Thank you all for being here this morning. So today it is my pleasure to introduce our speaker, Doctor Juanita Merchant. Juanita joined the faculty at the University of Arizona College of Medicine in Tucson in 2018 as a Professor of Medicine in the UA Department of Medicine and Chief of Division of Gastroenterology and Hepatology and is currently a member of the Cancer Biology Research Program and is currently.
also serving as Interim Cancer Center Director for the Arizona University of Arizona Cancer Center. She is coming back home as she earned her MDPHD from Yale School of Medicine, did her internship and residency in internal medicine at Boston, MA General Hospital before completing her Gastroenterology Fellowship at the University of California, Los Angeles. In 2008, Doctor Merchant was elected to the National Academy of Medicine and appointed a member of the National Institute of Health Council of Councils.
and in 2016 she also joined the Board of Scientific Counselors for the National Institute of Diabetes and Digestive and Kidney Disease, a unit of the NIH. Prior to joining UA, she was on the faculty of University of Michigan. She’s Board certified in Internal Medicine and Gastroenterology. She has written or Co written more than 165 pair reviewed research publication and is editor or Co editor of two books and several book chapters. She is a Co Pi on the NAHAG Forward.
00:01:51.346 --> 00:01:52.920 program which was developed
NOTE Confidence: 0.851591256666667
00:01:52.920 --> 00:01:55.700 to increase the number of
NOTE Confidence: 0.851591256666667
00:01:55.700 --> 00:01:56.812 academic gastroenterologists
NOTE Confidence: 0.851591256666667
00:01:56.812 --> 00:01:58.480 from underrepresented groups.
NOTE Confidence: 0.851591256666667
00:01:58.480 --> 00:02:00.500 Doctor Merchant has remained continuously
NOTE Confidence: 0.851591256666667
00:02:00.500 --> 00:02:03.473 funded by the Nah for her work in
NOTE Confidence: 0.851591256666667
00:02:03.473 --> 00:02:06.438 Gastric and newer endocrine tumors.
NOTE Confidence: 0.851591256666667
00:02:06.440 --> 00:02:08.216 Head Hodge signaling,
NOTE Confidence: 0.851591256666667
00:02:08.216 --> 00:02:09.400 gastric cancer,
NOTE Confidence: 0.851591256666667
00:02:09.400 --> 00:02:11.200 and transcriptional control
NOTE Confidence: 0.851591256666667
00:02:11.200 --> 00:02:13.600 mechanisms in colon cancer.
NOTE Confidence: 0.851591256666667
00:02:13.600 --> 00:02:16.733 Please join me in welcoming Dr.
NOTE Confidence: 0.851591256666667
00:02:16.733 --> 00:02:17.719 Monita Wharton.
NOTE Confidence: 0.46492526
00:02:21.720 --> 00:02:24.760 Great. Thank you, Doctor Rogers. Oh,
NOTE Confidence: 0.669391365
00:02:24.760 --> 00:02:25.680 here we get started.
NOTE Confidence: 0.633541566666667
00:02:26.880 --> 00:02:29.999 Well, like to present to you this plaque
in honor of your presentation.
PDE inhibitors, block MDSC metabolism in gastric endocarcinoma.
Oh, free. Thank you. OK.
So oh fixture, thank
Oh. Thank you.
Great. Well, I’m, I’m really excited to be here.
I’d probably come back
about once or twice a year.
I’ll be back in November for the
the Dean’s Advisory Committee,
but I’m excited to present to the
Cancer Center today because I
really love to get some feedback
from the esteemed oncologists and faculty here at Yale. So that so those of us, so as you know, I’m a practicing gastroenterologist and for those of us on the more the diagnostic side, gastrogatinal carcinoma is primarily initiated by an infectious Organism which I’ll review on the next slide and therefore is largely preventable. There obviously are some caveats particularly in underrepresented minorities, but here is the basic summary.
we used to say the second or third most frequent cancer, but has now dropped to about the 5th, probably because more intensive, particularly in Asia, in terms of screening and surveillance. But still, if that type of diagnosis is still associated with a high mortality rate, about 27,000 cases, new cases will be identified in the US and it’s about 11,000 deaths. The important point here with respect to prevention is the infectious component that can initiate this cancer.
And the person Barry Marshall and Robin Warren got the Nobel Prize for discovering the association of Helicobacter pylori first with ulcers but then made the association with gastric cancer. But also there we need to think about dietary components such as high salt nitrates. The other less frequent infection is a viral infection with Epstein Barr virus or EBV. So the in the US the prevalence of gastric cancer has declined. From probably from about the 1920s to about the 1960s, gastric cancer was probably the second most frequent cancer,
but then with an improvement in sanitation, which has not occurred in certain places. Being in Arizona, this is a big issue on the Native American reservations, but that was what was probably driving the decline in the US because Helicobacter is found in the water table basically. However, it is continuing to rise in minorities and immigrant communities. The other interesting issue with respect to gastric cancer, so that’s really the distal cancer.
and I don’t remember if I think I do include a picture of the stomach for those not familiar about thinking about the different regions of the stomach. But the cancers that are arising in the cardia are rising and are thought to be more associated with the maybe increase in use of Ppis. I’ll come back to that point in a little bit. But in general the needle really has been moved more in Asia where Helicobacter is pretty much endemic in the population and certainly on the West Coast where you see more of the Asian immigrants. It’s still fairly prevalent with
the first degree relatives, but you can see the highest incidence tends to be in East Asia. China, Japan and Korea per capita is actually Korea’s the highest. So what I’ll be covering today is how we ended up starting to address this issue or why we started looking at this question in terms of what is driving the inflammation to change the mucosa from chronic inflammation to the metaplastic changes in the stomach. And we came at this or I came at this from the the Hedgehog signaling pathway which I’ll show you why in a few minutes.
And then we started asking questions in terms of translating from our mouse models to what can we do in people and in moving from Michigan to Arizona, have been fortunate to start to collaborate with some of the oncologists there to begin a phase two clinical trial which we're very excited about based upon some of the findings in our mouse models. So again from a gastroenterologist perspective, you know what we typically are seeing in many instances patients really don't even come to be seen by the physician and already have metaplasia. So I scoped many patients that
just have chronic gastritis, sometimes metaplasia, but no helicobacter is nowhere to be found. What is the connection between metaplasia in the stomach and the esophagus? And when I looked into the history of this term, metaplasia, what's interesting is that, so if people remember, you basically have that goblet cell you basically have that goblet cell that's normally in the small intestine, that's normally in the small intestine, but it's showing up in the stomach or in the esophagus. So I like to the pathologist like to say a normal cell in the wrong place,
but that’s signifying that the mucosa is starting to move more toward cancer, we think maybe more directly in response to the immune microenvironment as opposed to the bug being there. The issue of metaplasia and the link of metaplasia to a cancer was really initially identified in the esophagus with Barrett’s esophagus. So that metaplastic change in the esophagus is a precursor lesion and we actually have surveillance approaches for patients that have Barrett’s esophagus. So the debate in the GI field is you know is it going to be worth to
00:09:04.551 --> 00:09:06.587 actually start doing surveillance

00:09:06.587 --> 00:09:08.932 for gastric cancer based upon

00:09:08.932 --> 00:09:10.556 identification of intestinal metaplasia

00:09:10.556 --> 00:09:12.480 and the jury still out.

00:09:12.480 --> 00:09:15.000 I’m actually going to an NCI think tank next week we’re we’re going to be

00:09:18.272 --> 00:09:21.157 discussing this whether we can change the recommendations for gastric metaplasia.

00:09:23.760 --> 00:09:25.797 But the reason why that that is,

00:09:25.800 --> 00:09:27.416 is because the question

00:09:29.440 --> 00:09:31.800 when do we survey them and how often,

00:09:31.800 --> 00:09:35.200 which is where the cost comes in.

00:09:35.200 --> 00:09:37.824 So this is a picture of for those

00:09:37.824 --> 00:09:40.065 not familiar with the four regions

00:09:40.065 --> 00:09:40.065
of the stomach, the cardia, which is where you see the incidence of this cardiac cancer is higher. And this was a nice review article by the Gastroenterology Group from UCSD, where cancer at the cardia tends to be higher in lice and less so in minorities. And it’s more strongly associated with GERD and obesity and not with socioeconomic status. Whereas the traditional cancer that is associated with Helicobacter pylori infection tends to be in the body of the stomach where the parietal cells sit. And the antrum of the stomach,
00:10:18.800 --> 00:10:20.662 which is another name for is the endocrine part of the stomach,
00:10:20.662 --> 00:10:22.160 which is where the G cells that produce gastro.
00:10:22.160 --> 00:10:23.777 And the reason I’ll be coming back to that in a second.
00:10:23.777 --> 00:10:25.182 And so it’s this region here that then it’s connected to the small intestine.
00:10:25.182 --> 00:10:26.160 And so definitely in Arizona, we’re seeing high incidence in Hispanics and the Native American population.
00:10:26.160 --> 00:10:29.120 And so so representing the socio economic status with socio increase in,
00:10:29.120 --> 00:10:31.520 decrease in socio economic status
00:10:31.520 --> 00:10:33.685 So this is more strongly associated with socio increase in,
00:10:33.685 --> 00:10:36.244 decrease in socio economic status
00:10:36.244 --> 00:10:38.899 and underrepresented minorities.
00:10:38.899 --> 00:10:41.959 And so definitely in Arizona, we’re seeing high incidence in Hispanics
00:10:41.960 --> 00:10:44.200 and the Native American population.
So as I mentioned earlier, there's an increase in interest in the tumor microenvironment, which I think is well known to the oncology group here, which is a very heterogeneous environment that is comprised of stromal cells, neuronal endothelial. But you know most of the work now is really focused on the immune cells because this is a target for the checkpoint inhibitors. And so we are coming at this from the approach that if we can decipher a bit more about the tumor microenvironment particularly in gastric cancer where
we already know that the initiation of the inflammation is from an infectious agents most of the time that we can develop better targets for treatment and biomarkers. So this is actually an example of the what we call the Correa paradigm which Playa Correa who say epidemiologist who initially was in Columbia, South America and made the observation and published in The Lancet in 75. This observation that there was chronic inflammation in the stomach that progressed on to cancer and this is looking at obviously if
This is an epidemiologic study looking at people over time. But he noticed that there was sort of this intermediate stage where some people had loss of the acid secreting portion of the stomach and a substitution of the normal epithelium of the stomach with this mucous phenotype, what we call metaplasia. In humans, the pathologists will read out intestinal metaplasia, an example here where you see goblet cells. This is the intestinal type.
The colonic type is actually more strongly associated with gastric cancer and it’s more of this foamy type of mucous metaplasia that one sees in the stomach. About half the world is infected with Helicobacter and may develop this chronic gastritis, but then about 10% will go on to develop metaplasia and 1 to 3% gastric carcinoma. The thought is, that this the tipping point in with respect to the progression toward the likelihood of progression toward cancer is at this step where
there is atrophy and metaplasia. And we started asking the question whether hedgehog signaling might be important in this transition of the mucosa from chronic inflammation to the metaplastic change. And I’ve added here to emphasize that the Pilea Correa, the paradigm was basically formulated before the discovery of Helicobacter and then once Helicobacter was identified a link was then made to this chronic and then atrophic gastritis. But we asked the question about hedgehog signaling and really is it was because of an incidental finding by Andy
00:14:09.592 --> 00:14:12.640 McMahon’s group at Harvard in 2000,
NOTE Confidence: 0.83324198
00:14:12.640 --> 00:14:14.914 where they published a paper saying
NOTE Confidence: 0.83324198
00:14:14.914 --> 00:14:16.949 that the Sonic Hedgehog knockout
NOTE Confidence: 0.83324198
00:14:16.949 --> 00:14:18.760 mouse resulted in gastric metaplasia.
NOTE Confidence: 0.83324198
00:14:18.760 --> 00:14:21.320 It turned out that probably wasn’t accurate,
NOTE Confidence: 0.83324198
00:14:21.320 --> 00:14:24.240 it probably was more hyperproliferation,
NOTE Confidence: 0.83324198
00:14:24.240 --> 00:14:26.688 but they didn’t have any GI
NOTE Confidence: 0.83324198
00:14:26.688 --> 00:14:28.320 pathologists reviewing those slides.
NOTE Confidence: 0.94402933
00:14:30.840 --> 00:14:34.828 So, but based upon that MO El Zatari
NOTE Confidence: 0.94402933
00:14:34.828 --> 00:14:36.800 at the time who was in my lab,
NOTE Confidence: 0.94402933
00:14:36.800 --> 00:14:40.916 we actually obtained the mice that
NOTE Confidence: 0.94402933
00:14:40.920 --> 00:14:43.496 are in which the Laxi reporter is
NOTE Confidence: 0.94402933
00:14:43.496 --> 00:14:46.299 knocked into the locus of the Glee
NOTE Confidence: 0.94402933
00:14:46.299 --> 00:14:48.324 one which is the transcription
NOTE Confidence: 0.94402933
00:14:48.324 --> 00:14:50.942 factor and is the transcriptional
NOTE Confidence: 0.94402933
readout for Hedgehog signaling.

So we obtained these mice, so the knock in of the Laxi molecule, we're able to maintain these mice in the heterozygous or homozygous state. But essentially you have a total body knockout and he infected those mice with helicobacter.

Now we typically use Feliz in the mice because you get a much more aggressive inflammatory response sooner. We were hoping to save a little bit of money and see changes, you know, and not have to wait six months. But with pylori itself using the human pathogen,
NOTE Confidence: 0.94402933
00:15:26.680 --> 00:15:28.880 it can take a lot longer and the
NOTE Confidence: 0.94402933
00:15:28.880 --> 00:15:31.478 inflammatory response is not as robust.
NOTE Confidence: 0.94402933
00:15:31.480 --> 00:15:35.180 So he looked at the mice
NOTE Confidence: 0.94402933
00:15:35.180 --> 00:15:37.680 at the time of infection,
NOTE Confidence: 0.94402933
00:15:37.680 --> 00:15:39.480 two months after infection and
NOTE Confidence: 0.94402933
00:15:39.480 --> 00:15:41.280 then six months after infection.
NOTE Confidence: 0.94402933
00:15:41.280 --> 00:15:45.426 And so you can actually identify
NOTE Confidence: 0.94402933
00:15:45.426 --> 00:15:47.935 fluorescently using an antibody.
NOTE Confidence: 0.94402933
00:15:47.935 --> 00:15:52.025 So in the uninfected mice you see that
NOTE Confidence: 0.94402933
00:15:52.025 --> 00:15:54.155 the alpha smooth muscle positive cells,
NOTE Confidence: 0.94402933
00:15:54.160 --> 00:15:57.023 which are the mild fibroblasts in the
NOTE Confidence: 0.94402933
00:15:57.023 --> 00:15:59.238 stomach are positive for Glee one,
NOTE Confidence: 0.94402933
00:15:59.240 --> 00:16:02.264 and pretty much those were the only
NOTE Confidence: 0.94402933
00:16:02.264 --> 00:16:04.808 cells that were expressing Glee one.
NOTE Confidence: 0.94402933
00:16:04.808 --> 00:16:05.640 So again,
Glee one is in the stroma and typically what happens is that the epithelial cells such as the parietal cells will make the ligand Sonic hedgehog and it’s received by the stromal cells. So in the uninfected mice, it’s the alpha smooth muscle positive cells. But during after two months of infection with Helicobacter, not surprisingly you have a pro inflammatory situation where you have an infiltration of inflammatory cells and those cells are positive for Glee one. And when we did flow cytometry, this is published several years ago now mostly myeloid cells but
not T or B cells were the cells that were expressing Glee one. And so just to summarize that basically we then we’re asking the question well what are these Glee 1 positive immune cells doing. And what was interesting is that when we infected again wild type mice and now what you see here is an immunofluorescent stain for the different cell populations in the corpus of or the body of the stomach intrinsic factor in the mice mark the chief cells whereas in human it’s normally in the parietal cells,
HKTPAS marks the parietal cells and GS2 lectin marks a mucous cell. So here in the mice that are infected for six months you see that they’re developing A metaplasia and atrophy. These this region should show parietal cells, which would be the orange stain, but you can see here they’re kind of moved off to the side because they’re starting to disappear and show atrophy and being replaced by this mucus phenotype. However, in the cells that were heterozygous for the
Glee, one deletion or because of the Laxi insertion into the locus or homozygous, they maintain the normal architecture of the stomach. And so we were really surprised by that because as I mentioned earlier, it’s the stromal cell that’s expressing Glee one. So this was telling us right there that there was something going on in the micro environment, in the immune environment, that was affecting the mucosa. So to try to summarize this quickly, so we started looking at hedgehog.
signaling in this transition
from gastritis to metaplasia.
I should also mention one of the issues with working with the mouse models is they never progressed to dysplasia and cancer, just with an infection from Helicobacter. So we could only look at this step.
So I’ve just shown you that Glee one is important in the meta formation of metaplasia.
So this like I said is you know 2015 sixteen we were doing this so we did microarrays and we identified this molecule Schlafen 4. There certainly were quite a few other genes,
but this one was interesting because there were some papers of both Schlafen 2 and Four I should mention. But the reason why we didn’t pursue the pathogenesis related to Schlafen 2 is because Two does not have a ortholog in in humans. So there is an ortholog for Schlafen 4. So we wanted to be eventually be able to translate the work that we were doing in mice into humans. So that’s why we focus on Schlafen 4. This was the further analysis of this locus which we identified in the array of the mice.
Comparing wild type mice to the Glee One knockout mice, you can see here that there’s a decrease in Schlafman 4, which suggested that this gene was regulated by hedgehog signaling. And so we did chromatin immunoprecipitation at the time to determine that indeed Schlafen 4 is a direct target of Glee one. So you can show that it does sit on the promoter of Schlafen Four. However, what exactly are these Schlafen’s. So the reason why we focused on them again is because there was a
NOTE Confidence: 0.779132571333333
00:20:49.042 --> 00:20:52.292 paper in immunity in 1999 that said
NOTE Confidence: 0.779132571333333
00:20:52.292 --> 00:20:55.007 that the Schlafen molecules were
NOTE Confidence: 0.779132571333333
00:20:55.007 --> 00:20:58.041 involved in both T cell and and
NOTE Confidence: 0.779132571333333
00:20:58.041 --> 00:20:59.620 and myeloid cell differentiation.
NOTE Confidence: 0.779132571333333
00:20:59.620 --> 00:21:01.894 So that's why we thought, well,
NOTE Confidence: 0.779132571333333
00:21:01.894 --> 00:21:04.512 you know if we're looking at Hedgehog
NOTE Confidence: 0.779132571333333
00:21:04.512 --> 00:21:06.343 signaling and it's rolling the
NOTE Confidence: 0.779132571333333
00:21:06.343 --> 00:21:08.860 stroma and its effect in mediating
NOTE Confidence: 0.779132571333333
00:21:08.860 --> 00:21:11.360 the this metaplast gastritis and
NOTE Confidence: 0.779132571333333
00:21:11.360 --> 00:21:13.023 metaplastic transition that that
NOTE Confidence: 0.779132571333333
00:21:13.023 --> 00:21:14.793 would be a a good target.
NOTE Confidence: 0.779132571333333
00:21:14.800 --> 00:21:16.320 So actually I'm gonna just
NOTE Confidence: 0.779132571333333
00:21:16.320 --> 00:21:17.840 give you a quick primer.
NOTE Confidence: 0.779132571333333
00:21:17.840 --> 00:21:19.540 The Schlafen locus however is
NOTE Confidence: 0.779132571333333
00:21:19.540 --> 00:21:21.240 fairly complicated and this is
NOTE Confidence: 0.779132571333333
what I was kind of getting at.

So we identified Schlafen 4.

There’s actually quite a bit of information from one group that’s looking at Schlafen 2.

We did see this go up in mice, but you can see that it doesn’t have its ortholog in humans.

Just plant that seed in your brain.

These are what are called the intermediate schloffens.
And the reason why that’s important is because the longer schloffens ones in green have another domain that’s a helicase domain that’s thought to bind to nucleic acids. I will be coming back to this point later, and so coming back to the mouse model, we’re interested in that gastritis to metaplastic change and we’ve identified these immune cells that are Schlafen positive. And so to understand more...
of what they did, we created a very fancy mouse model. And I know some people are not as familiar with some of these you know, kind of mouse tricks that we do. But essentially we took the mouse promoter, it was a large back trans gene. We hook it up to inducible Cree recombinase. We breed this mouse line to a reporter mouse line TD tomato. So this hybrid mouse is expressing or can be expressed in the presence when we give it tamoxifen this reporter so turning the cells red. But what we also did is to do a bone marrow transplant and put the
note confidence: 0.779132571333333
00:23:20.547 --> 00:23:23.766 bone marrow from these mice into a
note confidence: 0.779132571333333
00:23:23.766 --> 00:23:26.081 radiated mice so that essentially
note confidence: 0.779132571333333
00:23:26.081 --> 00:23:28.795 only the immune cells are going
note confidence: 0.779132571333333
00:23:28.795 --> 00:23:31.273 to be labeled with TD tomato.
note confidence: 0.779132571333333
00:23:31.280 --> 00:23:32.760 and ask the question,
note confidence: 0.779132571333333
00:23:32.760 --> 00:23:35.280 can we lineage trace this Schlopfen
note confidence: 0.779132571333333
00:23:35.280 --> 00:23:38.400 positive cell from the bone marrow
note confidence: 0.779132571333333
00:23:38.400 --> 00:23:41.118 of these mice that have recovered
note confidence: 0.779132571333333
00:23:41.120 --> 00:23:43.265 and infected with Helicobacter in
note confidence: 0.779132571333333
00:23:43.265 --> 00:23:45.920 waiting four to six months to see
note confidence: 0.779132571333333
00:23:45.920 --> 00:23:48.120 you know how they get to the stomach.
note confidence: 0.779132571333333
00:23:48.120 --> 00:23:50.592 again this is was published in 2016,
note confidence: 0.779132571333333
00:23:50.592 --> 00:23:53.088 but i just wanted to show you that
note confidence: 0.779132571333333
00:23:53.088 --> 00:23:55.237 it’s really been a very powerful
note confidence: 0.779132571333333
00:23:55.237 --> 00:23:57.669 tool for us because you can see
note confidence: 0.779132571333333
here like stars in the sky and what
I'm showing you here is a wild type mouse infected with Helicobacter.
But I'm taking we're taking these mice at four months before we have seen the cells actually arrive in the stomach.
However, if we breed those mice onto a background where they're where the Sonic Hedgehog,
the ligand signal is goosed up.
So it was pretty easy.
It was just a PCMV Sonic Hedgehog Transgene.
We breed those mice, you know,
with the TD tomato signal and you can see at four months there are these cells that are TD tomato positive.
Here’s a high-powered view. Since they’re fluorescent, we can pull them out. You can see they have a granulocytic nucleus and they are exhibiting markers of a granulocyte. Even better. You can certainly do all sorts of arrays, which I’ll get into a little bit later. But more importantly, we could actually isolate these cells from the infected stomach and show that they had T cell suppressor activity. So we did the Co culture and show that they were really functionally.
T myeloid derived suppressor cells.

So I wanted to show you well what’s the connection between Hedgehog and how this gene is regulated and why I think we were happy that we decided to kind of stick with this even though nobody’s heard of Stroffen. So what you see here is where you can isolate these cells from. We basically know create a pus situation by injecting them with thioglycolate, take the peritoneal cells and then we can culture them and incubate them with recombinant Sonic hedgehog. About a fivefold induction of
Sonic hedgehog message.

Helicobacter alone threefold but the two together synergize but more importantly interferon alpha.

So type 1 interferons, 800 fold induction, This gene is and that locus is very strongly induced by type 1 interferons.

However, if you isolate those cells from a Glee One null mouse, you can see that This is why this locus is still dependent upon hedgehog.

You can get a little bit of induction, but essentially it’s a dead promoter. So it’s like you need 2 keys to unlock.
this gene and follow it and we mapped the,
so essentially the hedgehog signal
Glee one is a constitutive signal.
The inducible signal is through
type 1 interferons.
And so then we asked the question,
well you know in the infected Mao
interferons coming from and it turns
out that and we’ve done some later
work that was published in 2022.
But basically plasma cytoid dendritic
cells are sort of resident dendritic
cells that are the most the cell
population that is probably sensing
the debris field there chronically
and why probably why it takes time for this to develop. So really putting all together and you may want to look at our gastro paper in 2022, what we’re saying is that Helicobacter infection is detected not only by the epithelium, so the epithelial cells will also produce type 1 interferon, but sort of PER on a per cell basis, it’s the plasma cytoid dendritic cell. There’s a certain pathway with activation of the interferon response factors, which are the factors.
transcription factors that bind to the type 1 interferon promoters releasing type 1 interferons that then will polarize what we now think is a neutrophil or granulocytic cell that has been sitting there and had was recruited to the stomach at some point in time. But then this debris field and threshold must be reached over time. So these cells are PDL 1 positive and we were able to show as I mentioned earlier that they do have T cell suppressor function. But analysis of these cells also reveals that they are producing other cytokines not surprisingly some
of which that were of particular interest to us or was IO 1A and Beta.

And we think that and that’s why we think that it’s the immune cells that are really picking up the baton and really pushing the mucosa more toward cancer as opposed to the bug itself.

And recently and I didn’t put the reference in here, we actually had for other reasons had generated a triple transgenic mouse where we can inducibly over express I-1 beta in the antrum. So you may ask, why would I bother to do that?
And it’s because the Helicobacter infection, whether it’s Feliz or Pylori, when we infect the mouse, because the mouse stomach is actually aph of three or four compared to our stomachs, which is pH of one, the Organism tends to only infect the corpus, not the antrum where traditionally you see it in people. So we really wanted to understand distal gastric cancer where we can drive a, much more aggressive tumor in the antrum of the stomach. And so we took the gastroin,
we made a gastroin Cree ERT two,
crossed it to a TET activator, RTTA.
So these are three different mice that have to be all bred together.
And then this mouse is then bred to a Tet on where we’ve inserted the IO1 beta,
where it’ll generate A secreted form.
And so you give the mice tamoxifen.
So the the TET TET activator will sit in the cytoplasm until we give
the mice doxycycline in the water.
And so we keep them on doxycycline and after about six months we about
40% of the mice will develop these.
ugly dysplastic looking tumors. I caution to call it cancer because the mouse models never metastasize. I have yet even the colon, all the models that people talk about, they never metastasize. So you know you can kind of quibble about what you want to call that. But I'll just say you can see there is they're pretty ugly looking cells and more importantly these cells are so they do have and have recruited the Schlafen for positive MDS, CS into the tumor. So at least we now have a sort of a pre clinical model to actually study.
So going back again in time a little bit,

So 2020 we started to do bulk RNAC which we did with these mice that were TD Tomato positive.

What I want to point out here that was quite interesting and coming back to the Type 1 interferon theme is that a lot of the genes that we identified, the heat map happened to be interferon, strongly interferon regulated and were these guanalite binding proteins or GTP aces.

This is our the changes in the heat map, binding proteins or GTP aces, GBP, 2G VIN and they’re of the dynamin class of GTP aces.

This is our the changes in the heat map, 2G VIN and they’re of the dynamin class of GTP aces.

This is our the changes in the heat map,
the full log pole change.

But I am comparing it to a paper in 2019 where it was really elegant study of both a mouse and human lung cancer.

So there were seven patients with lung cancer and they had a mouse model using Ras and I want to say P53.

There was another gene where they were able to generate lung cancer and they had a mouse model using Ras and I want to say P53.

There was another gene where they were able to generate lung cancer and they did a complete analysis by single cell sequencing of the tumor microenvironment what they call tumor associated neutrophils or Tans.

They had the same gene profile.
that we identified in our Schloffen MDSCS and I highlight here
that their mouse into was positive for Schloffen 4 and here this is the human counterpart for seven patients.
I think one of the problems they I didn’t see Schloffen 12 but again when you move to human you’ve got a whole variety of stages,
you’ve got a whole variety of stages, tumor types etcetera.
And so we we they did not observe it in that but all the other genes were similar. We’ve also gone on to show that using proteomic analysis and using the Schloffen 4 antibody that we can actually
pull down and show that Schloffen 4, which I didn’t mention is actually a cytoplasmic. It’s actually an ER membrane endoplasmic reticular membrane protein. So I’ll come back to that. So that even adds to the complexity what are we dealing with. But interestingly it forms a complex with at least when we pull down with many of these genes that we identified in the bulk RNA seq. A little bit of a complicated slide here again it’s published for those that are interested. So if we take that pull down using
Schlafmann for antibody and we wanted to know whether it had Gtpas activity. So we take that complex where we pulled it down and actually show that it can hydrolyze GTP and so shown here and it does that here higher levels in blue of GTP bold change and the interferon treated cells where we do the pull down versus we have also made recently a Schlafmann for knockout mouse model. So if we isolate cells from those versus sildenafil, now why did I use sildenafil? I kind of skipped over that and
that’s because some of the genes

also were these G cyclic GMP related phosphodiesterases.

So we already were starting to think,

there’s already an off the shelf.

there’s already an off the shelf.

there’s already an off the shelf.

inhibitor of phosphodiesterases,

plus I’m sure the oncologists

are very familiar with,

particularly from the multiple

myeloma field where you can use these

phosphodiesterase 5-6 inhibitors

as a sort of neoadjuvant.

So that was one of the reasons
00:35:48.430 --> 00:35:49.806 why we thought, oh,

00:35:49.806 --> 00:35:51.836 let’s see whether this works.

00:35:51.840 --> 00:36:00.155 And indeed it also knocks down the

00:36:00.160 --> 00:36:04.222 ability of the this complex to form GTP.

00:36:04.222 --> 00:36:07.776 So we put together this model which I’m

00:36:07.776 --> 00:36:10.016 showing you here that interferon will induce.

00:36:10.016 --> 00:36:12.200 Because remember it’s a very

00:36:12.200 --> 00:36:14.984 strong inducer of Schlopfen,

00:36:14.984 --> 00:36:16.978 so we can mark these cells but along

00:36:16.978 --> 00:36:19.564 with Schlopfen there are other

00:36:19.564 --> 00:36:22.504 very important type 1 interferon

00:36:22.504 --> 00:36:25.319 regulated genes that appear to

00:36:25.320 --> 00:36:28.632 be somewhere in this pathway.

00:36:28.632 --> 00:36:31.317 of a model because essentially what
these myeloid derived suppressor cells their ability to inhibit T cells has to do with their being able to gobble up L arginine so that the T cells can’t proliferate. But what are these myeloid derived suppressor cells are actually using that L arginine themselves to generate reactive oxygen species. Here are some of the pathways. So arginase making nitric oxide or NO2 make making nitric oxide, which happens to be a cofactor for soluble guanillate cyclase.
So guanulate cyclase generates cyclic GMP.

Cyclic GMP, if it hangs around is a cofactor for protein kinase G, which can in some cell populations trigger the cells to undergo cell death.

So if you have high levels of something that’s going to break down cyclic GMP, you’re going to move the cells away from apoptosis, regenerate this the sort of backbone for regenerating GTP. And so that’s why we think and I’ve shown you that Schlafen 4 is at least in a complex with these Guanali binding proteins which you know need this GTP.
So we think that there’s a whole

nother pathway or metabolism that pulls the substrate away from maintaining high levels of cyclic GMP and you can essentially accelerate that and we’ll get back to that, oops, going too fast if we inhibit phosphodiesterases.

So you can imagine if we block phosphodiesterases here, this is going to build up and you can trigger the cells to undergo apoptosis.

So that’s kind of the hypothesis that I want to keep in mind.

OK, so let’s move on.
We’ve moved to the next era where we’re now using single cell sequencing. And I want to point out again that we’re reinforcing what we initially observed and I just want you to this is published, but you can see here in our go enrichment for this is the Gtpas activity, GTP binding. So again a lot of the genes even in the doing the single cell sequencing seem to take us to these Gtpas types of proteins.
I want to highlight though this region here which kind of didn’t blow up quite as big as it should. But what we were kind of surprised about is that there’s really three groups, low, medium, medium, high and high expressors of Schlafen. And this is what we’re finding many times as you start to get into single cell sequencing is that many of these cells exist in sort of different activation states. I we haven’t quite gotten to the pseudo trajectory. Somebody’s working on that ’cause you need,
you need a different program. But what you can kind of see is that the Low Expressors Group 3, which is this blue, actually it has more of the neutrophil genotype, so that would be I guess no. Anyways, I won’t point it. I guess this group here. And whereas the higher expressing ones, there’s one group number two that tends to be and so that’s this cluster here higher in nitric oxide which is actually a different group than that express arginate. So this is just the mouse.
So even that mouse cluster that we are,

we're already thinking that we're

derived suppressor cells from

They actually have different sort of

activation states or different gene

clusters that you can now identify

by single cell sequencing. OK.

So I've given you a lot of information.

So essentially from the mouse model,

what we're saying is that,

and I didn’t really give you

the sort of how this all begins,

but essentially when Helicobacter

infests the stomach, it can,
the dying parietal cells or intraparietal cells actually can release Sonic Hedgehog into the plasma. Some of the papers that I didn’t talk about in detail actually you can pick up Sonic Hedgehog in the plasma of the mice within two or three days these cells track to the stomach. But the first two months or so of the infection it’s we’re still in more of the pro inflammatory stage. It’s not till about when we did a formal time course about five and a half, six months of a Helicobacter infection in mice.
Do you actually see these cells

Do you actually generate enough interferon

That and I'm the reason

why I'm crossing that out,

is that we actually infuse interferon

antibody in our 2022 paper to show

that we could actually block the

polarization of the Schloffen for MDS,

CS and we did not get the spim.

Is the the term metaplasia that

we use for the mice,

it stands for spasmolytic

polypeptide expressing metaplasia,

but we just call it SPM because in the

mice you actually don’t see the goblet cells.
So they had to come up with another way to market.

And so again what we’re proposing is that if we block the phosphodiesterases and maybe these along with the GTP Azes that we can do the same thing.

So what I’ve shown you is more in vitro data, but now I’m going to show you what it looks like with the knockout.

So as I mentioned this is a normal mouse and like I said we can goose up the the whole signal and and get the metaplastic change faster if we over express with Sonic Hedgehog.

So the green staining you saw before
00:43:17.345 --> 00:43:19.839 is the metaplastic change in the mice.
NOTE Confidence: 0.931444244761905
00:43:19.840 --> 00:43:24.480 And when we do the conditional deletion
NOTE Confidence: 0.931444244761905
00:43:24.480 --> 00:43:27.077 and we’re deleting it using Glee one,
NOTE Confidence: 0.931444244761905
00:43:27.080 --> 00:43:28.223 Cree ERT two.
NOTE Confidence: 0.931444244761905
00:43:28.223 --> 00:43:30.890 So we’re deleting it in that those
NOTE Confidence: 0.931444244761905
00:43:30.979 --> 00:43:33.829 myeloid cells that we originally
NOTE Confidence: 0.931444244761905
00:43:33.829 --> 00:43:35.588 identified the Schlafen cells in.
NOTE Confidence: 0.931444244761905
00:43:35.588 --> 00:43:38.561 And you can see that you start to read
NOTE Confidence: 0.931444244761905
00:43:38.561 --> 00:43:40.799 the normal architecture of the stomach.
NOTE Confidence: 0.931444244761905
00:43:40.800 --> 00:43:43.480 The parietal cells are shown here in white,
NOTE Confidence: 0.931444244761905
00:43:43.480 --> 00:43:46.360 are starting to come back.
NOTE Confidence: 0.931444244761905
00:43:46.360 --> 00:43:47.998 What about Sildenafil?
NOTE Confidence: 0.931444244761905
00:43:47.998 --> 00:43:49.636 Didn’t take much.
NOTE Confidence: 0.931444244761905
00:43:49.640 --> 00:43:52.760 We did two injections of sildenafil,
NOTE Confidence: 0.931444244761905
00:43:52.760 --> 00:43:53.460 same thing.
NOTE Confidence: 0.931444244761905
And here I’m showing you an H&E where you can really see the parietal cells, which I’m used to looking at it. But these big pink cells are your parietal cells, starting to return in the presence of just after two injections of sildenafil and very recently within the last couple of months going back to our aisle 1 overexpressing mice with those big ugly tumors. So here you can see in this low power view. Here is the villi of the intestine. Here is the pyloris. Here is the villi of the intestine. Here is the pyloris.
the junction between the stomach or the antrum and the in the small intestine.

These are Bruner’s glands. Here the tumors develop and we’re able to accelerate it if you give it the MNU nitrosamine. So instead of 40% of the mice alone, we get about 60% of the mice we’ll develop these ugly dysplastic tumors. But two injections of SILDENAFIL were able to melt those tumors down. And the reason why I put this in here, this is again kind of hot off the presses. I want to come back to, OK, I told you that I mean Schlafen
00:45:09.434 --> 00:45:12.758 is AER protein, well guess what,
00:45:12.760 --> 00:45:15.040 it’s an RNA binding protein.
00:45:15.040 --> 00:45:20.136 And so we actually have recently done a
00:45:20.136 --> 00:45:23.272 pull down again with the Schlafen antibody.
00:45:23.280 --> 00:45:27.445 These are transfer RNAs and what’s very
00:45:27.445 --> 00:45:30.638 interesting is that it actually binds to
00:45:30.638 --> 00:45:33.880 very specifically in an inducible manner,
00:45:33.880 --> 00:45:37.678 glycine and tyrosine specific transfer RNAs.
00:45:37.680 --> 00:45:39.440 I don’t have time to get into it right now,
00:45:39.440 --> 00:45:41.160 but we can come back to it at the end.
00:45:41.160 --> 00:45:43.923 But I just wanted to start to close the
00:45:43.923 --> 00:45:46.354 loop of this is very interesting protein
00:45:46.354 --> 00:45:49.399 and why is it so important and why an
00:45:49.400 --> 00:45:51.680 ERRNA binding protein is involved.
00:45:51.680 --> 00:45:54.837 OK. I’m going to because of time,
I’m going to come back to this diagram which I know is pretty complicated because I wanted to show you our phase two clinical trial. So this is a collaboration with primarily a really talented junior faculty in oncology, Junaid Arshad, Rosten Schroff is our Chief of he Monk and Aaron Scott are the trio of GI oncologists. And so when I presented this to them, they Janae suggested, well you know why not just try, let’s try and see if we can set up a window trial. And so essentially I didn’t know what a window trial was,
but he said you know what we can do because most of these patients are going to have to go to receive standard of care, flot therapy and then go for a gastrectomy if we if there's stage one to three. So we're only dealing with stage one to three, well 1B to three and so these are the window trial objectives. So the primary objective and I didn’t realize this, we can’t just give patients to Dalafail even though it the safety profile we know is pretty good. I’m not supposed to I guess because of CME,
I'm not supposed to say the trade name. But anyway, so they're just focused on this feasibility and safety and but the secondary objectives shown here, to see whether there's some pathologic response. But my interest in what I'll show you because the study is still ongoing, I'll just show you that of what we're looking at in terms of does tadalafil in a patient with actual gastric cancer do anything, right. So remember we've got to follow L these are the exclusion criteria,
study feasibility.

So we’ve been going for about a year. We’ve got six patients enrolled, 2 patients have finished the study.

But what I want to show, so we’re our goal is to enroll 10 patients.

When I moved to Arizona, I set up a repository for our endoscopy lab. I or one of the other endoscopists will do a if if the patient is not referred in at their not referring him from the outside that becomes a problem because we actually want to try to do single cell sequencing at each of these intervals.

So that really means that we
have to do the endoscopy here.

So just with the biopsies, jumbo biopsies we can do single cell sequencing.

And I just wanted to show you that even in a gastric cancer patient, we can stain for Schlaf and 12 L So it’s there in the immune cells in the lamina propria of these tumors.

So I’ll just show you a little bit of, let’s see and I’m sorry it ends up going counterclockwise because of the way the data gets uploaded into the cloud for the 10X genomic analysis. So what we are able to do because obviously we run into problems with we’re
able to capture the 2nd interval endoscopy, so this one, but if the patient comes in from the outside, we basically do single cell sequencing. We have plenty of normal referrals to endoscopy that there’s nothing there. They don’t have gastritis and so we can do single cell sequencing on those patients. So what I have circled here is the Myeloid cluster in a normal patient, one of my patients that I had referred for endos because I knew that I was having trouble eradicating Helicobacter. So they had Helicobacter gastritis and here intestinal metaplasia.
And then this was one of the patients that the first patient that was enrolled in the study. And so they actually have a lot more of these Schlafen 12 L positive cells, which if you look at the just that gene, you can see here that this is where the myeloid cells are. But look at the normal gastritis intestinal atrophy, I'm sort of going in order. Sorry, I didn’t give you the preya paradigm, but Schlafen 12 L doesn’t come on until you very strong, strongly, maybe a little bit in the metaplastic stage, but until these patients are actually,
you actually have gastric cancer

now you’re gonna say, well,

what are these other cells?

They’re T cells.

So this was the big surprise as we move and not surprisingly when you move from mouse models to people,

you know, sometimes all bets are off.

So we now also have to understand what’s going on in these T cells because you can see again Schlafen 12 LS picked up in the T cells and gastritis and intestinal metaplasia.

Now I think I have one slide here.

It turns out that
00:51:27.400 --> 00:51:30.600 in the actual cancer,
NOTE Confidence: 0.9529675
00:51:30.600 --> 00:51:32.938 the T cells that are most prominent
NOTE Confidence: 0.9529675
00:51:32.938 --> 00:51:35.349 that you don’t see in the other
NOTE Confidence: 0.9529675
00:51:35.349 --> 00:51:37.353 groups are the exhausted T cells.
NOTE Confidence: 0.9529675
00:51:37.360 --> 00:51:40.555 So that’s going to be a whole other project
NOTE Confidence: 0.9529675
00:51:40.555 --> 00:51:43.584 to understand what is this molecule
NOTE Confidence: 0.9529675
00:51:43.584 --> 00:51:48.960 doing in terms of the metabolism of T cells.
NOTE Confidence: 0.9529675
00:51:48.960 --> 00:51:50.517 So we have our work cut out for us.
NOTE Confidence: 0.9529675
00:51:50.520 --> 00:51:54.400 I finally wanted to show you what happens
NOTE Confidence: 0.9529675
00:51:54.400 --> 00:51:58.920 with Tadalafil and so here’s a cancer,
NOTE Confidence: 0.9529675
00:51:58.920 --> 00:52:01.179 so this was one of the patients where we
NOTE Confidence: 0.9529675
00:52:01.179 --> 00:52:03.000 the first, this was our first patient.
NOTE Confidence: 0.9529675
00:52:03.000 --> 00:52:05.760 were referred in from the outside.
NOTE Confidence: 0.9529675
00:52:05.760 --> 00:52:11.120 So we only had slides and so this
NOTE Confidence: 0.9529675
00:52:11.120 --> 00:52:13.850 is sustaining for CD11B myeloid
marker and our Schlofen 12 L Co localized here in the merge view, but here the high-powered view in the cancer, but with Cialis, oh sorry, Tadalafil that we are eliminating these Schlafen positive MDSCS. OK. So the take away there seems to be overlap between the pathways regulating Schlafen 4 and we also believe 12 L and cyclic GMP dependent phosphodiesterases and these inhibitors allow cyclic GMP to accumulate and induce MDSE apoptosis and that’s the mechanism. Their elimination we think is we can at least see it in our mouse model.
The big question will be as we expand this trial and get past the safety stage that this potentially may be a neoadjuvant for gastric. But again these cells are in a lot of cancers and we need to think about it in several cancers. So that I just want to certainly acknowledge linding who moved with me from Michigan and has really carried out all of these studies. Acknowledge again our HE monk GI group division and our targets by a repository and to thank the patients for their participation. Thank you.
I'll take any questions and I should, yes, there's also two questions online. So thank you for this education. But my question is the degree of expression in the myeloid cells. Is it something innate or is it acquired? Do we know? Is it like, is it something that's hereditary tendency to have higher expression in certain individuals and lower in others.
00:54:28.080 --> 00:54:30.960 people. So your question is whether people are predisposed because they have snips or mutations.
00:54:32.690 --> 00:54:35.160 I’m just saying is it does it take like a two hit phenomena where you have H pylori infection but there is innate over expression of certain of these proteins and then that’s when cancer happens? And I also had a second question. Do you think that some of the same pathways are involved in other types of gastric cancer like smoking related or others.
00:55:00.640 --> 00:55:02.860 So this pathway I think and
00:55:04.840 --> 00:55:04.840 is similar. I shouldn’t because of

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the type 1 interferon regulation, is it similar to like the Sting C gas pathway?

I haven’t looked to see where the parallel and the overlap is, but I would emphasize that you know DAMPS, but probably even Pamps certainly can activate these plasma cytodendritic cells. The reason why I I like focusing on the Schlafen is because we’re able to take it all the way down to the promoter. We know why that promoter and those cells get marked. So it suggests that you really need a very strong induction.
of Type 1 interferons or maybe there's mutations in those Irfs, etcetera constitutive.
I mean it gets pretty complicated where whether people may be predisposed or not, we are some of the endpoints that we're looking at it are so TLR 9 mainly because there is already information actually related to gastric cancer and Helicobacter that patients that have mutations in TLR 9 May have a more aggressive response to an infection with Helicobacter. So that's we're starting with more upstream. Yes, thank you. That was a great talk. So Tadalphil is,
00:56:32.720 --> 00:56:34.220 you know, prescribed for
00:56:34.220 --> 00:56:35.720 other things as well.
00:56:35.720 --> 00:56:36.920 Do you, have you considered
00:56:36.920 --> 00:56:38.184 doing like a retrospective
00:56:38.184 --> 00:56:40.080 study and looking at you know,
00:56:40.080 --> 00:56:42.360 maybe stomach cancer versus people
00:56:42.360 --> 00:56:44.640 who’ve been prescribed to Dalafil,
00:56:46.040 --> 00:56:47.900 a retrospective study? Yeah.
00:56:47.900 --> 00:56:51.440 So in other words, it’s in wide use.
00:56:53.800 --> 00:56:55.655 The problem is and I think we’re
00:56:55.655 --> 00:57:00.055 going to need AI to do these kinds of
00:56:58.165 --> 00:57:00.055 things because it’s it’s really being
00:57:00.055 --> 00:57:02.680 someone has to mine the clinical data.
00:57:02.680 --> 00:57:04.145 I’d have to see whether
00:57:04.145 --> 00:57:05.317 it’s already out there.
Most likely it’s not for gastric cancer maybe for one of the bigger cancers like lung or colon cancer. But I still think it’s going to take some energy to pull it out of the clinical records and really analyze it. I really offhand I haven’t seen any papers really looking at that but it’s that’s an excellent question.

Thank you, Clara. Hello. 2 questions. One, back to the inflammatory cytokine role. And you showed that overexpressing is sufficient to contribute. But if you sort of throw in inhibitors or utilize cell specific
00:57:44.290 --> 00:57:46.600 deletion of Aisle 1 beta TNF,

00:57:46.600 --> 00:57:48.679 you had a range of different cytokines.

00:57:48.680 --> 00:57:51.394 What’s the effect of deletion in

00:57:51.394 --> 00:57:53.038 your model on the end outcome?

00:57:53.040 --> 00:57:57.080 So deletion of like PNF or we we haven’t,

00:57:57.760 --> 00:57:58.960 yeah, we haven’t gone there.

00:58:00.920 --> 00:58:04.384 You can imagine how many mice my

00:58:04.384 --> 00:58:06.600 yeah, we haven’t gone there.

00:58:06.600 --> 00:58:08.240 I just it’s reading all of those

00:58:08.240 --> 00:58:10.800 yeah mice onto those backgrounds

00:58:10.800 --> 00:58:12.800 the antibodies a little cheaper.

00:58:12.800 --> 00:58:14.478 And then I guess the second question

00:58:14.880 --> 00:58:16.714 is a little bit related to the

00:58:16.714 --> 00:58:18.120 spectrum of Schlafen expression.

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If I know you mentioned in the mouse model that you can’t, you don’t see progression to the to cancer, it’s more than metaplasia. But in human if you try and sort of consider the transition between metaplasia to gastric cancer. Well, I guess the first, can you speak to some of the things contributing to that enabling that transition into the in the 1 to 3% that sort of overlap with some of the pathways you’ve highlighted. In other words, you got the 10% that have metaplasia and then one to 3% actual gastric cancer,
And so in I guess in your studies that you’re doing where you’re looking at are you able to look at spectrum of Schlafen expression in that subset that goes on to gastric cancer relative to those that stay in metaplasia. So I’m trying to so have we looked at so you’re taking gastric cancer or you mean taking metaplasia patients with metaplasia, I mean in the pathway are you and and it can be Schlafen and can be you know for the full for the full pathway.
In general are you able to see that those that progress to cancer are fit on the higher end of or on the altered end of expression of the pathway relative to those that remain in metaplasia. I'm so I'm sorry altered of I'm so I'm sorry of your can you segregate like in any number of thing can you segregate the high versus low in the shop and pathway for those that progress versus that remain in metaplasia Oh so but in people or or in the in from bulk RNA or from other types of data sets that may
NOTE Confidence: 0.753566116666667
01:00:07.096 --> 01:00:08.440 have been done in the stomach.
NOTE Confidence: 0.8566534925
01:00:09.520 --> 01:00:11.960 You know it just really hasn’t been done.
NOTE Confidence: 0.8566534925
01:00:11.960 --> 01:00:16.979 We haven’t really segregated the subtypes of
NOTE Confidence: 0.8566534925
01:00:16.979 --> 01:00:21.840 Schlafen 12 L cells in the patients at all.
NOTE Confidence: 0.8566534925
01:00:21.840 --> 01:00:25.412 I I mean it it’s we’re just a lot of
NOTE Confidence: 0.8566534925
01:00:25.412 --> 01:00:28.500 it is just numbers and we’re just happy
NOTE Confidence: 0.8566534925
01:00:28.592 --> 01:00:31.328 to be able to to well you know with
NOTE Confidence: 0.8566534925
01:00:31.328 --> 01:00:33.353 the repository the logistics is not
NOTE Confidence: 0.8566534925
01:00:33.353 --> 01:00:36.127 as tricky because I have to have our
NOTE Confidence: 0.8566534925
01:00:36.127 --> 01:00:38.710 inpatient teams let us know there’s a
NOTE Confidence: 0.8566534925
01:00:38.710 --> 01:00:41.012 patient in house patient has to agree
NOTE Confidence: 0.8566534925
01:00:41.012 --> 01:00:43.335 many times they don’t come or they
NOTE Confidence: 0.8566534925
01:00:43.335 --> 01:00:45.267 come and they we’ve already gotten
NOTE Confidence: 0.8566534925
01:00:45.267 --> 01:00:47.428 the biopsies and our hospital will
NOTE Confidence: 0.8566534925
01:00:47.428 --> 01:00:49.730 not release that the even the tissue.
NOTE Confidence: 0.8566534925
So we have a lot of just sort of logistical issues but I'll keep that in mind as we or I can send you the data.

Yeah, the data sets out there from the stomach and then look at high versus, you know the high versus lower expressors. Yeah, ends of the persistent hedgehog sibling in the MDSC cells required to maintain the dysplastic tumors and if so is
there a role for smoothened inhibitors?

Oh that Doctor Kaplan had raised that issue yesterday.

We could think about Vesmotogib to revisit that I it’s a good question and I I mean I would try it out in our mouse models probably first I I do know that.

So if you use a a Glee one null, we we didn’t, we haven’t used any hedgehog inhibitors but basically you as I showed you with the in vitro data, you need hedgehog.

some kind of hedgehog signalling for that promoter to come on.

The assumption is that the
cells aren’t or polarizing.
but we haven’t gone back
to really explore that.
Oh, they were listening.
Oh, OK. I thought there was,
there was two things on the line,
but it was just the CME.
OK, no questions. All right.