PEDIAPOD JANUARY 2023 TRANSCRIPT

Geoff Marsh
Hello and welcome back to Pediapod for January 2023. This month, sex-specific effects of prenatal opioid exposure.

Neonatal opioid withdrawal syndrome is a condition seen in infants born to mothers who have used opioids during their pregnancy. It represents a major public health problem in the US and has a high socioeconomic burden. The pathophysiology of neonatal opioid withdrawal syndrome is not yet fully understood. Data from animal models have shown that opioids modulate brain reward signaling via an inflammatory cascade. However, no such data exist for opioid-exposed neonates.
In this episode of Pediapod we meet this month's spotlighted Early Career Investigator, Elizabeth Yen. Elizabeth is an Assistant Professor of Pediatrics at Tufts University School of Medicine, and she recently published a pilot study which looked at the effects of prenatal opioid exposure on gene expression and white matter injury. She started off by telling me about where her career got started.

Elizabeth Yen
So I grew up in Indonesia and I had always wanted to become a physician but because of the cultural restrictions back then, it was very difficult for me to pursue this path. So I did get an opportunity to come to the United States. I then studied psychology and I got a Master's degree working on school psychology. I wanted to work with children helping this population. I ended up getting my first job at the New York Foundling, where I worked in a Foster boarding home. And that's where I saw the impact of the opioid epidemic. I got really interested in the medical part of it because of the withdrawal signs in this population. I then pursued my postbaccalaureate study at Columbia and then proceeded to my medical school at Rutgers University, New Jersey. And then once I graduated, I became really interested in research. So in combination with Dr. Johnathan Davis who is an expert in neonatal abstinence syndrome and my mentor, Dr. Jill Maron, who is the expert in salivary diagnostics, we then pursued this path where we wanted to really understand how the opioid that mothers use during pregnancy affects the feeding behavior in children or in babies by using drops of saliva.

Geoff Marsh
Is that linked with the reward seeking behaviors that we often hear about with regards to opioids' effect on the brain?

Elizabeth Yen
Yes, absolutely. So again, Geoff, just looking at these babies, it's very interesting to see how early on, we cannot really see which babies will have more severe withdrawal. We know they will have certain withdrawal signs, including affected feeding behavior, but some of them later on will have actually this hyperphagia down the road where they actually really eat so much more- twice, or even two and a half times the caloric intake of the normal newborns. So looking into these withdrawal signs, and in particular the feeding behavior, we wanted to see which among all these newborns will eventually have this more severe withdrawal. This early saliva collection is the most important formation

Geoff Marsh

Because within the salivary sample, you can have a sort of non-invasive way of looking at gene expression?

Elizabeth Yen
Correct. And going back to your question about that reward, I really wanted to look into the impact of prenatal opioids on the hypothalamic feeding regulation as well as one of the key markers for the reward signaling which is the dopamine receptor.

Geoff Marsh
And we'll get to your study in just a second, but perhaps first we should mention some of the physical structural changes that we see in the brain of opioid exposed neonates?

Elizabeth Yen
Yes, absolutely. The salivary biomarkers are a way to understand why these babies have dysregulated feeding. But then I also wanted to look non-invasively into their brain. So then I created another study by which I concurrently looked into their brains using a non-invasive method called magnetic resonance imaging or MRI. And the idea initially was to look into the functional magnetic resonance imaging or fMRI. Before we even got to the functional MRI which is still being analyzed, interestingly, my radiologist called me and he said ‘this baby's brain had white matter hyperintensity’. And when I heard that I was like, ‘oh, interesting’. So I looked back at the clinical information, and I saw that this baby was indeed exposed. So then when he called for the second one I thought this had to be more than coincidence. So I got really interested in the structural, or more like the larger picture, even though my initial goal was to look into the functional.

Geoff Marsh
What was your hypothesis going into this in terms of the effects of prenatal opioid exposure?

Elizabeth Yen
So my hypothesis really was geared toward the impact- specifically sex-specific impact. We haven't talked about the sex component yet. But the impact of prenatal opioids on the brain area that regulates feeding. I do believe that there is a direct impact of the prenatal opioids on the brain center that regulates feeding. There is now a very interesting evolution of this research by which I not only see the interaction of the opioids and the reward, but also now inflammation on top of this. So it looks like prenatal opioids are pro-inflammatory and that affects the feeding area of the brain that then causes like an upregulated reward. In addition to that, the males and females they're quite different. And often as a clinician we treat them all the same. But this is the first time using the salivary biomarkers and brain MRI that I actually saw the difference between males and females. It looks like the opioid-exposed males, they have upregulated reward signaling and this is interesting because clinically, we do see that males are the ones who are known to have worse withdrawal and that the reward signaling in my previous paper did correlate with the amount of their feeding.

Geoff Marsh
And so that's the males showing these higher levels of reward-seeking gene expression. But females presented differently as well in terms of their inflammatory response, right?

Elizabeth Yen
Correct. Even though as a clinician we often say females do better because they're quiet, they go home, they don't have any withdrawal, they're not treated, interestingly in this study, using this early saliva in the first 48 hours of life, there is already evidence that opioid-expose females have higher inflammation markers in their saliva. So in other words, all these female babies who may not even get treatment, they ended up having much higher inflammation compared to the males.

Geoff Marsh 07:49
I guess that sort of makes sense because the males having higher expression of these genes that are presumably controlling their feeding behavior, that has this very clear phenotype, doesn't it? Whereas inflammation on the brain, you don't necessarily find that unless you go looking for it?

Elizabeth Yen
Correct. So again, the males with the higher reward dopamine expression, they're much more ravenous in their feeding behavior. They're the ones where we often think, oh- they're gonna need to be followed up. But now through this pilot study research, the females actually have higher inflammation and the impact of long-term, pro-inflammatory effects of prenatal opioids? What does that mean in the long run?

Geoff Marsh
What happened when you looked at the MRI scores?

Elizabeth Yen
So the brain MRI of the opioid-expose population, the exposed ones had a higher incidence of that white matter hyperintensity,

Geoff Marsh
Like white matter injury?

Elizabeth Yen
That white matter injury. We don't know if this is completely related to opioids. A lot of these babies are born smaller compared to the non opioid-exposed. So whether or not this is directly opioids that cause them to have a smaller size because again, of that inflammation, and this affects also on their brain development and creates these MRI changes.

Geoff Marsh
And of those children that did present with white matter injury, were there sex-specific differences there?

Elizabeth Yen
Yes. So more females had white matter hyperintensity compared to the males. So out of these 11, six of them had white matter hyperintensity, and four of them were females.

Geoff Marsh

Thinking about the opioid-exposed females in your study that had higher levels of inflammation and higher levels of white matter injury, does that tell you a story about the role of inflammation in the effect of opioids on the developing brain?

Elizabeth Yen
Yes, so animal studies have actually looked into this and there's a lot more literature available for the animal studies, whereby the idea of the opioid binding onto the TLR4 or the toll-like receptor type four on microglia…

Geoff Marsh 10:01
…they're like the resident immune cells in our brains aren't?

Elizabeth Yen
Correct. So interestingly, in animal studies they were able to delineate how the opioids bind to this TLR4, then through the NF-κB pathway then send all these pro-inflammatory cytokines and chemokines. And I think this is a really novel area, not yet done in neonates, because we tend to clinically monitor them and if they're doing well we discharge them. But on the molecular level, this opioid binding to the TLF on the microglia- how they send all these cytokines and chemokines- I believe that will have a long-term impact on their neurodevelopmental outcomes.

Geoff Marsh
One of the obvious lessons from your results, even if it is just a pilot study, is that young female neonates shouldn't be ignored. I wonder, do you think it presents a therapeutic target perhaps to alleviate some of those negative effects of prenatal opioid exposure?

Elizabeth Yen
Absolutely. My research is evolving. I'm looking to see what is the next step. With this research showing that opioids can be pro-inflammatory, I think that brought me to the next thing- what can we do in the future in terms of the treatment strategies. Whether or not we can use anti-inflammatory agents in addition to these opioids or psychotropic medications that we use in these babies.

Geoff Marsh
And what about the young male neonates with their increased reward-seeking behaviors- is that mediated by inflammation as well? It doesn't doesn't sound like your results suggest that.

Elizabeth Yen
Not yet at this point, because, again, we see more inflammation in the female group. But that begs the question of whether this reward-signaling and reward-seeking behavior with their hyperphagia down the road creates another inflammation pathway, let's say through obesity or metabolic syndrome, because of their very highly primed feeding dysregulation.

Geoff Marsh
It would be interesting to follow up the children in this study.

Elizabeth Yen
Yes. So my studies actually are starting to look into serial saliva samples. When they are just born, we had collected them and I have also started postdischarge saliva collections because I wanted to see whether or not this inflammatory and reward gene changes after they go home, after treatment, as well as trying to get the babies who had the brain MRI when they were just born to see the progression of this brain MRI at two months, for instance, whether or not those white matter changes still persist.