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Epsiode name: 'Fatness and the Body' Episode 4: 15(+!) ways estrogens influence adipose tissues

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Transcript

Good morning from El Paso, TX. In case you don't know where it is, we're located right on the Mexico border. You almost can see the US Mexico border outside my window. So that just sort of gives you a little landscape. Actually it's snowing today and in a very arid area. So it's a little just unusual but that's, you see snow. That's actually what's happening. And so I'm so pleased to share my passion with all of you. I have been working on sex hormones and metabolism. And so let me just go ahead and get started.

And so this is sort of the beginning of my slide and this is just to bring us all onto the same page and that is to recognize that men and women indeed are very different. I think that I understood this, but I don't think that I understood it as importantly as I understand now and what really I think, it's so important, is that when we look at experiments or the research or the literature, and we find that there's a study that says there's absolutely no differences between men and women, and then we actually look at, OK, how did they conduct that study? And we see that they enrolled women who were the age from 18 to 82. I think that we have to then step back and pause and question whether that's really true or not, because women across our life cycle, we're very, very different. And so to clump us all in together and just say women, it's a huge disservice. And so I really want to emphasize that point today and talk specifically about one of the female hormones, which is estrogen. Sex is a biological variable, and yet we all appreciate this, but it wasn't necessarily truly appreciated at the National Institutes of Health until about 2016, and this becomes important because my very first experiment - I actually was a dietician - I decided to get my doctorate degree. I went to my mentor. My mentor handed me a gigantic fat rat and said that I was going to put a brain cannula in this rat and off I was gone. My mentor had all of this data that he was waiting to submit a grant to the National Institutes of Health. He knew

exactly the way the study was supposed to turn out. So off I went undaunted. I went ahead and I did the brain surgeries. I went ahead and ran the experiment, and then we tabulated the results and the results were exactly opposite of what he predicted. And this was quite some time ago, right around the time when this was actually before we were thinking about sex as a biological variable. And so he was livid with me and wanted to figure out what I had done with wrong with the experiment. And what we discovered was that I had actually used the female rats instead of the male. That's, and it was at that point in my life and my career, that gigantic oops, that I discovered that indeed very little had been done with respect to sex as a biological variable. Females were excluded from most of all the studies. I'll elaborate that in a bit. We could all well appreciate that every cell actually has a sex. But sex was underrepresented in medicine as well as in science. And yet we strive in the United States, and I'm sure that you all do as well, to personalize medicine. And this is what represented by the dotted line, which is we're all looking for personalized medicine. But yet medicine hadn't been personalized by sex and it's still pretty much lags with respect to how well we personalize medicine with respect to sex.

And this is just a snapshot to sort of understand that with cell based assays I would hazard to say that where this indicates 75% or 76% of the cell based assays, sex is not even specified. I'd say that number might even be actually larger. We know that in the United States, and I suspect also where you all are, that predominantly we utilize male rodents for all of our research and we do that because the female rodent goes through her cycle every three to four days, making her hormonally completely challenging and different based on her phase of her cycle. And so obviously then people like to have their data much tidier, and so they've been utilizing male rodents. Men are predominantly utilized in all of our human clinical trials. Women are excluded during their reproductive age, and so oftentimes we see more of the clinical trials including men. And yet women make a predominant amount of the healthcare decisions. So this is one of the just a snapshot there. I could find a million different slides that show you that this is important because we can all appreciate that there are sex differences with with respect to disease prevalence.

There are data upon data upon data that suggest that women are relatively protected from cardiovascular disease, diabetes and the like. Prior to menopause and after menopause, all bets are off, the risk reduction is reduced. And so this is just a chronology of estrogens in our understanding. So we got back in the 1980s, we could all appreciate that estrogens might be involved in body mass index. And then in the 1990s, we started becoming slightly more mechanistic. In the 2000s, we started to do human genetic studies, and suffice it to say, we've really progressed a long way. Originally, we just thought of estrogen as just being this reproductive hormone and that's basically all it did. And today I'm going to try to

highlight how many other phenomenal roles estrogens play with respect to our metabolism.

And why am I focusing on estrogens? Well, one reason is that the National Institutes of Health mandates that we focus. So I had to quickly decide after I looked at all of the demographics and saw that there were sex differences in disease. I had to decide, well, which one of the sex hormones was I going to focus on to begin to understand what was the driver of these sex differences? And so I focused on estrogens. And one of the reasons I did this is because, indeed, women spend half of their life in the menopausal state. And so half of their life is spent without this incredible hormone, estrogen. And I wanted to understand more of what happened when women transitioned through menopause.

Estrogen plays a predominant role in regulating metabolic health as well as immune and vascular actions. As such, menopausal induced decreases in estrogen increase women's risk of developing obesity, diabetes, cardiovascular disease and many different forms of cancer. In the United States, almost half of the women who are aged greater than 40 are obese and this is largely thought to be due to estrogen deficiency. And this is also there's increased adiposity that's associated with that menopausal transition and it's thought to be very detrimental with respect to metabolic and inflammatory perturbations. I love this slide because it fascinates me. I hope that none of you were the author of this article. But my mom just turned 80 yesterday 82, so I guess all of her estrogen is in her feet. I am 60. So I suppose that most of my estrogen is in my thighs but it this is just it. It makes me laugh every time I see this slide. But it's just to highlight again that the fact that that women hormonally are very different across these different decades of our lives. And estrogens again, I could go on and on and on about all of their the effects that they have on all of the different types of tissues that you see within this slide. But in this presentation, I'm going to predominantly focus on the role of estrogens in adipose tissues. And so this is actually the title of my slide that was presented to you. And that is that I'm going to talk to you about 15 different ways that estrogens influence body fat distribution and adipose tissue function.

A sort of an observational determination, and that is sex related body fat distribution, that predominantly men have more of their adipose tissue in the visceral depot, where women have more of their adipose tissue in the subcutaneous depot. And this was done for teleological reasons. We know that women when breastfeeding, we actually mobilize the fat that's located in our subcutaneous depot to supply the energy that's needed for reproduction and breast feeding. Men, on the other hand, predominantly store more of their fat in the visceral depot, and this was due to the hunter gatherer days where men needed a readily mobilizable fat store that could be utilized on a quick energy demand.

And that's why they predominantly stored their adipose tissue within the visceral adipose tissue depot.

Now just to remind you that the subcutaneous and the visceral depots are very, very different. The subcutaneous adipose tissue depot is located outside of the abdominal wall. And again this is predominantly where women deposit most of their fat, where the visceral adipose tissue depot is located inside the abdominal wall. And this becomes really important because it allows me to introduce the concept of an expandable fat cell, and I'm going to utilize this diagram to sort of help elucidate this point. That is, that these are two, this is two different sexes, a male and a female. They're actually matched for body mass index and body adiposity. Yet you can observe that the female has more of her adipose tissue that's located in her hips, her thighs, whereas the male has more of his fat that's located inside of his abdominal region. If we were to dive down and look at that adipose tissue specifically, what we would see is that the female adipose tissue has these large adipocytes, but they are not encased in what you can see in the male slide. In the male adipose tissue it is actually encased in fibrosis fibrotic cells that are in and around the adipose tissue. And why this becomes important is that what happens is that those adipocytes within that male visceral depot have no ability or very little ability to expand to take up more lipid. Once that adipocyte takes up as much lipid as it possibly can, then that lipid starts to become deposited within organs such as the liver or the pancreas the heart, any of those internal abdominal organs actually then become a lipid reservoir. Once that visceral adipose tissue can no longer take up more of those adipocytes or more of the lipids, and so this is sort of the way that I like to characterize this male adipose tissue. As more like wool and that is that again these are these adipocytes, they have a finite ability to expand due to all of this connective and fibrotic tissue that surrounds the male adipocyte. Female adipose tissue is more like spandex. You can cram a lot of lipids into those adipocytes for the females and we do this specifically because we have to store adipose tissue in a very, very healthy way. If we didn't, while we were gaining weight during pregnancy, that would be a metabolic challenge. So females are specifically designed to have these beautiful, incredibly expandable fat cells. That can take up more lipid that those fat cells are not inflammatory and they're very, very healthy.

But this is just sort of a concept that I wanted to put forward. So the next concept about what estrogens do is actually we can appreciate that there are sex differences with respect to the amount of circulating estrogens and between males and females. What we often forget is that estrogens are actually a family of compounds. There's estrone, estradiol, estriol and estriol. Oftentimes we only think of estradiol and yet this is a disservice because there are all of these other estrogenic components. I think that there is a huge amount of literature that that needs to be embarked upon to begin to understand what

these other estrogens besides 17 beta estradiol are doing in our adipose tissues. And we'll talk more about that in a moment. But this just only represents that E3 and E4 are present in significant quantities during pregnancy.

But this is more of the areas that I think that we need to dive more into, and that is that E1 is the predominant estrogen in circulating in post menopausal women whereas E2 is the parent compound estradiol that we all think of when we're talking about estrogens is predominantly produced by the ovaries and is predominantly seen in circulation and premenopausal. There are three known estrogen receptors. However, this area of literature is expanding greatly. We know that there are important functions with estrogen receptor alpha, which I will articulate more in a moment. Estrogen receptor beta also has an important role and now people are focusing on GPR 30 in its role as an estrogen binding receptor, playing most of the significant roles with respect to metabolism and inflammatory processes. So next I think that estrogens actually influence overall body fat distribution is not necessarily estrogens per se, but oftentimes we forget about the sex chromosomes. We have to remind ourselves that absolutely every cell has sex chromosomes. Every cell has a sex. And this is important because we're now beginning to understand that these sex chromosomes are also driving where we deposit fat as well as the overall health of those adipose tissues. And this is just a cartoon that I want to utilize to sort of represent that. And this is some of the work that I embarked upon when we talk about the terminology that we're utilizing. These sex chromosomes are circulating or are being exposed to suit to hormonal milieu where testosterone is greater than estrogen. And I'll highlight the importance of this right now and that as I did this is another one of Debbie's oops in her career. So I decided that I had determined that there is an amazing amount of wealth with respect to rodent models and understanding how sex hormones influence adipose tissue health. And I wanted to do a translational experiment, so off I went and I actually derived cells from males specifically. And then I put them into a cell culture media and then I wanted to see if I could replicate some of the findings that I'd had in my in my rodent studies with my male rodents and what I instantly found was that I took out these XY male cells and I put them into the typical cell culture media. But the cell culture media is loaded with estrogenic compounds, non steroidal estrogen. The plastic ware contains polystyrene, which is an estrogen. The serum contains estrogens and you're not even sure exactly what hormones you're actually taking out. And this is just sort of a cautionary tale for those of you who are doing cell culture models, remember that your cell culture models may be changing the hormonal milieu for which you're actually exploring those cells. And what I discovered quickly was that I actually had embarked upon a transgender experiment. I had taken a male cell which had an XY sex chromosomes, but now I put those XY sex chromosomes in a hormonal menu where estrogen was greater than

testosterone. This is exactly the type of experiment that transgender individuals are embarking on almost a daily basis when they utilize cross-hormone therapy and the same would be true for a transgender male who sits with a 2X sex chromosomes. But now those X chromosomes are in a hormonal milieu where testosterone is greater than estrogen. We've written a few papers on this on this topic, and there's much more now in the literature about trying to understand what happens when you take these sex hormone or these cells that have the sex and you put them into a hormonal milieu which is opposite that what they're typically used to seeing.

This is one of my favorite studies. I didn't do it. I'm actually was a runner. I still am, but I'm not quite to the extent that I was in the past and I read this this article in Runners World, which is a popular magazine for those of us who are runners. Have you ever wondered how much faster or slower you might be if you ran and you were the opposite sex? And this is a story about Janet Furman Bowman, who might be the one of the few runners, and now there's many, many more runners who actually are able to answer this question. So this is a story of Jim and Janet. So Jim would go to this place in Los Angeles, run with his running male buddies. And was a phenomenal runner, but Jim recognized that he had gender dysphoria. He transitioned into Janet by utilizing cross hormone therapy. Now, as Janet, Janet had the same body habitus. Same running style, same running everything, same training program. Now, just with the advent of the addition of estrogens, now Janet, when she crosses the finish line, she's more than two minutes per mile slower than she was as Jim. When you take those sex chromosomes and you put them in unusual milieu, there are some really amazing metabolic consequences associated with that. And so while I was doing while I was partaking and and thinking about these types of studies, I was asked then to actually go to a transplant meeting and the transplant meeting was because they the people at the transplant meeting were asking the question “Do we pay attention to sex chromosomes when we actually do a living transplant?” And the answer is that this data is really, really sparse, and the question is if all of a sudden “I take I need a kidney and I am now given a male kidney, how does that male kidney respond in my body where that male kidney again has an XY chromosome?” But now it's in a body that has more estrogens than testosterone. Does it function in the same way as if I had gotten a female kidney? Again, I think these are phenomenal, interesting research questions to Understand, so the next area of how and where sex hormones influence adipose tissue function is through specifically sex hormone receptor expression and as well as action. As I alluded to, there are now many different estrogen receptors that we've identified. There's estrogen receptor alpha, beta GPR30 and the like, and we used to think that these were just nuclear hormone receptors. But now what we understand is there's also estrogen receptors that have non-genomic action. They act inside the membrane as well as outside the membrane. There's a

whole host of different types of function that these estrogen receptors play specifically within adipose tissues. I focused my attention on estrogen receptor alpha because I was quite interested in understanding again what's the mechanisms by which estrogens may influence overall metabolism and adipose tissue health. And I was struck by this paper in the New England Journal of Medicine, which really sort of honed in my research, which identified that in an individual, this individual happened to live in Cincinnati, where I was doing my postdoctoral fellowship focusing on this research. This individual actually lacked the estrogen receptor alpha and had the complete metabolic syndrome. This was a male. He was obese, had insulin resistance, glucose intolerance and cardiovascular disease, and it was based on this that I determined that, oh, perhaps an estrogen receptor alpha is important not only for females but also for males. I went on then and looked at the data with respect to the mouse models with respect to estrogen receptor alpha and found that there was an estrogen receptor alpha, total body knockout mouse that completely phenocopies the human individual.

I then went on and explored further about the level of estrogen receptor expression within adipose tissues and found again that there were sex differences with respect to estrogen receptor alpha and that is that females specifically have higher expression of estrogen receptor alpha in their adipose tissue than do males. And specifically I went on to explore that estrogen receptor alpha is required for that expandable fat cell that we talked about earlier in the presentation. Additionally, there have been data now with respect to the GWAS studies that are determining that there are genetic loci that are actually determined where sex where body fat is determined, where body fat is distributed. Specifically, as I identify that there are sex differences with respect to body fat distribution between males and females, and that the GWAS data now has identified several different genetic loci with important sexual dimorphisms. So again, this is just again highlighting the fact that how we deposit fat has been designed by evolution over a large period of time and it is really determined not only by our sex hormones, our sex chromosomes, but also by different genetic loci. There's also sex differences with respect to single nucleotide polymorphisms. Specifically, there are now data to suggest that premenopausal women who have a gene polymorphism which have differences in overall body fat distribution as well as metabolic health. So these SNPs are actually also determining our overall fat distribution as well as how healthy our adipose tissue is within these different adipose tissue depots. Estrogens also affect the adipocyte lineage. Specifically, adipocytes stem cells can generate committed progenitor cells which undergo adipogenesis and become lipid laden adipocyte. Estrogens, specifically, have been shown to regulate various aspects of this adipocyte lineage. So not only are estrogens influencing that the estrogen complement within the tissues, which we'll talk about more in a moment, but they're even influencing

the overall lineage of that adipocyte. And there's been some phenomenal papers that have come out recently. about looking at the adipocyte stem cell, how estrogens are influencing that adipocyte stem cell and overall again the ability of that adipocyte to take up more lipid and to be a relatively healthy adipocyte and have and be more of like that spandex adipocyte with that expandable capabilities.

Estrogens also appear to have an impact on epigenetic regulation, specifically within adipose tissue. Estrogen receptor alpha, as well as beta, influence DNA methylation through several different mechanisms. And again, the thought is that through epigenetic regulation, even within adipose tissues, estrogens are influencing the overall health of adipose tissue as well as adipocytes through changes in DNA methylation status. Estrogens also influence the microenvironment within the adipose tissue. Again, another area that has been largely underrepresented and under studied, but I think this is a really, really promising area and that is we can all appreciate that aromatase is an important enzyme in this steroid pathway that's responsible for the conversion of testosterone.

In human males and premenopausal women, the primary site of aromatase activity is actually within the gonads. However, during the transition to menopause, women start to produce more of their estrogens due to the lack of ovarian production of estrogens, they start to produce more of their estrogens than within their visceral adipose tissue because of the aromatase gene expression in that area. So oftentimes what you see is that as women transition through menopause, there's a shift in their overall body fat distribution. So now they begin to accrue more of their body fat distribution within the visceral depot, and the reason I believe that they do that is that this is the primary site of estrogen production in postmenopausal woman. So this is, you know, post menopausal women are often will come to me and say, oh, I have this visceral adipose tissue. We've got this belly. How do I get rid of it? Well, it would be lovely if we could, but it also provides us a source of estrogen. But it's within those adipose tissues in the visceral depot that where estrogen is being made. There's so much that we don't understand about what's happening within that adipose tissue. Is it possible that we could upregulate estrogen production and other adipose tissues either in the premenopausal state or the postmenopausal state, through upregulation of the aromatase gene expression? Really interesting areas of research that I think that hopefully we will be able to embark upon in the future. And it's also been recently shown that in adipose tissue there's three times higher estrogen level in the adipose tissue than circulating estrogens in post menopausal women. So again, that adipose tissue is becoming that source of estrogen and so much so, more so that there's so much more estrogen in that adipose tissue than in circulation. And I would love to be able to explore again more of what this is actually doing within that adipose tissue.

So this is just another cartoon highlighting what I just spoke about and that is that again the adipose tissue becomes a source or a sink of estrogens. And what's that adipose tissue doing when you have this ability to make estrogen within that adipose tissue? I think there's so much more that we can learn. There's also sex differences with respect to the innervation of the adipose tissues. This was work that I I did early on, as well as many of my other colleagues, where we actually found that if you knock out one of the estrogen receptors, specifically estrogen receptor alpha from brain regions, you actually see a shift in overall body fat distribution. You see a change in overall energy expenditure and you also see an overall change again in just the innervation of the adipose tissue per se. So this is just to remind us that the innervation of these depots is really, really important, and it too is influenced by estrogens as well as by estrogen receptor alpha. Also, this is a slide to demonstrate the different ways that estrogens influence adipose tissue is through direct regulation of the adrenergic receptors. Again, this is an area that much more research is needed, but estradiol increases the expression of A2 adrenergic receptors within the human adipocytes through upregulation or activity through the estrogen receptor alpha, so again estrogen receptor alpha is even influencing the rate of lipolysis and lipogenesis through direct activation of these adrenergic receptors, which are located within the adipose tissues.

Not only are estrogens influencing adrenergic receptors, but they're also influencing the expression of the genes associated with lipolysis and lipogenesis. This is another area which is a little bit messy. There's differences with respect to the rodents and that are different from humans, but to suffice it to say there are data. This suggests that estradiol acutely inhibits whole body lipid oxidation and attenuates lipolysis specifically in the subcutaneous depot. Can you imagine then that estrogens are driving lipolysis or lipogenesis differentially, depending on the different adipose tissues that they're actually influencing? And this is happening both in the premenopausal woman, the postmenopausal woman, as well as in males. It's another area of interest that I think again that we need much more research to be able to dive in and understand their true influence of estrogens on lipolysis and lipogenesis. The 13th way that estrogens influence adipose tissue is not on the white adipose tissue. Estrogens actually have a profound influence on the brown adipose tissue. This was a relatively recent paper that came out in the New England Journal of Medicine to demonstrate that indeed, women have more brown adipose tissue than do men. Through estrogen receptors, again in the brain, we get different levels of brown adipose tissue activation, which drives overall adipose tissue function and metabolism. Again, this is an area that has become hotly investigated because trying to understand if you can generate more brown adipose tissue in individuals to increase overall metabolic rate if increasing brown adipose tissue has that influence on metabolism

in anything other than a rodent is an area that people are actively pursuing with respect to investigation. But this is again to suggest that estrogens are not only influencing white adipose tissue, but they're also influencing brown adipose tissue.

We wanted to understand if estrogens also influence another type of adipose tissue and this is the beige adipose tissue. So this is not a white adipose tissue, it's not a brown adipose tissue. It's sort of a hyper morph. It's the beige adipose tissue, the beige adipose tissue actually can be located within the subcutaneous or the visceral depot. It is an adipocyte that has much more mitochondria, and therefore it actually burns more calories than it actually stores. And so through a series of investigations in my lab and as well as now has been replicated in other labs, we were able to demonstrate that estrogens, through activation of estrogen receptor alpha, stimulates the beiging of adipose tissue. Again, this is going to be an incredible area to continue to investigate because if we're able then to up regulate the beiging of adipose tissues, would this be a way that we could combat some of the obesity or improve the overall metabolic health of the adipose tissues independent of where they're actually deposited? Estrogens are actually upregulating mitochondria remodeling within the adipose tissues. Just to suggest that through changes in mitochondrial activity within the adipose tissues, you might be able to enhance overall adipose tissue health and that would change overall insulin sensitivity in the entire individual, which might be really, really interesting to look at pharmacology that could upregulate this process.

And then lastly, I just wanted to cover that there, are estrogenic compounds that are out there within our environment and I think this is another area of research that we need to start paying much more attention to. These are the xenoestrogens. These can come in two different forms. They can come in the natural form of xenoestrogens as well as the synthetic form. The natural form of xenoestrogens are your phytoestrogens. These are the phytoestrogens that you might get from soybeans from lignins from a whole host host of our foods. Oftentimes postmenopausal women will revert to taking these phytoestrogens if they are not on an estrogen supplementation regimen in order to provide the body with an external source of estrogens or an exogenous source of estrogens. Again, there are data to suggest that some phytoestrogens might be beneficial. They might be beneficial in reducing overall menopausal symptoms such as hot flashes as well as perhaps influencing adipose tissue health. But again, I think that there is so much more data because it's the amount of phytoestrogen, the type of phytoestrogen and as well as what estrogen receptor the phytoestrogens are binding to, to provide some of the beneficial effects women are seeking. Data to date suggest that soybeans are actually activating estrogen receptor beta, which I didn't spend a lot of time talking to you about today. I worry that in post

menopausal women who are not utilizing estrogen supplementation, they may not be getting the full benefit of estrogens with respect to adipose tissue function in health.

But probably even more concerning to me are the xenoestrogens. These are the synthetic estrogens that we are exposed to now quite readily within our environment. Where do we get these xenoestrogens? Well, they come from pesticides. They come from metals, they come from pharmaceuticals, they come from solvents, their ingredients, household products as well as materials. We have absolutely no idea how much we're actually being exposed to these xenoestrogens, and this becomes a problem because these xenoestrogens come from all different sources such as cosmetics, plastics, plants, industrial byproducts. These xenoestrogens actually mimic the chemical structure of estrogens, and they can actually even hijack estrogen receptors in ways that we don't fully understand. So I begin to wonder whether we are actually now having xenoestrogens that are sitting within our adipose tissues. Perhaps they are hijacking the estrogen receptor alpha pathways, for example, which are normally very, very beneficial. They have all these beneficial effects with expandability, beiging of the mitochondria. Perhaps, though, these xenoestrogens are binding to estrogen receptor alpha and blocking their ability to activate those very beneficial pathways and perhaps are making our adipose tissues less healthy than we would otherwise expect.

Additionally, estrogens as you know, can weakly act as a compounds that actually interfere with our normal hormonal balance. Specifically, perhaps then all of a sudden the adipose tissue, which is normally making aromatase, now all of a sudden it senses that there are these xenoestrogens, it cuts back on the amount of estrogen production because it senses that there's estrogen already being that's located in within those tissues and that might be also a detriment to the overall health of our adipose tissues. Xenoestrogens exert tissue specific and non-genomic actions with respect to estrogen concentrations, especially when estrogen levels are low. So I worry about postmenopausal women who might be exposed in their environment to these xenoestrogens, perhaps these xenoestrogens are being taken up in higher concentrations because there's lower levels of estrogens within these tissues.

And so with that, I just want to summarize and then take some of your questions. I went through this relatively fast, but what I wanted to do was just highlight to you that here is a sex hormone, estrogen, that we predominantly originally thought was only responsible for reproduction. And now we understand there's a whole host of different ways that estrogens influence our adipose tissue health. I probably could have this presentation and focus on all of the ways estrogen influences the heart or the pancreas or any of the other tissues. But today I focussed on my work that we literally looked at the adipose tissues per

se. And that is that we understand now that there are teleological reasons why sex hormones, make differences in these patternings of body fat distribution. We understand now that circulating estrogens and cross hormone therapy, even in transgender individuals, are actually influencing the health and the biology of our adipose tissue. We also understand now and in greater ways, that sex chromosomes, not only the sex hormones, are influencing fat distribution, and again, trying to understand that estrogen to sex chromosome interaction, what does that mean? How does that influence tissue health? Again, I think these are areas for further investigation. Also, we are able to understand that there are sex differences in the expression patterns of these estrogen receptors. I focus predominantly on my discussion today on estrogen receptor alpha because that's what I know and love and have done most of my research on. But other estrogen receptors are important in adipose tissue health as well. We've looked a little bit at the GWAS studies, which I think have begun to enlighten us as to how these genetic loci are influencing fat distribution and how this is influenced by sex hormones as well as sex chromosomes. We've also looked at these single nucleotide paramorphisms and how they're actually influencing overall waist to hip ratio. And the influence of sex hormones. We talked a little bit about estrogens, which now have been identified to actually influence the adipocyte lineage. They're taking these preadipocytes and transforming them into these healthy adipocytes or not, depending on the location of this estrogenic pathway. Estrogens are also able to influence DNA methylation and also turn on or off different types of genes that might be located within the adipose tissues that may be beneficial or detrimental to overall adipose tissue health. I have talked a little bit about the microenvironment that aromatase is located within adipose tissues and actually were able to make estrogens within that adipose tissue. We now know a little bit more about how 17 beta estrodiol is being made in the adipose tissue. But now as we're beginning to understand and being able to interrogate and determine the level of the other estrogens which are which are in adipose tissue, I think we need to understand what happens with estrone and estriol 17A estradiol. How is that influencing overall adipose tissue? We can now also understand that there are sex differences in the direct sympathetic innervation of adipose tissue. We were able to determine this through selectively moving the estrogen receptor from different brain loci, which actually changed in the sympathetic innervation of the adipose tissue and body fat distribution. Estrogens also appear to regulate the synthesis and the control of alpha adrenergic receptors within the adipose tissue, and this is specifically done through estrogen receptor alpha. Estrogen receptors can also influence lipolysis as well as inhibit lipogenesis through different levels of estrogen receptor alpha through regulation of different gene transcripts. Estrogens can also influence brown adipose tissue, specifically through mitochondrial remodeling, as well as influencing the overall amount in sympathetic innervation to the brown adipose tissue

depot. Estrogens now can also be demonstrated to influence the beiging of white adipose tissue that is the transference of white adipose tissue, increasing the level of mitochondria within that white adipose tissue to influence overall adipose tissue health. And lastly, just a comment or concern about what happens now when we're exposed environmentally to these different xenoestrogens or phytoestrogens, how do they influence our overall adipose tissue function and health? Again, I think this is an area that we need to spend a lot more time in researching. So with that, I'm just going to end you with this last sort of cartoon and I can stop sharing my screen and take your questions, if you happen to have any. But thank you so very much for listening to me and talking about my interest in estrogens and adipose tissue. Thank you.