WEBVTT

NOTE duration:"01:00:50.2400000"

NOTE language:en-us

NOTE Confidence: 0.72860694

00:00:00.000 --> 00:00:03.717 Of hosting today and we are incredibly

NOTE Confidence: 0.72860694

00:00:03.717 --> 00:00:06.308 thrilled and delighted that we

NOTE Confidence: 0.72860694

00:00:06.308 --> 00:00:09.026 were able to induce Doctor Gigi

NOTE Confidence: 0.72860694

00:00:09.026 --> 00:00:11.848 Lissonota to be our speaker today.

NOTE Confidence: 0.72860694

00:00:11.850 --> 00:00:14.300 Doctor Lozano graduated from Pan

NOTE Confidence: 0.72860694

00:00:14.300 --> 00:00:16.260 American University in Texas,

NOTE Confidence: 0.72860694

00:00:16.260 --> 00:00:19.200 did some work at Oakridge Laboratories,

NOTE Confidence: 0.72860694

00:00:19.200 --> 00:00:21.650 got her PhD at Rutgers,

NOTE Confidence: 0.72860694

00:00:21.650 --> 00:00:24.100 and then went to Princeton,

NOTE Confidence: 0.72860694

00:00:24.100 --> 00:00:28.020 where she was a postdoc with Arnie Levine.

NOTE Confidence: 0.72860694

00:00:28.020 --> 00:00:29.490 Immediately following that,

NOTE Confidence: 0.72860694

00:00:29.490 --> 00:00:32.430 she was recruited to MD Anderson,

NOTE Confidence: 0.72860694

00:00:32.430 --> 00:00:38.118 and in her long tenure there she has done.

NOTE Confidence: 0.72860694

00:00:38.120 --> 00:00:40.540 Path breaking work on P53.

NOTE Confidence: 0.72860694

00:00:40.540 --> 00:00:43.095 She's recognized for enormous contributions

NOTE Confidence: 0.72860694

00:00:43.095 --> 00:00:45.850 that include the recognition that P.

NOTE Confidence: 0.72860694

00:00:45.850 --> 00:00:48.748 53 works as a transcriptional activator.

NOTE Confidence: 0.72860694

00:00:48.750 --> 00:00:51.996 Many, many findings regarding the ways

NOTE Confidence: 0.72860694

00:00:51.996 --> 00:00:56.619 that MDM two and MDM three regulate P 53.

NOTE Confidence: 0.72860694

00:00:56.620 --> 00:00:59.200 Extensive use of transgenic models

NOTE Confidence: 0.72860694

00:00:59.200 --> 00:01:01.264 to understand these mechanisms.

NOTE Confidence: 0.72860694

00:01:01.270 --> 00:01:03.910 Better definition of specific P53

NOTE Confidence: 0.72860694

00:01:03.910 --> 00:01:07.145 loss and gain of function mutations

NOTE Confidence: 0.72860694

00:01:07.145 --> 00:01:10.151 and and their effects on P53's

NOTE Confidence: 0.72860694

00:01:10.151 --> 00:01:13.514 biology as well as studies of

NOTE Confidence: 0.72860694

00:01:13.514 --> 00:01:15.866 the P53 transcriptional program.

NOTE Confidence: 0.72860694

00:01:15.870 --> 00:01:18.250 She's a member of the National Academy

NOTE Confidence: 0.72860694

00:01:18.250 --> 00:01:20.432 of Sciences and the National Academy

NOTE Confidence: 0.72860694

00:01:20.432 --> 00:01:22.616 of Medicine and at MD Anderson.

NOTE Confidence: 0.72860694

00:01:22.620 --> 00:01:25.105 She is now chair of the Department

NOTE Confidence: 0.72860694

00:01:25.105 --> 00:01:26.879 of Genetics and Hubert L.

NOTE Confidence: 0.72860694

00:01:26.880 --> 00:01:28.042 Olive Stringer,

NOTE Confidence: 0.72860694

00:01:28.042 --> 00:01:30.366 distinguished chair in oncology.

NOTE Confidence: 0.72860694

00:01:30.370 --> 00:01:32.278 In the division of Basic Science

NOTE Confidence: 0.72860694

00:01:32.278 --> 00:01:33.550 Research at MD Anderson,

NOTE Confidence: 0.72860694

00:01:33.550 --> 00:01:36.043 so I don't want to talk for a long

NOTE Confidence: 0.72860694

00:01:36.043 --> 00:01:37.785 time because I'm really eager

NOTE Confidence: 0.72860694

00:01:37.785 --> 00:01:40.230 to hear what you have to say.

NOTE Confidence: 0.72860694

00:01:40.230 --> 00:01:42.590 So thank you very much for joining us

NOTE Confidence: 0.72860694

00:01:42.590 --> 00:01:45.320 and I will just say to the audience.

NOTE Confidence: 0.72860694

00:01:45.320 --> 00:01:47.714 You can use the chat function to

NOTE Confidence: 0.72860694

00:01:47.714 --> 00:01:49.450 enter your questions or we can.

NOTE Confidence: 0.72860694

00:01:49.450 --> 00:01:51.394 I believe we can unmute people

NOTE Confidence: 0.72860694

00:01:51.394 --> 00:01:53.579 at the end if we need to.

NOTE Confidence: 0.72860694

00:01:53.580 --> 00:01:55.170 So with that do welcome.

NOTE Confidence: 0.8811495

00:01:56.070 --> 00:01:58.758 Thank you, thank you very much,

NOTE Confidence: 0.8811495

00:01:58.760 --> 00:02:00.935 Barbara. For that introduction I

NOTE Confidence: 0.8811495

00:02:00.935 --> 00:02:03.690 hopefully now I'm sharing my screen.

NOTE Confidence: 0.67953175

00:02:07.070 --> 00:02:08.210 Can you see it?

NOTE Confidence: 0.83147156

00:02:10.390 --> 00:02:12.268 Weekend OK super Alright so

NOTE Confidence: 0.83147156

00:02:12.268 --> 00:02:13.766 anyway thank you Barbara.

NOTE Confidence: 0.83147156

00:02:13.766 --> 00:02:16.554 It's it's fun to always visit a place

NOTE Confidence: 0.83147156

00:02:16.554 --> 00:02:19.238 and now during covid we're doing these

NOTE Confidence: 0.83147156

00:02:19.238 --> 00:02:21.740 virtual seminars but hopefully at the

NOTE Confidence: 0.83147156

00:02:21.740 --> 00:02:24.703 end I'll be able to address some of

NOTE Confidence: 0.83147156

00:02:24.703 --> 00:02:26.865 the questions that people might have.

NOTE Confidence: 0.83147156

00:02:26.865 --> 00:02:29.853 What I thought I would do today is

NOTE Confidence: 0.83147156

00:02:29.853 --> 00:02:31.995 described some of the models that

NOTE Confidence: 0.83147156

00:02:31.995 --> 00:02:35.083 I think have to find some of the

NOTE Confidence: 0.83147156

00:02:35.083 --> 00:02:37.018 basic real basic understanding of

NOTE Confidence: 0.83147156

00:02:37.020 --> 00:02:39.290 the P53 tumor suppressor pathway.

NOTE Confidence: 0.83147156

00:02:39.290 --> 00:02:41.796 So I'll get started with my disclosures.

NOTE Confidence: 0.83147156

00:02:41.800 --> 00:02:44.626 I am on the Scientific Advisory

NOTE Confidence: 0.83147156

00:02:44.626 --> 00:02:46.510 Board for PMV Pharma.

NOTE Confidence: 0.83147156

00:02:46.510 --> 00:02:48.580 So the P 53 pathway.

NOTE Confidence: 0.83147156

00:02:48.580 --> 00:02:51.356 This is my myopic view of the pathway

NOTE Confidence: 0.83147156

00:02:51.356 --> 00:02:53.550 and it really highlights.

NOTE Confidence: 0.83147156

00:02:53.550 --> 00:02:56.034 I think some of the critical

NOTE Confidence: 0.83147156

00:02:56.034 --> 00:02:57.690 features of the pathway.

NOTE Confidence: 0.83147156

00:02:57.690 --> 00:03:00.511 First and foremost people decree is present

NOTE Confidence: 0.83147156

00:03:00.511 --> 00:03:03.478 in very low levels in normal cells,

NOTE Confidence: 0.83147156

00:03:03.480 --> 00:03:05.550 but any kind of abnormality

NOTE Confidence: 0.83147156

00:03:05.550 --> 00:03:07.206 that the South senses,

NOTE Confidence: 0.83147156

00:03:07.210 --> 00:03:08.674 hypoxia, DNA damage,

NOTE Confidence: 0.83147156

00:03:08.674 --> 00:03:10.626 inappropriate activation of an

NOTE Confidence: 0.83147156

00:03:10.626 --> 00:03:12.485 oncogene will stabilize that

NOTE Confidence: 0.83147156

00:03:12.485 --> 00:03:14.760 P53 protein and then P53 in turn

NOTE Confidence: 0.83147156

00:03:14.760 --> 00:03:16.979 functions as a transcription factor.

NOTE Confidence: 0.83147156

00:03:16.980 --> 00:03:18.880 To activate hundreds of genes

NOTE Confidence: 0.83147156

00:03:18.880 --> 00:03:21.585 and I'll show you some of those

NOTE Confidence: 0.83147156

00:03:21.585 --> 00:03:23.775 experiments in in a little bit,

NOTE Confidence: 0.83147156

00:03:23.780 --> 00:03:26.426 but some of the genes that P.

NOTE Confidence: 0.83147156

00:03:26.430 --> 00:03:29.823 53 is known to activate is P21 for example,

NOTE Confidence: 0.83147156

00:03:29.830 --> 00:03:32.854 which is a inhibitor of the South cycle.

NOTE Confidence: 0.83147156

00:03:32.860 --> 00:03:34.835 It also induces senescence program

NOTE Confidence: 0.83147156

00:03:34.835 --> 00:03:37.768 P53 can activate a slew of gene sick,

NOTE Confidence: 0.83147156

00:03:37.770 --> 00:03:40.786 initiate a pop ptosis and it also activates.

NOTE Confidence: 0.83147156

00:03:40.790 --> 00:03:43.445 Genes are involved in changing

NOTE Confidence: 0.83147156

00:03:43.445 --> 00:03:46.640 the metabolic functions of a cell.

NOTE Confidence: 0.83147156

00:03:46.640 --> 00:03:49.286 Now when P 53 is activated,

NOTE Confidence: 0.83147156

00:03:49.290 --> 00:03:52.377 if the cell is allowed to survive

NOTE Confidence: 0.83147156

00:03:52.377 --> 00:03:53.259 and proceed,

NOTE Confidence: 0.83147156

00:03:53.260 --> 00:03:55.900 P53 has to activate this protein

NOTE Confidence: 0.83147156

00:03:55.900 --> 00:03:57.220 called MDM 2,

NOTE Confidence: 0.83147156

00:03:57.220 --> 00:03:59.620 which is an E3 ubiquitin ligase

NOTE Confidence: 0.83147156

00:03:59.620 --> 00:04:01.814 that targets P53 for degradation

NOTE Confidence: 0.83147156

00:04:01.814 --> 00:04:04.389 and basically removes the Peachtree

NOTE Confidence: 0.83147156

00:04:04.389 --> 00:04:07.070 levels back down to normal.

NOTE Confidence: 0.83147156

00:04:07.070 --> 00:04:08.530 So if you just.

NOTE Confidence: 0.83147156

00:04:08.530 --> 00:04:11.329 Think about what people think we can do.

NOTE Confidence: 0.83147156

00:04:11.330 --> 00:04:13.994 We can do so much in getting arrested,

NOTE Confidence: 0.83147156

00:04:14.000 --> 00:04:15.436 phallic it, killed itself,

NOTE Confidence: 0.83147156

00:04:15.436 --> 00:04:18.027 and it can also induce its own

NOTE Confidence: 0.83147156

00:04:18.027 --> 00:04:20.575 inhibitor to allow the cell to survive.

NOTE Confidence: 0.83147156

00:04:20.580 --> 00:04:22.452 And even though there's so much

NOTE Confidence: 0.83147156

00:04:22.452 --> 00:04:24.529 work in the pizza tree killed,

NOTE Confidence: 0.83147156

00:04:24.530 --> 00:04:26.952 we still don't understand all of the

NOTE Confidence: 0.83147156

00:04:26.952 --> 00:04:29.238 cues that determine which of these

NOTE Confidence: 0.83147156

00:04:29.238 --> 00:04:31.208 pathways Peabody Creek and activate.

NOTE Confidence: 0.83147156

00:04:31.210 --> 00:04:33.536 But because it has all these functions,

NOTE Confidence: 0.83147156

00:04:33.536 --> 00:04:35.528 it is a critical tumor suppressor,

NOTE Confidence: 0.83147156

00:04:35.530 --> 00:04:37.858 and it is the most doctor

NOTE Confidence: 0.83147156

00:04:37.858 --> 00:04:39.410 mutated gene human cancers.

NOTE Confidence: 0.83147156

00:04:39.410 --> 00:04:42.274 So what I showed here is what the

NOTE Confidence: 0.83147156

00:04:42.274 --> 00:04:44.559 field called the Manhattan Plot,

NOTE Confidence: 0.83147156

00:04:44.560 --> 00:04:47.325 and it was developed by Magali Olivia.

NOTE Confidence: 0.83147156

00:04:47.330 --> 00:04:49.906 So across this axis are 125 genes

NOTE Confidence: 0.83147156

00:04:49.906 --> 00:04:52.258 that are commonly mutated and cancers

NOTE Confidence: 0.83147156

00:04:52.258 --> 00:04:54.988 and then across this axis here are

NOTE Confidence: 0.83147156

00:04:55.062 --> 00:04:57.227 36 different types of cancers,

NOTE Confidence: 0.83147156

00:04:57.230 --> 00:04:59.995 and there's some features that stand out.

NOTE Confidence: 0.83147156

00:05:00.000 --> 00:05:02.920 But this is the one I want to

NOTE Confidence: 0.83147156

00:05:02.920 --> 00:05:05.148 highlight here across the board.

NOTE Confidence: 0.83147156

00:05:05.150 --> 00:05:07.526 These are mutations in the P53

NOTE Confidence: 0.83147156

00:05:07.526 --> 00:05:08.318 tumor suppressor.

NOTE Confidence: 0.83147156

00:05:08.320 --> 00:05:10.540 So almost all cancers mutate.

NOTE Confidence: 0.83147156

00:05:10.540 --> 00:05:15.467 53 but P 53 pathway is inactivated by

NOTE Confidence: 0.83147156

00:05:15.467 --> 00:05:17.970 multiple mechanisms and I show here

NOTE Confidence: 0.83147156

00:05:17.970 --> 00:05:20.525 in some of the different cancers and

NOTE Confidence: 0.83147156

00:05:20.607 --> 00:05:23.349 how they inactivate piece of debris.

NOTE Confidence: 0.83147156

00:05:23.350 --> 00:05:24.613 So high grade,

NOTE Confidence: 0.83147156

00:05:24.613 --> 00:05:25.876 serious ovarian carcinomas,

NOTE Confidence: 0.83147156

00:05:25.880 --> 00:05:28.834 mutations in P53 are the most common.

NOTE Confidence: 0.83147156

00:05:28.840 --> 00:05:29.262 However,

NOTE Confidence: 0.83147156

00:05:29.262 --> 00:05:32.638 in liposarcomas it is upregulation of MDM 2,

NOTE Confidence: 0.7966159

00:05:32.640 --> 00:05:37.280 the P fifty 383 if it could be like a San.

NOTE Confidence: 0.7966159

00:05:37.280 --> 00:05:40.472 About 100% of these liposarcomas inactivate

NOTE Confidence: 0.7966159

00:05:40.472 --> 00:05:42.900 the pathway by overexpressing MD.

NOTE Confidence: 0.7966159

00:05:42.900 --> 00:05:45.190 In the new Asia glioblastoma,

NOTE Confidence: 0.7966159

00:05:45.190 --> 00:05:47.938 big Sweet come interested in glioblastomas.

NOTE Confidence: 0.7966159

00:05:47.940 --> 00:05:51.139 Recently, the P 53 gene is altered,

NOTE Confidence: 0.7966159

00:05:51.140 --> 00:05:53.455 deleted in approximately 1/3 of

NOTE Confidence: 0.7966159

00:05:53.455 --> 00:05:56.640 glioblastomas in the MDM to an Indian.

NOTE Confidence: 0.7966159

00:05:56.640 --> 00:05:58.620 For genes are upregulated in

NOTE Confidence: 0.7966159

00:05:58.620 --> 00:06:01.814 about it order and this is a

NOTE Confidence: 0.7966159

00:06:01.814 --> 00:06:03.506 mutually exclusive relationship.

NOTE Confidence: 0.7966159

00:06:03.510 --> 00:06:05.800 So if P53 is mutant,

NOTE Confidence: 0.7966159

00:06:05.800 --> 00:06:10.469 MDM 2 doesn't have to be upregulated.

NOTE Confidence: 0.7966159

00:06:10.470 --> 00:06:12.614 And the other thing I want to point

NOTE Confidence: 0.7966159

00:06:12.614 --> 00:06:15.010 out in glioblastomas is that we have

NOTE Confidence: 0.7966159

00:06:15.010 --> 00:06:17.243 about half of these tumors that

NOTE Confidence: 0.7966159

00:06:17.243 --> 00:06:19.128 have neither mutations and piece

NOTE Confidence: 0.7966159

00:06:19.128 --> 00:06:21.657 of D3 or upregulation of the MDM.

NOTE Confidence: 0.7966159

00:06:21.657 --> 00:06:24.030 Two and MDM four inhibitors of P53.

NOTE Confidence: 0.7966159

00:06:24.030 --> 00:06:26.032 And so since I really believe that

NOTE Confidence: 0.7966159

00:06:26.032 --> 00:06:28.753 the P 53 pathway has to be undermined

NOTE Confidence: 0.7966159

00:06:28.753 --> 00:06:31.150 in the development of all cancers,

NOTE Confidence: 0.7966159

00:06:31.150 --> 00:06:33.926 I think that there's a big hole here

NOTE Confidence: 0.7966159

00:06:33.926 --> 00:06:37.386 that we have to understand in more detail.

NOTE Confidence: 0.7966159

00:06:37.390 --> 00:06:39.497 So today's start is really going to

NOTE Confidence: 0.7966159

00:06:39.497 --> 00:06:41.280 concentrate on just a few proteins,

NOTE Confidence: 0.7966159

00:06:41.280 --> 00:06:43.366 and I'm not going to have time

NOTE Confidence: 0.7966159

00:06:43.366 --> 00:06:45.758 to show you a whole lot of data,

NOTE Confidence: 0.7966159

00:06:45.760 --> 00:06:48.736 but I thought I would use this slide

NOTE Confidence: 0.7966159

00:06:48.736 --> 00:06:51.227 to highlight some of the important.

NOTE Confidence: 0.7966159

00:06:51.230 --> 00:06:53.576 Functions that I of these proteins

NOTE Confidence: 0.7966159

00:06:53.576 --> 00:06:56.310 that I will discuss with you today.

NOTE Confidence: 0.7966159

00:06:56.310 --> 00:06:59.438 So MDM two is an inhibitor of P53.

NOTE Confidence: 0.7966159

00:06:59.440 --> 00:07:01.786 It's an E3 ubiquitin ligase and

NOTE Confidence: 0.7966159

00:07:01.786 --> 00:07:03.350 target speakers for degradation.

NOTE Confidence: 0.7966159

00:07:03.350 --> 00:07:07.130 MDM Four also inhibits P 53.

NOTE Confidence: 0.7966159

00:07:07.130 --> 00:07:09.888 It doesn't have any E3 ligase function,

NOTE Confidence: 0.7966159

00:07:09.890 --> 00:07:12.110 but it actually facilitates and makes

NOTE Confidence: 0.7966159

00:07:12.110 --> 00:07:15.010 MDM two or better yet riveting ligase,

NOTE Confidence: 0.7966159

00:07:15.010 --> 00:07:17.100 although it also has independent

NOTE Confidence: 0.7966159

00:07:17.100 --> 00:07:19.735 functions of MDM two and can

NOTE Confidence: 0.7966159

00:07:19.735 --> 00:07:22.549 actually bind and inhibit the pizza

NOTE Confidence: 0.7966159

00:07:22.549 --> 00:07:24.550 guy free transactivation domain.

NOTE Confidence: 0.7966159

00:07:24.550 --> 00:07:27.546 This relationship does MDM 2 New Four

NOTE Confidence: 0.7966159

00:07:27.546 --> 00:07:30.280 also conform hetero dimer and that

NOTE Confidence: 0.7966159

00:07:30.280 --> 00:07:33.028 header dimer is critical in embryo

NOTE Confidence: 0.7966159

00:07:33.028 --> 00:07:35.440 development to inhibit P 53 and then,

NOTE Confidence: 0.7966159

00:07:35.440 --> 00:07:36.700 as I indicated,

NOTE Confidence: 0.7966159

00:07:36.700 --> 00:07:39.220 and will discuss in some detail,

NOTE Confidence: 0.7966159

00:07:39.220 --> 00:07:41.626 P53 can activate the Indian two

NOTE Confidence: 0.7966159

00:07:41.626 --> 00:07:44.173 promoter so it can up regulate

NOTE Confidence: 0.7966159

00:07:44.173 --> 00:07:46.759 MDM two in inhibited so levels.

NOTE Confidence: 0.7966159

00:07:46.760 --> 00:07:48.432 Another important concept that

NOTE Confidence: 0.7966159

00:07:48.432 --> 00:07:50.104 I'll mention very briefly,

NOTE Confidence: 0.7966159

00:07:50.110 --> 00:07:52.952 maybe at the end is that MDM

NOTE Confidence: 0.7966159

00:07:52.952 --> 00:07:55.070 two can also inhibit.

NOTE Confidence: 0.7966159

00:07:55.070 --> 00:07:57.779 A mutant P53 protein and that's because

NOTE Confidence: 0.7966159

00:07:57.779 --> 00:07:59.677 these mutant proteins have mutations

NOTE Confidence: 0.7966159

00:07:59.677 --> 00:08:02.029 in the DNA binding domain but retain

NOTE Confidence: 0.7966159

00:08:02.029 --> 00:08:04.589 a transcriptional activation domain.

NOTE Confidence: 0.7966159

00:08:04.590 --> 00:08:08.270 But the important point that I want to

NOTE Confidence: 0.7966159

00:08:08.270 --> 00:08:11.587 make here is even though this mutant

NOTE Confidence: 0.7966159

00:08:11.587 --> 00:08:15.228 can be targeted by MD M2 and MD M4.

NOTE Confidence: 0.7966159

00:08:15.230 --> 00:08:18.230 It is mutant in so can no longer

NOTE Confidence: 0.7966159

00:08:18.230 --> 00:08:19.610 feed back it up.

NOTE Confidence: 0.7966159

00:08:19.610 --> 00:08:21.898 Regulate MDM two so with time in our

NOTE Confidence: 0.7966159

00:08:21.898 --> 00:08:24.333 in vivo models we find that these

NOTE Confidence: 0.7966159

00:08:24.333 --> 00:08:26.629 people period this can become stabilized

NOTE Confidence: 0.7966159

00:08:26.629 --> 00:08:28.809 'cause there's insufficient MDM two

NOTE Confidence: 0.7966159

00:08:28.809 --> 00:08:31.348 to down modulate the protein levels

NOTE Confidence: 0.7966159

00:08:31.348 --> 00:08:34.500 and in a few minutes you'll see how

NOTE Confidence: 0.7966159

00:08:34.585 --> 00:08:37.110 important it down modulation is.

NOTE Confidence: 0.7966159

00:08:37.110 --> 00:08:39.374 So the outline of my talk today is

NOTE Confidence: 0.7966159

00:08:39.374 --> 00:08:41.820 I'm going to talk about some of the

NOTE Confidence: 0.7966159

00:08:41.820 --> 00:08:43.833 models that showed us how exquisitely

NOTE Confidence: 0.7966159

00:08:43.833 --> 00:08:48.500 sensitive P 53 is to inhibition via video 2.

NOTE Confidence: 0.7966159

00:08:48.500 --> 00:08:51.230 To tell you what the molecular

NOTE Confidence: 0.7966159

00:08:51.230 --> 00:08:53.561 responses to people three activation

NOTE Confidence: 0.7966159

00:08:53.561 --> 00:08:56.543 in vivo and last but not least,

NOTE Confidence: 0.79513806

00:08:56.550 --> 00:08:59.694 I'm going to describe some of the new

NOTE Confidence: 0.79513806

00:08:59.694 --> 00:09:02.551 cancer models that were working with

NOTE Confidence: 0.79513806

00:09:02.551 --> 00:09:05.036 that expressed mutant P53 proteins.

NOTE Confidence: 0.79513806

00:09:05.040 --> 00:09:07.994 So let's first talk about the MDM

NOTE Confidence: 0.79513806

00:09:07.994 --> 00:09:10.400 proteins and their innovation P.

NOTE Confidence: 0.79513806

00:09:10.400 --> 00:09:14.848 53. So along time ago and now we.

NOTE Confidence: 0.79513806

00:09:14.850 --> 00:09:17.412 Attempted to make an Indian to know

NOTE Confidence: 0.79513806

00:09:17.412 --> 00:09:19.708 Mouse and it's just not possible

NOTE Confidence: 0.79513806

00:09:19.708 --> 00:09:22.319 and the reason is not possible is

NOTE Confidence: 0.79513806

00:09:22.401 --> 00:09:25.320 because an MDM two null embryo just

NOTE Confidence: 0.79513806

00:09:25.320 --> 00:09:27.765 prior to implantation is APOP totic.

NOTE Confidence: 0.79513806

00:09:27.765 --> 00:09:29.740 This embryo stained with the

NOTE Confidence: 0.79513806

00:09:29.740 --> 00:09:31.658 tunnel essay in every salad.

NOTE Confidence: 0.79513806

00:09:31.660 --> 00:09:34.630 This embryo is is dead.

NOTE Confidence: 0.79513806

00:09:34.630 --> 00:09:37.521 And at the time we knew that

NOTE Confidence: 0.79513806

00:09:37.521 --> 00:09:39.470 MDM two interacted with P.

NOTE Confidence: 0.79513806

00:09:39.470 --> 00:09:42.627 53, but we really didn't know how

NOTE Confidence: 0.79513806

00:09:42.627 --> 00:09:44.765 important interaction was and what

NOTE Confidence: 0.79513806

00:09:44.765 --> 00:09:46.877 we did was test the importance

NOTE Confidence: 0.79513806

00:09:46.877 --> 00:09:49.608 of P53 in this little embryo by

NOTE Confidence: 0.79513806

00:09:49.608 --> 00:09:52.136 crossing 2P53 miles and we completely

NOTE Confidence: 0.79513806

00:09:52.136 --> 00:09:53.864 rescue this phenotype.

NOTE Confidence: 0.79513806

00:09:53.864 --> 00:09:56.744 These Meister born and are

NOTE Confidence: 0.79513806

00:09:56.744 --> 00:09:59.058 perfectly normal except now because

NOTE Confidence: 0.79513806

00:09:59.058 --> 00:10:01.860 they lack P 53 they have it.

NOTE Confidence: 0.79513806

00:10:01.860 --> 00:10:04.476 So with with this experiment indicates

NOTE Confidence: 0.79513806

00:10:04.476 --> 00:10:08.072 is that what MDM two is doing in

NOTE Confidence: 0.79513806

00:10:08.072 --> 00:10:10.277 these embryos is upregulating P53,

NOTE Confidence: 0.79513806

00:10:10.280 --> 00:10:12.490 which is preventing the normal

NOTE Confidence: 0.79513806

00:10:12.490 --> 00:10:14.258 development of these embryos.

NOTE Confidence: 0.79513806

00:10:14.260 --> 00:10:16.480 MDM fours are related MDM,

NOTE Confidence: 0.79513806

00:10:16.480 --> 00:10:19.138 two protein that aren't Johansson discovered.

NOTE Confidence: 0.79513806

00:10:19.140 --> 00:10:22.868 And since MDM two has such a unique

NOTE Confidence: 0.79513806

00:10:22.868 --> 00:10:25.469 relationship with P53 we decided that

NOTE Confidence: 0.79513806

00:10:25.469 --> 00:10:28.449 we would make the MDM for knockout

NOTE Confidence: 0.79513806

00:10:28.449 --> 00:10:31.539 but weren't sure what to expect.

NOTE Confidence: 0.79513806

00:10:31.540 --> 00:10:34.532 But in MD for knockout is also embryo

NOTE Confidence: 0.79513806

00:10:34.532 --> 00:10:37.316 lethal a few days after the Indian

NOTE Confidence: 0.79513806

00:10:37.316 --> 00:10:40.232 to know but again that phenotype is

NOTE Confidence: 0.79513806

00:10:40.232 --> 00:10:43.305 rescued by deletion of P53 and we've

NOTE Confidence: 0.79513806

00:10:43.305 --> 00:10:47.470 also made mice that have no MDM to know

NOTE Confidence: 0.79513806

00:10:47.470 --> 00:10:50.988 Indian for Nokia 53 and these mice.

NOTE Confidence: 0.79513806

00:10:50.990 --> 00:10:54.206 The viable they have Pizza 3 two or

NOTE Confidence: 0.79513806

00:10:54.206 --> 00:10:56.207 phenotypes because they lack P 53.

NOTE Confidence: 0.79513806

00:10:56.210 --> 00:10:57.698 So at least physiologically,

NOTE Confidence: 0.79513806

00:10:57.698 --> 00:10:58.814 in the mouse,

NOTE Confidence: 0.79513806

00:10:58.820 --> 00:11:01.532 the two most important functions of

NOTE Confidence: 0.79513806

00:11:01.532 --> 00:11:04.598 these empty in proteins is to keep

NOTE Confidence: 0.79513806

00:11:04.598 --> 00:11:06.950 P 53 levels low during homeostasis.

NOTE Confidence: 0.79513806

00:11:06.950 --> 00:11:07.283 OK,

NOTE Confidence: 0.79513806

00:11:07.283 --> 00:11:09.614 so we have these MDM two heterozygous

NOTE Confidence: 0.79513806

00:11:09.614 --> 00:11:12.289 in these MDM for headers I customized.

NOTE Confidence: 0.79513806

00:11:12.290 --> 00:11:14.330 They have only one allele of

NOTE Confidence: 0.79513806

00:11:14.330 --> 00:11:16.210 each of these two genes.

NOTE Confidence: 0.79513806

00:11:16.210 --> 00:11:17.985 In these mice are perfectly

NOTE Confidence: 0.79513806

00:11:17.985 --> 00:11:19.405 normal and running around,

NOTE Confidence: 0.79513806

00:11:19.410 --> 00:11:21.546 but as I indicated in my

NOTE Confidence: 0.79513806

00:11:21.546 --> 00:11:22.970 in my introductory slide.

NOTE Confidence: 0.7945916

00:11:25.790 --> 00:11:28.726 P 53 is a DNA damage response protein,

NOTE Confidence: 0.7945916

00:11:28.730 --> 00:11:31.215 and so we wondered if there was

NOTE Confidence: 0.7945916

00:11:31.215 --> 00:11:33.149 any phenotypes in these mice.

NOTE Confidence: 0.7945916

00:11:33.150 --> 00:11:34.618 If we irradiated them,

NOTE Confidence: 0.7945916

00:11:34.618 --> 00:11:37.564 if we damaged their DNA, and sure enough,

NOTE Confidence: 0.7945916

00:11:37.564 --> 00:11:39.404 we saw a beautiful phenotype.

NOTE Confidence: 0.7945916

00:11:39.410 --> 00:11:41.979 So the MDM two in the MDM,

NOTE Confidence: 0.7945916

00:11:41.980 --> 00:11:44.283 two Ambien for headers agasse mice are

NOTE Confidence: 0.7945916

00:11:44.283 --> 00:11:46.770 sensitive to low dose ionizing radiation.

NOTE Confidence: 0.7945916

00:11:46.770 --> 00:11:49.086 So in this experiment what we've

NOTE Confidence: 0.7945916

00:11:49.086 --> 00:11:51.373 done is irradiated mice with six

NOTE Confidence: 0.7945916

00:11:51.373 --> 00:11:53.389 Gray and the black line above.

NOTE Confidence: 0.7945916

00:11:53.390 --> 00:11:55.770 Here is a normal mouse.

NOTE Confidence: 0.7945916

00:11:55.770 --> 00:11:59.556 That for 50 days just ignores.

NOTE Confidence: 0.7945916

00:11:59.560 --> 00:12:01.708 Six grade radiation, but the headers.

NOTE Confidence: 0.7945916

00:12:01.710 --> 00:12:02.548 I guess.

NOTE Confidence: 0.7945916

00:12:02.548 --> 00:12:06.456 Mice MDM four in Red and MDM two and

NOTE Confidence: 0.7945916

00:12:06.456 --> 00:12:10.088 blue are dead by three weeks of age.

NOTE Confidence: 0.7945916

00:12:10.090 --> 00:12:10.475 Importantly,

NOTE Confidence: 0.7945916

00:12:10.475 --> 00:12:13.555 if we now move P53 from this system,

NOTE Confidence: 0.7945916

00:12:13.560 --> 00:12:15.490 we completely rescue these phenotypes.

NOTE Confidence: 0.7945916

00:12:15.490 --> 00:12:17.420 So this is the rescue.

NOTE Confidence: 0.7945916

00:12:17.420 --> 00:12:19.736 The Indium MDM, two heterozygous mouse.

NOTE Confidence: 0.7945916

00:12:19.740 --> 00:12:22.398 And here's the rescue of the

NOTE Confidence: 0.7945916

00:12:22.398 --> 00:12:24.170 Indian for headers ecospace.

NOTE Confidence: 0.7945916

00:12:24.170 --> 00:12:25.484 So even though.

NOTE Confidence: 0.7945916

00:12:25.484 --> 00:12:28.550 The MDM two and MDM for heterozygotes

NOTE Confidence: 0.7945916

00:12:28.635 --> 00:12:32.275 mice have sufficient levels of the Zambian

NOTE Confidence: 0.7945916

00:12:32.275 --> 00:12:35.909 proteins to maintain homeostasis with damage.

NOTE Confidence: 0.7945916

00:12:35.910 --> 00:12:39.970 There's just not enough of these proteins

NOTE Confidence: 0.7945916

00:12:39.970 --> 00:12:44.017 to return degree back to normal levels.

NOTE Confidence: 0.7945916

00:12:44.020 --> 00:12:46.533 The next experiment that I want to

NOTE Confidence: 0.7945916

00:12:46.533 --> 00:12:48.944 tell you about is the importance

NOTE Confidence: 0.7945916

00:12:48.944 --> 00:12:50.620 of this feedback loop.

NOTE Confidence: 0.7945916

00:12:50.620 --> 00:12:52.172 So as I indicated,

NOTE Confidence: 0.7945916

00:12:52.172 --> 00:12:54.500 MDM two is regulated by P53.

NOTE Confidence: 0.7945916

00:12:54.500 --> 00:12:56.525 There are two peaks decree

NOTE Confidence: 0.7945916

00:12:56.525 --> 00:12:59.370 binding sites in the P2 promoter.

NOTE Confidence: 0.7945916

00:12:59.370 --> 00:13:00.312 There are people,

NOTE Confidence: 0.7945916

00:13:00.312 --> 00:13:00.940 three dependent,

NOTE Confidence: 0.7945916

00:13:00.940 --> 00:13:03.045 so people free byansi sequences

NOTE Confidence: 0.7945916

00:13:03.045 --> 00:13:04.729 and activates MDM 2.

NOTE Confidence: 0.7945916

00:13:04.730 --> 00:13:07.187 And So what we decided to do is to

NOTE Confidence: 0.7945916

00:13:07.187 --> 00:13:09.771 ask how important was this feedback

NOTE Confidence: 0.7945916

00:13:09.771 --> 00:13:12.001 loop in regulating P53 levers?

NOTE Confidence: 0.7945916

00:13:12.010 --> 00:13:12.300 OK,

NOTE Confidence: 0.7945916

00:13:12.300 --> 00:13:14.620 so we made point mutations and I show

NOTE Confidence: 0.7945916

00:13:14.620 --> 00:13:16.572 here the different point mutations

NOTE Confidence: 0.7945916

00:13:16.572 --> 00:13:18.960 because we didn't want to disrupt

NOTE Confidence: 0.7945916

00:13:19.025 --> 00:13:21.110 the architecture of the promoter.

NOTE Confidence: 0.7945916

00:13:21.110 --> 00:13:24.438 We just wanted to disrupt MD PhD degree

NOTE Confidence: 0.7945916

00:13:24.438 --> 00:13:27.597 binding to the end game to promote it.

NOTE Confidence: 0.7945916

00:13:27.600 --> 00:13:30.253 So these experiments in the bottom bar

NOTE Confidence: 0.7945916

00:13:30.253 --> 00:13:32.822 chip data that basically show that

NOTE Confidence: 0.7945916

00:13:32.822 --> 00:13:35.540 P53 cannot find this mutant promoter,

NOTE Confidence: 0.7945916

00:13:35.540 --> 00:13:37.180 which we call P2P2,

NOTE Confidence: 0.7945916

00:13:37.180 --> 00:13:39.640 and this is different promoter that

NOTE Confidence: 0.7945916

00:13:39.715 --> 00:13:43.067 shows our assays are working in P53 combined.

NOTE Confidence: 0.7945916

00:13:43.070 --> 00:13:44.742 The pull up remote.

NOTE Confidence: 0.7945916

00:13:44.742 --> 00:13:45.160 OK,

NOTE Confidence: 0.7945916

00:13:45.160 --> 00:13:47.560 so we made these point mutations

NOTE Confidence: 0.7945916

00:13:47.560 --> 00:13:50.108 and we were surprised that are

NOTE Confidence: 0.7945916

00:13:50.108 --> 00:13:51.840 most was perfectly normal.

NOTE Confidence: 0.7945916

00:13:51.840 --> 00:13:53.870 We really thought that this

NOTE Confidence: 0.7945916

00:13:53.870 --> 00:13:56.416 feedback loop is going to be

NOTE Confidence: 0.7945916

00:13:56.416 --> 00:13:58.586 critical for regulation of P53.

NOTE Confidence: 0.7945916

00:13:58.590 --> 00:14:00.990 The mice are fine, but again,

NOTE Confidence: 0.7945916

00:14:00.990 --> 00:14:02.990 as in the previous case,

NOTE Confidence: 0.7945916

00:14:02.990 --> 00:14:04.990 their exquisitely sensitive to radiation,

NOTE Confidence: 0.7945916

00:14:04.990 --> 00:14:08.686 so this is the same experiment that I

NOTE Confidence: 0.7945916

00:14:08.686 --> 00:14:12.593 showed you before we rated it the we

NOTE Confidence: 0.7945916

00:14:12.593 --> 00:14:15.579 irradiated the mice with six grade.

NOTE Confidence: 0.7945916

00:14:15.580 --> 00:14:17.532 Normal mice.

NOTE Confidence: 0.7945916

00:14:17.532 --> 00:14:20.460 MDM two heterozygotes.

NOTE Confidence: 0.7945916

00:14:20.460 --> 00:14:22.310 Don't care about this dose,

NOTE Confidence: 0.7945916

00:14:22.310 --> 00:14:25.710 but you can see that the P2P2 homozygous

NOTE Confidence: 0.7945916

00:14:25.710 --> 00:14:28.770 mice that no longer have this feedback

NOTE Confidence: 0.7945916

00:14:28.770 --> 00:14:31.830 loop are dead for the most part.

NOTE Confidence: 0.7945916

00:14:31.830 --> 00:14:34.080 So what is the phenotype?

NOTE Confidence: 0.7945916

00:14:34.080 --> 00:14:36.004 These animals are actually

NOTE Confidence: 0.7945916

00:14:36.004 --> 00:14:38.409 dying because of the complete

NOTE Confidence: 0.7945916

00:14:38.409 --> 00:14:40.380 ablation of the ball mirror,

NOTE Confidence: 0.7945916

00:14:40.380 --> 00:14:43.104 so here's a heterozygous irradiated mice

NOTE Confidence: 0.7945916

00:14:43.104 --> 00:14:47.339 and you can see that at 12 days you have

NOTE Confidence: 0.7945916

00:14:47.339 --> 00:14:50.280 some disruption of bone marrow function,

NOTE Confidence: 0.7945916

00:14:50.280 --> 00:14:51.992 but it's still viable,

NOTE Confidence: 0.7945916

00:14:51.992 --> 00:14:54.560 whereas in the mice that lack

NOTE Confidence: 0.7945916

00:14:54.647 --> 00:14:56.129 the feedback loop,

NOTE Confidence: 0.7945916

00:14:56.130 --> 00:14:57.930 there's a complete ablation

NOTE Confidence: 0.7945916

00:14:57.930 --> 00:14:59.280 of Humana Pelisses,

NOTE Confidence: 0.79331917

00:14:59.280 --> 00:15:02.500 and this is again a P53 dependent.

NOTE Confidence: 0.79331917

00:15:02.500 --> 00:15:05.734 Process so if we take out

NOTE Confidence: 0.79331917

00:15:05.734 --> 00:15:08.330 just one illegal appeal 53.

NOTE Confidence: 0.79331917

00:15:08.330 --> 00:15:10.784 We completely rescued this phenotype so

NOTE Confidence: 0.79331917

00:15:10.784 --> 00:15:13.847 we can rescue this phenotype with complete

NOTE Confidence: 0.79331917

00:15:13.847 --> 00:15:17.004 deletion of P53 or header zygosity 5053.

NOTE Confidence: 0.79331917

00:15:17.010 --> 00:15:20.202 So with these experiments are beginning

NOTE Confidence: 0.79331917

00:15:20.202 --> 00:15:24.238 to tell us is that there's this.

NOTE Confidence: 0.79331917

00:15:24.240 --> 00:15:26.070 There is this important relationship

NOTE Confidence: 0.79331917

00:15:26.070 --> 00:15:27.900 between MDM two and P53,

NOTE Confidence: 0.79331917

00:15:27.900 --> 00:15:29.715 and then there's an important

NOTE Confidence: 0.79331917

00:15:29.715 --> 00:15:32.081 balance that has to be maintained

NOTE Confidence: 0.79331917

00:15:32.081 --> 00:15:34.466 for survival after DNA damage.

NOTE Confidence: 0.79331917

00:15:34.470 --> 00:15:36.786 The last experiment that I'll show

NOTE Confidence: 0.79331917

00:15:36.786 --> 00:15:40.293 you here is our attempts to try to

NOTE Confidence: 0.79331917

00:15:40.293 --> 00:15:42.177 understand which pathway downstream

NOTE Confidence: 0.79331917

00:15:42.177 --> 00:15:45.668 of P53 is important for this phenotype.

NOTE Confidence: 0.79331917

00:15:45.670 --> 00:15:49.072 So I've already told you that heterozygosity

NOTE Confidence: 0.79331917

00:15:49.072 --> 00:15:51.340 Peachtree rescues the phenotype.

NOTE Confidence: 0.79331917

00:15:51.340 --> 00:15:54.460 We also generated mice with deletion of P.

NOTE Confidence: 0.79331917

00:15:54.460 --> 00:15:57.085 21, which is the cell cycle inhibitor

NOTE Confidence: 0.79331917

00:15:57.085 --> 00:16:00.310 and that had no effect on the phenotype.

NOTE Confidence: 0.79331917

00:16:00.310 --> 00:16:02.260 Those mice are also very

NOTE Confidence: 0.79331917

00:16:02.260 --> 00:16:03.430 sensitive to radiation,

NOTE Confidence: 0.79331917

00:16:03.430 --> 00:16:05.770 and then we also deleted Puma,

NOTE Confidence: 0.79331917

00:16:05.770 --> 00:16:08.110 which is one of the APOP

NOTE Confidence: 0.79331917

00:16:08.110 --> 00:16:09.670 totic targets of P53.

NOTE Confidence: 0.79331917

00:16:09.670 --> 00:16:11.590 And here you can see that

NOTE Confidence: 0.79331917

00:16:11.590 --> 00:16:13.458 there was a complete rescue

NOTE Confidence: 0.79331917

00:16:13.458 --> 00:16:15.518 of this hematopoietic defects.

NOTE Confidence: 0.79331917

00:16:15.520 --> 00:16:17.722 So in this scenario it appears

NOTE Confidence: 0.79331917

00:16:17.722 --> 00:16:20.799 that it is the APOP totic function

NOTE Confidence: 0.79331917

00:16:20.799 --> 00:16:23.344 opekta degree that is killing.

NOTE Confidence: 0.79331917

00:16:23.350 --> 00:16:26.618 These hematopoietic stem cells.

NOTE Confidence: 0.79331917

00:16:26.620 --> 00:16:29.861 So those are just a couple of

NOTE Confidence: 0.79331917

00:16:29.861 --> 00:16:31.872 the numerous experiments being

NOTE Confidence: 0.79331917

00:16:31.872 --> 00:16:34.532 done to evaluate the relationship

NOTE Confidence: 0.79331917

00:16:34.532 --> 00:16:36.660 between MDM24 and P53,

NOTE Confidence: 0.79331917

00:16:36.660 --> 00:16:39.666 and it's just an exquisite relationship.

NOTE Confidence: 0.79331917

00:16:39.670 --> 00:16:45.074 You need sufficient MDM 2 for survival.

NOTE Confidence: 0.79331917

00:16:45.080 --> 00:16:47.040 And.

NOTE Confidence: 0.79331917

00:16:47.040 --> 00:16:49.544 And if you have too much and you

NOTE Confidence: 0.79331917

00:16:49.544 --> 00:16:52.531 too MDM for deletion of 53 deletion

NOTE Confidence: 0.79331917

00:16:52.531 --> 00:16:55.433 of downstream effectors of P53 can

NOTE Confidence: 0.79331917

00:16:55.433 --> 00:16:57.629 rescue those lethal phenotypes.

NOTE Confidence: 0.79331917

00:16:57.630 --> 00:16:59.952 Now come. Because.

NOTE Confidence: 0.79331917

00:16:59.952 --> 00:17:04.596 MDM two is so lethal early.

NOTE Confidence: 0.79331917

00:17:04.600 --> 00:17:06.067 During embryo Genesis,

NOTE Confidence: 0.79331917

00:17:06.067 --> 00:17:09.001 one of the experiments we wanted

NOTE Confidence: 0.79331917

00:17:09.001 --> 00:17:12.232 to do is really ask about an adult

NOTE Confidence: 0.79331917

00:17:12.232 --> 00:17:16.032 mouse and how important is MDM two in

NOTE Confidence: 0.79331917

00:17:16.032 --> 00:17:18.602 different tissues at different times,

NOTE Confidence: 0.79331917

00:17:18.610 --> 00:17:20.218 and we've used,

NOTE Confidence: 0.79331917

00:17:20.218 --> 00:17:22.362 we've generated this conditional

NOTE Confidence: 0.79331917

00:17:22.362 --> 00:17:25.828 allele of MDM two and this is using

NOTE Confidence: 0.79331917

00:17:25.828 --> 00:17:29.167 The LOX P system so these two lacks

NOTE Confidence: 0.79331917

00:17:29.167 --> 00:17:32.119 besides encompass two of the accents

NOTE Confidence: 0.79331917

00:17:32.119 --> 00:17:35.350 that code for the major finding.

NOTE Confidence: 0.79331917

00:17:35.350 --> 00:17:37.240 Region 2P53 so this conditional

NOTE Confidence: 0.79331917

00:17:37.240 --> 00:17:39.841 Leo then allows us to delete MDM

NOTE Confidence: 0.79331917

00:17:39.841 --> 00:17:42.401 two in any tissue that we want to.

NOTE Confidence: 0.79331917

00:17:42.410 --> 00:17:44.175 And we've generated number of

NOTE Confidence: 0.79331917

00:17:44.175 --> 00:17:45.940 tissues and experiments that way.

NOTE Confidence: 0.79331917

00:17:45.940 --> 00:17:48.996 But what I want to show you is

NOTE Confidence: 0.79331917

00:17:48.996 --> 00:17:51.263 what happens when you globally

NOTE Confidence: 0.79331917

00:17:51.263 --> 00:17:54.560 remove MDM two in the whole months.

NOTE Confidence: 0.79331917

00:17:54.560 --> 00:17:56.390 So as the title says,

NOTE Confidence: 0.79331917

00:17:56.390 --> 00:17:58.567 Indian too lost in the adult is

NOTE Confidence: 0.79331917

00:17:58.567 --> 00:18:00.780 results in a lethal phenotype.

NOTE Confidence: 0.79331917

00:18:00.780 --> 00:18:02.985 So what we've done here is we've

NOTE Confidence: 0.79331917

00:18:02.985 --> 00:18:05.212 used a Cree transgene that is

NOTE Confidence: 0.79331917

00:18:05.212 --> 00:18:07.217 tamoxifen inducible so this is

NOTE Confidence: 0.79331917

00:18:07.217 --> 00:18:10.296 this is a mouse that has one of

NOTE Confidence: 0.79331917

00:18:10.296 --> 00:18:12.126 the conditional alleles and it

NOTE Confidence: 0.79331917

00:18:12.126 --> 00:18:13.956 has the other legalism missing,

NOTE Confidence: 0.79331917

00:18:13.960 --> 00:18:16.324 so it's single recombination event is

NOTE Confidence: 0.79331917

00:18:16.324 --> 00:18:19.807 going to create an M2 normals or not sell in.

NOTE Confidence: 0.79331917

00:18:19.810 --> 00:18:22.650 All we do is inject tamoxifen and then

NOTE Confidence: 0.79331917

00:18:22.650 --> 00:18:25.376 we look at what happens to these.

NOTE Confidence: 0.79331917

00:18:25.380 --> 00:18:25.734 Phenotypes,

NOTE Confidence: 0.79331917

00:18:25.734 --> 00:18:28.566 and I think you can see from this

NOTE Confidence: 0.79331917

00:18:28.566 --> 00:18:31.326 graph here on the right that within 5

NOTE Confidence: 0.79331917

00:18:31.326 --> 00:18:34.070 days of treating the mice with tamoxifen,

NOTE Confidence: 0.79331917

00:18:34.070 --> 00:18:35.813 they're all dead.

NOTE Confidence: 0.79331917

00:18:35.813 --> 00:18:36.394 So,

NOTE Confidence: 0.79331917

00:18:36.394 --> 00:18:40.914 so losing MDM two and it's a

NOTE Confidence: 0.79331917

00:18:40.914 --> 00:18:43.638 Peachtree dependent process.

NOTE Confidence: 0.8166339

00:18:43.640 --> 00:18:49.320 Causes this enormous physiological response.

NOTE Confidence: 0.8166339

00:18:49.320 --> 00:18:52.792 These are some of the pathologies we

NOTE Confidence: 0.8166339

00:18:52.792 --> 00:18:56.919 see in these mice the hippocampus has.

NOTE Confidence: 0.8166339

00:18:56.920 --> 00:18:59.992 Less number of cells, the retina is is

NOTE Confidence: 0.8166339

00:18:59.992 --> 00:19:02.044 comprised of multiple beautiful layers

NOTE Confidence: 0.8166339

00:19:02.044 --> 00:19:06.164 and you can see that it all of these cases

NOTE Confidence: 0.8166339

00:19:06.164 --> 00:19:09.069 that it's it's a decreased cell numbers.

NOTE Confidence: 0.8166339

00:19:09.070 --> 00:19:11.905 There's some differences in in the liver,

NOTE Confidence: 0.8166339

00:19:11.910 --> 00:19:13.935 and it's actually if you

NOTE Confidence: 0.8166339

00:19:13.935 --> 00:19:15.150 measure liver function.

NOTE Confidence: 0.8166339

00:19:15.150 --> 00:19:16.770 Liver function is compromised

NOTE Confidence: 0.8166339

00:19:16.770 --> 00:19:19.200 when you have deletions of MDM.

NOTE Confidence: 0.8166339

00:19:19.200 --> 00:19:21.630 Two kidney has all these protein

NOTE Confidence: 0.8166339

00:19:21.630 --> 00:19:23.250 casts and dilated tubules,

NOTE Confidence: 0.8166339

00:19:23.250 --> 00:19:27.121 and then in the spleen we have

NOTE Confidence: 0.8166339

00:19:27.121 --> 00:19:29.560 complete absence of white.

NOTE Confidence: 0.8166339

00:19:29.560 --> 00:19:31.404 And as I indicated,

NOTE Confidence: 0.8166339

00:19:31.404 --> 00:19:34.170 these phenotypes are all P53 dependent.

NOTE Confidence: 0.8166339

00:19:34.170 --> 00:19:35.232 So it just.

NOTE Confidence: 0.8166339

00:19:35.232 --> 00:19:37.344 I mean, I think the important

NOTE Confidence: 0.8166339

00:19:37.344 --> 00:19:39.885 aspect of this slide is that in

NOTE Confidence: 0.8166339

00:19:39.885 --> 00:19:42.391 some of the previous slide is it

NOTE Confidence: 0.8166339

00:19:42.391 --> 00:19:44.195 highlights some of the pathologies

NOTE Confidence: 0.8166339

00:19:44.195 --> 00:19:47.088 that we might see when we use MDM.

NOTE Confidence: 0.8166339

00:19:47.088 --> 00:19:50.784 Two inhibitors in the clinic to it.

NOTE Confidence: 0.8166339

00:19:50.790 --> 00:19:53.622 To inhibit the tumors that have

NOTE Confidence: 0.8166339

00:19:53.622 --> 00:19:56.300 high levels of empathy into it.

NOTE Confidence: 0.8166339

00:19:56.300 --> 00:19:58.060 And the hematopoietic defense

NOTE Confidence: 0.8166339

00:19:58.060 --> 00:20:00.260 is actually observed in humans.

NOTE Confidence: 0.8166339

00:20:00.260 --> 00:20:03.508 It is treated with MDM two inhibitors.

NOTE Confidence: 0.8166339

00:20:03.510 --> 00:20:03.854 Uhm?

NOTE Confidence: 0.8166339

00:20:03.854 --> 00:20:06.950 But now I want to use this model system

NOTE Confidence: 0.8166339

00:20:07.029 --> 00:20:09.465 to understand what piece of degree

NOTE Confidence: 0.8166339

00:20:09.465 --> 00:20:12.579 is doing in these different tissues.

NOTE Confidence: 0.8166339

00:20:12.580 --> 00:20:14.974 In you know one reason for doing

NOTE Confidence: 0.8166339

00:20:14.974 --> 00:20:17.777 this is we would like to be

NOTE Confidence: 0.8166339

00:20:17.777 --> 00:20:19.897 able to reactivate P53 somehow,

NOTE Confidence: 0.8166339

00:20:19.900 --> 00:20:22.210 and it's kind of hard people.

NOTE Confidence: 0.8166339

00:20:22.210 --> 00:20:24.905 A lot of people are trying to

NOTE Confidence: 0.8166339

00:20:24.905 --> 00:20:26.829 to reactivate people to agree,

NOTE Confidence: 0.8166339

00:20:26.830 --> 00:20:29.063 but what we were hoping is that

NOTE Confidence: 0.8166339

00:20:29.063 --> 00:20:31.501 we might be able to identify

NOTE Confidence: 0.8166339

00:20:31.501 --> 00:20:33.369 downstream pathways to P53.

NOTE Confidence: 0.8166339

00:20:33.370 --> 00:20:35.850 There would be better targets

NOTE Confidence: 0.8166339

00:20:35.850 --> 00:20:37.338 for reactivation tours.

NOTE Confidence: 0.8166339

00:20:37.340 --> 00:20:40.916 So let me show you what we did.

NOTE Confidence: 0.8166339

00:20:40.920 --> 00:20:43.608 So again we used our MDM,

NOTE Confidence: 0.8166339

00:20:43.610 --> 00:20:45.750 two conditional mouse and we

NOTE Confidence: 0.8166339

00:20:45.750 --> 00:20:48.989 deleted MDM two in the adult mouse.

NOTE Confidence: 0.8166339

00:20:48.990 --> 00:20:52.166 But we did this acutely and we actually

NOTE Confidence: 0.8166339

00:20:52.166 --> 00:20:55.375 chose a 24 time our time point to

NOTE Confidence: 0.8166339

00:20:55.375 --> 00:20:58.102 ask what P53 targets are regulated

NOTE Confidence: 0.8166339

00:20:58.102 --> 00:21:01.348 in different issues that lead to

NOTE Confidence: 0.8166339

00:21:01.348 --> 00:21:05.319 these pathologies in the adults.

NOTE Confidence: 0.8166339

00:21:05.320 --> 00:21:05.669 OK,

NOTE Confidence: 0.8166339

00:21:05.669 --> 00:21:08.112 so this is now all the different

NOTE Confidence: 0.8166339

00:21:08.112 --> 00:21:10.029 issues that we initially looked

NOTE Confidence: 0.8166339

00:21:10.029 --> 00:21:12.682 at and what I'm showing you here

NOTE Confidence: 0.8166339

00:21:12.756 --> 00:21:14.868 is the percent recombination.

NOTE Confidence: 0.8166339

00:21:14.870 --> 00:21:17.258 So once we treat with tamoxifen,

NOTE Confidence: 0.8166339

00:21:17.260 --> 00:21:20.578 we induce recombination of the locus.

NOTE Confidence: 0.8166339

00:21:20.580 --> 00:21:23.177 And you can see that the pancreas,

NOTE Confidence: 0.8166339

00:21:23.180 --> 00:21:24.668 the heart being tested,

NOTE Confidence: 0.8166339

00:21:24.668 --> 00:21:26.900 had the highest level of recombination.

NOTE Confidence: 0.8166339

00:21:26.900 --> 00:21:27.269 Again,

NOTE Confidence: 0.8166339

00:21:27.269 --> 00:21:31.000 I will point out that we only have one MDM,

NOTE Confidence: 0.8166339

00:21:31.000 --> 00:21:31.742 two allele.

NOTE Confidence: 0.8166339

00:21:31.742 --> 00:21:33.968 The other allele is an allele,

NOTE Confidence: 0.8166339

00:21:33.970 --> 00:21:35.758 so single recombination event

NOTE Confidence: 0.8166339

00:21:35.758 --> 00:21:37.546 will activate P 53.

NOTE Confidence: 0.8166339

00:21:37.550 --> 00:21:40.030 And then on this axis we chose to

NOTE Confidence: 0.8166339

00:21:40.030 --> 00:21:42.419 look at P53 activation by measuring

NOTE Confidence: 0.8166339

00:21:42.419 --> 00:21:45.466 the expression of P 21 which is

NOTE Confidence: 0.8166339

00:21:45.466 --> 00:21:47.606 encodes a cell cycle inhibitor.

NOTE Confidence: 0.8166339

00:21:47.610 --> 00:21:50.319 So you can see in this experiment

NOTE Confidence: 0.8166339

00:21:50.319 --> 00:21:51.480 at the kidney,

NOTE Confidence: 0.8166339

00:21:51.480 --> 00:21:53.550 the pancreas in the intestine

NOTE Confidence: 0.8166339

00:21:53.550 --> 00:21:55.620 where the tissues that expressed

NOTE Confidence: 0.8166339

00:21:55.687 --> 00:21:57.287 the highest levels of P.

NOTE Confidence: 0.8166339

00:21:57.290 --> 00:21:57.898 21.

NOTE Confidence: 0.8166339

00:21:57.898 --> 00:22:01.546 And we were thinking the highest

NOTE Confidence: 0.8166339

00:22:01.546 --> 00:22:04.000 levels of of P53.

NOTE Confidence: 0.8166339

00:22:04.000 --> 00:22:06.317 So we've we've looked at these mice,

NOTE Confidence: 0.8166339

00:22:06.320 --> 00:22:08.630 so in 24 hours we see no.

NOTE Confidence: 0.8166339

00:22:08.630 --> 00:22:11.609 So let me back up just for one second,

NOTE Confidence: 0.8166339

00:22:11.610 --> 00:22:13.596 I hope I can do that,

NOTE Confidence: 0.8166339

00:22:13.600 --> 00:22:15.180 so that issues that we

NOTE Confidence: 0.8166339

00:22:15.180 --> 00:22:16.760 decided to look at where

NOTE Confidence: 0.84411275

00:22:16.830 --> 00:22:18.432 the kidney, the pancreas,

NOTE Confidence: 0.84411275

00:22:18.432 --> 00:22:20.587 the intestine in the heart.

NOTE Confidence: 0.84411275

00:22:20.590 --> 00:22:22.876 And the ovary in the ovary.

NOTE Confidence: 0.84411275

00:22:22.880 --> 00:22:25.652 Just because P 53 is mutant in

NOTE Confidence: 0.84411275

00:22:25.652 --> 00:22:27.469 ovarian carcinoma is at 95%.

NOTE Confidence: 0.86477065

00:22:29.560 --> 00:22:32.230 95% frequency and so we wondered

NOTE Confidence: 0.86477065

00:22:32.230 --> 00:22:37.008 if we might be able to to begin to

NOTE Confidence: 0.86477065

00:22:37.008 --> 00:22:39.324 understand that mutation frequency.

NOTE Confidence: 0.86477065

00:22:39.330 --> 00:22:42.354 So for the heart for the ovary,

NOTE Confidence: 0.86477065

00:22:42.360 --> 00:22:45.888 we saw absolutely no phenotypes after

NOTE Confidence: 0.86477065

00:22:45.888 --> 00:22:49.220 24 hours post deletion of MDM 2.

NOTE Confidence: 0.86477065

00:22:49.220 --> 00:22:52.184 In the intestine we saw a

NOTE Confidence: 0.86477065

00:22:52.184 --> 00:22:53.666 fascinating phenotype which

NOTE Confidence: 0.86477065

00:22:53.666 --> 00:22:56.640 is descript dropout phenotype.

NOTE Confidence: 0.86477065

00:22:56.640 --> 00:22:57.666 In yellow here,

NOTE Confidence: 0.86477065

00:22:57.666 --> 00:22:59.866 I've outlined the ***** of the

NOTE Confidence: 0.86477065

00:22:59.866 --> 00:23:02.757 intestine and one of the phenotypes is

NOTE Confidence: 0.86477065

00:23:02.757 --> 00:23:06.035 the complete absence of of the crypt.

NOTE Confidence: 0.86477065

00:23:06.040 --> 00:23:08.356 Sydney in the lab is quantified

NOTE Confidence: 0.86477065

00:23:08.356 --> 00:23:10.698 the number of ***** in these

NOTE Confidence: 0.86477065

00:23:10.698 --> 00:23:12.693 different animals and you can

NOTE Confidence: 0.86477065

00:23:12.693 --> 00:23:15.298 see the mice that have no MDM.

NOTE Confidence: 0.86477065

00:23:15.300 --> 00:23:17.586 2 have about little more than

NOTE Confidence: 0.86477065

00:23:17.586 --> 00:23:20.332 little but half of the number of

NOTE Confidence: 0.86477065

00:23:20.332 --> 00:23:24.020 ***** is in normal control mouse.

NOTE Confidence: 0.86477065

00:23:24.020 --> 00:23:26.150 The kidney also had some phenotypes

NOTE Confidence: 0.86477065

00:23:26.150 --> 00:23:28.714 at 24 hours and it had twice

NOTE Confidence: 0.86477065

00:23:28.714 --> 00:23:30.499 the number of protein casts,

NOTE Confidence: 0.86477065

00:23:30.500 --> 00:23:32.300 so you can see here.

NOTE Confidence: 0.86477065

00:23:32.300 --> 00:23:35.540 So this is an early phenotype in the kidney.

NOTE Confidence: 0.86477065

00:23:35.540 --> 00:23:36.012 Again,

NOTE Confidence: 0.86477065

00:23:36.012 --> 00:23:39.316 this is the normal kidney control experiment.

NOTE Confidence: 0.86477065

00:23:39.320 --> 00:23:42.648 And then the pancreas had to be a

NOTE Confidence: 0.86477065

00:23:42.648 --> 00:23:44.345 fascinating phenotype which will

NOTE Confidence: 0.86477065

00:23:44.345 --> 00:23:47.110 delve into a little bit more deeply.

NOTE Confidence: 0.86477065

00:23:47.110 --> 00:23:52.166 But we saw in the pancreas is this?

NOTE Confidence: 0.86477065

00:23:52.170 --> 00:23:53.922 Acinar to ductal metaplasia,

NOTE Confidence: 0.86477065

00:23:53.922 --> 00:23:57.220 so here's a normal pancreas in the top,

NOTE Confidence: 0.86477065

00:23:57.220 --> 00:24:00.146 and here's what the pancreas looks like

NOTE Confidence: 0.86477065

00:24:00.146 --> 00:24:03.958 in the in the animals that have no MDM.

NOTE Confidence: 0.86477065

00:24:03.960 --> 00:24:06.907 2 Here we've stained with keratin 19,

NOTE Confidence: 0.86477065

00:24:06.910 --> 00:24:11.214 which is a marker for a ductal cell.

NOTE Confidence: 0.86477065

00:24:11.220 --> 00:24:13.926 And here we've measured the Metaplastic

NOTE Confidence: 0.86477065

00:24:13.926 --> 00:24:17.161 area and we also see an immune

NOTE Confidence: 0.86477065

00:24:17.161 --> 00:24:19.849 infiltration in these in these mice.

NOTE Confidence: 0.86477065

00:24:19.850 --> 00:24:23.314 So within 24 hours we saw this huge

NOTE Confidence: 0.86477065

00:24:23.314 --> 00:24:26.657 plasticity in the pancreas from you know,

NOTE Confidence: 0.86477065

00:24:26.660 --> 00:24:29.960 this acinar to ductal metaplasia.

NOTE Confidence: 0.86477065

00:24:29.960 --> 00:24:30.357 OK,

NOTE Confidence: 0.86477065

00:24:30.357 --> 00:24:32.342 so we've taken these five

NOTE Confidence: 0.86477065

00:24:32.342 --> 00:24:33.930 tissues an we've done.

NOTE Confidence: 0.86477065

00:24:33.930 --> 00:24:36.709 We've looked for expression of P53 targets,

NOTE Confidence: 0.86477065

00:24:36.710 --> 00:24:39.770 so on the left here I show you all of

NOTE Confidence: 0.86477065

00:24:39.859 --> 00:24:43.219 the genes that were regulated in these

NOTE Confidence: 0.86477065

00:24:43.219 --> 00:24:46.240 different issues in our RNA seek data.

NOTE Confidence: 0.86477065

00:24:46.240 --> 00:24:48.879 The dark region is the region that

NOTE Confidence: 0.86477065

00:24:48.879 --> 00:24:51.400 is upregulated in the lighter color,

NOTE Confidence: 0.86477065

00:24:51.400 --> 00:24:54.028 shows the regions that were downregulated

NOTE Confidence: 0.86477065

00:24:54.028 --> 00:24:56.840 in each of these five tissues.

NOTE Confidence: 0.86477065

00:24:56.840 --> 00:24:59.536 On the right here I show the percent

NOTE Confidence: 0.86477065

00:24:59.536 --> 00:25:01.840 of these these dysregulated genes

NOTE Confidence: 0.86477065

00:25:01.840 --> 00:25:04.420 that are actually P53 targets.

NOTE Confidence: 0.86477065

00:25:04.420 --> 00:25:06.670 They have a pizza degree binding

NOTE Confidence: 0.86477065

00:25:06.670 --> 00:25:09.655 site and we used data from the

NOTE Confidence: 0.86477065

00:25:09.655 --> 00:25:12.000 literature to identify these tools.

NOTE Confidence: 0.86477065

00:25:12.000 --> 00:25:13.680 With P53 binding sites.

NOTE Confidence: 0.86477065

00:25:13.680 --> 00:25:15.360 So in the intestine,

NOTE Confidence: 0.86477065

00:25:15.360 --> 00:25:16.202 for example,

NOTE Confidence: 0.86477065

00:25:16.202 --> 00:25:19.572 I think that number is 69% of the

NOTE Confidence: 0.86477065

00:25:19.572 --> 00:25:22.098 channels had pizza pre binding sites.

NOTE Confidence: 0.86477065

00:25:22.100 --> 00:25:24.020 So the the most.

NOTE Confidence: 0.86477065

00:25:24.020 --> 00:25:26.420 Most of the genes dysregulated

NOTE Confidence: 0.86477065

00:25:26.420 --> 00:25:29.405 it in the system by deletion

NOTE Confidence: 0.86477065

00:25:29.405 --> 00:25:32.390 of MDM 2RP53 targets the ****.

NOTE Confidence: 0.86477065

00:25:32.390 --> 00:25:35.096 ** the other hand had a

NOTE Confidence: 0.86477065

00:25:35.096 --> 00:25:36.449 huge physiological response,

NOTE Confidence: 0.86477065

00:25:36.450 --> 00:25:39.156 600 for jeans that were dysregulated

NOTE Confidence: 0.86477065

00:25:39.156 --> 00:25:42.010 but only 16% repeated different targets.

NOTE Confidence: 0.86477065

00:25:42.010 --> 00:25:45.010 So what we're capturing here at

NOTE Confidence: 0.86477065

00:25:45.010 --> 00:25:47.913 24 hours is not just you know

NOTE Confidence: 0.86477065

00:25:47.913 --> 00:25:50.429 activation of P53 and P3 targets,

NOTE Confidence: 0.86477065

00:25:50.430 --> 00:25:53.785 But the downstream responds to

NOTE Confidence: 0.86477065

00:25:53.785 --> 00:25:55.798 that P53 activation.

NOTE Confidence: 0.86477065

00:25:55.800 --> 00:25:58.383 So this is now compilation of all

NOTE Confidence: 0.86477065

00:25:58.383 --> 00:26:00.343 those five different tissues to

NOTE Confidence: 0.86477065

00:26:00.343 --> 00:26:03.059 examine the overlap in P53 target genes.

NOTE Confidence: 0.86477065

00:26:03.060 --> 00:26:06.084 And as you can see from this figure

NOTE Confidence: 0.86477065

00:26:06.084 --> 00:26:10.130 on the left there were only 7 jewels

NOTE Confidence: 0.86477065

00:26:10.130 --> 00:26:12.258 that were commonly regulated.

NOTE Confidence: 0.80729765

00:26:12.260 --> 00:26:15.564 By MDM 2 lost that repeat 53 targets.

NOTE Confidence: 0.80729765

00:26:15.570 --> 00:26:19.107 So for example here in the pink is we

NOTE Confidence: 0.80729765

00:26:19.107 --> 00:26:22.655 have 206 genes, 135 of the P53 targets

NOTE Confidence: 0.80729765

00:26:22.655 --> 00:26:25.780 are specific to the pancreas and seven

NOTE Confidence: 0.80729765

00:26:25.780 --> 00:26:29.007 were shared with the other four tissues.

NOTE Confidence: 0.80729765

00:26:29.010 --> 00:26:32.490 So these seven jeans are MDM two cycling,

NOTE Confidence: 0.80729765

00:26:32.490 --> 00:26:35.970 G1 MDM two as we as I mentioned,

NOTE Confidence: 0.80729765

00:26:35.970 --> 00:26:38.195 the very beginning is regulated

NOTE Confidence: 0.80729765

00:26:38.195 --> 00:26:41.189 by P53 and we expect it well.

NOTE Confidence: 0.80729765

00:26:41.190 --> 00:26:44.670 We didn't expect them to be able to,

NOTE Confidence: 0.80729765

00:26:44.670 --> 00:26:48.492 but it's not a surprising result because

NOTE Confidence: 0.80729765

00:26:48.492 --> 00:26:51.789 the promoter is intact in MDM 2.

NOTE Confidence: 0.80729765

00:26:51.790 --> 00:26:55.446 Three of these genes segment you one GST,

NOTE Confidence: 0.80729765

00:26:55.450 --> 00:26:58.570 one piece art, one or cell cycle regulators

NOTE Confidence: 0.80729765

00:26:58.570 --> 00:27:01.838 to these jeans are transcription factors,

NOTE Confidence: 0.80729765

00:27:01.840 --> 00:27:05.039 and this gene EDA 2R herself directions.

NOTE Confidence: 0.80729765

00:27:05.040 --> 00:27:07.638 So these are the six Peachtree

NOTE Confidence: 0.80729765

00:27:07.638 --> 00:27:10.070 targets that have a common.

NOTE Confidence: 0.80729765

00:27:10.070 --> 00:27:13.297 They represent the common signature of of

NOTE Confidence: 0.80729765

00:27:13.297 --> 00:27:16.470 upregulated genes in in these three tissues.

NOTE Confidence: 0.80729765

00:27:16.470 --> 00:27:19.314 We wanted to validate the signature

NOTE Confidence: 0.80729765

00:27:19.314 --> 00:27:22.199 to make sure that they were.

NOTE Confidence: 0.80729765

00:27:22.200 --> 00:27:24.340 Truly, a P53 targets physiologically.

NOTE Confidence: 0.80729765

00:27:24.340 --> 00:27:28.060 And So what we did is we did our our DNA

NOTE Confidence: 0.80729765

00:27:28.060 --> 00:27:30.760 damage ionizing radiation experiment.

NOTE Confidence: 0.80729765

00:27:30.760 --> 00:27:32.895 We treated the whole animal

NOTE Confidence: 0.80729765

00:27:32.895 --> 00:27:34.176 with ionizing radiation,

NOTE Confidence: 0.80729765

00:27:34.180 --> 00:27:38.032 and here's the data for two of the jeans,

NOTE Confidence: 0.80729765

00:27:38.040 --> 00:27:40.650 and we've done it for all

NOTE Confidence: 0.80729765

00:27:40.650 --> 00:27:43.170 of seven cycling G1 E DA2R.

NOTE Confidence: 0.80729765

00:27:43.170 --> 00:27:45.310 Here's the wild type levels

NOTE Confidence: 0.80729765

00:27:45.310 --> 00:27:47.450 of expression of these genes.

NOTE Confidence: 0.80729765

00:27:47.450 --> 00:27:48.590 If we irradiate,

NOTE Confidence: 0.80729765

00:27:48.590 --> 00:27:52.260 you can see that these genes are upregulated.

NOTE Confidence: 0.80729765

00:27:52.260 --> 00:27:55.143 In in both cases,

NOTE Confidence: 0.80729765

00:27:55.143 --> 00:27:58.230 and if we irradiate a P53 null,

NOTE Confidence: 0.80729765

00:27:58.230 --> 00:28:01.317 you see you see no up regulation.

NOTE Confidence: 0.80729765

00:28:01.320 --> 00:28:04.248 So these are P3 target genes that are

NOTE Confidence: 0.80729765

00:28:04.248 --> 00:28:06.383 being upregulated following punishing

NOTE Confidence: 0.80729765

00:28:06.383 --> 00:28:09.930 radiation. So these experiments.

NOTE Confidence: 0.80729765

00:28:09.930 --> 00:28:12.830 Highlight this incredible repertoire.

NOTE Confidence: 0.80729765

00:28:12.830 --> 00:28:15.730 Transcriptional targets that P53

NOTE Confidence: 0.80729765

00:28:15.730 --> 00:28:17.979 physiologically regulates the vivo

NOTE Confidence: 0.80729765

00:28:17.979 --> 00:28:20.513 and I think it also suggests that

NOTE Confidence: 0.80729765

00:28:20.513 --> 00:28:22.676 maybe these specific targets can

NOTE Confidence: 0.80729765

00:28:22.676 --> 00:28:25.256 be used to understand in vivo.

NOTE Confidence: 0.80729765

00:28:25.260 --> 00:28:26.688 If you have.

NOTE Confidence: 0.80729765

00:28:26.688 --> 00:28:30.020 If you can reactivate piece of D3

NOTE Confidence: 0.80729765

00:28:30.127 --> 00:28:33.439 or convert mutant and wild type,

NOTE Confidence: 0.80729765

00:28:33.440 --> 00:28:37.142 these might be great markers to

NOTE Confidence: 0.80729765

00:28:37.142 --> 00:28:40.070 look at for activation 53.

NOTE Confidence: 0.80729765

00:28:40.070 --> 00:28:40.551 OK,

NOTE Confidence: 0.80729765

00:28:40.551 --> 00:28:43.918 I want to now just briefly discuss

NOTE Confidence: 0.80729765

00:28:43.918 --> 00:28:47.090 this this encrypted hypothesis.

NOTE Confidence: 0.80729765

00:28:47.090 --> 00:28:49.398 Acinar ductal hyperplasia that

NOTE Confidence: 0.80729765

00:28:49.398 --> 00:28:53.380 we see within 24 hours in the.

NOTE Confidence: 0.80729765

00:28:53.380 --> 00:28:54.916 In the pancreas.

NOTE Confidence: 0.80729765

00:28:54.916 --> 00:28:58.500 So one of the other experiments that

NOTE Confidence: 0.80729765

00:28:58.600 --> 00:29:02.232 Sidney Moyer in my lab did is we

NOTE Confidence: 0.80729765

00:29:02.232 --> 00:29:05.560 obtained these mice MST one missed one.

NOTE Confidence: 0.80729765

00:29:05.560 --> 00:29:06.736 Cree are OK,

NOTE Confidence: 0.80729765

00:29:06.736 --> 00:29:09.088 so missed one is an SNR

NOTE Confidence: 0.80729765

00:29:09.088 --> 00:29:11.400 specific transcription factor.

NOTE Confidence: 0.80729765

00:29:11.400 --> 00:29:14.809 An upstream of the missed one promoter.

NOTE Confidence: 0.7735238

00:29:16.910 --> 00:29:18.730 There is a criar transgene,

NOTE Confidence: 0.7735238

00:29:18.730 --> 00:29:21.320 which means that you can express create

NOTE Confidence: 0.7735238

00:29:21.320 --> 00:29:24.770 only in the *** and ourselves with the.

NOTE Confidence: 0.7735238

00:29:24.770 --> 00:29:28.553 Pancreas and so Sydney worked out the

NOTE Confidence: 0.7735238

00:29:28.553 --> 00:29:31.688 tamoxifen conditions that gave you

NOTE Confidence: 0.7735238

00:29:31.688 --> 00:29:34.838 a similar percent recombination as

NOTE Confidence: 0.7735238

00:29:34.838 --> 00:29:37.598 our previous experiments with MDM.

NOTE Confidence: 0.7735238

00:29:37.600 --> 00:29:40.996 Two position in the entire pancreas.

NOTE Confidence: 0.7735238

00:29:41.000 --> 00:29:44.474 OK, so similar percentage of recombination

NOTE Confidence: 0.7735238

00:29:44.474 --> 00:29:47.820 and similar activation of P3 targets.

NOTE Confidence: 0.7735238

00:29:47.820 --> 00:29:52.356 So here we use two of our targets,

NOTE Confidence: 0.7735238

00:29:52.360 --> 00:29:54.528 EADE, DA2RG, STT, SC1.

NOTE Confidence: 0.7735238

00:29:54.528 --> 00:29:57.780 To measure people to the activation

NOTE Confidence: 0.7735238

00:29:57.875 --> 00:30:00.955 and you can see in both my sweet.

NOTE Confidence: 0.7735238

00:30:00.960 --> 00:30:02.970 MDM two deletion happens in the

NOTE Confidence: 0.7735238

00:30:02.970 --> 00:30:05.038 home pancreas or mice where it

NOTE Confidence: 0.7735238

00:30:05.038 --> 00:30:07.030 only happens in the acinar cells.

NOTE Confidence: 0.7735238

00:30:07.030 --> 00:30:08.715 You have similar activation of

NOTE Confidence: 0.7735238

00:30:08.715 --> 00:30:10.400 these three of these targets,

NOTE Confidence: 0.7735238

00:30:10.400 --> 00:30:14.360 so we felt we could do.

NOTE Confidence: 0.7735238

00:30:14.360 --> 00:30:15.920 We could actually compare deletion

NOTE Confidence: 0.7735238

00:30:15.920 --> 00:30:18.450 of MDM two in the whole pancreas.

NOTE Confidence: 0.7735238

00:30:18.450 --> 00:30:20.982 The deletion of MDM two jestoni

NOTE Confidence: 0.7735238

00:30:20.982 --> 00:30:21.826 Essen ourselves.

NOTE Confidence: 0.7735238

00:30:21.830 --> 00:30:25.430 And we have absolutely no phenotype.

NOTE Confidence: 0.7735238

00:30:25.430 --> 00:30:28.568 So these pancreas look completely normal.

NOTE Confidence: 0.7735238

00:30:28.570 --> 00:30:32.238 Here we're measuring just we're looking at.

NOTE Confidence: 0.7735238

00:30:32.240 --> 00:30:33.288 I mean,

NOTE Confidence: 0.7735238

00:30:33.288 --> 00:30:37.480 I ageny sections in here in the right.

NOTE Confidence: 0.7735238

00:30:37.480 --> 00:30:39.576 We're measuring the immune

NOTE Confidence: 0.7735238

00:30:39.576 --> 00:30:42.196 component and these these pancreas.

NOTE Confidence: 0.7735238

00:30:42.200 --> 00:30:46.260 These pancreatic perfectly normal. So.

NOTE Confidence: 0.7735238

00:30:46.260 --> 00:30:49.487 The take home message here is that.

NOTE Confidence: 0.7735238

00:30:49.490 --> 00:30:51.700 This esnard ductal hyperplasia that

NOTE Confidence: 0.7735238

00:30:51.700 --> 00:30:55.830 we see is a P53 specific hyperplasia.

NOTE Confidence: 0.7735238

00:30:55.830 --> 00:30:58.920 But it's it's arising from signals

NOTE Confidence: 0.7735238

00:30:58.920 --> 00:31:01.630 outside of the acinar cells.

NOTE Confidence: 0.7735238

00:31:01.630 --> 00:31:03.487 So to me,

NOTE Confidence: 0.7735238

00:31:03.487 --> 00:31:06.582 this is a fascinating experiment

NOTE Confidence: 0.7735238

00:31:06.582 --> 00:31:10.507 because no one's ever noted that.

NOTE Confidence: 0.7735238

00:31:10.510 --> 00:31:13.906 That the environment can can affect

NOTE Confidence: 0.7735238

00:31:13.906 --> 00:31:16.170 the pizza delivery response,

NOTE Confidence: 0.7735238

00:31:16.170 --> 00:31:20.790 and so we'll be delving into understanding

NOTE Confidence: 0.7735238

00:31:20.790 --> 00:31:24.898 this phenotype a little bit better.

NOTE Confidence: 0.7735238

00:31:24.900 --> 00:31:28.862 Pancreas is is one of the tumors

NOTE Confidence: 0.7735238

00:31:28.862 --> 00:31:32.180 with 7075% mutations in P53 an and

NOTE Confidence: 0.7735238

00:31:32.180 --> 00:31:35.066 it always has this very compromised

NOTE Confidence: 0.7735238

00:31:35.066 --> 00:31:38.738 stromal component and so maybe by

NOTE Confidence: 0.7735238

00:31:38.738 --> 00:31:43.319 understanding what P 53 is doing is

NOTE Confidence: 0.7735238

00:31:43.319 --> 00:31:45.305 physiologically important Organism,

NOTE Confidence: 0.7735238

00:31:45.310 --> 00:31:49.377 we might be able to impact our

NOTE Confidence: 0.7735238

00:31:49.377 --> 00:31:52.929 understanding of Peter mutations in

NOTE Confidence: 0.7735238

00:31:52.929 --> 00:31:56.360 pancreatic cancer. OK, so the let out.

NOTE Confidence: 0.7735238

00:31:56.360 --> 00:31:58.944 OK so I'm just going to check my

NOTE Confidence: 0.7735238

00:31:58.944 --> 00:32:01.880 Clock to see how much time I'm doing.

NOTE Confidence: 0.7735238

00:32:01.880 --> 00:32:02.542 Well, OK,

NOTE Confidence: 0.7735238

00:32:02.542 --> 00:32:05.190 so I've shown you a lot of data

NOTE Confidence: 0.7735238

00:32:05.276 --> 00:32:07.817 where we deleted MDM two and an

NOTE Confidence: 0.7735238

00:32:07.817 --> 00:32:10.611 I didn't show you data for MDM 4

NOTE Confidence: 0.7735238

00:32:10.611 --> 00:32:12.928 but you you see these people three

NOTE Confidence: 0.7735238

00:32:12.928 --> 00:32:13.975 dependent physiological phenotypes

NOTE Confidence: 0.7735238

00:32:13.975 --> 00:32:16.020 and that's all fine and good.

NOTE Confidence: 0.7735238

00:32:16.020 --> 00:32:18.360 It showed us how important this

NOTE Confidence: 0.7735238

00:32:18.360 --> 00:32:20.820 relationship is between these proteins but.

NOTE Confidence: 0.7735238

00:32:20.820 --> 00:32:23.346 What happens in human cancers is

NOTE Confidence: 0.7735238

00:32:23.346 --> 00:32:25.383 you've got high expressions of

NOTE Confidence: 0.7735238

00:32:25.383 --> 00:32:27.343 MDM two and this is just yes,

NOTE Confidence: 0.7735238

00:32:27.350 --> 00:32:29.294 mean Valentina Vega in the lab

NOTE Confidence: 0.7735238

00:32:29.294 --> 00:32:31.597 a number of years ago looked at

NOTE Confidence: 0.7735238

00:32:31.597 --> 00:32:33.774 Indian 2 levels in head and neck

NOTE Confidence: 0.7735238

00:32:33.843 --> 00:32:36.038 squamous carcinomas and these are

NOTE Confidence: 0.7735238

00:32:36.038 --> 00:32:38.233 some of her beautiful pictures.

NOTE Confidence: 0.7735238

00:32:38.240 --> 00:32:39.179 So here's MDM,

NOTE Confidence: 0.7735238

00:32:39.179 --> 00:32:41.860 two expressed a very highly in a small

NOTE Confidence: 0.7735238

00:32:41.860 --> 00:32:44.779 region of this squamous cell carcinoma here.

NOTE Confidence: 0.7735238

00:32:44.780 --> 00:32:46.705 6 expressed almost across the

NOTE Confidence: 0.7735238

00:32:46.705 --> 00:32:49.071 entire tissue and then here is

NOTE Confidence: 0.7735238

00:32:49.071 --> 00:32:50.926 an interesting example of MDM.

NOTE Confidence: 0.7735238

00:32:50.930 --> 00:32:53.228 To be expressed in the cytoplasm,

NOTE Confidence: 0.7735238

00:32:53.230 --> 00:32:54.355 not the nucleus.

NOTE Confidence: 0.7735238

00:32:54.355 --> 00:32:56.230 So we really don't understand

NOTE Confidence: 0.7735238

00:32:56.230 --> 00:32:58.589 what it's doing in the cytoplasm,

NOTE Confidence: 0.7735238

00:32:58.590 --> 00:33:02.033 but not in all three of these experiments, P.

NOTE Confidence: 0.7735238

00:33:02.033 --> 00:33:03.948 53 is 1 type OK,

NOTE Confidence: 0.7735238

00:33:03.950 --> 00:33:06.866 so I think with this experiment

NOTE Confidence: 0.7735238

00:33:06.866 --> 00:33:08.810 in many others that

NOTE Confidence: 0.8437264

00:33:08.909 --> 00:33:13.180 people have done. Again.

NOTE Confidence: 0.8437264

00:33:13.180 --> 00:33:16.636 Again, show that what MDM two is doing in

NOTE Confidence: 0.8437264

00:33:16.636 --> 00:33:19.967 these tissues is inhibiting P53 activity.

NOTE Confidence: 0.8437264

00:33:19.970 --> 00:33:23.466 Now the I also don't want to leave

NOTE Confidence: 0.8437264

00:33:23.466 --> 00:33:26.876 you with the notion that MDM too.

NOTE Confidence: 0.8437264

00:33:26.880 --> 00:33:30.760 The P53 is the only MDM to target.

NOTE Confidence: 0.8437264

00:33:30.760 --> 00:33:32.708 Physiologically is the most

NOTE Confidence: 0.8437264

00:33:32.708 --> 00:33:35.630 relevant target because of the cell

NOTE Confidence: 0.8437264

00:33:35.717 --> 00:33:38.037 lethal phenotypes that we see,

NOTE Confidence: 0.8437264

00:33:38.040 --> 00:33:41.603 but in several experiments that my lab

NOTE Confidence: 0.8437264

00:33:41.603 --> 00:33:44.830 and Carol previous labs have done is,

NOTE Confidence: 0.8437264

00:33:44.830 --> 00:33:47.700 we've tried to overexpress MDM two in

NOTE Confidence: 0.8437264

00:33:47.700 --> 00:33:50.613 normal cells to understand what it's

NOTE Confidence: 0.8437264

00:33:50.613 --> 00:33:53.817 actually doing in with regards to

NOTE Confidence: 0.8437264

00:33:53.817 --> 00:33:55.979 transformation and tumor evolution.

NOTE Confidence: 0.8437264

00:33:55.980 --> 00:33:57.788 So here's what happens.

NOTE Confidence: 0.8437264

00:33:57.788 --> 00:34:01.501 So this is a normal control and the

NOTE Confidence: 0.8437264

00:34:01.501 --> 00:34:04.665 left these are mouse cells express a

NOTE Confidence: 0.8437264

00:34:04.665 --> 00:34:07.790 normal number of mouse chroma zones,

NOTE Confidence: 0.8437264

00:34:07.790 --> 00:34:11.042 and when we overexpressed MDM two

NOTE Confidence: 0.8437264

00:34:11.042 --> 00:34:14.180 we see this incredibly abnormal.

NOTE Confidence: 0.8437264

00:34:14.180 --> 00:34:16.655 Chromosome instability we can quantify

NOTE Confidence: 0.8437264

00:34:16.655 --> 00:34:19.558 the numbers of fusions here and

NOTE Confidence: 0.8437264

00:34:19.558 --> 00:34:21.987 we have a huge number of fusions.

NOTE Confidence: 0.8437264

00:34:21.990 --> 00:34:26.645 We also have a lot of fragments.

NOTE Confidence: 0.8437264

00:34:26.650 --> 00:34:29.140 So in data from multiple labs,

NOTE Confidence: 0.8437264

00:34:29.140 --> 00:34:31.954 if you overexpress MDM two in a

NOTE Confidence: 0.8437264

00:34:31.954 --> 00:34:34.538 normal cell the cell just dies.

NOTE Confidence: 0.8437264

00:34:34.540 --> 00:34:37.210 It can continue to grow.

NOTE Confidence: 0.8437264

00:34:37.210 --> 00:34:39.496 So and you know some experiments

NOTE Confidence: 0.8437264

00:34:39.496 --> 00:34:42.118 that are ongoing in the lab is OK,

NOTE Confidence: 0.8437264

00:34:42.120 --> 00:34:43.503 trying to understand.

NOTE Confidence: 0.8437264

00:34:43.503 --> 00:34:43.964 OK,

NOTE Confidence: 0.8437264

00:34:43.964 --> 00:34:46.730 so if we can't overexpress MDM

NOTE Confidence: 0.8437264

00:34:46.819 --> 00:34:48.549 two in a normal cell?

NOTE Confidence: 0.8437264

00:34:48.550 --> 00:34:51.105 Why do tumors have very high levels

NOTE Confidence: 0.8437264

00:34:51.105 --> 00:34:54.068 of ambient 2IN in one idea that we're

NOTE Confidence: 0.8437264

00:34:54.068 --> 00:34:56.765 working with is that there are some

NOTE Confidence: 0.8437264

00:34:56.765 --> 00:34:59.261 other rotation in those tumors that

NOTE Confidence: 0.8437264

00:34:59.261 --> 00:35:01.636 allows those tumors to survive with

NOTE Confidence: 0.8437264

00:35:01.636 --> 00:35:05.110 high levels of MDM two and so if we

NOTE Confidence: 0.8437264

00:35:05.110 --> 00:35:08.190 could understand what else MDM two is doing,

NOTE Confidence: 0.8437264

00:35:08.190 --> 00:35:12.474 we might be able to obtain a.

NOTE Confidence: 0.8437264

00:35:12.480 --> 00:35:16.188 A window of vulnerability to try to get the

NOTE Confidence: 0.8437264

00:35:16.188 --> 00:35:19.857 MDM two overexpressing cells to implode.

NOTE Confidence: 0.8437264

00:35:19.860 --> 00:35:22.878 But the screens that we're doing

NOTE Confidence: 0.8437264

00:35:22.878 --> 00:35:24.890 currently are are ongoing.

NOTE Confidence: 0.8437264

00:35:24.890 --> 00:35:28.768 OK then for the last few minutes

NOTE Confidence: 0.8437264

00:35:28.768 --> 00:35:31.758 of my lecture I want to.

NOTE Confidence: 0.8437264

00:35:31.760 --> 00:35:35.918 Move over to our understanding of P53

NOTE Confidence: 0.8437264

00:35:35.918 --> 00:35:39.270 mutations in breast cancer models.

NOTE Confidence: 0.8437264

00:35:39.270 --> 00:35:42.357 Didn't tell you earlier, but pizza degree.

NOTE Confidence: 0.8437264

00:35:42.360 --> 00:35:44.880 Why did tell you the people

NOTE Confidence: 0.8437264

00:35:44.880 --> 00:35:47.210 limitations for the most common,

NOTE Confidence: 0.8437264

00:35:47.210 --> 00:35:49.946 but really it speak into three

NOTE Confidence: 0.8437264

00:35:49.946 --> 00:35:52.672 missense mutations that are is the

NOTE Confidence: 0.8437264

00:35:52.672 --> 00:35:55.144 most common type of genetic lesion,

NOTE Confidence: 0.8437264

00:35:55.150 --> 00:35:59.119 and so my lab and that of Tyler left.

NOTE Confidence: 0.8437264

00:35:59.120 --> 00:36:02.326 Tyler Jacks is lab have made germline

NOTE Confidence: 0.8437264

00:36:02.326 --> 00:36:05.042 mutations in P53IN animal models and

NOTE Confidence: 0.8437264

00:36:05.042 --> 00:36:08.380 we show that these mice are tumor pro.

NOTE Confidence: 0.8437264

00:36:08.380 --> 00:36:11.250 But more importantly in contrast.

NOTE Confidence: 0.8437264

00:36:11.250 --> 00:36:12.408 Green or mice?

NOTE Confidence: 0.8437264

00:36:12.408 --> 00:36:15.110 These mice have a high metastatic capability,

NOTE Confidence: 0.8437264

00:36:15.110 --> 00:36:17.750 so this here is our data from the

NOTE Confidence: 0.8437264

00:36:17.750 --> 00:36:19.888 172 mutation corresponds to the

NOTE Confidence: 0.8437264

00:36:19.888 --> 00:36:21.289 origin 175 mutation,

NOTE Confidence: 0.8437264

00:36:21.290 --> 00:36:23.775 which is one of the hot spot

NOTE Confidence: 0.8437264

00:36:23.775 --> 00:36:25.530 mutations in human cancers.

NOTE Confidence: 0.8437264

00:36:25.530 --> 00:36:28.232 And here you can see a metastasis

NOTE Confidence: 0.8437264

00:36:28.232 --> 00:36:29.390 to the liver,

NOTE Confidence: 0.8437264

00:36:29.390 --> 00:36:32.085 and here stained with the P53 antibody,

NOTE Confidence: 0.8437264

00:36:32.090 --> 00:36:35.020 a metastasis to the brain.

NOTE Confidence: 0.8437264

00:36:35.020 --> 00:36:37.953 And this is in contrast to mice

NOTE Confidence: 0.8437264

00:36:37.953 --> 00:36:39.869 that have deletions of 353,

NOTE Confidence: 0.8437264

00:36:39.870 --> 00:36:42.705 so this really was the first example

NOTE Confidence: 0.8437264

00:36:42.705 --> 00:36:44.384 that suggested that expressing

NOTE Confidence: 0.8437264

00:36:44.384 --> 00:36:46.736 a mutant P53 was much more.

NOTE Confidence: 0.8372316

00:36:48.880 --> 00:36:50.872 Much more aggressive than not having

NOTE Confidence: 0.8372316

00:36:50.872 --> 00:36:53.259 people to create and and in the field.

NOTE Confidence: 0.8372316

00:36:53.260 --> 00:36:55.444 We call this a gain of function.

NOTE Confidence: 0.8372316

00:36:55.450 --> 00:36:57.962 Mutant P 53 is doing something in these

NOTE Confidence: 0.8372316

00:36:57.962 --> 00:37:00.500 cells to make them highly metastatic.

NOTE Confidence: 0.8372316

00:37:00.500 --> 00:37:03.636 So these are germline mice and what we

NOTE Confidence: 0.8372316

00:37:03.636 --> 00:37:06.966 wanted to do is to generate semantic

NOTE Confidence: 0.8372316

00:37:06.966 --> 00:37:10.018 models because the these germline models

NOTE Confidence: 0.8372316

00:37:10.018 --> 00:37:13.623 represent Lee from Many syndrome which is

NOTE Confidence: 0.8372316

00:37:13.623 --> 00:37:16.480 an inheritance of people to mutations.

NOTE Confidence: 0.8372316

00:37:16.480 --> 00:37:20.324 But that's a rare syndrome and we really

NOTE Confidence: 0.8372316

00:37:20.324 --> 00:37:22.894 wanted to understand this metastatic

NOTE Confidence: 0.8372316

00:37:22.894 --> 00:37:26.237 phenotype in a system where the the

NOTE Confidence: 0.8372316

00:37:26.237 --> 00:37:29.336 specific cell type has a Peach limitation

NOTE Confidence: 0.8372316

00:37:29.336 --> 00:37:31.400 and surrounding normal environment.

NOTE Confidence: 0.8372316

00:37:31.400 --> 00:37:33.680 To feel yourself to catch neutral

NOTE Confidence: 0.8372316

00:37:33.680 --> 00:37:36.612 goes to T cells are all wild type

NOTE Confidence: 0.8372316

00:37:36.612 --> 00:37:39.470 for P53 so that mouse did not exist.

NOTE Confidence: 0.8372316

00:37:39.470 --> 00:37:41.682 Tyler Jacks made a beautiful mouse that

NOTE Confidence: 0.8372316

00:37:41.682 --> 00:37:44.336 has been used extensively in the literature

NOTE Confidence: 0.8372316

00:37:44.336 --> 00:37:46.802 that basically is heterozygous for P53.

NOTE Confidence: 0.8372316

00:37:46.810 --> 00:37:49.826 So the entire mouse is missing 1P53 allele

NOTE Confidence: 0.8372316

00:37:49.826 --> 00:37:52.367 and Natalie all can be converted to a

NOTE Confidence: 0.8372316

00:37:52.367 --> 00:37:55.258 mutant P53 in a tissue specific fashion.

NOTE Confidence: 0.8372316

00:37:55.260 --> 00:37:58.837 So we didn't think that that was.

NOTE Confidence: 0.8372316

00:37:58.840 --> 00:38:01.138 Adequate enough to study the tumor,

NOTE Confidence: 0.8372316

00:38:01.140 --> 00:38:03.060 stroma tumor immune interactions because

NOTE Confidence: 0.8372316

00:38:03.060 --> 00:38:05.370 of heterozygosity of the P53 locus.

NOTE Confidence: 0.8372316

00:38:05.370 --> 00:38:08.234 So let me tell you a little bit

NOTE Confidence: 0.8372316

00:38:08.234 --> 00:38:11.567 about how we generate these mice and

NOTE Confidence: 0.8372316

00:38:11.567 --> 00:38:14.450 what our breast tumor phenotype is.

NOTE Confidence: 0.8372316

00:38:14.450 --> 00:38:17.738 OK, so here's how we generated these alleles.

NOTE Confidence: 0.8372316

00:38:17.740 --> 00:38:20.715 So we call these WM allele for

NOTE Confidence: 0.8372316

00:38:20.715 --> 00:38:23.078 wild type to mute P 53.

NOTE Confidence: 0.8372316

00:38:23.080 --> 00:38:25.432 So I'll show you in a minute this

NOTE Confidence: 0.8372316

00:38:25.432 --> 00:38:28.234 is a wild type allele normally and

NOTE Confidence: 0.8372316

00:38:28.234 --> 00:38:30.943 it's wild type because we earned

NOTE Confidence: 0.8372316

00:38:30.943 --> 00:38:33.423 started seeding a sequence upstream

NOTE Confidence: 0.8372316

00:38:33.423 --> 00:38:36.643 of the point mutation and this is

NOTE Confidence: 0.8372316

00:38:36.643 --> 00:38:38.698 this example is the Argentine,

NOTE Confidence: 0.8372316

00:38:38.700 --> 00:38:40.344 once it imitation which

NOTE Confidence: 0.8372316

00:38:40.344 --> 00:38:41.577 we generated previously.

NOTE Confidence: 0.8372316

00:38:41.580 --> 00:38:45.342 So what you have is a wild type P50.

NOTE Confidence: 0.8372316

00:38:45.350 --> 00:38:47.438 Three years of pollination site and

NOTE Confidence: 0.8372316

00:38:47.438 --> 00:38:49.611 then in a cream immediate fashion

NOTE Confidence: 0.8372316

00:38:49.611 --> 00:38:52.451 you can remove the wild type C DNA

NOTE Confidence: 0.8372316

00:38:52.521 --> 00:38:54.976 and basically reconstruct the locus.

NOTE Confidence: 0.8372316

00:38:54.980 --> 00:38:57.948 The arm and Cindy to Locust that we

NOTE Confidence: 0.8372316

00:38:57.948 --> 00:39:00.749 had studied forever in the germ line.

NOTE Confidence: 0.8372316

00:39:00.750 --> 00:39:03.646 With these mice and because it took a

NOTE Confidence: 0.8372316

00:39:03.646 --> 00:39:06.529 very long time to make these animals,

NOTE Confidence: 0.8372316

00:39:06.530 --> 00:39:09.085 we actually decided to make a second

NOTE Confidence: 0.8372316

00:39:09.085 --> 00:39:11.055 hotspot mutation and that's the

NOTE Confidence: 0.8372316

00:39:11.055 --> 00:39:13.070 Argentine 2.5 to double mutation,

NOTE Confidence: 0.8372316

00:39:13.070 --> 00:39:15.455 which corresponds to the 248

NOTE Confidence: 0.8372316

00:39:15.455 --> 00:39:16.409 hotspot mutation.

NOTE Confidence: 0.8372316

00:39:16.410 --> 00:39:18.530 Uhm?

NOTE Confidence: 0.8372316

00:39:18.530 --> 00:39:20.658 So this is just showing me wild type

NOTE Confidence: 0.8372316

00:39:20.658 --> 00:39:22.630 initially and then we committed fashion.

NOTE Confidence: 0.8372316

00:39:22.630 --> 00:39:25.134 You make them into protein and you can

NOTE Confidence: 0.8372316

00:39:25.134 --> 00:39:27.677 make it in akhri dependente manner.

NOTE Confidence: 0.8372316

00:39:27.680 --> 00:39:30.403 So this just shows you how normal

NOTE Confidence: 0.8372316

00:39:30.403 --> 00:39:31.570 those mice are.

NOTE Confidence: 0.8372316

00:39:31.570 --> 00:39:34.608 So here we're comparing wild type 2

NOTE Confidence: 0.8372316

00:39:34.608 --> 00:39:37.407 heterozygous mice with the 172 or the 2.5.

NOTE Confidence: 0.8372316

00:39:37.410 --> 00:39:39.355 We stabilized the mutant protein

NOTE Confidence: 0.8372316

00:39:39.355 --> 00:39:41.300 in response to DNA damage.

NOTE Confidence: 0.8372316

00:39:41.300 --> 00:39:44.037 And when we look at the activation

NOTE Confidence: 0.8372316

00:39:44.037 --> 00:39:47.128 of the three targets be 21 in Puma,

NOTE Confidence: 0.8372316

00:39:47.130 --> 00:39:51.108 they were activated to similar levels.

NOTE Confidence: 0.8372316

00:39:51.110 --> 00:39:54.365 We looked at the ability of these.

NOTE Confidence: 0.8372316

00:39:54.370 --> 00:39:56.757 Of DNA damage to induce labor ptosis?

NOTE Confidence: 0.8372316

00:39:56.760 --> 00:39:58.240 No difference between these

NOTE Confidence: 0.8372316

00:39:58.240 --> 00:40:00.460 two alleles in wild type mice.

NOTE Confidence: 0.8372316

00:40:00.460 --> 00:40:03.220 And again for the ability to rest the

NOTE Confidence: 0.8372316

00:40:03.220 --> 00:40:05.860 cycle in mouse embryo fibroblasts,

NOTE Confidence: 0.8372316

00:40:05.860 --> 00:40:07.030 there's no difference,

NOTE Confidence: 0.8372316

00:40:07.030 --> 00:40:08.980 so these mutant alleles really

NOTE Confidence: 0.8372316

00:40:08.980 --> 00:40:10.839 represent these condition alleles.

NOTE Confidence: 0.8372316

00:40:10.840 --> 00:40:12.910 That column you Tilly's really

NOTE Confidence: 0.8372316

00:40:12.910 --> 00:40:14.566 represent wild type allele.

NOTE Confidence: 0.749259

00:40:14.570 --> 00:40:17.790 They can become a moot.

NOTE Confidence: 0.749259

00:40:17.790 --> 00:40:20.184 And this is the last experiment I'll

NOTE Confidence: 0.749259

00:40:20.184 --> 00:40:22.828 show you about the the actual alleles.

NOTE Confidence: 0.749259

00:40:22.830 --> 00:40:24.840 What we've done is what we've

NOTE Confidence: 0.749259

00:40:24.840 --> 00:40:27.022 done here is compared the 172

NOTE Confidence: 0.749259

00:40:27.022 --> 00:40:28.947 heterozygous mice at 245 headers.

NOTE Confidence: 0.749259

00:40:28.950 --> 00:40:31.590 I guess mice and wild type mice to

NOTE Confidence: 0.749259

00:40:31.590 --> 00:40:33.976 each other over more than two years

NOTE Confidence: 0.749259

00:40:33.976 --> 00:40:36.510 to look at the tumor phenotypes.

NOTE Confidence: 0.749259

00:40:36.510 --> 00:40:37.890 Mice is the age.

NOTE Confidence: 0.749259

00:40:37.890 --> 00:40:39.615 Just like people will sporadically

NOTE Confidence: 0.749259

00:40:39.615 --> 00:40:42.198 get tumors, but what you can see is

NOTE Confidence: 0.749259

00:40:42.198 --> 00:40:43.878 that there's absolutely no statistical

NOTE Confidence: 0.749259

00:40:43.878 --> 00:40:46.228 difference between the three alleles.

NOTE Confidence: 0.749259

00:40:46.230 --> 00:40:48.160 So for all practical purposes.

NOTE Confidence: 0.749259

00:40:48.160 --> 00:40:50.794 This new allele that we generated

NOTE Confidence: 0.749259

00:40:50.794 --> 00:40:53.590 expresses a wild type P53 protein.

NOTE Confidence: 0.749259

00:40:53.590 --> 00:40:58.198 OK, so let me tell you bout two experiments.

NOTE Confidence: 0.749259

00:40:58.200 --> 00:41:01.974 One is our semantic breast semantic

NOTE Confidence: 0.749259

00:41:01.974 --> 00:41:06.463 model and what we did is we did

NOTE Confidence: 0.749259

00:41:06.463 --> 00:41:10.310 use the R172H and the 245 alleles.

NOTE Confidence: 0.749259

00:41:10.310 --> 00:41:13.526 We injected Adna virus Cree into the duct

NOTE Confidence: 0.749259

00:41:13.526 --> 00:41:16.930 of the mammary gland and so and then.

NOTE Confidence: 0.749259

00:41:16.930 --> 00:41:19.513 In addition, we use this TV tomato

NOTE Confidence: 0.749259

00:41:19.513 --> 00:41:22.319 allele which is also create dependent.

NOTE Confidence: 0.749259

00:41:22.320 --> 00:41:25.057 So when we inject adeno Korea not

NOTE Confidence: 0.749259

00:41:25.057 --> 00:41:28.108 show you a picture in a minute.

NOTE Confidence: 0.749259

00:41:28.110 --> 00:41:31.308 We basically make a mutant P

NOTE Confidence: 0.749259

00:41:31.308 --> 00:41:35.139 53 and we label the cell red.

NOTE Confidence: 0.749259

00:41:35.140 --> 00:41:37.988 So here is the.

NOTE Confidence: 0.749259

00:41:37.990 --> 00:41:40.852 Control experiments to show that we

NOTE Confidence: 0.749259

00:41:40.852 --> 00:41:43.693 do get recombination when we inject

NOTE Confidence: 0.749259

00:41:43.693 --> 00:41:46.231 the cream expressing adeno virus here

NOTE Confidence: 0.749259

00:41:46.231 --> 00:41:49.267 on the left is a low titer injection

NOTE Confidence: 0.749259

00:41:49.267 --> 00:41:52.368 and you can see the red cells here.

NOTE Confidence: 0.749259

00:41:52.368 --> 00:41:56.988 1 to 5% of the ductal cells become infected.

NOTE Confidence: 0.749259

00:41:56.990 --> 00:42:00.320 And then here on the right you can see.

NOTE Confidence: 0.749259

00:42:00.320 --> 00:42:02.770 A high titer virus was used was

NOTE Confidence: 0.749259

00:42:02.770 --> 00:42:05.290 injected into this gland in about 50 to

NOTE Confidence: 0.749259

00:42:05.290 --> 00:42:08.720 70% of the ductal cells are checked.

NOTE Confidence: 0.749259

00:42:08.720 --> 00:42:11.772 I also want to note that these

NOTE Confidence: 0.749259

00:42:11.772 --> 00:42:13.497 mice are 50% balzi.

NOTE Confidence: 0.749259

00:42:13.497 --> 00:42:16.416 Normally the P53 field is used to

NOTE Confidence: 0.749259

00:42:16.416 --> 00:42:20.908 see 57 black, 6 string to study.

NOTE Confidence: 0.749259

00:42:20.910 --> 00:42:22.854 Peabody create tumor phenotypes,

NOTE Confidence: 0.749259

00:42:22.854 --> 00:42:25.770 but that strain is resistant to

NOTE Confidence: 0.749259

00:42:25.846 --> 00:42:28.196 breast cancers for some reason,

NOTE Confidence: 0.749259

00:42:28.200 --> 00:42:30.996 and the beltsy component brings in

NOTE Confidence: 0.749259

00:42:30.996 --> 00:42:33.550 more sensitivity to breast cancers,

NOTE Confidence: 0.749259

00:42:33.550 --> 00:42:37.318 and we really don't know the

NOTE Confidence: 0.749259

00:42:37.318 --> 00:42:39.830 genetic reasons for this.

NOTE Confidence: 0.749259

00:42:39.830 --> 00:42:42.236 OK, so here is the data.

NOTE Confidence: 0.749259

00:42:42.240 --> 00:42:45.040 Let me go through it in detail.

NOTE Confidence: 0.749259

00:42:45.040 --> 00:42:47.602 So this is the 172 mutation expressed

NOTE Confidence: 0.749259

00:42:47.602 --> 00:42:50.259 only in a few mammary glands.

NOTE Confidence: 0.749259

00:42:50.260 --> 00:42:52.188 Memory cells, low titer.

NOTE Confidence: 0.749259

00:42:52.188 --> 00:42:54.598 We didn't see any tumors.

NOTE Confidence: 0.749259

00:42:54.600 --> 00:42:55.592 High titer,

NOTE Confidence: 0.749259

00:42:55.592 --> 00:42:58.568 we actually didn't see any tumors.

NOTE Confidence: 0.749259

00:42:58.570 --> 00:43:01.050 This one tumor showed up.

NOTE Confidence: 0.749259

00:43:01.050 --> 00:43:04.440 Post that to your end time

NOTE Confidence: 0.749259

00:43:04.440 --> 00:43:06.700 point of the experiment.

NOTE Confidence: 0.749259

00:43:06.700 --> 00:43:09.283 Because we weren't sure we were going

NOTE Confidence: 0.749259

00:43:09.283 --> 00:43:12.037 to get any tumors by just making

NOTE Confidence: 0.749259

00:43:12.037 --> 00:43:14.930 people scream Mutant in a few cells.

NOTE Confidence: 0.749259

00:43:14.930 --> 00:43:17.110 We also.

NOTE Confidence: 0.749259

00:43:17.110 --> 00:43:19.270 Irradiated with the sub lethal

NOTE Confidence: 0.749259

00:43:19.270 --> 00:43:20.566 dose of radiation.

NOTE Confidence: 0.749259

00:43:20.570 --> 00:43:23.587 This is not lethal to the mouse,

NOTE Confidence: 0.749259

00:43:23.590 --> 00:43:26.558 but it does cause damage and if the

NOTE Confidence: 0.749259

00:43:26.558 --> 00:43:30.005 damage is in the right or wrong genes

NOTE Confidence: 0.749259

00:43:30.005 --> 00:43:32.660 that contributes to tumor phenotype.

NOTE Confidence: 0.749259

00:43:32.660 --> 00:43:36.980 So with a low titer we begin to see tumors.

NOTE Confidence: 0.749259

00:43:36.980 --> 00:43:40.004 We sell one at a higher titer.

NOTE Confidence: 0.749259

00:43:40.010 --> 00:43:43.082 We now solve 4 tumors and

NOTE Confidence: 0.749259

00:43:43.082 --> 00:43:44.618 one tumor metastasized.

NOTE Confidence: 0.749259

00:43:44.620 --> 00:43:47.455 The 2.5 allele was a much stronger,

NOTE Confidence: 0.749259

00:43:47.460 --> 00:43:49.480 had a much stronger tumor,

NOTE Confidence: 0.749259

00:43:49.480 --> 00:43:51.100 phenotype with low titer.

NOTE Confidence: 0.749259

00:43:51.100 --> 00:43:54.799 We saw four tumors and one of them was

NOTE Confidence: 0.749259

00:43:54.799 --> 00:43:57.578 meta static with the high tier tighter.

NOTE Confidence: 0.7843864

00:43:57.580 --> 00:44:00.820 We saw nine tumors, so this is about

NOTE Confidence: 0.7843864

00:44:00.820 --> 00:44:04.058 75% and more than half were meta static.

NOTE Confidence: 0.7843864

00:44:04.060 --> 00:44:07.300 So let me kind of summarize all the data

NOTE Confidence: 0.7843864

00:44:07.300 --> 00:44:10.140 that we've done with these animals.

NOTE Confidence: 0.7843864

00:44:10.140 --> 00:44:12.564 So first let's just look at

NOTE Confidence: 0.7843864

00:44:12.564 --> 00:44:14.875 the tumor incidence. The R 172.

NOTE Confidence: 0.7843864

00:44:14.875 --> 00:44:17.430 Each allele all by itself in in.

NOTE Confidence: 0.7843864

00:44:17.430 --> 00:44:20.316 In these experiments, we only use

NOTE Confidence: 0.7843864

00:44:20.316 --> 00:44:23.699 one copy because we didn't want to.

NOTE Confidence: 0.7843864

00:44:23.700 --> 00:44:26.388 We didn't want to do LOH and

NOTE Confidence: 0.7843864

00:44:26.388 --> 00:44:27.900 we didn't want to.

NOTE Confidence: 0.7843864

00:44:27.900 --> 00:44:30.273 We just wanted to figure out what

NOTE Confidence: 0.7843864

00:44:30.273 --> 00:44:32.271 would happen with the minimal

NOTE Confidence: 0.7843864

00:44:32.271 --> 00:44:33.630 number of alterations.

NOTE Confidence: 0.7843864

00:44:33.630 --> 00:44:37.212 So if you compare the ones need two hitters

NOTE Confidence: 0.7843864

00:44:37.212 --> 00:44:40.129 Vegas to the 2.5 low and high titer,

NOTE Confidence: 0.7843864

00:44:40.130 --> 00:44:43.559 there's a huge number of tumors in the 248.

NOTE Confidence: 0.7843864

00:44:43.560 --> 00:44:46.616 Nice if we irradiated the ones only two,

NOTE Confidence: 0.7843864

00:44:46.620 --> 00:44:48.900 we got increased tumor incidence and

NOTE Confidence: 0.7843864

00:44:48.900 --> 00:44:51.355 then this is the experiment where

NOTE Confidence: 0.7843864

00:44:51.355 --> 00:44:53.935 we did mutate the other allele.

NOTE Confidence: 0.7843864

00:44:53.940 --> 00:44:56.598 So existing law supporters I got

NOTE Confidence: 0.7843864

00:44:56.598 --> 00:44:58.840 city and we can see.

NOTE Confidence: 0.7843864

00:44:58.840 --> 00:45:01.584 Increased tumor phenotype with the high dose.

NOTE Confidence: 0.7843864

00:45:01.590 --> 00:45:03.854 And this is a comparison on in the

NOTE Confidence: 0.7843864

00:45:03.854 --> 00:45:06.167 middle panel of the metastatic phenotype,

NOTE Confidence: 0.7843864

00:45:06.170 --> 00:45:08.185 and again the 245 documentation

NOTE Confidence: 0.7843864

00:45:08.185 --> 00:45:09.797 was the most metastatic.

NOTE Confidence: 0.7843864

00:45:09.800 --> 00:45:14.088 And then we looked at lots of header

NOTE Confidence: 0.7843864

00:45:14.088 --> 00:45:17.629 zygosity so the 245 mutation had.

NOTE Confidence: 0.7843864

00:45:17.630 --> 00:45:19.880 Variations in terms of LOH.

NOTE Confidence: 0.7843864

00:45:19.880 --> 00:45:22.967 About 50% of the mice showed LOH

NOTE Confidence: 0.7843864

00:45:22.967 --> 00:45:25.734 and then others retained some or

NOTE Confidence: 0.7843864

00:45:25.734 --> 00:45:27.974 all of the P3 alleles.

NOTE Confidence: 0.7843864

00:45:27.980 --> 00:45:30.680 The irradiated are once again 28.

NOTE Confidence: 0.7843864

00:45:30.680 --> 00:45:35.316 Showed 100%. LOH so to us.

NOTE Confidence: 0.7843864

00:45:35.316 --> 00:45:37.950 While we don't understand why we

NOTE Confidence: 0.7843864

00:45:38.055 --> 00:45:40.568 see 100% outrage with this allele,

NOTE Confidence: 0.7843864

00:45:40.568 --> 00:45:41.879 would it says?

NOTE Confidence: 0.7843864

00:45:41.880 --> 00:45:44.386 Is it that wild type allele is

NOTE Confidence: 0.7843864

00:45:44.386 --> 00:45:46.602 very strong at inhibiting tumors

NOTE Confidence: 0.7843864

00:45:46.602 --> 00:45:49.770 in this winsome need to background?

NOTE Confidence: 0.7825957

00:45:52.030 --> 00:45:54.697 These are the breast tumor subtypes that

NOTE Confidence: 0.7825957

00:45:54.697 --> 00:45:58.053 we saw the irradiated once in need two

NOTE Confidence: 0.7825957

00:45:58.053 --> 00:46:00.790 we saw mostly triple negative breast.

NOTE Confidence: 0.7825957

00:46:00.790 --> 00:46:02.875 There are 172 with the

NOTE Confidence: 0.7825957

00:46:02.875 --> 00:46:04.960 that lacks wild type P53.

NOTE Confidence: 0.7825957

00:46:04.960 --> 00:46:09.104 We sell mostly luminal B and then here

NOTE Confidence: 0.7825957

00:46:09.104 --> 00:46:13.558 with the 2.5 mutation we saw all three.

NOTE Confidence: 0.7825957

00:46:13.560 --> 00:46:16.815 Molecular subtypes to me this is fascinating

NOTE Confidence: 0.7825957

00:46:16.815 --> 00:46:19.500 experiment because we've made one mutation,

NOTE Confidence: 0.7825957

00:46:19.500 --> 00:46:23.840 we made a P53 mutation and yet here in this

NOTE Confidence: 0.7825957

00:46:23.941 --> 00:46:28.180 sample with the 2:45 we see off the tumor,

NOTE Confidence: 0.7825957

00:46:28.180 --> 00:46:31.484 molecular subtypes evolving and so one of

NOTE Confidence: 0.7825957

00:46:31.484 --> 00:46:34.592 the experiments that we're doing now is

NOTE Confidence: 0.7825957

00:46:34.592 --> 00:46:37.780 trying to understand with this 245 mutation,

NOTE Confidence: 0.7825957

00:46:37.780 --> 00:46:40.065 what are the triggers to

NOTE Confidence: 0.7825957

00:46:40.065 --> 00:46:41.436 these different subtypes?

NOTE Confidence: 0.7825957

00:46:41.440 --> 00:46:43.408 Triple negative breast cancer

NOTE Confidence: 0.7825957

00:46:43.408 --> 00:46:45.868 is very hard to treat.

NOTE Confidence: 0.7825957

00:46:45.870 --> 00:46:47.139 But for example,

NOTE Confidence: 0.7825957

00:46:47.139 --> 00:46:49.677 here to enrich tumors you can,

NOTE Confidence: 0.7825957

00:46:49.680 --> 00:46:52.634 you can treat with her two antibodies,

NOTE Confidence: 0.7825957

00:46:52.640 --> 00:46:54.765 so we're trying to understand

NOTE Confidence: 0.7825957

00:46:54.765 --> 00:46:56.465 basically the tumor evolution

NOTE Confidence: 0.7825957

00:46:56.465 --> 00:46:59.355 that initiates with this one, P.

NOTE Confidence: 0.7825957

00:46:59.355 --> 00:47:01.740 53 missense mutation.

NOTE Confidence: 0.7825957

00:47:01.740 --> 00:47:05.030 We've also wanted to so the data

NOTE Confidence: 0.7825957

00:47:05.030 --> 00:47:08.048 I just showed you says that

NOTE Confidence: 0.7825957

00:47:08.048 --> 00:47:11.048 the 248 mutation is much more.

NOTE Confidence: 0.7825957

00:47:11.050 --> 00:47:11.690 Dramatic.

NOTE Confidence: 0.7825957

00:47:11.690 --> 00:47:14.250 Then the 175 mutation.

NOTE Confidence: 0.7825957

00:47:14.250 --> 00:47:17.338 So what I showed here is a comparison

NOTE Confidence: 0.7825957

00:47:17.338 --> 00:47:19.929 of ovarian lung and breast tumors

NOTE Confidence: 0.7825957

00:47:19.929 --> 00:47:22.996 from people and just looking at the

NOTE Confidence: 0.7825957

00:47:22.996 --> 00:47:25.642 kind of mutation that they have and

NOTE Confidence: 0.7825957

00:47:25.642 --> 00:47:29.220 you can see in people that the 248

NOTE Confidence: 0.7825957

00:47:29.220 --> 00:47:32.740 mutation is has the worst outcomes.

NOTE Confidence: 0.7825957

00:47:32.740 --> 00:47:35.314 We couldn't do these data just

NOTE Confidence: 0.7825957

00:47:35.314 --> 00:47:37.922 for breast because the number of

NOTE Confidence: 0.7825957

00:47:37.922 --> 00:47:40.346 samples out there was not enough

NOTE Confidence: 0.7825957

00:47:40.346 --> 00:47:42.540 to give us significance.

NOTE Confidence: 0.7825957

00:47:42.540 --> 00:47:42.929 OK,

NOTE Confidence: 0.7825957

00:47:42.929 --> 00:47:45.652 so the one of the last experiments

NOTE Confidence: 0.7825957

00:47:45.652 --> 00:47:49.489 I'll show you here is just trying to

NOTE Confidence: 0.7825957

00:47:49.489 --> 00:47:51.469 understand tumor evolution because

NOTE Confidence: 0.7825957

00:47:51.553 --> 00:47:54.277 we made a semantic model that

NOTE Confidence: 0.7825957

00:47:54.277 --> 00:47:56.551 develop different kinds of breast

NOTE Confidence: 0.7825957

00:47:56.551 --> 00:47:58.806 cancers that were highly metastatic.

NOTE Confidence: 0.7825957

00:47:58.810 --> 00:48:01.378 And so I'm really interested in

NOTE Confidence: 0.7825957

00:48:01.378 --> 00:48:04.591 understanding the task sees in an in

NOTE Confidence: 0.7825957

00:48:04.591 --> 00:48:06.495 vivo physiologically relevant system.

NOTE Confidence: 0.7825957

00:48:06.500 --> 00:48:07.826 So we did.

NOTE Confidence: 0.7825957

00:48:07.826 --> 00:48:10.920 Here is we took the 2:45 mutant

NOTE Confidence: 0.7825957

00:48:11.032 --> 00:48:13.140 animals and we took.

NOTE Confidence: 0.7825957

00:48:13.140 --> 00:48:15.108 UH-22 memory tumors from these mice

NOTE Confidence: 0.7825957

00:48:15.108 --> 00:48:17.224 we sequenced them in three different

NOTE Confidence: 0.7825957

00:48:17.224 --> 00:48:19.069 regions trying to understand a

NOTE Confidence: 0.7825957

00:48:19.069 --> 00:48:21.299 little bit about the heterogeneity,

NOTE Confidence: 0.7825957

00:48:21.300 --> 00:48:23.525 and then we sequenced three

NOTE Confidence: 0.7825957

00:48:23.525 --> 00:48:26.380 metastases from each of these tumors.

NOTE Confidence: 0.7825957

00:48:26.380 --> 00:48:26.811 OK,

NOTE Confidence: 0.7825957

00:48:26.811 --> 00:48:29.397 and we sequence them to an

NOTE Confidence: 0.7825957

00:48:29.397 --> 00:48:30.690 incredible debt depth.

NOTE Confidence: 0.7825957

00:48:30.690 --> 00:48:34.338 So what we have here then is the

NOTE Confidence: 0.7825957

00:48:34.338 --> 00:48:37.295 comparison of the primary to the

NOTE Confidence: 0.7825957

00:48:37.295 --> 00:48:41.498 metastases and if we just look at mouse #4,

NOTE Confidence: 0.7825957

00:48:41.500 --> 00:48:45.056 there is some overlap in these these

NOTE Confidence: 0.7825957

00:48:45.056 --> 00:48:48.355 this overlap is considered an early

NOTE Confidence: 0.7825957

00:48:48.355 --> 00:48:52.254 gene signature and then you see this.

NOTE Confidence: 0.7825957

00:48:52.260 --> 00:48:54.440 Slate sequences that come up,

NOTE Confidence: 0.7825957

00:48:54.440 --> 00:48:55.745 which is how,

NOTE Confidence: 0.7825957

00:48:55.745 --> 00:48:57.050 which is what,

NOTE Confidence: 0.7825957

00:48:57.050 --> 00:48:59.486 where the metastases is now evolving

NOTE Confidence: 0.7825957

00:48:59.486 --> 00:49:02.699 when it gets into its metastatic site,

NOTE Confidence: 0.7825957

00:49:02.700 --> 00:49:06.424 which in this case was the lung.

NOTE Confidence: 0.7825957

00:49:06.430 --> 00:49:09.062 And so we can compare the early

NOTE Confidence: 0.7825957

00:49:09.062 --> 00:49:10.740 mutations of all three.

NOTE Confidence: 0.7825957

00:49:10.740 --> 00:49:13.092 The task season you see early

NOTE Confidence: 0.7825957

00:49:13.092 --> 00:49:14.660 mutations in both experiments,

NOTE Confidence: 0.7825957

00:49:14.660 --> 00:49:16.862 so these are the mutations that

NOTE Confidence: 0.7825957

00:49:16.862 --> 00:49:19.230 were acquired in the primary and

NOTE Confidence: 0.7825957

00:49:19.230 --> 00:49:21.714 metastatic lesion at the same time.

NOTE Confidence: 0.7825957

00:49:21.720 --> 00:49:24.457 But when you look at late mutations,

NOTE Confidence: 0.7825957

00:49:24.460 --> 00:49:27.330 here's all three mutations for mouse #4

NOTE Confidence: 0.7825957

00:49:27.330 --> 00:49:30.179 there's only one late mutation common.

NOTE Confidence: 0.7825957

00:49:30.180 --> 00:49:32.800 So with these sequencing data,

NOTE Confidence: 0.7825957

00:49:32.800 --> 00:49:35.120 indicate is that these matasa

NOTE Confidence: 0.7825957

00:49:35.120 --> 00:49:38.039 fees left the tumor very early

NOTE Confidence: 0.7825957

00:49:38.039 --> 00:49:40.574 during the metastatic process and

NOTE Confidence: 0.7825957

00:49:40.574 --> 00:49:42.602 then seated and had

NOTE Confidence: 0.7901794

00:49:42.695 --> 00:49:46.722 additional changes. So this was the

NOTE Confidence: 0.7901794

00:49:46.722 --> 00:49:49.712 first suggestion that maybe metastasis.

NOTE Confidence: 0.7901794

00:49:49.720 --> 00:49:51.632 Breast cancer metastasis driven

NOTE Confidence: 0.7901794

00:49:51.632 --> 00:49:55.749 by a new P53 is an early event.

NOTE Confidence: 0.7901794

00:49:55.750 --> 00:49:58.035 So to summarize, this model

NOTE Confidence: 0.7901794

00:49:58.035 --> 00:50:00.351 just briefly, we can make.

NOTE Confidence: 0.7901794

00:50:00.351 --> 00:50:04.303 P 3 point mutation in just a few

NOTE Confidence: 0.7901794

00:50:04.303 --> 00:50:07.717 cells that become a tumor that.

NOTE Confidence: 0.7901794

00:50:07.720 --> 00:50:09.502 Interessee migrate,

NOTE Confidence: 0.7901794

00:50:09.502 --> 00:50:13.957 proliferate and develop these metastases.

NOTE Confidence: 0.7901794

00:50:13.960 --> 00:50:16.312 Where we now have I called it a

NOTE Confidence: 0.7901794

00:50:16.312 --> 00:50:18.389 little factory but we just have

NOTE Confidence: 0.7901794

00:50:18.389 --> 00:50:20.164 these mice now developing tumors.

NOTE Confidence: 0.7901794

00:50:20.170 --> 00:50:22.459 We can isolate the circulating tumor cells,

NOTE Confidence: 0.7901794

00:50:22.460 --> 00:50:25.788 so we're trying to do is understand that

NOTE Confidence: 0.7901794

00:50:25.788 --> 00:50:28.620 the changes that occur for these cells

NOTE Confidence: 0.7901794

00:50:28.620 --> 00:50:31.658 to be able to survive in the blood.

NOTE Confidence: 0.7901794

00:50:31.660 --> 00:50:34.837 To home into an organ and then to develop.

NOTE Confidence: 0.7901794

00:50:34.840 --> 00:50:37.472 OK, if I have a few minutes which

NOTE Confidence: 0.7901794

00:50:37.472 --> 00:50:39.780 I have just a few minutes,

NOTE Confidence: 0.7901794

00:50:39.780 --> 00:50:42.104 I'm going to tell you about the

NOTE Confidence: 0.7901794

00:50:42.104 --> 00:50:44.608 other model that we made because we

NOTE Confidence: 0.7901794

00:50:44.608 --> 00:50:46.762 weren't sure that making a people

NOTE Confidence: 0.7901794

00:50:46.840 --> 00:50:49.311 communication in just a few cells was

NOTE Confidence: 0.7901794

00:50:49.311 --> 00:50:51.782 going to give us a tumor phenotype.

NOTE Confidence: 0.7901794

00:50:51.782 --> 00:50:53.900 So here we use K14 create,

NOTE Confidence: 0.7901794

00:50:53.900 --> 00:50:56.364 which expresses then a mutant P53 in all

NOTE Confidence: 0.7901794

00:50:56.364 --> 00:50:59.549 of the epithelial cells of the mammary gland,

NOTE Confidence: 0.7901794

00:50:59.550 --> 00:51:02.308 and this is a model that develops.

NOTE Confidence: 0.7901794

00:51:02.310 --> 00:51:03.582 Triple negative breast

NOTE Confidence: 0.7901794

00:51:03.582 --> 00:51:05.693 cancer 100% of the time.

NOTE Confidence: 0.7901794

00:51:05.693 --> 00:51:06.956 So again this.

NOTE Confidence: 0.7901794

00:51:06.960 --> 00:51:10.061 This suggests that you know South normal

NOTE Confidence: 0.7901794

00:51:10.061 --> 00:51:12.877 cell tumor cell interactions are altering

NOTE Confidence: 0.7901794

00:51:12.877 --> 00:51:16.146 the kinds of tumors that come up.

NOTE Confidence: 0.7901794

00:51:16.150 --> 00:51:19.102 And in this scenario we also had a

NOTE Confidence: 0.7901794

00:51:19.102 --> 00:51:22.720 cast 9 allele that is Creed dependent,

NOTE Confidence: 0.7901794

00:51:22.720 --> 00:51:26.131 so K14 CRV and allows us to make a

NOTE Confidence: 0.7901794

00:51:26.131 --> 00:51:29.270 mutant people degree in just a few

NOTE Confidence: 0.7901794

00:51:29.270 --> 00:51:32.083 in the epithelium of the mammary

NOTE Confidence: 0.7901794

00:51:32.083 --> 00:51:35.380 gland and to express castanon so we

NOTE Confidence: 0.7901794

00:51:35.380 --> 00:51:38.615 can use CRISPR technologies to to

NOTE Confidence: 0.7901794

00:51:38.615 --> 00:51:40.819 begin to address vulnerabilities.

NOTE Confidence: 0.7901794

00:51:40.820 --> 00:51:41.323 OK,

NOTE Confidence: 0.7901794

00:51:41.323 --> 00:51:44.341 so this vulnerability that we examine

NOTE Confidence: 0.7901794

00:51:44.341 --> 00:51:48.228 this in this model was whether these

NOTE Confidence: 0.7901794

00:51:48.228 --> 00:51:52.389 tumors were addicted to having immune to P53.

NOTE Confidence: 0.7901794

00:51:52.390 --> 00:51:52.905 OK,

NOTE Confidence: 0.7901794

00:51:52.905 --> 00:51:56.510 so here is the use that adnot

NOTE Confidence: 0.7901794

00:51:56.510 --> 00:51:58.700 associated virus that expresses

NOTE Confidence: 0.7901794

00:51:58.700 --> 00:52:02.382 a guide RNA that will delete P.

NOTE Confidence: 0.7901794

00:52:02.390 --> 00:52:02.951 53.

NOTE Confidence: 0.7901794

00:52:02.951 --> 00:52:06.317 So the tumor burden before treatment

NOTE Confidence: 0.7901794

00:52:06.317 --> 00:52:11.325 with a V in the control and the mute.

NOTE Confidence: 0.7901794

00:52:11.330 --> 00:52:12.634 Depleted tumors with similar,

NOTE Confidence: 0.7901794

00:52:12.634 --> 00:52:16.010 but you can see here in in the Purple line

NOTE Confidence: 0.7901794

00:52:16.010 --> 00:52:18.447 that those tumors that had depletion of

NOTE Confidence: 0.7901794

00:52:18.447 --> 00:52:20.813 you piece of degree survived much longer.

NOTE Confidence: 0.7901794

00:52:20.820 --> 00:52:24.220 This is just a picture of the tumor

NOTE Confidence: 0.7901794

00:52:24.220 --> 00:52:27.236 phenotypes that control you can see that.

NOTE Confidence: 0.7901794

00:52:27.240 --> 00:52:28.136 It's very.

NOTE Confidence: 0.7901794

00:52:28.136 --> 00:52:29.928 It's obviously a tumor.

NOTE Confidence: 0.7901794

00:52:29.930 --> 00:52:32.618 These mice die very quickly post

NOTE Confidence: 0.7901794

00:52:32.618 --> 00:52:34.410 identification of the tumors,

NOTE Confidence: 0.7901794

00:52:34.410 --> 00:52:38.258 and then you can see here with the

NOTE Confidence: 0.7901794

00:52:38.258 --> 00:52:40.853 depletion immunity theory this this

NOTE Confidence: 0.7901794

00:52:40.853 --> 00:52:43.937 this gland is looking more normal.

NOTE Confidence: 0.7901794

00:52:43.940 --> 00:52:44.304 OK,

NOTE Confidence: 0.7901794

00:52:44.304 --> 00:52:47.580 this is a whole bunch of data for the

NOTE Confidence: 0.7901794

00:52:47.676 --> 00:52:51.588 individual mice that this is a tumor volume.

NOTE Confidence: 0.7901794

00:52:51.590 --> 00:52:53.720 The controls and green here.

NOTE Confidence: 0.7901794

00:52:53.720 --> 00:52:55.840 Once we identify the tumor,

NOTE Confidence: 0.7901794

00:52:55.840 --> 00:52:58.360 they just keep growing the experimental

NOTE Confidence: 0.7901794

00:52:58.360 --> 00:53:01.005 cohort here in purple are animals

NOTE Confidence: 0.7901794

00:53:01.005 --> 00:53:03.240 that have recombined have basically

NOTE Confidence: 0.7901794

00:53:03.240 --> 00:53:05.827 deleted that mute people free protein

NOTE Confidence: 0.7901794

00:53:05.827 --> 00:53:08.167 in these mice live much longer.

NOTE Confidence: 0.7901794

00:53:08.170 --> 00:53:11.138 On the right is a tumor volume,

NOTE Confidence: 0.7901794

00:53:11.140 --> 00:53:14.458 so you can see that depletion of

NOTE Confidence: 0.7901794

00:53:14.458 --> 00:53:17.269 P53 affects the tumor volume.

NOTE Confidence: 0.7901794

00:53:17.270 --> 00:53:19.730 Greatly and then I'll just point

NOTE Confidence: 0.7901794

00:53:19.730 --> 00:53:22.210 out these two samples in Orange,

NOTE Confidence: 0.7901794

00:53:22.210 --> 00:53:24.320 which appeared not to respond

NOTE Confidence: 0.7901794

00:53:24.320 --> 00:53:26.430 to depletion of Mutant P

NOTE Confidence: 0.8528144

00:53:26.515 --> 00:53:28.810 53, and when we look at

NOTE Confidence: 0.8528144

00:53:28.810 --> 00:53:30.870 these two samples in detail,

NOTE Confidence: 0.8528144

00:53:30.870 --> 00:53:34.158 they did not express a stable mutant P.

NOTE Confidence: 0.8528144

00:53:34.160 --> 00:53:37.154 53, and so we think that

NOTE Confidence: 0.8528144

00:53:37.154 --> 00:53:40.260 these two tumors are actually.

NOTE Confidence: 0.8528144

00:53:40.260 --> 00:53:42.306 Since the P 53 isn't stable,

NOTE Confidence: 0.8528144

00:53:42.310 --> 00:53:44.058 the evolution that's occurring

NOTE Confidence: 0.8528144

00:53:44.058 --> 00:53:47.100 in these tumors is due to the

NOTE Confidence: 0.8528144

00:53:47.100 --> 00:53:49.116 absence of the P53 protein and

NOTE Confidence: 0.8528144

00:53:49.116 --> 00:53:51.528 not to a gain of function.

NOTE Confidence: 0.8528144

00:53:51.530 --> 00:53:53.861 OK, so I'm going to stop there

NOTE Confidence: 0.8528144

00:53:53.861 --> 00:53:56.535 and just I thrown a lot of data

NOTE Confidence: 0.8528144

00:53:56.535 --> 00:53:59.265 at you so I wanted to summarize

NOTE Confidence: 0.8528144

00:53:59.265 --> 00:54:02.439 briefly the first set of experiments

NOTE Confidence: 0.8528144

00:54:02.439 --> 00:54:04.296 really captured the exquisite

NOTE Confidence: 0.8528144

00:54:04.296 --> 00:54:06.286 sentence sensitivity of the MDM,

NOTE Confidence: 0.8528144

00:54:06.290 --> 00:54:08.290 two protein and P53 activity.

NOTE Confidence: 0.8528144

00:54:08.290 --> 00:54:11.578 We were able to.

NOTE Confidence: 0.8528144

00:54:11.580 --> 00:54:13.255 Identify this the molecular response

NOTE Confidence: 0.8528144

00:54:13.255 --> 00:54:15.394 to P53 activation in vivo identified

NOTE Confidence: 0.8528144

00:54:15.394 --> 00:54:17.848 numerous targets that are tissue specific.

NOTE Confidence: 0.8528144

00:54:17.850 --> 00:54:19.700 What are they all doing?

NOTE Confidence: 0.8528144

00:54:19.700 --> 00:54:22.164 I think we have our work cut out

NOTE Confidence: 0.8528144

00:54:22.164 --> 00:54:25.089 for us 'cause there's no way I can

NOTE Confidence: 0.8528144

00:54:25.089 --> 00:54:27.480 delete every one of these targets

NOTE Confidence: 0.8528144

00:54:27.480 --> 00:54:30.357 and see how important they are in

NOTE Confidence: 0.8528144

00:54:30.357 --> 00:54:32.978 vivo and then last but not least,

NOTE Confidence: 0.8528144

00:54:32.980 --> 00:54:35.500 I showed you the generation of of

NOTE Confidence: 0.8528144

00:54:35.500 --> 00:54:37.737 this novel mouse model that really

NOTE Confidence: 0.8528144

00:54:37.737 --> 00:54:40.257 allows us now to make a semantic

NOTE Confidence: 0.8528144

00:54:40.334 --> 00:54:41.918 point potential 53 in.

NOTE Confidence: 0.8528144

00:54:41.920 --> 00:54:45.000 Any cell of origin that we want

NOTE Confidence: 0.8528144

00:54:45.000 --> 00:54:48.637 to an in in the breast model.

NOTE Confidence: 0.8528144

00:54:48.640 --> 00:54:49.139 Specifically,

NOTE Confidence: 0.8528144

00:54:49.139 --> 00:54:52.632 we had a highly metastatic phenotype that

NOTE Confidence: 0.8528144

00:54:52.632 --> 00:54:55.839 we're trying to understand in more detail.

NOTE Confidence: 0.8528144

00:54:55.840 --> 00:54:56.291 OK,

NOTE Confidence: 0.8528144

00:54:56.291 --> 00:54:59.448 so my last slide is just the

NOTE Confidence: 0.8528144

00:54:59.448 --> 00:55:02.451 numerous people in the lab that

NOTE Confidence: 0.8528144

00:55:02.451 --> 00:55:04.956 have contributed to the studies.

NOTE Confidence: 0.8528144

00:55:04.960 --> 00:55:08.369 The Vinodh Pant did the feedback loop

NOTE Confidence: 0.8528144

00:55:08.369 --> 00:55:11.927 studies Roberto Navy MTM Tunicate Johnny DMD.

NOTE Confidence: 0.8528144

00:55:11.930 --> 00:55:12.810 Or not,

NOTE Confidence: 0.8528144

00:55:12.810 --> 00:55:15.450 God Tamera did all the studies

NOTE Confidence: 0.8528144

00:55:15.450 --> 00:55:18.120 with the radiation and with the

NOTE Confidence: 0.8528144

00:55:18.120 --> 00:55:21.590 node to look at the P3 response,

NOTE Confidence: 0.8528144

00:55:21.590 --> 00:55:24.200 Sidney generated the conditional mouse to

NOTE Confidence: 0.8528144

00:55:24.200 --> 00:55:27.568 look at the acute activation of people.

NOTE Confidence: 0.8528144

00:55:27.570 --> 00:55:30.090 Three targets you in terrific postdoc

NOTE Confidence: 0.8528144

00:55:30.090 --> 00:55:34.050 in the lab now has her own independent

NOTE Confidence: 0.8528144

00:55:34.050 --> 00:55:36.246 position generated that conditional

NOTE Confidence: 0.8528144

00:55:36.246 --> 00:55:39.772 mood P53 allele and Donata is the

NOTE Confidence: 0.8528144

00:55:39.772 --> 00:55:42.112 one who's studying the addiction.

NOTE Confidence: 0.8528144

00:55:42.120 --> 00:55:44.135 We're really wondering what the

NOTE Confidence: 0.8528144

00:55:44.135 --> 00:55:45.747 mechanisms is acquire these

NOTE Confidence: 0.8528144

00:55:45.747 --> 00:55:47.919 tumors imploding when they no

NOTE Confidence: 0.8528144

00:55:47.919 --> 00:55:49.627 longer have communities industry.

NOTE Confidence: 0.8528144

00:55:49.630 --> 00:55:52.742 So with that, I'll end in, oh,

NOTE Confidence: 0.8528144

00:55:52.742 --> 00:55:56.414 I'm glad to answer any questions.

NOTE Confidence: 0.8528144

00:55:56.420 --> 00:55:56.820 Thank

NOTE Confidence: 0.8138522

00:55:56.820 --> 00:55:58.011 you so much.

NOTE Confidence: 0.8138522

00:55:58.011 --> 00:55:59.996 That was a wonderful talk.

NOTE Confidence: 0.8138522

00:56:00.000 --> 00:56:02.835 I'm going to ask people to put

NOTE Confidence: 0.8138522

00:56:02.835 --> 00:56:05.179 questions in the in the chat,

NOTE Confidence: 0.8138522

00:56:05.180 --> 00:56:08.756 but I I wanted to ask you, sort of,

NOTE Confidence: 0.8138522

00:56:08.756 --> 00:56:10.746 from from the therapeutics perspective,

NOTE Confidence: 0.8138522

00:56:10.750 --> 00:56:13.246 people have been very interested in

NOTE Confidence: 0.8138522

00:56:13.246 --> 00:56:15.637 compounds like Prima and Cody that

NOTE Confidence: 0.8138522

00:56:15.637 --> 00:56:18.245 assist with re re folding of P53 with

NOTE Confidence: 0.8138522

00:56:18.317 --> 00:56:20.502 disruptive mutation and yet clinically

NOTE Confidence: 0.8138522

00:56:20.502 --> 00:56:25.380 those have been a little bit disappointing.

NOTE Confidence: 0.8138522

00:56:25.380 --> 00:56:29.060 Is is much known about whether or not

NOTE Confidence: 0.8138522

00:56:29.060 --> 00:56:31.667 those refolded P 53's are better,

NOTE Confidence: 0.8138522

00:56:31.670 --> 00:56:35.702 worse the same as as substrates for the MDM?

NOTE Confidence: 0.7994716

00:56:35.710 --> 00:56:37.950 Two MDM four? Yeah, so,

NOTE Confidence: 0.7994716

00:56:37.950 --> 00:56:41.532 so we've done a few studies using some of

NOTE Confidence: 0.7994716

00:56:41.532 --> 00:56:46.650 the drugs that are available, not many.

NOTE Confidence: 0.7994716

00:56:46.650 --> 00:56:49.240 My lab is focused on the genetics

NOTE Confidence: 0.7994716

00:56:49.240 --> 00:56:52.740 because if we take out him to an MP4 we

NOTE Confidence: 0.7994716

00:56:52.740 --> 00:56:55.470 see people three different phenotypes,

NOTE Confidence: 0.7994716

00:56:55.470 --> 00:56:57.876 but you know it's very different.

NOTE Confidence: 0.7994716

00:56:57.880 --> 00:56:59.388 Genetic told us mechanisms,

NOTE Confidence: 0.7994716

00:56:59.388 --> 00:57:02.690 but the drugs are really as you indicated.

NOTE Confidence: 0.7994716

00:57:02.690 --> 00:57:04.695 They're going to tell us

NOTE Confidence: 0.7994716

00:57:04.695 --> 00:57:06.700 whether they work or not.

NOTE Confidence: 0.7994716

00:57:06.700 --> 00:57:10.291 So I agree with you, I don't think

NOTE Confidence: 0.7994716

00:57:10.291 --> 00:57:13.453 that drugs are working very well.

NOTE Confidence: 0.7994716

00:57:13.460 --> 00:57:14.600 And you know,

NOTE Confidence: 0.7994716

00:57:14.600 --> 00:57:16.880 I don't know enough about those

NOTE Confidence: 0.7994716

00:57:16.880 --> 00:57:18.832 experiments to know you know how

NOTE Confidence: 0.7994716

00:57:18.832 --> 00:57:21.127 often you know how often the drug

NOTE Confidence: 0.7994716

00:57:21.127 --> 00:57:22.937 with the level of activation.

NOTE Confidence: 0.7994716

00:57:22.940 --> 00:57:25.908 I think that our.

NOTE Confidence: 0.7994716

00:57:25.910 --> 00:57:28.430 Our.

NOTE Confidence: 0.7994716

00:57:28.430 --> 00:57:31.398 That that that 7 gene signature that

NOTE Confidence: 0.7994716

00:57:31.398 --> 00:57:33.970 we identified would really help in

NOTE Confidence: 0.7994716

00:57:33.970 --> 00:57:36.418 those studies to try to understand

NOTE Confidence: 0.7994716

00:57:36.418 --> 00:57:39.030 what is the pika degree response?

NOTE Confidence: 0.7994716

00:57:39.030 --> 00:57:42.498 I also think that the people

NOTE Confidence: 0.7994716

00:57:42.498 --> 00:57:44.232 agree response required.

NOTE Confidence: 0.7994716

00:57:44.240 --> 00:57:46.230 Will vary in different tissues.

NOTE Confidence: 0.84930223

00:57:48.590 --> 00:57:51.371 We just know from our MDM two studies that

NOTE Confidence: 0.84930223

00:57:51.371 --> 00:57:53.845 some tissues are just much more sensitive

NOTE Confidence: 0.84930223

00:57:53.845 --> 00:57:56.690 to increase P 53 levels versus others.

NOTE Confidence: 0.84930223

00:57:56.690 --> 00:57:59.840 So I think that there's just a whole lot

NOTE Confidence: 0.84930223

00:57:59.840 --> 00:58:03.072 more work to be in to do in the clinic to

NOTE Confidence: 0.84930223

00:58:03.153 --> 00:58:06.184 be able to to understand that response.

NOTE Confidence: 0.8373146

00:58:07.290 --> 00:58:10.680 Yeah, yeah, I I think having a common set of.

NOTE Confidence: 0.8373146

00:58:10.680 --> 00:58:12.984 I mean I think people have just looked

NOTE Confidence: 0.8373146

00:58:12.984 --> 00:58:16.308 at P 21 over and over again and it's

NOTE Confidence: 0.8373146

00:58:16.308 --> 00:58:17.796 probably very inadequate, right?

NOTE Confidence: 0.8373146

00:58:17.796 --> 00:58:19.812 I see a couple questions and

NOTE Confidence: 0.8373146

00:58:19.812 --> 00:58:21.868 we have like a minute left.

NOTE Confidence: 0.8373146

00:58:21.870 --> 00:58:25.447 So first Jeff Townsend wants to know.

NOTE Confidence: 0.8373146

00:58:25.450 --> 00:58:27.640 Whether or not you've considered

NOTE Confidence: 0.8373146

00:58:27.640 --> 00:58:30.356 looking at sequencing of much larger

NOTE Confidence: 0.8373146

00:58:30.356 --> 00:58:33.068 cohorts of tumors and multi sample

NOTE Confidence: 0.8373146

00:58:33.068 --> 00:58:35.512 datasets to understand the temporal

NOTE Confidence: 0.8373146

00:58:35.512 --> 00:58:37.536 order of mutation appearances,

NOTE Confidence: 0.8373146

00:58:37.540 --> 00:58:38.938 that's exactly what

NOTE Confidence: 0.8160114

00:58:38.940 --> 00:58:42.603 we're doing right now I have a postdoc in

NOTE Confidence: 0.8160114

00:58:42.603 --> 00:58:46.519 a graduate student who just generated.

NOTE Confidence: 0.8160114

00:58:46.520 --> 00:58:48.904 A cohort of 100. Nice 'cause we want

NOTE Confidence: 0.8160114

00:58:48.904 --> 00:58:50.927 to understand the sequence of events.

NOTE Confidence: 0.8160114

00:58:50.930 --> 00:58:52.290 We want to understand the

NOTE Confidence: 0.8160114

00:58:52.290 --> 00:58:54.113 different events that lead to the

NOTE Confidence: 0.8160114

00:58:54.113 --> 00:58:55.337 different molecular subtypes.

NOTE Confidence: 0.8160114

00:58:55.340 --> 00:58:57.636 So we are in the midst of those

NOTE Confidence: 0.8160114

00:58:57.636 --> 00:58:59.750 experiments and we're going to do RNA

NOTE Confidence: 0.8160114

00:58:59.750 --> 00:59:01.999 seek and in DNA sequencing to understand

NOTE Confidence: 0.8160114

00:59:01.999 --> 00:59:04.153 both what happens if Arnie level.

NOTE Confidence: 0.8160114

00:59:04.160 --> 00:59:06.239 I think that it's critical we need

NOTE Confidence: 0.8160114

00:59:06.239 --> 00:59:07.482 to understand what's happening

NOTE Confidence: 0.8160114

00:59:07.482 --> 00:59:09.526 at the DNA level because I think

NOTE Confidence: 0.8160114

00:59:09.526 --> 00:59:11.921 that that's what gives rise to the

NOTE Confidence: 0.8160114

00:59:11.921 --> 00:59:12.974 different molecular subtypes.

NOTE Confidence: 0.8160114

00:59:12.980 --> 00:59:15.074 But I think it's the expression

NOTE Confidence: 0.8160114

00:59:15.074 --> 00:59:17.060 that's really going to tell us.

NOTE Confidence: 0.8160114

00:59:17.060 --> 00:59:19.880 What's happening to those cells once

NOTE Confidence: 0.8160114

00:59:19.880 --> 00:59:23.557 they reach home to deliver the line so?

NOTE Confidence: 0.8160114

00:59:23.560 --> 00:59:27.264 So we got all those are in progress.

NOTE Confidence: 0.7847016

00:59:28.410 --> 00:59:30.786 Then, Karen Anderson, who's my Co.

NOTE Confidence: 0.7847016

00:59:30.790 --> 00:59:32.770 Host for having invited you,

NOTE Confidence: 0.7847016

00:59:32.770 --> 00:59:34.978 wants to ask what your thoughts

NOTE Confidence: 0.7847016

00:59:34.978 --> 00:59:37.817 might be for a pro TEC directed

NOTE Confidence: 0.7847016

00:59:37.817 --> 00:59:40.686 against MDM two try to grade that

NOTE Confidence: 0.7847016

00:59:40.686 --> 00:59:42.670 as a therapeutic strategy. I

NOTE Confidence: 0.7847016

00:59:42.670 --> 00:59:44.645 think there's two Protex we

NOTE Confidence: 0.7847016

00:59:44.645 --> 00:59:46.225 should be thinking about.

NOTE Confidence: 0.7847016

00:59:46.230 --> 00:59:49.523 One is MDM 2. Although.

NOTE Confidence: 0.7847016

00:59:49.523 --> 00:59:52.974 Somehow I think for an MDM two

NOTE Confidence: 0.7847016

00:59:52.974 --> 00:59:55.957 inhibitor I think you've got to,

NOTE Confidence: 0.7847016

00:59:55.960 --> 00:59:57.935 you've got to target it

NOTE Confidence: 0.7847016

00:59:57.935 --> 01:00:00.470 to the to the tumor cell.

NOTE Confidence: 0.7847016

01:00:00.470 --> 01:00:02.690 Better just because of the

NOTE Confidence: 0.7847016

01:00:02.690 --> 01:00:04.910 hematopoetic toxic cities that have

NOTE Confidence: 0.7847016

01:00:04.982 --> 01:00:07.558 been seen with the MTM 2 inhibitors.

NOTE Confidence: 0.7847016

01:00:07.560 --> 01:00:09.990 But I also think we should

NOTE Confidence: 0.7847016

01:00:09.990 --> 01:00:11.610 start thinking about potentially

NOTE Confidence: 0.7847016

01:00:11.682 --> 01:00:13.398 doing degrading mutant P.

NOTE Confidence: 0.7847016

01:00:13.400 --> 01:00:16.417 53 Taking it out of the picture

NOTE Confidence: 0.7847016

01:00:16.417 --> 01:00:19.609 to see to see what happens.

NOTE Confidence: 0.7847016

01:00:19.610 --> 01:00:21.890 Particularly with those gain

NOTE Confidence: 0.7847016

01:00:21.890 --> 01:00:23.600 of function mutations,

NOTE Confidence: 0.7847016

01:00:23.600 --> 01:00:24.740 exactly exactly.

NOTE Confidence: 0.8569164

01:00:28.210 --> 01:00:30.520 We are over there more questions but

NOTE Confidence: 0.8569164

01:00:30.520 --> 01:00:33.657 we are over the time so I want to be

NOTE Confidence: 0.8569164

01:00:33.657 --> 01:00:36.200 respectful of very appreciative of of you,

NOTE Confidence: 0.8569164

01:00:36.200 --> 01:00:38.198 having joined us today and if

NOTE Confidence: 0.8569164

01:00:38.198 --> 01:00:40.530 anybody wants to ask me a question,

NOTE Confidence: 0.8569164

01:00:40.530 --> 01:00:43.104 email that they should feel free to I think

NOTE Confidence: 0.8569164

01:00:43.104 --> 01:00:45.190 allowed to answer additional questions.

NOTE Confidence: 0.8188419

01:00:47.520 --> 01:00:50.240 Super very much.