Beverly Biller:
Hello, I'm Beverly Biller, an adult endocrinologist at the Neuroendocrine and Pituitary Tumor Clinical Center here at Massachusetts General Hospital in Boston. And I am joined today by my colleague, Maria Fleseriu because we are going to share with you some new information from the Pituitary Society Guidelines update on Cushing’s Disease. Maria, would you like to say hello before I get started?

Maria Fleseriu:
Hello, everyone. It's a pleasure to be here and listening to diagnosis and comorbidities in Cushing's disease.

Beverly Biller:
So I’m going to talk about this topic. And maybe once or twice, Maria, I will ask you what you would do. Let's get started. Of potential conflicts of interests over the past two years, you see occasional consulting and research support that I have received. Let's start with the basic definitions. As you know, Cushing's syndrome simply means the clinical features associated with too much cortisol. And the most common cause is us, iatrogenic. We give cortisol for many disorders and that's exogenous Cushing's, but that is not what we’re talking about today. We will be focused on endogenous Cushing’s, that is cortisol excess due to a tumor somewhere within the body that is either making excess ACTH or excess cortisol. The tumors that make excess ACTH, either in the pituitary gland, which represent the majority of these cases, or in an ectopic location, such as a lung tumor, fall under the category of ACTH dependent Cushing's.

In contrast, if there's a tumor on the adrenal gland secreting solely cortisol, we call that ACTH independent Cushing's. When Harvey Cushing, a neurosurgeon first described this condition, he was talking about tumors of the pituitary. So when we say Cushing's disease, we are specifically referring to pituitary Cushing's. This is a rare disease, perhaps three, maybe four cases per million people, and it's more common in females, except in the pediatric age range, where boys and girls have fairly similar prevalence. Pituitary Cushing's is nearly always benign, but as I will talk about momentarily, it's associated with high morbidity and increased mortality. The good news though, is that it's very treatable, with surgery, with medication, sometimes with radiation. And when cortisol is controlled, patients can recover.

So, I'd like to tell you about this patient. She was a 36 year old woman who was pregnant and developed facial rounding hypertension and fungal infections. After delivery, Cushing’s syndrome was diagnosed with clearly high urine free cortisols.

Her ACTH was not suppressed, so she had ACTH dependent Cushing's. The head MRI did not show a macro adenoma. There was a question of a possible small lesion on the right. And because that wasn't definitive, she underwent bilateral inferior petrosal sinus sampling, which clearly centralized, and all of that points to the diagnosis of Cushing's syndrome and the suggestion that it's located in the pituitary gland. We know that when we are faced with a patient like this who has the clinical features of Cushing's syndrome, there are three steps that we need to take in order.

First, we need to answer the question, does this patient have endogenous Cushing's syndrome? Do they have pathological exogenous, sorry, endogenous cortisol excess? And only if the answer is yes should we go on to looking for where in the body this tumor might lie, or the differential diagnosis? Is it pituitary or adrenal, if it's ACTH independent? Is it pituitary or ectopic, if it is ACTH dependent? Once we determine whether the patient is likely to have pituitary adrenal or ectopic cushings based on the biochemical tests, that is the time when we move on to treatment, because we need to know, are we calling an
expert pituitary surgeon, a thoracic or abdominal surgeon, or an adrenal surgeon, before we move the patient on to their definitive treatment.

We're going to focus on talking about how to make the diagnosis to answer the question, does this patient have endogenous cortisol excess? And so I want to tell you a little bit about the Pituitary Society Consensus meeting that I mentioned a few minutes ago, co-chaired by Dr. Fleseriu. She put together a group of over 50 academic investigators and clinical Cushing's disease experts who gathered and reviewed data since the last 2015 Endocrine Society Consensus guidelines.

This consisted of a two day virtual meeting and discussions where recommendations for diagnosis and management were agreed upon. And this presentation that I will give reflects those recommendations. What you see here is the algorithm for diagnosis. Now of course, I don't expect you to be able to see all of this on the screen, but I show it to you so that you're aware that there's a very detailed algorithm in this consensus document, in Lancet Diabetes and Endocrinology. And for those of you who enjoy algorithms, you might want to take a look at this. So, when we think about the diagnostic tests that we can use for a patient who might have Cushing’s, we always start with considering the clinical features. One of the things that I find so interesting about Cushing's syndrome is how very many different body systems are affected by this disorder.

You can look at this diagram and pick out almost any organ system and see how it is affected by too much cortisol. Whether it's the many skin changes, the psychiatric features, the problems with the musculoskeletal system, with muscle weakness and osteoporosis, or whether it's the many cardiovascular features that are associated with Cushing's, this disorder really has a big impact on the body. It includes a predisposition for infections and sepsis, and both cardiovascular risk and sepsis are also causes of the mortality associated with this disorder.

So, this brings me to my first question that I would like to ask Dr. Fleseriu. So, you have published about the risk of clotting from the thrombotic events that occur in these patients. So, I’m wondering how do you address this clinically? And do you anticoagulate your patients? So, we're interested in hearing what you do. And maybe you can also give a mention to what was discussed at the consensus meeting.

Maria Fleseriu:

Thank you so much, Beverly, for bringing this up. And as you remember, and all our colleagues do, this was very contested, because yes, we know that increased thromboembolic risk is present in almost all patients with Cushing's. We also know that this persists even after the disease cure and sometimes even years after that. And somehow, we don't really know we if we should anticoagulate everybody, and furthermore, for how long? So, what I usually do, I look at the patient and decide in my mind the risks. Though we have published it, the classical risk factors are not exactly the same as in other patients. So yes, we know that the surgeon can increase risk, immobility can increase risk. However, in several of the studies did not link directly. Does the severity of the hypercortisolism link? Possibly. So, in my mind, in any patient with severe hypercortisolism, unless there are directly a contraindications, I do anticoagulate. Now, for how long really depends on immobility of the patient type of procedures.

And in some patients, especially if they undergo bilateral [inaudible 00:08:55], we should probably anticoagulate for two to three months. And this has been shown that possibly will decrease risk. Though, us and others have shown that even with anticoagulation, the risk does not go to zero. So, this is a very interesting area, and we also put it in the future directions. We really need more studies. Each of us are doing this differently, and we should focus on which are the really best procedures to also decrease the risk, but not in just bleeding either.

Beverly Biller:
Yes, I do remember that this was one of the more contentious areas of discussion, because there were many strong opinions at both extremes, always anticoagulate, never anticoagulate. And probably the right answer is somewhere in the middle, but I totally agree that there are many unanswered questions about the type and duration and timing of anticoagulation. And it is an area where investigation is really seriously needed. Thanks for sharing your thoughts with us.

Now, back to thinking about the clinical features of Cushing's. Some patients look obviously Cushingoid. You can see in these patients, hirsutism. This patient on the left has clear superclavicular fat. Facial rounding and plethora are visible here. These are remarkable striae, as we are used to seeing in our patients with Cushing's, although of course, not every patient has every clinical feature of the disorder. Spontaneous ecchymoses, the opposite side of the coin from clotting can also be seen in patients with Cushing's.

And in this patient, you see hyperpigmentation of a scar on her knee where she had fallen while rollerblading. And so if we see patients who have clinical features of endogenous Cushing's syndrome, the first thing to do is to make sure they're not taking exogenous steroids, because as I mentioned, that is the most common cause of Cushing's syndrome. They're getting it somewhere, from perhaps another doctor. And often, we just ask if they're taking glucocorticoids. Are they taking prednisone, dexamethasone, hydrocortisone, for some reason? We need to remember that steroids can also be given orally, they can be given injected, they can be given inhaled, and they can also be topical. And a common one is patients who are getting joint injections but don't think to bring that up when we ask if they're taking any steroids. So, we really need to go through all the methods of delivery to find out whether our patients have been exposed to exogenous steroids.

But in patients like these, if they have not been given topical, inhaled injected or oral steroids, then they do need a workup. It's not always so obvious. This is a patient who really felt like something was wrong. She was having to exercise more and eat less, and yet was still gaining weight, and said, "There's just something not right about how I'm thinking." She was finding that she was up all night and sort of hyper and thought something had changed, but she didn't look obviously Cushingoid. So, how can we tell if patients like these need evaluation? One of the helpful things can be to think about whether they have the features that have been shown to be without any predictive significance. That is the simple presence of obesity or hypertension, glucose intolerance, irregular periods or plethora are not predictive that that patient is more likely than the next person to have Cushing's. But there are some clinical features that increase the probability of Cushing's. They're not a guarantee, but they increase the probability.

And those include spontaneous bruises, where the patient doesn't even remember getting injured, the wide stretch marks, such as the ones I just showed you, thinning of the bones at an earlier age than you'd expect in a woman or in men, or spontaneous hypokalemia, a very central pattern to obesity, or proximal myopathy and objective weakness. If these things are present, that increases the probability and suggests a workup might be needed. The other thing we can do is ask the patient for old pictures. And when we asked the patient I just showed you for old pictures, we could see that she was right. She really had changed, and she had developed a significant amount of superclavicular fat. And so this is a patient where we would conduct a workup.

So, then what do we do? What's the workup? Well, if there is one thing that you remember from my entire presentation today, this is what it should be. Look carefully at the screen and watch what I'm going to do about serum cortisol. I am crossing it off. Please resist the urge to draw a serum cortisol, and teach your colleagues in other specialties to resist that urge. And here's why. You know this. Cortisol is released in a very pulsatile manner. Here is a pattern of cortisol secretion in a healthy normal adult who was admitted to the clinical research center and had ACTH and cortisol drawn every 10 minutes for 24 hours, so that's 145 samples of blood across the 24 hour period. And you can see that eight in the
morning, even earlier than eight on the second part of the day, early in the morning before the patient wakes up, between six and eight, there are more and higher amplitude pulses of cortisol.

Beverly Biller: And as the day goes on, there are fewer and lower amplitude pulses. The same protocol was conducted in a patient proven to have pituitary Cushing's. And you can see that this patient has more pulses and they're more frequent, and some of them are higher. There is also no time of the day that the patient with Cushing's drops down to low levels. But the point of this is that if we sent the patient to the lab, at any of the times circled in green, the level in the healthy person and the level in the patient is exactly the same. A random serum cortisol is not diagnostic because there's too much overlap with healthy normals. Notice that the greatest separation is late at night. And we'll come back to that when we talk about late night salivary cortisol as being a good time to test for Cushing's.

What about the one milligram overnight dexamethasone suppression test? This is a test that the Endocrine Society guidelines suggested for screening, and the pituitary society guidelines have also confirmed that this is a good test, as long as we bear in mind the caveats. A normal test pretty much rules out the diagnosis. The false negative rate is very low, if we use the low cut point of 1.8 micrograms per deciliter or 50 nanomoles per liter. The trouble with this is if the test is abnormal, that is it fails to suppress with levels above those cutoffs, we are not sure. The patient may have Cushing's or they may have one of the causes of a false positive. And those could be many things. It could be a lab or patient error. The patient may not have taken the dexamethasone when we told them to, may not have gone to the lab at the hour they were supposed to.

Obesity itself causes a false positive rate in patients who may or may not have Cushing's. And then there's a whole host of causes of hypercortisolism without Cushing's, such as pregnancy or depression or intense exercise or tight control of diabetes with hypoglycemic episodes. So, we have to ask about and think about all these things. In a patient on estrogen, cortisol binding globulin is increased. And so that means measured total cortisol is also increased, but the patient doesn't have excess free cortisol circulating. So, we need to think about that as well. And finally, drugs that speed up the metabolism of dexamethasone through the liver, for example, mean that, by the next morning, there may not be enough dexamethasone to cause suppression of cortisol. So often, a dex level is also measured, particularly if we're repeating this test. And so we have to be aware of all of these caveats, particularly in healthy young women who may be on estrogen.

And so if there is an abnormal dex test, further confirmatory testing is needed. And one of the tests that can be done is a 24 hour urine free cortisol. This basically provides us the area under the curve, that red curve that I showed you, because it's an integrated assessment of how much cortisol is being secreted over a 24 hour period. And the word free is referring to the fact that it's not protein bound, so it is not affected by any changes in cortisol binding globulin. For that, there's not a specific cutoff, but rather it depends on your own lab's normal range of assays. And it's a good test in moderate to severe Cushing's, but we also have to be aware of some caveats of this test too. We have to be sure the patient really collected 24 hours worth of urine. And so there we look at creatinine to make sure that's an appropriate amount.

But it turns out that volume is also important. There was a really interesting study done at the NIH, where Veronica Merick asked her colleagues in the lab to drink five liters of water over a 24-hour period. And she measured urine volume, which no surprise went way up to four liters or more. But the point is that urine free cortisol also went up. You can see that most of her healthy normal subjects ended up with urine free cortisol levels above the normal range depicted in the hatched bars at the bottom. So, we have to think about that and counsel our patients not to drink too much fluid the day of the test. The other question is, how much does urine free cortisol vary from one sample to the next? We had the
chance to look at that in over 150 adults who were enrolling in a study for patients with confirmed Cushing’s disease, and they collected four 24 hour urines over a two week period.

They were done in central labs. The patients were given the same written and oral instructions, and they had assays collected over this time period. This graph is showing you the first sample and the second sample in individual patients. So, each dot is a single patient’s first and second sample depicted on the graph. And the good news is that if you look at the R value, it’s highly correlated, almost 0.8. So, that’s good news because we know that we want the first and second test to give us about the same answer, and it did. And especially, that was true with the mild elevations in urine free cortisol. But notice that the higher the urine free cortisol, the first and second values being more different from each other. So, if we picked patients down here, near the intersection of the two a axis, on each of their samples, their urine free cortisol was about three times the upper limit of normal.

So, that’s really great. But we can also pick a couple of patients where they have two very different results in their urine free cortisol. One sample is nearly normal. There’s one patient sitting very close to the normal range, which is the shaded blue bars across the axis, and the other is elevated manyfold. And it turned out in this study that the within patient variability over four collections was just over 50%. And that is why we suggest getting at least two samples, so that at least we have some idea by taking a mean of what the overall excess cortisol production is over time in these patients. And so both the previous guideline and the new pituitary society guideline suggest getting at least two samples if we’re doing urine free cortisol. What about late night salivary cortisol? Patients really like this test because they can do it at home and it does not involve a jug of urine, and it’s very sensitive and specific.

It’s especially helpful in early Cushing’s, and also in recurrences. I’ll show you shortly. So, patients who have normal levels are quite unlikely to have this disorder. It’s a test that should be done before the patient does their dental hygiene, and it should be done at their natural bedtime. They chew on a cotton, put it in a tube, and either mail it in or bring it into the lab. As with the 24 hour urine, we suggest at least two samples, because we know that healthy normal people occasionally have an elevated late night salivary cortisol. The biggest caveat about this test is if a patient has a switch in their day night cycle, such as a shift worker whose predominant time of work is at nighttime instead of in the daytime. So, this slide summarizes for you the Pituitary Societal guidelines about the diagnosis of Cushing’s. And after excluding exogenous steroids, there are three tests that can be used, the late night salivary that I just talked about, the 24 hour urines, in both cases, at least two, and the one milligram overnight dex test with all of the caveats that we talked about.

Now just as an aside, I will say that I’ve been talking about a patient who presented with symptoms of Cushing’s syndrome. If a patient comes in from a different direction, from the direction of presenting because an adrenal nodule was incidentally found, then the first and best test in that case is the one milligram overnight dex test, because there, what you’re looking for is whether the nodule seen on scan is autonomous. But when we interpret these tests, we need to think about the causes of non neoplastic hypercortisolism that I’ve mentioned and consider ruling them out if needed. So, what happened with this patient after she was diagnosed? She underwent what is considered the first therapeutic choice that is pituitary surgery by an expert surgeon.

She made a very good clinical improvement. And fortunately, all of her pituitary function was normal. She was euthyroid, eugonadal, had normal growth hormone axis, and was able to be titrated off replacement glucocorticoids in about a year with proof on testing that her cortisol system had recovered. A number of years went by, she stayed in remission, she was doing well, and then she moved away to Arizona. So, think to yourself, what do you think are her chances of recurrence? I will tell you that we used to say, in fact we published that the recurrence rate was five to 10%, but we were wrong.
It's actually higher than that. These are data that show you the remission rate at expert pituitary surgical centers, and you see that it's somewhere between 70 and 90%.

That's great. The majority of patients with micro adenomas, in the hands of an expert surgeon, can be placed in remission. But the point I want to make from these papers is that there was also a significant recurrence rate as high as nearly a third. And recent studies have also shown even higher values. I think the longer we follow patients, the more we are going to see who recur. So, a number of years later, she developed new diabetes, weight gain, and hypertension, and she asked her doctor if she might have Cushing's disease again. She was told no. Because metformin controlled the diabetes, she was able to lose weight with diet and exercise, and her serum cortisol was normal. So, that reminds me to say that the guidelines I've just shown you, both the Endocrine Society guidelines and the recent Pituitary Society guidelines are not being followed.

In fact, in nearly 80% of cases where a patient presented with hypertension and was screened for Cushing's, the initial test was serum cortisol. Remember, that is not the ideal test. As an aside, mentioning adrenal incidentalomas, where I told you the best test is a one milligram overnight dex test, only about a third of patients were tested using that. So, I think it's important that we publicize the results of the guidelines, and we should also be teaching our colleagues about the right test to use to make this diagnosis.

So, let me step back and ask Dr. Fleseriu, based on what you've heard about this patient so far, her doctor telling her that she doesn't have Cushing's because their serum cortisol was normal, what would you do? What would you think about in terms of making a diagnosis of possible recurrence in this patient?

Maria Fleseriu:

Thank you for your question, Beverly. In general, for recurrence, if a patient would confirm Cushing's disease comes back and tells me I think that the Cushing's is back, I think also that the questions is back. Patients know the symptoms that they had before, and sometimes they know way longer than we think that we have the numbers to prove that. And I think one of the reasons is, in this case, was serum cortisol that we shouldn't check or rely on separately for that. But we know now that the salivary cortisol for example, and we need several, could become abnormal, and in the beautiful study from MGH that your group published, even a year before other tests. So, I usually do, for every patient, the moment they're off hydrocortisone, every year I do salivary cortisol, even if the patient has no symptoms. Of course with symptoms, I do more than one, and then do further testing.

But clearly, the salivary cortisol is the best test for recurrence, and in has proven on and on, of course, in a good lab. Sometimes the urinary cortisol will become abnormal. The overnight dex can become abnormal. Sometimes in few studies, even before the urine, but I don't think we have very good data to differentiate between them. But for the salivary cortisol, there's a first one for occurrence we have now enough studies, and that's what I do. I follow the guidelines.

Well I'm glad you follow the guidelines that you were the organizer for. I do agree with you that patients themselves are often the first to know. And years ago, when we only had urine free cortisol, we would measure urine cortisol and a patient had thought they had Cushing's again, and it would be normal. And we'd say, "No, nope, you're okay." We don't say that anymore, because many of those patients really did have early recurrences. And now, in fact, we can make the diagnosis. So, I'll go on to tell you how that was done in this patient. She actually still had family in Boston. And since she really thought she might have Cushing's, she asked if she could come back and be seen again. And of course, we said sure. Eight out of eight of her urine free cortisols were normal, and she looked pretty healthy, as you can see here.
But two thirds, 66% of her late night salivary cortisols were high. And in fact, since she hadn't had an MRI for a decade, we did an MRI, since there had been a question of something on the right. And in fact, it was pretty clear that this was recurrent Cushing's. Not only because the majority of her late night salivaries were high, but also because she had an abnormal scan. She is a great example of a really typical recurrence. That is mild clinical features with a few new comorbidities, easy to control diabetes, in her case, hypertension, weight gain, mild biochemical abnormalities with a normal urine free cortisol. And yet the majority of her late night salivaries were elevated. So, she's a great example of someone we would have predicted would have a very unlikely chance of recurring, because she was so profoundly adrenal insufficient postop.

And she's a reminder that even patients with a low probability of recurrence may recur if we followed them long enough. And there's a lot of data now that show us recurrences can be many years after surgery. Between six months and 12 years have been reported in a number of series. This patient recurred at 21 years after her initial remission. And the longest I've seen is 27 years. I don't know if Maria has seen a longer period of time, but it can take many decades. And we know, as you were just mentioning actually, that the sequence of hormone changes in a patient who's recurring starts with a rise in late night cortisol. And so that means that the late night salivary cortisol will be the first test to become abnormal. It usually precedes the elevation in urine free cortisol by more than a year. So, the first test to become abnormal is late night salivary, then the one milligram overnight dex test, and only later does the urine free cortisol become abnormal in a large cohort that was studied.

So, these data tell us that we really have to follow every patient in remission, and that we cannot rely on urine free cortisol alone. We have to use the late night salivary test, as you said you would do. Or in the cases where that's not available, first of all, fight to get it available, but secondly, in the meantime, use an overnight dex test. The late night salivary is the most sensitive test. These are some really nice data that followed a cohort of patients after surgery, showing you a series of late night salivary levels that were measured over time in individual patients. They call them sequences. So, the vertical dots is a single patient doing late night salivary cortisols periodically over time. In the patients in remission shown in this part of the graph, almost all of their late night salivaries were normal with just a couple of exceptions.

Now, look at the cohort of patients they followed who turned out to recur. In those patients, the majority of the little dots in the sequences are above that green normal zone. That is they have elevated late night salivary cortisols. They're not all elevated. Just like our patient, she had two thirds of her tests above normal. So, it doesn't have to be that all of the tests are abnormal. I think a majority being consistently abnormal is enough to say this is back and we have to start thinking about how we should treat the patient. So, there's a separate session about treatment. But while we're thinking about this patient, I will tell you what happened to her. She had all of the secondary treatments offered to her, and she decided to undergo a second operation, because she couldn't believe that she would only have to spend a day in the hospital. When she was originally operated on, she had to spend a week in the hospital.

I would be happy with another 20 year remission by spending just one day in the hospital. And indeed, she was placed into remission, and her diabetes and hypertension both resolved, so she was able to come off those medications. But one of the risks of a second surgery is hypopituitarism, and she did develop some pituitary hormone deficiencies. So now, instead of taking diabetes and hypertension meds, she's taking some pituitary hormone replacements. But those have been adjusted and she feels well. Here, you see her most recent picture. And in case you're wondering why she's tugging on her shirt, when I took this picture, she said, if you're going to show this when you teach about Cushing's, I want them to see that I have clavicles again.
And so in summary, we know that we must treat the comorbidities while we're also looking to put the patients in remission. We need to confirm pathologic andogenous cortisol excess before we start looking for lesion, because we might find an unrelated incidentaloma. I hope I've convinced everyone to teach your colleagues to resist the urge to measure a serum cortisol that is not suggested by either the old or the new guidelines, to be aware of the caveats with each test so that we can interpret them correctly, and to know that the late night salivary cortisol is the most sensitive test and should be our first choice, except in people with day night switch. All patients in remission from Cushing's disease should have lifelong monitoring for recurrence. Thank you.