease and the PSA often go down. That phase of disease when there is only biochemical progression, no growth, metastatic disease visualized on any scans—that is called PSA progression. PSA progression can be controlled when you introduce hormonal therapy. Invariably at some time, very often what happens is that over time the hormonal control of the cancer starts to fail. And you could have progression of disease. That may be still with biochemical only progression or may be with actually growth disease visualized on a scan, such as a bone scan.

SLIDE #16

That stage of progression is called hormone refractory disease or androgen-insensitive disease when hormonal maneuvers such as gonadal suppression, GNRH suppression doesn't control the disease. That term such as hormone-refractory disease or androgen-insenstive disease is introduced.

SLIDE #17

Again as I mentioned, this may be biochemical only, such as when the PSA starts to rise again after a time interval when the PSA becomes undetectible due to hormonal therapies such as injections of Lupron, Zoladex, etc. Or it may be hormone refractory or androgen insensitive with the presence of organ or bone metastasis. Most often, the majority of the time, prostate cancer is bone seeking and there will be evidence of bone metastasis, but not always. Pretty much the scientific community now is trying to get away from the term of hormone refractory and instead preferring the term androgen insensitive or castrate resistant as the more appropriate medical terms.

SLIDE #18

Ultimately at some point, you start to visual the presence of cancer within organs, such as can be detected by bone scans, x-rays, CAT scans. And that used to be called D3 disease, D2 disease, metastatic disease. We have gotten away from the D staging, and we pretty much call that Stage IV, metastatic prostate cancer.

SLIDE #19

Ultimately after some interval of disease control, which can be highly variable but at some point that hormonally responsive disease, even metastatic disease, unfortunately starts to not remain hormonally responsive and can progress further, where chemotherapy is being studied and is currently utilized for management of growth of metastatic disease.

SLIDE #20

So in monitoring disease through all of the life cycle from initial diagnosis to response to initial therapy, whether it be surgery or radiation or bead implants or anything else that is undertaken, to subsequent management of when the disease has come back, and then how well the disease responds to the next line of therapy, such as either salvage radiation to the prostatic bed or use of hormonal therapy, these various diagnostic tests become useful, PSA obviously being the most common one utilized. And after disease relapse for quite a while PSA becomes very informative in understanding the biology of disease and response to therapy. Again, those have to be used in context of how aggressive the disease was, how organ confined it was, and individual situations need to be best discussed with your own physician.

SLIDE #21

A closer look at some of the tools—we use imaging such as scans to check the extent of threat of cancer to adjacent tissue, such as lymph glands, lymph nodes, as well as to bone. PSA, which we all know about. Also very important modalities in testing is the amount of testosterone and dihydrotestosterone that is a growth promoter for cancer, particularly prostate adenocarcinoma. Other cellular assays or molecular analysis may be undertaken. And a big area of research is trying to understand the gene expression of individual tumors. But that is still in the research arena and not everyday common practice to submit tumor for genomic analysis. But I visualize the day where beyond just Gleason scoring, we will enter the era where we will be able to submit one patient's tumor for more accurate genomic analysis to understand what genes are up or down regulated so that we may more intelligently predict biology and equally importantly be able to determine what is the best modality of therapy initially based on genome expression and genomic profiling. That is coming around in some other cancers such as breast cancer, but we are not there yet in the arena of prostate cancer. Lastly, another interesting and very useful modality, particularly when people start to have metastatic disease, is a diagnostic tool called a circulating tumor cells that are becoming more common practice in medical oncology but not useful and not utilized yet for the initial diagnosis of cancer. And I will have a few slides to talk about circulating tumor cells as well.

SLIDE #22

In terms of imaging or x-rays, there are various types of x-rays utilized—magnetic resonance imaging or MRI is particularly good at imaging soft tissues, such as muscle, ligaments, lymph glands, and is often utilized initially maybe to look at if the cancer has broken through the prostate capsule and if the lymph glands around the prostate show signs of growth or abnormal size that would make a surgeon worry that the cancer has already spread outside the prostate, in which case undertaking surgery may not really best control the disease. So MRI is utilized for answering very specific questions like that. CAT scans are also utilized, but they do not image soft tissue as well as MRI does. Ultrasound can also be utilized, which you have found ways to (extent demarked) disease or abnormality. Ultrasound is not…it is great for

localizing an image grossly of the prostate and where for example biopsies should be done to cover all areas of the prostate on both sides of the gland. But ultrasound is not that useful as MRI to look at the lymph nodes around the prostate. So again each test has its positives and its negatives as to how much information it can provide, as well as how aggressive the testing is. So ultrasound, for example, is a great test (and a rectal) sometimes to help the surgeon localize which parts of the prostate to biopsy, but it is not that helpful as MRI to see the characteristics and size of the lymph glands around the prostate. PET scan is an up and coming modality and very useful. More studied and used for more distant disease to make sure that the cancer has not gone outside. But it has limited utilized initially currently in initial prostate cancer diagnosis. Particularly low grade tumors such as Gleason score 2, 3, 4 often can come negative in the PET scan. PET scan uses an injection of glucose that is radiolabeled. Tissues that are fast growing, such as aggressive tumors, but not just tumors—abcesses or infection or inflammation, can also light up on a PET scan. Last but not least is a modality which is often used called a bone scan where a tracer or material that particularly deposits in bone is injected. Then images are obtained after a time interval. And for example, the image that is shown in the bottom box shows in the pelvis the areas of bone where the arrows reveal that the uptake is more on that side of the pelvic bones than on the other side, demarkating that there is some abnormality and higher metabolic activity within the bone. Now again, not all uptake is from cancer. For example, arthritis and joints or trauma or injury from whatever cause can also light up on bone scans. So it all has to be put in clinical context. But bone scan is very useful to ask some of these questions and to check if the cancer has already spread to bone.

Some of the tests that used to be used that are not on this slide, such as the prostacin scan, which is a PSA honing scan, have a lot of false-positives and false-negatives and has fallen out of favor. But there are some centers still that utilize it. So I will not critique and talk extensively about prostacin scans, but they are in all the literature and not much utilized now because the test hasn't been that useful in most centers.

SLIDE #23

A quick word about testosterone and DHT. Prostate cancer cells, most of them, are driven by what is called the androgen receptor, which binds to testosterone or dihydrotestosterone. And these two male hormones are like fertilizers that make the seeds of the prostate cancer grow. And very often treatment attempts to bring the levels of these two hormones way down. But a lot of the side effects of hormonal therapy from fatigue and bone mass loss and loss of sexual function, anemia—a lot of side effects are driven by the loss of these two important hormones as part of suppressing testosterone. Testosterone, particularly in the prostatic tissue, is converted to the more active form, dihydrotestosterone, DHT. And these tests are also utilized both to obtain a baseline level to assess efficacy of hormonal therapy. And then very often when sometimes you do a stop-and-go approach, what is called intermittent androgen deprivation to minimize the side effects of bringing the testosterone levels down…so sometimes we give hormonal therapy for awhile, get the PSA to undetectible, and then patients go off hormonal therapy for awhile. These hormone levels are useful to monitor the state of the gonadal axis as it is called, to see if patients really are in an endocrine-suppressed state or a relapse state.

SLIDE #24

As I had mentioned already, but PSA is very helpful for monitoring disease over time to make sure that there is not biochemical relapse or metastatic disease that has come back. If a PSA continues to rise, it is very important to follow up with your physician. Just a single PSA rise is not diagnostic of cancer recurring. But the pace of rise and another term which I will talk about—PSA doubling time—becomes very important at that phase of disease. As I mentioned, other things beyond cancer, such as prostatic tissue that has been left behind that starts to grow or inflammation can also lead to a rise in PSA. And that is what makes the management of prostate cancer fairly challenging.

SLIDE #25

One concept I talked about was PSA doubling time. It is simply the time interval it takes for PSA to double in value, for example go from a value of 1 to 2 or 2 to 4. PSA doubling time is very important, specifically when we are monitoring recurrent prostate cancer. A doubling time that is below 3 months, that means that every 3 months the PSA is doubling, is a harbinger of more likely recurrent disease and aggressive disease with a poorer outcome. And a PSA doubling time over 15 months has been validated as a more favorable type of recurrent disease. Then a PSA doubling time that is in-between those two intervals isn't immediate. Other parameters such as how long after initial therapy the PSA came back, what was the initial Gleason score—those kind of parameters also are very useful in overall determining the prognosis or outcome.

A very useful tool that is available on the Memorial-Sloan-Kettering website to plug in PSAs and dates when they were obtained to more accurately determine the doubling time is available on the Memorial-Sloan-Kettering website. MSKCC.org. They actually have a search box on the main page. If you simply type in "PSA doubling time", or if you go to common search engines on the Internet, such as Yahoo!, Google, and type in "Memorial-Sloan-Kettering" and PSA doubling time tool, you can come up with this tool box that is very helpful in determining the doubling time. Doubling time is sometimes initially problematic because not every PSA doubles at a constant rate. Sometimes you can have PSAs that plateau and even drop before they rise again. So it is not simple plotting of months. You have to put in multiple values and the doubling time is more accurate not just from…you need a minimum of 3 measurements. With two you cannot determine the doubling time. You at least need 3 different PSA values over a month a part or a longer time interval apart. But then the more values you have, the more accurate the doubling time will be. And very often initially when the PSA starts to rise, this may be an important reason why the physician involved in your care might not automatically start some therapy but will want to wait for a couple of time measurements of PSA to more actively determine the PSA doubling time.

In the time interval that we have left of about 10 minutes, I wanted to also take some time to discuss something that is less discussed in most prostate cancer support groups I have gone to and is a very useful new test, especially in the medical oncology area when people have presence of more advanced Stage IV disease, what is called the circulating tumor cell. So I do appreciate that from the little that I have presented there are a lot of questions you might as an audience have about the dynamics of PSA, PSA doubling time, what should you do based on that value. And sometimes it is hard to just make a blanket recommendation because it is specific to an individual's risk parameters. At least through this I hope

I can at least search on some of the terms that are utilized in everyday practice so you can have a base for asking those questions from your physicians.

SLIDE #26

So what is a circulating tumor cell? Circulating tumor cells is actually measurable, visualizable cell that has broken away and detached from the mass bulk of the solid tumor and invades to the basement membrane of wherever the tumor is sitting and enters circulation into the blood stream and starts to enter circulation. Believe it or not, we've known about circulating tumor cells for over 100 years when physicians particularly with melanoma, when patients had very advanced melanoma, took the patients' blood and studied that under the microscope and they actually found dark, black, ugly-looking cells in circulation just because melanoma makes a lot of melanoma black pigment. Even before the era and arena of modern, scientific tools people were able under the microscope, looking at a slide or a film of blood, be able to see these cancer cells in circulation. So circulating tumor cells have been known for a long time. But how to scientifically utilized a quantitative methodology when they are present and understand what do they mean over response to therapy or what do they mean for the life of a patient has only come about in the last decade or so, particularly for prostate cancer, the last 5 years.

SLIDE #27

Why circulating tumor cells are important is because they are rarely if ever found in healthy patients. There is a cutoff of circulating a certain number of tumor cells after which pretty much they are only found in patients with cancer. Low levels like 1 or 2 circulating cells, epithelial cells, can be present due to a phenomenon called benign epithelial transport where epithelial cells from different organs in the body can enter the blood stream and circulate. But pretty much after that, circulating tumor cells are only present in cancer patients. For example in this slide, there is all these dots that you can see on the graph. Let me walk you through that. The very first set of slides…there is a green bunch of dots that show healthy people, those that we know do not have cancer, with the best testing that we know. It appears that there were some patients, such as 3 percent out of 295 that had 1 to 2 circulating tumor cells. But pretty much after 3, there were no healthy patients who had an elevated circulating tumor cell count. If you look at the lighter green color, you see that people with benign diseases such as fibrocystic disease of the breast or inflamed prostate are a little more likely to have a little higher CTC or circulating tumor cell count. But when you start to use a cutoff of 3, you see that in actual cancer patients, such as those in the two slides of the purple, which is metastatic breast cancer, those in the two bars with the blue, which is metastatic…in this case it is the last 2 bars, the brown, are prostate cancer. You start to see the presence of high circulating tumor cells only in the presence of metastatic disease. The blue is colorectal cancer. Those are patients with colorectal cancer that had metastacized. So now this technology has been validated for metastatic breast, metastatic colorectal; and the last two bars we see in the brown are patients with metastatic prostate cancer, who start to exhibit a fairly high presence of circulating tumor cells. And these cells are very helpful and they are prognostic at the very first time of measurement, and equally very helpful in determining when a new specific line of therapy is started for patients if they are going down or not.

SLIDE #28

How are these measured? Just like you get a blood test done, circulating tumor cells are collected in a specific tube only meant for that assay because they stabilize and prevent these cells from bursting open. If blood has just left in any test tube lying around, it has a special buffer that prevents these cells from bursting open. The medical term we use is lysing. Then this sample is then subjected to reagents that remove presence of white cells in the blood and other elements and particularly look for epithelial cells or cancer cells. Then they are run through a machine called (flow phetometry) that tries to label these cells. And you can actually visualize, like in the bottom most panel on the left, you see the green and purple boxes. Those are specific circulating tumor cells that the machine is able to detect from a vial of blood and not just detect but give you a numeric count of how many of these are present in a specific volume of blood to give you a circulating tumor cell count.

SLIDE #29

What does the circulating tumor cell count mean? So if we particularly look at patients with prostate cancer and we take the very first time that the cancer has metastacized and do this assay, do this cell count, we can divide up patients into those who have a low circulating cell count of less than 5—that's the green bar; and those patients who may have an elevated circulating tumor cell count, such as those in the red bar. So if you have a low circulating tumor cell count, like in the green bar, the curve—the diagram on the left—shows that if we go out in time since the very first month when we did this assay, patients who have a very low circulating tumor cell count below 5 live a lot longer and have a more favorable biology of disease than the unfortunate…you know, compared to the green ones, those who have an elevated circulating tumor cell count—those patients unfortunate appear to have a more aggressive type of disease and can have a more rapidly progressing course of disease as compared to patients who may not have a detectible PSA at initial time of diagnosis of metastatic disease.

Equally important is that the circulating tumor cells are just not static. They respond to change over time. And you can actually measure if a patient is responding to whatever therapy has been started, like hormonal therapy or chemotherapy with metastatic disease by seeing of the circulating tumor cell counts change. So what this slide or diagram is predicting is 4 different groups of folks. If we focus on the green line, that is Group 1. These are patients who start off with a low count below 5 and through the entire course of the disease continue to have a low count. Those patients have more indulent and longer control of disease and live longer. They have longer probability of survival. That is why they are progressing. Each drop in that curve is more people who progress on. So they have a more prolonged survival. And in the red bar are people who unfortunately have a high count. But in spite of whatever therapy you use, if they continue to display continuously through the course of the disease a very high circulating tumor cell or CTC count—unfortunately that cohort or that group of people have more aggressive disease that does not respond to therapy and have shortened median survival, overall survival. So circulating tumor cells can predict survival both at the very first time interval.

SLIDE #30

But now the interesting new curves that come up in this slide compared to the prior are the blue and the orange group. The blue group is people who initially started off with a high circulating tumor cell count but because of therapy the circulating tumor cell count starts to turn

negative. They have essentially moved from the red line probability of survival and more towards the green line because of responding to therapy and have a better outcome. And on the flip side, we have the orange line of people who initially started off with a low count, but because of either the biology of disease or over time the cancer becoming more aggressive, stopped responding to therapy, and now over time start to have a more measurable or higher level of circulating tumor cells. Unfortunately for this group of patients, they appear to progress more rapidly. And even if they started, such as those in the green line—they then drop off and start to behave like patients who have a very high circulating tumor cell count all through, such as those on the red line. So what this shows us is that circulating tumor cells are helpful both as a one-time measurement at the time of metastatic disease when it is diagnosed. But not just then but over the course of therapy over time interval these are informative in determining the biology of disease and how well the patients will do over time.

SLIDE #31

So in summary for circulating tumor cell or CTC assay is a monitoring tool for people with metastatic prostate cancer. But as I mentioned, it is also utilized and useful in breast and colorectal cancer. It is a simple blood test that can be drawn through one vial of tube. It has information at initial time of diagnosis of metastatic disease. But also useful when any line of therapy is changed. One thing I failed to monitor is that CTC assays are not always end all and be all for there are some patients who always have a low or undetectible CTC count but may obviously progress to disease radiographically (incomprehensible) metastatic disease with the CTC staying negative. The CTC saying negative always does not mean that people are not progressing. All it means is that they have a more favorable biology of disease. So CTCs can't completely abolish the need for scans such as bone scans, CAT scans, but they just predict that people are going to progress more slowly. And also monitoring of CTCs over time can predict if people are responding to therapy.

Limitations of CTC is that it has no value in first making a diagnosis of prostate cancer. So CTC should not be done for healthy individuals to just obtain a CTC and ask the question like we do for PSA. *Do I have prostate cancer?* CTCs are no value there. People with metastatic disease—you really should discuss with your doctor the value of CTC testing and how often it should be done and what utility it has in overall monitoring of disease. But as I shared with you, CTCs do provide a very useful value at the first time point of diagnosis of Stage IV metastatic disease, but equally importantly to see if a therapy you start… a month later the CTCs go from positive to negative, clearly prognosticates that that therapy is being effective. And then also I showed you the curves where people can start off with an undetectible value and turn positive and vice-versa.

SLIDE #32

Important things that these patients can do and providers can do is keeping an eye on all indicators and new science that is coming around and bringing it to the attention of your doctor to have informed discussion about the value of any of these tests that have come around.

Other things to focus on that are very important in this journal are obtaining optimal nutrition and diet. We don't want people to become malnourished or have low diet; while at the same time, there is a larger

body of literature coming about that prostate cancer may have some pathogenic role in people who have obesity. It appears that people who have obesity appear to have a little more worse outcomes than people who don't from prostate cancer parameters. There is a large role of exercise to play to stay fit, particularly when patients get on to androgen deprivation or hormonal therapy that can lead to bone mass loss and muscle mass loss from long-term androgen deprivation. It is very important to keep fit and exercise to avoid the muscle mass loss that will come about by medical therapy. Maintaining weight, an ideal body weight. It is very important to discuss incontinence and sexual function with your doctor, as well, because those are intimately tied with the diagnosis of prostate cancer and its treatment ever since diagnosis. So it is very important to discuss that with your doctor.

SLIDE #33

Also very importantly, there is a very useful publication available from the Us TOO organization. You can also find it on their Website. It's called Signposts, which talks about the utility of various modalities of diagnostic testing, whether it be done through scans or blood tests, such as PSA or free PSA or circulating tumor cells that I've touched about. So utilizing a resource like this to keep yourself up to date can be very useful.

I will now conclude my formal presentation.

Pam Barrett: Thanks, Dr. Bhandari. Right now, Dr. Bhandari, we have all of the questions are in the chat box. So if you want to start with those.

SLIDE #34

Dr Bhandari: Okay. Let me take a good very relevant question that comes from the Website and let me for everybody's benefit read that. Do most labs do CTC tests and are these covered by insurance? So as I explained to you, there is a propriety technology that has been developed after years of research and validation by Veridex that uses a specific sample collection buffer to keep these circulating tumor cells from bursting open and lysing. So there are many national reference labs, such as Quest Diagnostics (inaudible) that I know that do undertake CTC testing. You just have to ask your physician. Very often …I believe many, many medical oncologists are well educated and do these tests. And in terms of coverage by insurance, that is specific to insurance. I do know that for the diagnosis of metastatic breast, colorectal, and more importantly prostate cancer, all of these disease states—Medicare covers payment for this. In my personal experience, most insurances do. But obviously you should check with your insurance carrier and have your physician's office verify that so you're sure that your insurance will cover that testing. It will not be covered, as I mentioned, it is not indicated at the time of initial prostate-confined prostate cancer or for monitoring. PSA is our initial therapy. It only has a role in the advanced setting of advanced disease or Stage IV disease. I will also say for our members on the line today from Veridex—if you have other information that you can share, please…Katy or anybody else, please go ahead and add to what I've just shared with you.

There is another good question. What are the confounders of PSA tests? The participant asks examples prostatitis, recent sexual activity, digital rectal exam. That's a good question. The confounders….the established known confounders of PSA tests are a couple of important medical conditions beyond cancer. For example, we all know that an

enlarged prostate, a condition called benign prostatic hypertrophy, when the prostate gland starts to get larger as happens over the natural course of living or aging, most men start to see that the volume of the prostate gland itself gets larger over a time interval. So simple benign prostatic hypertrophy or having more normal prostate tissue can give you a rise in PSA, what we call BPH. Absolutely infection or inflammation of the prostate, such as prostatitis, can also give you some rise in the PSA marker. The amount of PSA that goes up from recent sexual activity or direct manipulation of the prostate itself is usually not that much. It might be simply .2 or so but should not give a very dramatic rise in PSA as the other two conditions can, such as BPH or prostatitis, but can also …yes…do lead to a little bit of a PSA rise. So generally that is what we believe in the field. And if that is a question at the time of your testing, you should also discuss this specifically with your urologist because they are the ones who most closely follow determining that level of PSA rise in a given clinical context.

Another good question is here that I read: Why aren't more directed biopsies in use? Is MRI with DCE useful in predicting tumor location? Good questions. I am not the absolute authority on PSA biopsies being a medical oncologist, given the fact that I don't do these myself like the urologist does. The problem with MRI is that it is hard to biopsy right when you are in a tube of an MRI machine. Ultrasound is a lot more portable, available in every doctor's office or biopsy suite and does a pretty good job at localizing at least where to put the needle in to get a biopsy, specifically for nodule or abnormalities detected within the prostate gland, but harder to do under MRI. But I think this is an active area of research. There might be centers that do it. At least in, for example, breast cancer, they are starting to do MRI-guided biopsies. But I haven't heard of that extensively in prostate cancer arena.

Why don't we open this up to the phone lines for some of our participants who may not be able to submit questions over the internet.

Pam Barrett: There is a question from Henry from Texas. Doctor, when you talk about the CTCs…he is wondering why when a biopsy is done, why a ploidy analysis is not automatically done. And does this….will this give the same results as the CTC analysis?

Dr Bhandari: I think they are very complimentary and different questions. Ploidy, if I understand correctly what you mean, is to determine the genetic make up. And a lot of aggressive cancers do not normally divide up their DNA content into (dotter) cells. And some of the (dotter) cells (maybe) can have extra chromosomal cells that leads to ploity. And ploity is a marker in a lot of cancers for more aggressive biology of disease. But I think that would correlate with the Gleason score. The Gleason does a fairly good job in determining the aggressive or more indulent biology of disease. CTC I think is in my mind complimentary and has never been compared directly with ploity. CTC is picking up another agressiveness biology of the cancer but through the blood stream. So there you are actually looking at cancer cells that have left (wherever) the bulk of the tumor, whether in the prostate or bones or to metastasize in the lymph glands. So these cancer cells are actually leaving where the tumor is sitting and entering the circulation in a very detectible manner. And I think that that provides complimentary information but not the exact same information as ploidy. So I think that the more directly comparable

(square of) ploity would be to correlate with the Gleason score. But I think these two provide complimentary information. Good question.

SLIDE #35

Pam Barrett: Dr. Bhandari, I think we are out of time. Thank you so very much. And thanks to everyone for all of your questions. In addition to the Signpost brochure that Dr. Bhandari mentioned, Us TOO has a number of other educational materials available, such as those listed here, plus a few others. All are available for free on the Us TOO International Website at www.UsTOO.org under the helpful resources section on the right-hand side of the page. You may also want to continue discussion, learning, and sharing in your local support group or in some of the Us TOO online discussion communities where you can remain more anonymous. You can join any of our Inspire or Prostate pointers, discussion groups on the Us TOO Website. Look under the chapters and support groups header.

Thanks again to Dr. Bhandari and our sponsor, Veridex, for making these event possible. And thanks especially to all of you for your time and interest. We hope you found this Web and our teleconference helpful to you and your family.