Luisa, maybe we can take a step back first and just tell us a little bit about yourself and what you do. I am a cancer scientist. I basically try to understand how cancer cells work?

I am originally born and raised in Columbia, South America, but I always had a passion to come to the US to train in cancer biology and therapy. And this was based on an

I basically try to understand at the molecular cell level, how cancer cells work?
0:01:14.332 –> 0:01:15.892 inspiration because my mom is
0:01:15.892 –> 0:01:18.489 also a cancer scientist and she
0:01:18.489 –> 0:01:20.957 inspired me from a young
0:01:20.957 –> 0:01:23.198 age to become a cancer scientist.
0:01:23.2 –> 0:01:25.258 So Fast forward a few years I
0:01:25.258 –> 0:01:27.61 came here 10 years ago with this
0:01:27.61 –> 0:01:29.71 big dream to make a difference
0:01:29.78 –> 0:01:32.132 for cancer and especially for
0:01:32.132 –> 0:01:34.034 the patients and their families.
0:01:34.034 –> 0:01:36.706 And recently a year ago I started my
0:01:36.706 –> 0:01:39.397 own lab here at Yale and in my lab we
0:01:39.397 –> 0:01:41.511 have different individuals that
0:01:41.511 –> 0:01:44.55 are training in research.
0:01:44.55 –> 0:01:46.65 So at this level we have
0:01:47.432 –> 0:01:48.214 graduate students and
0:01:49 –> 0:01:51.688 Master students and PhD students and
0:01:51.688 –> 0:01:54.637 we also have postdocs that come to
0:01:54.637 –> 0:01:57.217 train after their PhD level before
0:01:57.296 –> 0:01:59.6 they can launch their own lab.
0:01:59.6 –> 0:02:02.456 So my job as a mentor and as
0:02:02.46 –> 0:02:05.228 a leader is to manage all the
0:02:05.228 –> 0:02:07.396 research activity and programs that
0:02:07.396 –> 0:02:10.21 are being funded by different institutions,
0:02:10.21 –> 0:02:12.35 government or private
0:02:12.35 –> 0:02:15.331 institutions and it’s all with the hope
0:02:15.331 –> 0:02:17.641 that we can cure pancreatic cancer
0:02:17.641 –> 0:02:20.409 and change the course of this disease.
0:02:21.54 –> 0:02:24.888 Tell us more about that.
0:02:24.89 –> 0:02:27.938 It sounds like a lofty goal to
0:02:27.938 –> 0:02:31.64 find a cure for pancreatic cancer, and
0:02:31.64 –> 0:02:35.24 change the course of this disease. But how
exactly are you doing that? We try to understand this disease by using as many biological systems that we can, so we start by first understanding the tumors from the patients. So to do this, we dive into doing DNA sequencing, RNA sequencing proteomics to really understand the building blocks of these cells and from those analysis that we generate from the tumors, but also with clear understanding of the clinical need to develop new therapies to diagnose it early, that’s when we start combining how can we use the data that we’re receiving from the patients to answer these questions that the clinical field is being challenged with. Then we go and we start engineering different model systems where we tightly control the variables. So for example, in cells we can manipulate the expression of genes and proteins, or in mice we can actually introduce mutations to the mice in their pancreata and lead them to form tumors that
later we can use all these models combined to test different hypothesis related to the basic biology of the cancer cell or to test novel therapies that either we generated or a pharmaceutical company comes to us because they are interested in testing it in our models. So I guess what I’m trying to say is every time you’re going to learn such a complex disease as cancer, you need to take advantage and generate as many model systems to interrogate the hypothesis that is behind it. So we do this in a team based effort. In my group we not only have people interested in basic science, but we also have clinicians or individuals who are in clinical training. So we can bring all of these areas of thought into these questions and these experimental designs that we do. We also bring computational scientists. For example, there is a lot of data out there that has been derived from multiple institutions and across the world of sequencing from the patient samples. And sometimes we can build those databases in house, but we also take advantage of all of
0:04:56.536 –> 0:04:58.906 this data that is being deposited
0:04:58.906 –> 0:05:01.384 out there from other scientists too.
0:05:02.671 –> 0:05:05.66 So as a community we can actually
0:05:05.737 –> 0:05:08.557 develop and better
0:05:08.557 –> 0:05:10.437 understand these tumors
0:05:10.519 –> 0:05:12.039 and also come up with
0:05:12.04 –> 0:05:14.14 better ways to treat them.
0:05:14.14 –> 0:05:16.745 And so by sequencing you mean
0:05:16.745 –> 0:05:18.86 sequencing the genes of the
0:05:18.86 –> 0:05:20.084 tumor itself?
0:05:20.084 –> 0:05:23.472 Yes, so what we do is we sequence the
0:05:23.472 –> 0:05:26.1 whole genome of that tumor cell.
0:05:26.1 –> 0:05:28.722 So we’re looking at more than
0:05:28.722 –> 0:05:31.459 95,000 genes at the same time.
0:05:31.46 –> 0:05:34.04 And we are interrogating,
0:05:34.04 –> 0:05:36.62 are there mutations on these genes?
0:05:36.62 –> 0:05:39.665 How differently is a gene being turned
0:05:39.665 –> 0:05:42.838 on or turned off between normal cells and
0:05:42.838 –> 0:05:45.983 cancer cells and when we turn them
0:05:45.983 –> 0:05:48.734 on do they produce a single protein
0:05:48.734 –> 0:05:51.388 or do they produce multiple proteins
0:05:51.388 –> 0:05:54.644 from that same template of the DNA?
0:05:54.644 –> 0:05:57.255 And so that level of complexity and
0:05:57.26 –> 0:05:58.98 imagine all of this,
0:05:58.98 –> 0:06:01.13 all these 95,000 genes mutations,
0:06:01.13 –> 0:06:02.674 expressions on and off
0:06:02.674 –> 0:06:06.309 times the number of cells in a tumor and
0:06:06.31 –> 0:06:08.165 all the patients that are
0:06:08.165 –> 0:06:10.02 coming for us to analyze.
0:06:10.02 –> 0:06:12.533 So there is a lot of data
0:06:12.533 –> 0:06:14.1 analysis that goes on here.
But really what’s driving this analysis is the biological and clinical questions that we want to answer. And so as you look at all of this data, and you’re sequencing the genomes of these cancers and figuring out which genes are turned on and which ones are turned off. What’s the next step? I mean, what people really want to know is, can you prevent pancreatic cancer either by causing aberrant genes that should not be turned on to stay, or turn them off once they’re already there, so can you prevent cancers from forming? Or can you use some of what you’re learning in terms of the sequencing to actually treat these cancers? So how do you kind of get from understanding what genes are turned on and what genes are turned off to really having something that has clinical impact? That’s a very good question. So in the pancreatic cancer field there are two points of research that we’re trying to tackle. The first one is early diagnosis and then the second one is treatment.
My lab in particular is focused more on the treatment side, so when we start looking for what are we going to learn from all of these sequencing in terms to really come up with novel ways for therapeutic approaches for these patients that desperately need it, we take an approach where we start comparing the tumors from patients that we’re very aggressive versus those tumors from other patients that were maybe a little bit more responsive to therapy, and we try to understand how are these tumors different at the molecular level. The reason why we want to understand differences is because we don’t think that there is a single therapy that works for all of the tumors. We know that the mutations that the tumors carry makes them biologically different. So what I’m trying to say is, although they may have the same diagnosis, at the molecular level, they’re almost kind of oranges and apples, and so we’re trying to dissect out the therapy that goes for the oranges and the therapy that goes for the apples. What my lab is doing differently from what other labs have done is...
we look at the level of turning on or turning off genes at a level that it’s almost imagine 10 times deeper than what other scientists have covered so far. So let me tell you a little bit of how the genome works. We used to think that a gene would get transcribed into this MRNA and then the MRNA would form a single protein, and the proteins to remind everyone are the functional units of the cell. There is a pathway by which the cells actually form a single gene. They can produce up to 7 different MRNA’s, and each one of these MRNA’s can produce seven different proteins. So most of the time the scientists focus on just one of the forms of those proteins from that single gene, because probably it is the more abundant one. But it’s not until you start doing these analysis, that we do at the MRNA sequencing level that you start understanding that they’re not only genes that are being turned on or turned off, but that when some gene is turned is being turned on, maybe it’s producing protein A and maybe in
other tumors the gene is still turned on, but is producing protein B. A&B are so different, and this is what my lab tries to dissect out. A&B are protein isoforms, and these protein isoforms as I was mentioning, may have different functions, and because previously the technology or the methods that we had available could only tell us is the gene on or not, now we have the analytical tools in their technology to say it’s been on, but then it’s preferentially expressing the protein isoform A or the isoform B. And that uncovers a very new biology about cancer cells, but something that had not been seen before. Why is this important? It turns out that if we can dissect this complexity and diversity in pancreatic cancer, potentially this can lead us to new therapies. Actually, last year my work group published that pancreatic cancer is highly susceptible to any therapy that perturbs this system of producing protein isoform A versus protein isoform B,
suggeting that there is potentially a therapeutic opportunity to understand more of these tumors at the protein isoform level and to generate particular therapies for these different proteins that are being expressed. Let me make sure I've got this straight. So you've kind of discovered that various genes can, when turned on, will make different isoforms. And that these isoforms will respond differently to therapy. So then the question is, at the clinic level, is it possible to distinguish which are which? In other words, if there is a particular therapy that works better for protein isoform A versus B, is there a way to know whether a particular patient is producing protein isoform A or B? Yes, so basically we're trying to get at the point where we develop an isoform specific therapy and this will drive personalized therapy. We have developed in my lab a novel therapeutic mechanism to be able to switch and correct these isoform expression.
Let’s say that isoform B is the most aggressive one, and it’s the most tumorigenic we can actually correct that isoform and switch it to the form which is actually the less aggressive form. This can drastically impact the biology and the growth of the tumor. So we’re excited to see what was going to happen with this new therapy as we start moving it into clinical trials. We’re going to have to take a short break for a medical minute, but we’ll get back into that conversation right after this with my guest, doctor Luisa Escobar-Hoyos. Support for Yale Cancer Answers comes from AstraZeneca, working to eliminate cancer as a cause of death. Learn more at astrazeneca-us.com. This is a medical minute about pancreatic cancer, which represents about 3% of all cancers in the US and about 7% of cancer deaths. Clinical trials are currently being offered at federally designated comprehensive Cancer Centers for the treatment of advanced stage and metastatic pancreatic cancer using chemotherapy and other novel therapies.
Folfirinox, a combination of five different chemotherapies is the latest advance in the treatment of metastatic pancreatic cancer and research continues at centers around the world looking into targeted therapies. A recently discovered marker, HENT one, has been a medical minute brought to you as a public service by Yale Cancer Center. More information is available at yalecancercenter.org you’re listening to Connecticut Public Radio. Welcome back to Yale Cancer Answers. This is doctor Anees Chagpar and I’m joined tonight by my guest doctor Luisa Escobar-Hoyos. We’re talking about her recent research looking at pancreatic cancers and before the break she was telling us about how she’s looking at the genome of these cancers, finding out that it’s not just about genes being turned on and turned off, but what protein isoforms those genes that are turned on actually make? And some of those may be more aggressive than others. Luisa, before we dig more into your research and the idea that you could actually switch from
a protein isoform that is more aggressive to a protein isoform, that’s less aggressive. Maybe we can take a step back and you can tell us a little bit more about why you decided to look at pancreatic cancer to begin with. It’s certainly one of the most lethal cancers, but talk a little bit more about that. Yes, so it’s actually a personal journey. When I was a PhD student, I used to study cervical cancer, as we all know, is now not as lethal because we have controlled because we screened for this disease and there’s less cases that appear in the US. But after my PhD, I started thinking that I wanted to put all my effort to understanding a cancer that really needed our attention, that’s when pancreatic cancer came to my mind. Several reasons there is a clinical need that we need to meet in the last 40 years. We have not changed the five year survival of pancreatic cancer, although we have made big progress in understanding the genetics and also I wanted to be sure to bring
whatever I had learned from my understanding of cervical cancer and apply it into understanding this more aggressive disease. And that’s when I started training in pancreatic cancer at Memorial Sloan Kettering Cancer Center, under the mentorship of Stephen Leach, a world renowned pancreatic cancer scientist.

so we both kind of wanted to study a different level of gene expression by understanding isoform switching by more specifically understanding the RNA splicing pathway. So previously we were talking about 95,000 genes, and if we can now multiply that each one of those genes is going to produce at least five protein isoform versus another, this is what allows the cell to diversify the podium. So previously we were talking about 95,000 genes, and if we can now multiply that each one of those genes is going to produce at least five protein isoform versus another, this is what allows the cell to diversify the podium.
or seven different proteins.
Imagine how large and versatile the proteome of a cell becomes.
Why we wanted to study this pathway or why it came to our attention.
It was actually from patient derived data in 2016 when I decided to study this cancer.
There were many groups that were coming up with this hypothesis.
Pancreatic cancer comes into these two molecular subtypes.
And there is one subtype that is more lethal that different authors coined the term either basal or squamous subtype.
Then the less lethal form which the authors called it classical when we look back into the more aggressive form this basal squamous molecular subtype we were seeing that these tumors have a high expression of all of these genes that are going to encode the splicing machinery that actually allows the cells to produce the protein isoforms.
And we started wondering if the reason why these tumors are so aggressive is probably because could they be more versatile in switching from one isoform to another one, depending on whatever therapy we provide to the patient.
that they’re lancing to the tumor.
Is this why previously we had not been able to target the right protein isoforms because we had until this point ignored the importance of isoforms in this disease.
That’s an interesting concept, that certain cancer cells may have this splicing ability that helps them to switch from a given protein isoform to another protein isoform that may be more resistant to therapy when you look at these.
two different subtypes, are they different in terms of their aggressiveness?
Even before the therapy?
In other words, is it that these protein isoforms actually cause differences in the biology of the aggressiveness of the tumor itself, or is it really this ability to react to the treatment with a different isoform that is more resistant?
We think that this capability of being plastic, it appears in naive tumors, so meaning before any treatment.
But it also gets used once you challenge the tumor with different therapies,
so we think that this is kind of an active pathway that it allows the cells to transform and to become cancer cells during the pathogenesis and after the pathogenesis during treatment time. You were mentioning that you’ve come up with a way to block that splicing, block that switching. So that if you prevent the cancer cell from actually switching to a different isoform, then potentially that cell is going to be more responsive to therapy, or at least would not be able to produce a protein isoform that would be resistant to therapy. Is that right?

Yes, what we have learned so far from these therapies, that is actually very potent whenever you correct a splicing defect that they have in that they need to survive as soon as you corrected the cells become more sensitive to chemotherapeutic agents and or they just die on their own because they cannot tolerate losing that expression of a particular isoforms. The next question obviously
0:22:06.83 –> 0:22:09.46 is how exactly does that happen?
0:22:09.46 –> 0:22:11.735 I mean, because this splicing
0:22:11.735 –> 0:22:13.555 mechanism is presumably something
0:22:13.555 –> 0:22:16.22 that is intrinsic to that tumor cell.
0:22:16.22 –> 0:22:18.758 So in order to stop it,
0:22:18.76 –> 0:22:21.301 you would need to get something into
0:22:21.301 –> 0:22:23.932 that tumor cell that actually stops
0:22:28.49 –> 0:22:31.028 And has that been tested in
0:22:31.03 –> 0:22:34.446 humans?
0:22:34.45 –> 0:22:37.187 The cell in order to switch from
0:22:37.187 –> 0:22:39.45 one isoform to another one,
0:22:39.45 –> 0:22:42.288 the MRNA’s have different sequences
0:22:42.288 –> 0:22:44.615 or different signals that
0:22:44.615 –> 0:22:47.072 is going to tell a cell produce
0:22:47.072 –> 0:22:49.459 isoform A or produce isoform B.
0:22:49.46 –> 0:22:51.545 Once we have identified which
0:22:53.63 –> 0:22:56.549 What we do is we introduce these
0:22:56.55 –> 0:22:59.07 small pieces of RNA into
0:22:59.07 –> 0:23:02.074 a cell and what we’re going to
0:23:02.074 –> 0:23:04.528 do is we’re going to block
0:23:04.53 –> 0:23:06.43 signals that usually the
0:23:06.43 –> 0:23:08.87 cancer cell would read to produce
0:23:08.87 –> 0:23:10.71 the most lethal isoform,
0:23:10.71 –> 0:23:13.531 and we’re going to fool it to
0:23:13.531 –> 0:23:16.479 make sure that it doesn’t see it.
0:23:16.48 –> 0:23:19.07 To mask these sites and
0:23:19.07 –> 0:23:22.044 force it to produce the other form and
0:23:22.044 –> 0:23:25.181 this therapy because of the way that
it works, we called it SHOT. Actually giving SHOT to the cancer cells and shot stands for Splicing-Hit Oligonucleotide Therapy. So far we have not tested it in humans. All of our data comes so far from patient cells. Tumor patient tumor cells that we grow in the lab. We also have tested this in our genetically engineered mouse models and all of that has produced the preliminary data to start. Hopefully launching a clinical trial in the short future in the patients. So the next question is when you have this mechanism, this shot that presumably you’re giving it whether it’s IV or orally, somehow you’re trying to get this into tumor cells. Does it get into normal cells and does it have any effect on the normal cells as well? Or do normal cells not have this splicing mechanism? That’s a very important question, so far the therapy that we like, the first phase of this therapy, we know that it’s a specific for
cancer cells because it’s only going to correct splicing defect that appears only on cancer cells. It still gets into the normal cells. But it’s not active there. because the splicing defect is not present. So far we have managed to introduce the therapy into the cancer cells by directly injecting into the tumors of mice what we are excited right now is that we’re going to start coupling SHOT with another therapy delivery technology that has been developed here at Yale and is actually currently under clinical trial testing called FLIP and FLIP is almost like a bio syringe that is going to carry shot and once said it lands into the tumor that has this particularly low pH, at that time it will convert into a syringe. It will introduce shot into the cells that are in that tumor microenvironment. So in that tumor microenvironment you have cancer cells and you have cells that are non cancerous. But the specificity comes that shot would only be able to correct splicing defects in cells that have it, and those splicing defects are only present in cancer cells.
So I think the combination of flip and shot is going to be highly specific for tumor cells and is going to decrease the amount of side effects because this therapy is so specific.

One question is, if shot is so specific based on the fact that this slicing mechanism only exists in cancer cells, then I guess the next question is, do you really need flip to kind of take it to where the cancer cells are, which is a low pH area? Or can you just inject shot systemically and know that even if it were to circulate around, and get absorbed by other cells that it really wouldn’t cause any harm, the only harm it would cause is in the tumor cells.

It’s actually the latter. You would decrease the amount of shot that you would need so that you could more accurately target it to where the tumor actually is.
to go directly into the cancer cells. Because if we just put shot systemically without a delivery technology, it will start getting word out and the concentration is going to drop and by the time the little bit that reaches the tumor it might be too low to have a biological impact. And so has this combination of flip and shot been tried in mouse models? Were actually testing it and this is part of the one of the reasons why I wanted to come to Yale because I wanted to combine a very exciting therapy with other delivery technologies that were being developed here specifically for these therapies that modify the way that the cells express proteins and turn on genes, and so we are hoping that now that the research is ramping up after COVID that we can start testing, we cannot wait to collaborate and we’re already starting to synthesize the shot in combination with flip. Doctor Luisa Escobar-Hoyos is an assistant professor of therapeutic radiology at the Yale School of Medicine. If you have questions, the address is canceranswers@yale.edu
and past editions of the program are available in audio and written form at yalecancercenter.org. We hope you'll join us next week to learn more about the fight against cancer here on Connecticut Public Radio.