WEBVTT

NOTE duration:"00:47:22.1600000"

NOTE recognizability:0.788

NOTE language:en-us

NOTE Confidence: 0.759128495384615

00:00:00.000 --> 00:00:02.527 Friday, and thank you so much Harriet

NOTE Confidence: 0.759128495384615

 $00:00:02.527 \rightarrow 00:00:05.139$ and Marcus for your kind invitation.

NOTE Confidence: 0.759128495384615

 $00{:}00{:}05{.}140 \dashrightarrow 00{:}00{:}08{.}156$ I promise I'll go back in person and

NOTE Confidence: 0.759128495384615

 $00:00:08.156 \rightarrow 00:00:10.620$ hopefully with less technical issues.

NOTE Confidence: 0.759128495384615

00:00:10.620 --> 00:00:13.218 So as all of you know,

NOTE Confidence: 0.759128495384615

 $00:00:13.220 \longrightarrow 00:00:15.140$ probably even better than me,

NOTE Confidence: 0.759128495384615

 $00{:}00{:}15{.}140 \dashrightarrow 00{:}00{:}16{.}862$ Melanoma is the fifth most common

NOTE Confidence: 0.759128495384615

 $00:00:16.862 \dashrightarrow 00:00:18.859$ cancer in men and sixteen women.

NOTE Confidence: 0.759128495384615

00:00:18.860 --> 00:00:21.350 And can you please keep clicking?

NOTE Confidence: 0.759128495384615

 $00{:}00{:}21.350 \dashrightarrow 00{:}00{:}23.070$ I'm going to go fast through this first

NOTE Confidence: 0.759128495384615

00:00:23.070 --> 00:00:24.380 slides because it's just background.

NOTE Confidence: 0.759128495384615

 $00:00:24.380 \longrightarrow 00:00:26.036$ As you know, the incidence of

NOTE Confidence: 0.759128495384615

 $00{:}00{:}26.036 \dashrightarrow 00{:}00{:}27.140$ Melanoma continues to increase.

 $00:00:27.140 \longrightarrow 00:00:29.018$ It double s every 20 years

NOTE Confidence: 0.759128495384615

 $00:00:29.018 \longrightarrow 00:00:30.680$ and luckily this is like.

NOTE Confidence: 0.759128495384615

00:00:30.680 --> 00:00:31.218 Unfortunately,

NOTE Confidence: 0.759128495384615

 $00:00:31.218 \longrightarrow 00:00:34.446$ it's not a updated the mortality

NOTE Confidence: 0.759128495384615

 $00{:}00{:}34.446 \dashrightarrow 00{:}00{:}37.234$ associated with Melanoma is starting to

NOTE Confidence: 0.759128495384615

 $00:00:37.234 \longrightarrow 00:00:40.214$ decrease and this is you definitely to

NOTE Confidence: 0.759128495384615

 $00:00:40.214 \dashrightarrow 00:00:43.099$ the development of better the rapies.

NOTE Confidence: 0.759128495384615

 $00:00:43.100 \longrightarrow 00:00:45.680$ For the past 15 years,

NOTE Confidence: 0.759128495384615

 $00{:}00{:}45.680 \dashrightarrow 00{:}00{:}47.930$ Melanoma has become the poster

NOTE Confidence: 0.759128495384615

 $00:00:47.930 \rightarrow 00:00:50.180$ child for many targeted therapies

NOTE Confidence: 0.759128495384615

 $00:00:50.257 \rightarrow 00:00:52.437$ as well as immunotherapy space.

NOTE Confidence: 0.759128495384615

00:00:52.440 --> 00:00:53.649 Please keep clicking,

NOTE Confidence: 0.759128495384615

 $00:00:53.649 \longrightarrow 00:00:56.960$ but all of you know there is still

NOTE Confidence: 0.759128495384615

 $00:00:56.960 \rightarrow 00:01:00.570$ despite the success of this.

NOTE Confidence: 0.759128495384615

 $00{:}01{:}00{.}570 \dashrightarrow 00{:}01{:}01{.}170$ Uh,

NOTE Confidence: 0.759128495384615

 $00:01:01.170 \rightarrow 00:01:03.570$ we're developing immunotherapy treatments.

- NOTE Confidence: 0.759128495384615
- $00:01:03.570 \longrightarrow 00:01:05.730$ We still can keep clicking.
- NOTE Confidence: 0.759128495384615
- 00:01:05.730 --> 00:01:11.382 We are still facing significant resistance
- NOTE Confidence: 0.759128495384615
- $00{:}01{:}11{.}382 \dashrightarrow 00{:}01{:}17{.}870$ to some of these treatments or a.
- NOTE Confidence: 0.759128495384615
- $00{:}01{:}17.870 \dashrightarrow 00{:}01{:}20.330$ Year um side effects.
- NOTE Confidence: 0.759128495384615
- $00:01:20.330 \longrightarrow 00:01:22.910$ So approximately 40% of the
- NOTE Confidence: 0.759128495384615
- $00:01:22.910 \longrightarrow 00:01:24.370$ metastatic Melanoma patients have
- NOTE Confidence: 0.759128495384615
- $00{:}01{:}24.370 \dashrightarrow 00{:}01{:}26.189$ not benefited from the advances
- NOTE Confidence: 0.759128495384615
- $00:01:26.189 \longrightarrow 00:01:28.254$ that have been done over the past
- NOTE Confidence: 0.759128495384615
- $00:01:28.254 \rightarrow 00:01:30.419$ years in the treatment of Melanoma.
- NOTE Confidence: 0.759128495384615
- $00:01:30.420 \rightarrow 00:01:34.857$ So this takes me, can you please go on to my,
- NOTE Confidence: 0.759128495384615
- 00:01:34.857 00:01:37.479 the, the focus of my lab.
- NOTE Confidence: 0.759128495384615
- 00:01:37.480 --> 00:01:38.840 Can you go back?
- NOTE Confidence: 0.759128495384615
- 00:01:38.840 --> 00:01:39.180 Yeah,
- NOTE Confidence: 0.759128495384615
- $00{:}01{:}39{.}180 \dashrightarrow 00{:}01{:}43{.}220$ which evolves around these questions,
- NOTE Confidence: 0.759128495384615
- $00:01:43.220 \longrightarrow 00:01:45.320$ right, the fact that we still need
- NOTE Confidence: 0.759128495384615

 $00:01:45.320 \rightarrow 00:01:47.300$ better markers for patient selection,

NOTE Confidence: 0.759128495384615

 $00:01:47.300 \longrightarrow 00:01:47.603$ understand.

NOTE Confidence: 0.759128495384615

 $00:01:47.603 \rightarrow 00:01:49.724$ Where we check the patients that are

NOTE Confidence: 0.759128495384615

 $00:01:49.724 \rightarrow 00:01:52.170$ going to uh go on to recur after the

NOTE Confidence: 0.759128495384615

 $00:01:52.170 \dashrightarrow 00:01:53.630$ initial resection of their tumor,

NOTE Confidence: 0.759128495384615

 $00:01:53.630 \rightarrow 00:01:57.116$ which ones are going to metastasize.

NOTE Confidence: 0.759128495384615

00:01:57.120 --> 00:01:58.780 And also in particular,

NOTE Confidence: 0.759128495384615

 $00:01:58.780 \longrightarrow 00:02:00.025$ as Harriet mentioned,

NOTE Confidence: 0.759128495384615

 $00{:}02{:}00{.}030 \dashrightarrow 00{:}02{:}01{.}910$ we've developed an interest in

NOTE Confidence: 0.759128495384615

00:02:01.910 --> 00:02:04.510 understanding the biology of CNS metastases,

NOTE Confidence: 0.759128495384615

 $00:02:04.510 \longrightarrow 00:02:06.574$ particularly of brain metastasis

NOTE Confidence: 0.759128495384615

 $00:02:06.574 \rightarrow 00:02:09.670$ because they show less level responses

NOTE Confidence: 0.759128495384615

00:02:09.750 - 00:02:12.075 to immunotherapy and the incidence

NOTE Confidence: 0.759128495384615

 $00:02:12.075 \rightarrow 00:02:14.968$ is increasing as patients are living

NOTE Confidence: 0.759128495384615

 $00:02:14.968 \rightarrow 00:02:17.896$ longer and surviving from other systems.

NOTE Confidence: 0.759128495384615

 $00:02:17.900 \rightarrow 00:02:20.684$ Astasis, so my lab is focused

- NOTE Confidence: 0.759128495384615
- $00:02:20.684 \longrightarrow 00:02:22.540$ into aspects of metastasis,
- NOTE Confidence: 0.759128495384615
- $00:02:22.540 \longrightarrow 00:02:25.198$ the initial steps of early dissemination,
- NOTE Confidence: 0.759128495384615
- $00:02:25.200 \longrightarrow 00:02:27.750$ we now know that primary tumors,
- NOTE Confidence: 0.759128495384615
- $00:02:27.750 \longrightarrow 00:02:29.364$ especially in Melanoma.
- NOTE Confidence: 0.759128495384615
- 00:02:29.364 --> 00:02:30.978 Is that uh,
- NOTE Confidence: 0.759128495384615
- $00{:}02{:}30{.}980 \dashrightarrow 00{:}02{:}33{.}260$ disseminating start sharing cells that
- NOTE Confidence: 0.759128495384615
- $00:02:33.260 \rightarrow 00:02:35.540$ go into circulation and metastasize
- NOTE Confidence: 0.759128495384615
- $00:02:35.607 \rightarrow 00:02:37.515$ really early on in the process.
- NOTE Confidence: 0.759128495384615
- $00:02:37.520 \longrightarrow 00:02:38.960$ So we want to understand,
- NOTE Confidence: 0.759128495384615
- 00:02:38.960 --> 00:02:40.409 please click uh,
- NOTE Confidence: 0.759128495384615
- $00{:}02{:}40{.}409 \dashrightarrow 00{:}02{:}42{.}824$ which are the mechanisms that
- NOTE Confidence: 0.759128495384615
- $00{:}02{:}42.824 \dashrightarrow 00{:}02{:}45.695$ drive the metastatic behavior of
- NOTE Confidence: 0.759128495384615
- $00{:}02{:}45.695 \dashrightarrow 00{:}02{:}48.715$ certain melanomas and not others.
- NOTE Confidence: 0.759128495384615
- $00{:}02{:}48.720 \dashrightarrow 00{:}02{:}50.705$ We know that unity alterations
- NOTE Confidence: 0.759128495384615
- $00:02:50.705 \rightarrow 00:02:52.690$ don't explain this behavior because
- NOTE Confidence: 0.759128495384615

 $00:02:52.751 \rightarrow 00:02:54.706$ none of the well characterized

NOTE Confidence: 0.759128495384615

00:02:54.706 --> 00:02:56.270 genetic alterations of Melanoma

NOTE Confidence: 0.759128495384615

 $00{:}02{:}56{.}270 \dashrightarrow 00{:}02{:}58{.}465$ or combinations of those genetic

NOTE Confidence: 0.759128495384615

00:02:58.465 - 00:03:00.209 alterations can explain why.

NOTE Confidence: 0.759128495384615

00:03:00.210 --> 00:03:02.190 And primary tumors recurred,

NOTE Confidence: 0.759128495384615

 $00:03:02.190 \dashrightarrow 00:03:04.665$ metastasized and some others don't.

NOTE Confidence: 0.759128495384615

 $00{:}03{:}04.670 \dashrightarrow 00{:}03{:}06.800$ And also we want to understand

NOTE Confidence: 0.759128495384615

 $00{:}03{:}06{.}800 \dashrightarrow 00{:}03{:}08{.}936$ what is the contribution of

NOTE Confidence: 0.759128495384615

00:03:08.936 --> 00:03:10.228 intratumoral heterogeneity.

NOTE Confidence: 0.759128495384615

 $00{:}03{:}10.230 \dashrightarrow 00{:}03{:}12.624$ So we know now that despite the

NOTE Confidence: 0.759128495384615

 $00{:}03{:}12.624 \dashrightarrow 00{:}03{:}15.172$ fact that primary tumors can have

NOTE Confidence: 0.759128495384615

 $00:03:15.172 \rightarrow 00:03:17.100$ a relatively homogeneous genetic

NOTE Confidence: 0.759128495384615

 $00:03:17.100 \rightarrow 00:03:19.110$ profile inside those tumors,

NOTE Confidence: 0.759128495384615

 $00:03:19.110 \rightarrow 00:03:22.120$ we can recognize groups of cells with

NOTE Confidence: 0.759128495384615

 $00:03:22.120 \rightarrow 00:03:23.410$ different transcriptional programs,

NOTE Confidence: 0.759128495384615

 $00:03:23.410 \rightarrow 00:03:25.290$ what we call transcriptional states.

- NOTE Confidence: 0.759128495384615
- $00:03:25.290 \rightarrow 00:03:27.762$ And there is a recent search for what
- NOTE Confidence: 0.759128495384615
- $00{:}03{:}27.762 \dashrightarrow 00{:}03{:}30.240$ are the programs for their states.
- NOTE Confidence: 0.759128495384615
- $00{:}03{:}30{.}240 \dashrightarrow 00{:}03{:}32{.}130$ That drive these metastatic behavior
- NOTE Confidence: 0.759128495384615
- $00:03:32.130 \dashrightarrow 00:03:34.644$ and people have pointed to the neural
- NOTE Confidence: 0.759128495384615
- $00{:}03{:}34{.}644 \dashrightarrow 00{:}03{:}36{.}900$ Crest like or the EMT program on the
- NOTE Confidence: 0.8062973056
- $00:03:36.960 \longrightarrow 00:03:38.580$ other side of the equation.
- NOTE Confidence: 0.8062973056
- $00:03:38.580 \rightarrow 00:03:40.332$ We are also trying to understand
- NOTE Confidence: 0.8062973056
- $00:03:40.332 \longrightarrow 00:03:42.418$ what happens at the end of the
- NOTE Confidence: 0.8062973056
- $00:03:42.418 \longrightarrow 00:03:43.853$ process once the cancer cells,
- NOTE Confidence: 0.8062973056
- $00:03:43.860 \rightarrow 00:03:46.226$ in this case Melanoma cells have already
- NOTE Confidence: 0.8062973056
- $00{:}03{:}46.226 \dashrightarrow 00{:}03{:}48.039$ extravasated into those distal organs.
- NOTE Confidence: 0.8062973056
- $00:03:48.040 \dashrightarrow 00:03:50.231$ As you know Melanoma metastasis to the NOTE Confidence: 0.8062973056
- $00:03:50.231 \rightarrow 00:03:52.827$ lung delivered in the brain and we are
- NOTE Confidence: 0.8062973056
- 00:03:52.827 --> 00:03:54.840 interested in understanding what are the NOTE Confidence: 0.8062973056
- $00:03:54.840 \rightarrow 00:03:57.000$ site specific adaptations in those sites,
- NOTE Confidence: 0.8062973056

 $00:03:57.000 \rightarrow 00:04:00.829$ where are the type of metabolic changes.

NOTE Confidence: 0.8062973056

 $00:04:00.830 \longrightarrow 00:04:02.882$ The cell types of the cancer

NOTE Confidence: 0.8062973056

 $00:04:02.882 \longrightarrow 00:04:03.908$ cells are interacting.

NOTE Confidence: 0.8062973056

 $00{:}04{:}03{.}910 \dashrightarrow 00{:}04{:}05{.}625$ And what is this crosstalk between the

NOTE Confidence: 0.8062973056

 $00:04:05.625 \rightarrow 00:04:07.209$ cancer cells and their environment?

NOTE Confidence: 0.8062973056

00:04:07.210 --> 00:04:09.426 How is shaping the ability of animal cells NOTE Confidence: 0.8062973056

 $00:04:09.426 \rightarrow 00:04:11.727$ to adapt and grow in those environments?

NOTE Confidence: 0.8062973056

 $00{:}04{:}11.730 \dashrightarrow 00{:}04{:}14.550$ And I will have to stories

NOTE Confidence: 0.8062973056

 $00:04:14.550 \rightarrow 00:04:17.270$ presenting today if time permits,

NOTE Confidence: 0.8062973056

 $00:04:17.270 \longrightarrow 00:04:19.622$ one that focuses more in just the

NOTE Confidence: 0.8062973056

 $00{:}04{:}19.622 \dashrightarrow 00{:}04{:}21.609$ metastatic potential in general and how

NOTE Confidence: 0.8062973056

 $00{:}04{:}21{.}609 \dashrightarrow 00{:}04{:}24{.}147$ muscles cops some of the programs of the

NOTE Confidence: 0.8062973056

 $00:04:24.147 \dashrightarrow 00:04:26.464$ neural Crest cells from which they arise.

NOTE Confidence: 0.8062973056

00:04:26.470 --> 00:04:29.530 And also in the second part of the talk,

NOTE Confidence: 0.8062973056

 $00:04:29.530 \rightarrow 00:04:30.840$ I will talk more specifically.

NOTE Confidence: 0.8062973056

 $00:04:30.840 \rightarrow 00:04:34.774$ About to bring metastasis and how Melanoma

- NOTE Confidence: 0.8062973056
- $00{:}04{:}34{.}774 \dashrightarrow 00{:}04{:}37{.}714$ cells mimic some of the processes
- NOTE Confidence: 0.8062973056
- $00{:}04{:}37{.}714 \dashrightarrow 00{:}04{:}40{.}050$ that happen during neurodegeneration,
- NOTE Confidence: 0.8062973056
- $00:04:40.050 \rightarrow 00:04:41.860$ particularly to suppress new inflammation
- NOTE Confidence: 0.8062973056
- $00:04:41.860 \longrightarrow 00:04:44.739$ and be able to grow within the brain.
- NOTE Confidence: 0.8062973056
- $00:04:44.740 \longrightarrow 00:04:47.218$ So diving into the first story,
- NOTE Confidence: 0.8062973056
- $00{:}04{:}47{.}220 \dashrightarrow 00{:}04{:}49{.}460$ these are the four areas in which my
- NOTE Confidence: 0.8062973056
- $00:04:49.460 \dashrightarrow 00:04:51.940$ lab has been focused in recent years,
- NOTE Confidence: 0.8062973056
- $00:04:51.940 \longrightarrow 00:04:53.581$ epigenetic alterations, noncollinear,
- NOTE Confidence: 0.8062973056
- $00{:}04{:}53.581 \dashrightarrow 00{:}04{:}55.769$ a post translational modifications
- NOTE Confidence: 0.8062973056
- 00:04:55.769 --> 00:04:57.410 particularly like oscillation
- NOTE Confidence: 0.8062973056
- $00:04:57.469 \longrightarrow 00:04:59.077$ and site specific adaptations.
- NOTE Confidence: 0.8062973056
- 00:04:59.080 --> 00:05:00.514 So moving on,
- NOTE Confidence: 0.8062973056
- $00:05:00.514 \rightarrow 00:05:04.160$ can you please move these slides one more,
- NOTE Confidence: 0.8062973056
- $00{:}05{:}04{.}160 \dashrightarrow 00{:}05{:}07{.}292$ the first story that has been
- NOTE Confidence: 0.8062973056
- $00:05:07.292 \dashrightarrow 00:05:09.160$ recently accepted for publication
- NOTE Confidence: 0.8062973056

 $00:05:09.160 \longrightarrow 00:05:11.620$ is the work of three people.

NOTE Confidence: 0.8062973056

 $00:05:11.620 \longrightarrow 00:05:14.868$ It evolve over time and we have.

NOTE Confidence: 0.8062973056

00:05:14.870 --> 00:05:15.223 Uh,

NOTE Confidence: 0.8062973056

 $00{:}05{:}15{.}223 \dashrightarrow 00{:}05{:}17{.}694$ tightly muscles invoke and you like clear

NOTE Confidence: 0.8062973056

00:05:17.694 --> 00:05:19.990 sticky genetic program during metastasis.

NOTE Confidence: 0.8062973056

 $00{:}05{:}19{.}990$ --> $00{:}05{:}24{.}058$ So the genesis for this project was a very NOTE Confidence: 0.8062973056

 $00:05:24.058 \rightarrow 00:05:26.328$ simple premise and it's the fact that,

NOTE Confidence: 0.8062973056

 $00:05:26.330 \longrightarrow 00:05:26.580$ well,

NOTE Confidence: 0.8062973056

 $00{:}05{:}26.580 \dashrightarrow 00{:}05{:}28.580$ we all know that neural Crest cells are

NOTE Confidence: 0.8062973056

 $00:05:28.580 \rightarrow 00:05:30.466$ the cells of origin of melanocytes.

NOTE Confidence: 0.8062973056

 $00{:}05{:}30{.}470 \dashrightarrow 00{:}05{:}33{.}333$ And these are among the most to tipotent

NOTE Confidence: 0.8062973056

 $00{:}05{:}33{.}333 \dashrightarrow 00{:}05{:}36{.}247$ and among the most invasive and migratory

NOTE Confidence: 0.8062973056

 $00:05:36.247 \rightarrow 00:05:39.511$ cells of our body and is well known

NOTE Confidence: 0.8062973056

 $00:05:39.511 \rightarrow 00:05:42.490$ now if you keep clicking that Melanoma

NOTE Confidence: 0.8062973056

 $00:05:42.490 \rightarrow 00:05:45.130$ cells adopt programs characteristic.

NOTE Confidence: 0.8062973056

 $00:05:45.130 \rightarrow 00:05:47.538$ You address the cells during the progression

 $00:05:47.538 \rightarrow 00:05:49.670$ from primary to metastatic cancer.

NOTE Confidence: 0.8062973056

 $00:05:49.670 \rightarrow 00:05:52.028$ So we wanted to investigate which

NOTE Confidence: 0.8062973056

00:05:52.028 --> 00:05:53.600 epigenetic changes happen during

NOTE Confidence: 0.8062973056

00:05:53.663 --> 00:05:55.355 melanocyte differentiation from the NOTE Confidence: 0.8062973056

00:05:55.355 --> 00:05:57.893 neural Crest to the melanocyte that NOTE Confidence: 0.8062973056

 $00{:}05{:}57{.}955 \dashrightarrow 00{:}06{:}00{.}205$ potentially could be reversed in the

NOTE Confidence: 0.8062973056

 $00:06:00.205 \rightarrow 00:06:02.116$ transition from primary to metastasis.

NOTE Confidence: 0.8062973056

 $00:06:02.116 \longrightarrow 00:06:04.552$ And we did this by looking

NOTE Confidence: 0.8062973056

 $00:06:04.552 \rightarrow 00:06:06.690$ specifically at DNA methylation.

NOTE Confidence: 0.8062973056

 $00:06:06.690 \longrightarrow 00:06:08.838$ There are of course many other

NOTE Confidence: 0.8062973056

 $00:06:08.838 \dashrightarrow 00:06:10.270$ mechanisms of epigenetic regulation

NOTE Confidence: 0.8062973056

 $00{:}06{:}10.325 \dashrightarrow 00{:}06{:}12.167$ being the enumeration one of them.

NOTE Confidence: 0.8062973056

 $00:06:12.170 \longrightarrow 00:06:15.514$ So if you move to the next slide.

NOTE Confidence: 0.8062973056

 $00:06:15.520 \longrightarrow 00:06:17.466$ This is what we did basically was

NOTE Confidence: 0.8062973056

 $00{:}06{:}17.466 \dashrightarrow 00{:}06{:}19.218$ a comparison of four data sets.

00:06:19.220 --> 00:06:22.379 So in one hand I cannot use my pointer,

NOTE Confidence: 0.8062973056

 $00{:}06{:}22{.}380 \dashrightarrow 00{:}06{:}24{.}738$ but you can see that we have 4 columns.

NOTE Confidence: 0.8062973056

 $00{:}06{:}24.740 \dashrightarrow 00{:}06{:}27.008$ We have neural Crest cells that were

NOTE Confidence: 0.8062973056

 $00:06:27.008 \rightarrow 00:06:29.480$ obtained from a collaborator in France.

NOTE Confidence: 0.8062973056

 $00:06:29.480 \longrightarrow 00:06:30.908$ These are human cells.

NOTE Confidence: 0.8062973056

 $00{:}06{:}30{.}908 \dashrightarrow 00{:}06{:}33{.}050$ We also had the human melanocytes

NOTE Confidence: 0.8062973056

00:06:33.116 --> 00:06:34.520 from different donors.

NOTE Confidence: 0.8062973056

 $00:06:34.520 \longrightarrow 00:06:36.144$ And as you can see there is

NOTE Confidence: 0.8062973056

 $00:06:36.144 \longrightarrow 00:06:38.039$ kind of a mirroring pattern.

NOTE Confidence: 0.8062973056

 $00{:}06{:}38{.}040 \dashrightarrow 00{:}06{:}41{.}162$ We were looking for CPG islands that

NOTE Confidence: 0.8062973056

 $00:06:41.162 \rightarrow 00:06:43.424$ were either hypomethylated in the

NOTE Confidence: 0.8062973056

 $00:06:43.424 \rightarrow 00:06:45.549$ conversion from neural Crest cells.

NOTE Confidence: 0.8062973056

 $00:06:45.550 \rightarrow 00:06:48.232$ To Milano sites that were progressively

NOTE Confidence: 0.8062973056

 $00:06:48.232 \rightarrow 00:06:50.020$ hypermethylated from primary to

NOTE Confidence: 0.769571231333333

 $00:06:50.083 \rightarrow 00:06:52.348$ metastatic Melanoma or the converse,

NOTE Confidence: 0.769571231333333

 $00:06:52.350 \longrightarrow 00:06:53.706$ if you see at the bottom,

 $00:06:53.710 \longrightarrow 00:06:56.700$ we have some CPG islands.

NOTE Confidence: 0.769571231333333

 $00:06:56.700 \longrightarrow 00:06:58.210$ If you can click again,

NOTE Confidence: 0.769571231333333

00:06:58.210 - > 00:07:00.352 you will see there are some CPG

NOTE Confidence: 0.769571231333333

 $00:07:00.352 \rightarrow 00:07:02.349$ islands around the gene called NR

NOTE Confidence: 0.769571231333333

 $00{:}07{:}02{.}349 \dashrightarrow 00{:}07{:}04{.}371$ 22 that are hypomethylated in the

NOTE Confidence: 0.769571231333333

 $00{:}07{:}04.371 \dashrightarrow 00{:}07{:}06.905$ neural grass cells that become

NOTE Confidence: 0.769571231333333

 $00:07:06.905 \rightarrow 00:07:09.037$ hypermethylated in melanocytes and

NOTE Confidence: 0.769571231333333

 $00:07:09.037 \rightarrow 00:07:10.562$ progressively hypomethylated from

NOTE Confidence: 0.769571231333333

00:07:10.562 --> 00:07:12.370 primary to metastatic Melanoma.

NOTE Confidence: 0.769571231333333

 $00:07:12.370 \longrightarrow 00:07:14.602$ And this really call our attention

NOTE Confidence: 0.769571231333333

00:07:14.602 --> 00:07:16.090 because it really represented.

NOTE Confidence: 0.769571231333333

00:07:16.090 --> 00:07:19.205 Example of a potential gene or or

NOTE Confidence: 0.769571231333333

 $00{:}07{:}19.205 \dashrightarrow 00{:}07{:}21.820$ candidate program to be modulated

NOTE Confidence: 0.769571231333333

 $00{:}07{:}21.820 \dashrightarrow 00{:}07{:}24.196$ during neural Crest differentiation

NOTE Confidence: 0.769571231333333

 $00{:}07{:}24.196 \dashrightarrow 00{:}07{:}27.320$ to melanocytes that was reversed

 $00:07:27.320 \rightarrow 00:07:29.260$ during the progression from primary

NOTE Confidence: 0.769571231333333

 $00{:}07{:}29{.}260 \dashrightarrow 00{:}07{:}31{.}200$ to metastatic Melanoma and what

NOTE Confidence: 0.769571231333333

00:07:31.260 --> 00:07:32.940 is first of all an R2 of two,

NOTE Confidence: 0.769571231333333

 $00:07:32.940 \longrightarrow 00:07:35.380$ so it's a nuclear receptor

NOTE Confidence: 0.769571231333333

 $00{:}07{:}35{.}380 \dashrightarrow 00{:}07{:}36{.}796$ is also called cooked.

NOTE Confidence: 0.769571231333333

 $00{:}07{:}36.796 \dashrightarrow 00{:}07{:}38.920$ TF2 is an orphan nuclear receptor.

NOTE Confidence: 0.769571231333333

00:07:38.920 --> 00:07:40.260 We don't know still what

NOTE Confidence: 0.769571231333333

 $00:07:40.260 \rightarrow 00:07:41.332$ is the natural ligand,

NOTE Confidence: 0.769571231333333

 $00:07:41.340 \dashrightarrow 00:07:43.990$ although retinoic acid can bind

NOTE Confidence: 0.769571231333333

 $00:07:43.990 \longrightarrow 00:07:46.110$ it at high concentrations.

NOTE Confidence: 0.769571231333333

 $00:07:46.110 \longrightarrow 00:07:47.405$ This is the motif that it binds,

NOTE Confidence: 0.769571231333333

 $00{:}07{:}47.410 \dashrightarrow 00{:}07{:}50.850$ and as you can see the NRF 2 full

NOTE Confidence: 0.769571231333333

00:07:50.850 --> 00:07:53.970 isoform has a DNA binding domain.

NOTE Confidence: 0.769571231333333

 $00{:}07{:}53{.}970 \dashrightarrow 00{:}07{:}55{.}490$ It has a ligand domain.

NOTE Confidence: 0.769571231333333

 $00{:}07{:}55{.}490 \dashrightarrow 00{:}07{:}57{.}668$ So it's a conventional nuclear receptor

NOTE Confidence: 0.769571231333333

 $00:07:57.668 \dashrightarrow 00:08:00.794$ that can form a **** or heterodimers.

- NOTE Confidence: 0.769571231333333
- 00:08:00.794 --> 00:08:04.154 It is essential if you,
- NOTE Confidence: 0.769571231333333
- $00:08:04.160 \longrightarrow 00:08:04.626$ yeah,
- NOTE Confidence: 0.769571231333333
- $00:08:04.626 \rightarrow 00:08:06.956$ it's essential for development and
- NOTE Confidence: 0.769571231333333
- $00:08:06.956 \rightarrow 00:08:09.464$ particularly for the formation of blood
- NOTE Confidence: 0.769571231333333
- $00{:}08{:}09{.}464 \dashrightarrow 00{:}08{:}11{.}768$ vessels and it has been shown to be
- NOTE Confidence: 0.769571231333333
- $00{:}08{:}11.842 \dashrightarrow 00{:}08{:}14.597$ important for neural test differentiation.
- NOTE Confidence: 0.769571231333333
- $00{:}08{:}14.600 \dashrightarrow 00{:}08{:}17.365$ Now there have been already some studies
- NOTE Confidence: 0.769571231333333
- $00:08:17.365 \rightarrow 00:08:20.338$ showing the role of NRF 2IN cancer,
- NOTE Confidence: 0.769571231333333
- $00{:}08{:}20.338 \dashrightarrow 00{:}08{:}21.916$ particularly pancreatic and
- NOTE Confidence: 0.769571231333333
- 00:08:21.916 -> 00:08:23.494 prostate cancer metastasis,
- NOTE Confidence: 0.769571231333333
- $00:08:23.500 \longrightarrow 00:08:25.365$ whereas in breast cancer it
- NOTE Confidence: 0.769571231333333
- $00{:}08{:}25.365 \dashrightarrow 00{:}08{:}26.857$ had a controversial effects.
- NOTE Confidence: 0.769571231333333
- 00:08:26.860 --> 00:08:28.940 So you can say well you know this,
- NOTE Confidence: 0.769571231333333
- 00:08:28.940 --> 00:08:30.220 this may already be known
- NOTE Confidence: 0.769571231333333
- $00:08:30.220 \rightarrow 00:08:31.500$ that this factor is important.
- NOTE Confidence: 0.769571231333333

00:08:31.500 --> 00:08:32.354 Cancer, however,

NOTE Confidence: 0.769571231333333

 $00{:}08{:}32{.}354 \dashrightarrow 00{:}08{:}34{.}916$ if you click one more slide,

NOTE Confidence: 0.769571231333333

 $00:08:34.920 \longrightarrow 00:08:37.230$ what really cool our attention is

NOTE Confidence: 0.769571231333333

 $00:08:37.230 \dashrightarrow 00:08:40.300$ that the CPG islands that we found

NOTE Confidence: 0.769571231333333

 $00:08:40.300 \rightarrow 00:08:42.240$ to be differential differentially

NOTE Confidence: 0.769571231333333

00:08:42.240 --> 00:08:44.611 methylated in neural Crest cells

NOTE Confidence: 0.769571231333333

 $00:08:44.611 \rightarrow 00:08:46.716$ versus melanocytes and then later

NOTE Confidence: 0.769571231333333

00:08:46.716 --> 00:08:49.394 on in primary versus Melanoma,

NOTE Confidence: 0.769571231333333

 $00:08:49.394 \longrightarrow 00:08:52.929$ we're actually affecting exactly that

NOTE Confidence: 0.769571231333333

 $00{:}08{:}52{.}929 \dashrightarrow 00{:}08{:}55{.}728$ region in the transcription start

NOTE Confidence: 0.769571231333333

 $00{:}08{:}55{.}728 \dashrightarrow 00{:}08{:}58{.}338$ site that controls the expression

NOTE Confidence: 0.769571231333333

 $00:08:58.338 \rightarrow 00:09:01.680$ of an alternative isoform isoform 2.

NOTE Confidence: 0.769571231333333

 $00:09:01.680 \longrightarrow 00:09:03.850$ Which is not the one that has

NOTE Confidence: 0.769571231333333

 $00:09:03.850 \longrightarrow 00:09:04.780$ been commonly characterized.

NOTE Confidence: 0.769571231333333

 $00{:}09{:}04.780 \dashrightarrow 00{:}09{:}06.328$ So most studies have focused on

NOTE Confidence: 0.769571231333333

 $00:09:06.328 \rightarrow 00:09:07.766$ the full length isoform, isoform,

 $00:09:07.766 \rightarrow 00:09:10.608$ one that has the DNA binding domain,

NOTE Confidence: 0.769571231333333

 $00:09:10.610 \longrightarrow 00:09:12.574$ the like minded domain.

NOTE Confidence: 0.769571231333333

 $00:09:12.574 \dashrightarrow 00:09:15.692$ But this methylated region or the

NOTE Confidence: 0.769571231333333

 $00:09:15.692 \rightarrow 00:09:17.956$ methylated region is in atss that

NOTE Confidence: 0.769571231333333

 $00{:}09{:}17.956 \dashrightarrow 00{:}09{:}19.750$ gives rise to a truncated isoform

NOTE Confidence: 0.769571231333333

 $00:09:19.820 \dashrightarrow 00:09:21.896$ that lacks the DNA binding domain.

NOTE Confidence: 0.769571231333333

 $00{:}09{:}21{.}900 \dashrightarrow 00{:}09{:}25{.}356$ So on the right you can see that as

NOTE Confidence: 0.769571231333333

 $00{:}09{:}25.356 \dashrightarrow 00{:}09{:}29.558$ we had shown before the CPG islands

NOTE Confidence: 0.769571231333333

 $00:09:29.558 \rightarrow 00:09:32.393$ controlling this this nuclear receptor.

NOTE Confidence: 0.769571231333333

 $00:09:32.393 \rightarrow 00:09:36.110$ Networks to ISO two are hypomethylated and

NOTE Confidence: 0.769571231333333

 $00:09:36.193 \rightarrow 00:09:39.798$ they appear in green in cells and stem cells,

NOTE Confidence: 0.769571231333333

 $00:09:39.798 \dashrightarrow 00:09:41.160$ but completely hypermethylated

NOTE Confidence: 0.769571231333333

 $00:09:41.160 \longrightarrow 00:09:42.068$ in melanocytes,

NOTE Confidence: 0.769571231333333

 $00{:}09{:}42.070 \dashrightarrow 00{:}09{:}44.790$ and this is also described here at the

NOTE Confidence: 0.769571231333333

 $00{:}09{:}44.790 \dashrightarrow 00{:}09{:}47.345$ bottom where you can see the beta value.

 $00:09:47.350 \rightarrow 00:09:49.996$ The beta value of 1 means completely

NOTE Confidence: 0.769571231333333

 $00:09:49.996 \dashrightarrow 00:09:53.007$ methylated and a better value close to

NOTE Confidence: 0.769571231333333

 $00:09:53.007 \rightarrow 00:09:55.262$ 0 represents a hypomethylated gene.

NOTE Confidence: 0.769571231333333

 $00:09:55.270 \rightarrow 00:09:57.305$ This completely corresponds to gene

NOTE Confidence: 0.769571231333333

 $00{:}09{:}57{.}305 \dashrightarrow 00{:}09{:}59{.}757$ expression because you can see that

NOTE Confidence: 0.769571231333333

 $00:09:59.757 \dashrightarrow 00:10:02.186$ whereas isoform one is expressed in ESL.

NOTE Confidence: 0.769571231333333

00:10:02.190 --> 00:10:04.134 1000 monocytes isoform 2 is expressed

NOTE Confidence: 0.769571231333333

00:10:04.134 --> 00:10:06.620 in your cells and neural Crest cells,

NOTE Confidence: 0.769571231333333

 $00:10:06.620 \rightarrow 00:10:09.770$ but not expressed at all in melanocytes.

NOTE Confidence: 0.769571231333333

 $00:10:09.770 \longrightarrow 00:10:10.058$ OK.

NOTE Confidence: 0.769571231333333

 $00{:}10{:}10{.}058 \dashrightarrow 00{:}10{:}12.650$ So in when you look at the human samples,

NOTE Confidence: 0.769571231333333

 $00:10:12.650 \longrightarrow 00:10:14.778$ you can see and that even though

NOTE Confidence: 0.769571231333333

 $00:10:14.778 \longrightarrow 00:10:15.690$ it's not completely

NOTE Confidence: 0.880226573846154

00:10:15.753 --> 00:10:18.415 black and white, what we see is that

NOTE Confidence: 0.880226573846154

 $00{:}10{:}18{.}415 \dashrightarrow 00{:}10{:}20{.}641$ there is an increased percentage of

NOTE Confidence: 0.880226573846154

 $00:10:20.641 \rightarrow 00:10:23.227$ hypomethylation of isotope in the metastatic

- NOTE Confidence: 0.880226573846154
- $00:10:23.227 \rightarrow 00:10:25.788$ cases compared to the primary cases.
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}25{.}790 \dashrightarrow 00{:}10{:}29{.}356$ You can see there is from 30% approximately
- NOTE Confidence: 0.880226573846154
- $00:10:29.356 \longrightarrow 00:10:33.034$ to more than 50% of the samples have
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}33{.}034 \dashrightarrow 00{:}10{:}35{.}414$ hypomethylation for NR2ISO2 and if you
- NOTE Confidence: 0.880226573846154
- $00:10:35.414 \rightarrow 00:10:38.195$ keep clicking you will see that you can
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}38{.}195 \dashrightarrow 00{:}10{:}40{.}379$ see there is a progressive increase.
- NOTE Confidence: 0.880226573846154
- $00:10:40.380 \longrightarrow 00:10:42.924$ In the better value from Levi
- NOTE Confidence: 0.880226573846154
- $00:10:42.924 \rightarrow 00:10:44.620$ to primary and metastasis,
- NOTE Confidence: 0.880226573846154
- $00:10:44.620 \rightarrow 00:10:47.155$ so more hypomethylated and conversely
- NOTE Confidence: 0.880226573846154
- $00:10:47.155 \rightarrow 00:10:49.690$ increase expression from primary to
- NOTE Confidence: 0.880226573846154
- $00:10:49.762 \rightarrow 00:10:52.426$ metastasis with a very nice correlation
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}52.426 \dashrightarrow 00{:}10{:}55.390$ between M RNA expression and methylation.
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}55{.}390 \dashrightarrow 00{:}10{:}57{.}382$ Is exactly, if you look at
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}57{.}382 \dashrightarrow 00{:}10{:}58{.}378$ the protein levels,
- NOTE Confidence: 0.880226573846154
- $00{:}10{:}58.380 \dashrightarrow 00{:}11{:}00.368$ we were able to develop actually an
- NOTE Confidence: 0.880226573846154

 $00:11:00.368 \longrightarrow 00:11:01.769$ antibody specific for isoform 2,

NOTE Confidence: 0.880226573846154

00:11:01.770 --> 00:11:02.786 which was challenging because

NOTE Confidence: 0.880226573846154

 $00:11:02.786 \longrightarrow 00:11:04.660$ there are only 15 amino acid symbol

NOTE Confidence: 0.880226573846154

 $00:11:04.660 \rightarrow 00:11:06.160$ amino terminal that that specific.

NOTE Confidence: 0.880226573846154

 $00{:}11{:}06{.}160 \dashrightarrow 00{:}11{:}08{.}270$ But also two you can see that there is an

NOTE Confidence: 0.880226573846154

 $00{:}11{:}08{.}327 \dashrightarrow 00{:}11{:}10{.}385$ increase in the expression of isoform 2.

NOTE Confidence: 0.880226573846154

 $00{:}11{:}10.390 \dashrightarrow 00{:}11{:}12.812$ There are few cells that are positive

NOTE Confidence: 0.880226573846154

 $00:11:12.812 \rightarrow 00:11:15.214$ in the primary cases and this

NOTE Confidence: 0.880226573846154

 $00:11:15.214 \rightarrow 00:11:17.374$ population expands in the metastasis.

NOTE Confidence: 0.880226573846154

 $00{:}11{:}17{.}380 \dashrightarrow 00{:}11{:}20{.}173$ This is something that we are actually

NOTE Confidence: 0.880226573846154

 $00{:}11{:}20{.}173 \dashrightarrow 00{:}11{:}22{.}154$ trying to understand now whether

NOTE Confidence: 0.880226573846154

 $00{:}11{:}22.154 \dashrightarrow 00{:}11{:}24.660$ there is a selection for those cells

NOTE Confidence: 0.880226573846154

 $00:11:24.660 \rightarrow 00:11:27.116$ that expressed isoform 2 over time.

NOTE Confidence: 0.880226573846154

 $00{:}11{:}27{.}120 \dashrightarrow 00{:}11{:}29{.}780$ And again this is this is something

NOTE Confidence: 0.880226573846154

 $00:11:29.780 \rightarrow 00:11:31.858$ we're coming time to investigate but

NOTE Confidence: 0.880226573846154

 $00:11:31.858 \rightarrow 00:11:34.886$ if we can go on we we can see that

 $00{:}11{:}34.886 \dashrightarrow 00{:}11{:}37.529$ isoform one the one that is the full

NOTE Confidence: 0.880226573846154

 $00:11:37.529 \rightarrow 00:11:40.103$ length isoform is not modulated by

NOTE Confidence: 0.880226573846154

 $00:11:40.103 \rightarrow 00:11:41.870$ methylation is completely hypomethylated

NOTE Confidence: 0.880226573846154

00:11:41.870 --> 00:11:44.733 in ES cells you know cells melanocytes

NOTE Confidence: 0.880226573846154

 $00{:}11{:}44{.}733 \dashrightarrow 00{:}11{:}47{.}419$ and both in primary and metastatic

NOTE Confidence: 0.880226573846154

 $00{:}11{:}47{.}419 \dashrightarrow 00{:}11{:}50{.}260$ Melanoma and there is no correlation

NOTE Confidence: 0.880226573846154

 $00:11:50.260 \rightarrow 00:11:52.172$ between expression and methylation.

NOTE Confidence: 0.880226573846154

 $00:11:52.180 \longrightarrow 00:11:54.238$ So isoform one is always there,

NOTE Confidence: 0.880226573846154

 $00{:}11{:}54{.}240 \dashrightarrow 00{:}11{:}56{.}277$ but is isoform 2 the one that.

NOTE Confidence: 0.880226573846154

00:11:56.280 --> 00:11:58.386 Is normally not expressed in melanocytes,

NOTE Confidence: 0.880226573846154

 $00:11:58.390 \rightarrow 00:12:00.582$ but is increasingly demethylated

NOTE Confidence: 0.880226573846154

 $00{:}12{:}00{.}582 \dashrightarrow 00{:}12{:}03{.}322$ and expressed from primary to

NOTE Confidence: 0.880226573846154

00:12:03.322 --> 00:12:04.630 metastatic Melanoma.

NOTE Confidence: 0.880226573846154

 $00:12:04.630 \longrightarrow 00:12:06.280$ Now of course the question is,

NOTE Confidence: 0.880226573846154

 $00:12:06.280 \longrightarrow 00:12:08.255$ is this isoform tool doing

 $00:12:08.255 \rightarrow 00:12:09.835$ anything in Melanoma metastasis?

NOTE Confidence: 0.880226573846154

 $00:12:09.840 \longrightarrow 00:12:12.571$ And for that we had to move to cell

NOTE Confidence: 0.880226573846154

 $00{:}12{:}12{.}571 \dashrightarrow 00{:}12{:}14.650$ lines and again we found this pattern

NOTE Confidence: 0.880226573846154

 $00:12:14.713 \rightarrow 00:12:17.425$ in which melanocytes and some melanomas

NOTE Confidence: 0.880226573846154

 $00:12:17.425 \longrightarrow 00:12:19.233$ aliens had complete hypermethylation

NOTE Confidence: 0.880226573846154

 $00{:}12{:}19{.}293 \dashrightarrow 00{:}12{:}21{.}429$ and lack of expression as you can see

NOTE Confidence: 0.880226573846154

 $00:12:21.429 \longrightarrow 00:12:26.975$ here on the right and you can also see.

NOTE Confidence: 0.880226573846154

 $00:12:26.980 \longrightarrow 00:12:29.095$ Hypomethylation in some of the

NOTE Confidence: 0.880226573846154

 $00{:}12{:}29.095 \dashrightarrow 00{:}12{:}31.210$ cell lines and this corresponds

NOTE Confidence: 0.880226573846154

 $00:12:31.280 \rightarrow 00:12:33.429$ to expression and the same can be

NOTE Confidence: 0.880226573846154

 $00:12:33.429 \longrightarrow 00:12:35.578$ seen at the level of protein.

NOTE Confidence: 0.880226573846154

 $00{:}12{:}35{.}580 \dashrightarrow 00{:}12{:}37{.}476$ You can see isoform 2 expressed only in

NOTE Confidence: 0.880226573846154

 $00{:}12{:}37{.}476 \dashrightarrow 00{:}12{:}39{.}209$ those cells that have hypomethylation.

NOTE Confidence: 0.880226573846154

 $00:12:39.210 \rightarrow 00:12:41.266$ So this represents a good model to study

NOTE Confidence: 0.880226573846154

 $00:12:41.266 \rightarrow 00:12:43.448$ loss of function and general function.

NOTE Confidence: 0.880226573846154

 $00:12:43.450 \rightarrow 00:12:49.470$ But before I go into that you can see that.

- NOTE Confidence: 0.880226573846154
- $00{:}12{:}49{.}470 \dashrightarrow 00{:}12{:}51{.}546$ The methylation uh status can also
- NOTE Confidence: 0.880226573846154
- $00:12:51.546 \longrightarrow 00:12:53.880$ be seen in short term culture.
- NOTE Confidence: 0.880226573846154
- $00{:}12{:}53.880 \dashrightarrow 00{:}12{:}56.255$ These are cells isolated from
- NOTE Confidence: 0.880226573846154
- 00:12:56.255 -> 00:12:59.130 patients and again you have some,
- NOTE Confidence: 0.880226573846154
- $00{:}12{:}59{.}130 \dashrightarrow 00{:}13{:}01{.}265$ some of these short-term cultures
- NOTE Confidence: 0.880226573846154
- 00:13:01.265 -> 00:13:02.973 have hypomethylation and some
- NOTE Confidence: 0.880226573846154
- $00:13:02.973 \rightarrow 00:13:05.220$ of them have hypermethylation.
- NOTE Confidence: 0.880226573846154
- $00{:}13{:}05{.}220 \dashrightarrow 00{:}13{:}08{.}244$ So this is not an artifact of invitro
- NOTE Confidence: 0.880226573846154
- $00{:}13{:}08.250 \dashrightarrow 00{:}13{:}13.080$ culture and it happens in cells derive
- NOTE Confidence: 0.880226573846154
- $00:13:13.080 \rightarrow 00:13:15.786$ very shortly from from patients.
- NOTE Confidence: 0.880226573846154
- $00:13:15.786 \longrightarrow 00:13:19.658$ So if now we move into cells that.
- NOTE Confidence: 0.880226573846154
- $00:13:19.658 \rightarrow 00:13:21.778$ Have hypermethylation like mayor cells.
- NOTE Confidence: 0.880226573846154
- $00{:}13{:}21.780 \dashrightarrow 00{:}13{:}24.510$ If we treat them with editing agent
- NOTE Confidence: 0.880226573846154
- 00:13:24.510 --> 00:13:28.266 like 5 ASA you can see that there is
- NOTE Confidence: 0.880226573846154
- $00:13:28.266 \longrightarrow 00:13:31.185$ an induction of isoform 2 and this
- NOTE Confidence: 0.880226573846154

 $00{:}13{:}31{.}185 \dashrightarrow 00{:}13{:}33{.}915$ is seen also in short term cultures

NOTE Confidence: 0.675399618333333

 $00{:}13{:}33{.}920 \dashrightarrow 00{:}13{:}36{.}762$ in which treatment with FAFSA results in

NOTE Confidence: 0.675399618333333

 $00:13:36.762 \longrightarrow 00:13:38.745$ induction of isophorone to expression

NOTE Confidence: 0.675399618333333

 $00:13:38.745 \rightarrow 00:13:40.839$ with no change in isoform one.

NOTE Confidence: 0.675399618333333

 $00{:}13{:}40.840 \dashrightarrow 00{:}13{:}43.102$ Now when we silence isoform to

NOTE Confidence: 0.675399618333333

 $00:13:43.102 \rightarrow 00:13:45.061$ using the specific srnas against

NOTE Confidence: 0.675399618333333

 $00{:}13{:}45{.}061 \dashrightarrow 00{:}13{:}46{.}946$ these isoform that don't have

NOTE Confidence: 0.675399618333333

 $00:13:46.946 \rightarrow 00:13:49.489$ any impact on the other isoform.

NOTE Confidence: 0.675399618333333

 $00{:}13{:}49{.}490 \dashrightarrow 00{:}13{:}51{.}962$ Initially we did not see any

NOTE Confidence: 0.675399618333333

 $00:13:51.962 \rightarrow 00:13:53.610$ effect onto the proliferation,

NOTE Confidence: 0.675399618333333

 $00{:}13{:}53{.}610 \dashrightarrow 00{:}13{:}56{.}100$ but we observed a clear decrease

NOTE Confidence: 0.675399618333333

 $00:13:56.100 \longrightarrow 00:13:58.612$ in the ability to form colonies

NOTE Confidence: 0.675399618333333

 $00:13:58.612 \longrightarrow 00:14:01.668$ in soft Agar or to form a sphere

NOTE Confidence: 0.675399618333333

 $00:14:01.670 \rightarrow 00:14:03.170$ spheres upon single cells.

NOTE Confidence: 0.675399618333333

 $00:14:03.170 \longrightarrow 00:14:05.045$ So these are properties that

NOTE Confidence: 0.675399618333333

 $00{:}14{:}05{.}045 \dashrightarrow 00{:}14{:}07{.}107$ are characteristic of metastatic

- NOTE Confidence: 0.675399618333333
- $00{:}14{:}07{.}107 \dashrightarrow 00{:}14{:}08{.}982$ cells and basically measure the
- NOTE Confidence: 0.675399618333333
- $00:14:08.982 \rightarrow 00:14:11.545$ ability of the cells to grow under
- NOTE Confidence: 0.675399618333333
- 00:14:11.545 -> 00:14:13.027 very stressful conditions.
- NOTE Confidence: 0.675399618333333
- $00:14:13.030 \rightarrow 00:14:14.290$ Now if we overexpressed,
- NOTE Confidence: 0.675399618333333
- $00:14:14.290 \rightarrow 00:14:16.890$ sorry before we go into their expression.
- NOTE Confidence: 0.675399618333333
- $00:14:16.890 \rightarrow 00:14:19.610$ This is the experiment in this case.
- NOTE Confidence: 0.675399618333333
- $00:14:19.610 \longrightarrow 00:14:21.932$ We injected on the cancer cells
- NOTE Confidence: 0.675399618333333
- $00{:}14{:}21{.}932 \dashrightarrow 00{:}14{:}23{.}480$ intracardiac in a conventional
- NOTE Confidence: 0.675399618333333
- $00:14:23.545 \longrightarrow 00:14:25.303$ in a model of metastasis that
- NOTE Confidence: 0.675399618333333
- $00:14:25.303 \rightarrow 00:14:27.230$ we use very often in the lab.
- NOTE Confidence: 0.675399618333333
- $00:14:27.230 \longrightarrow 00:14:29.710$ And you can see that the silencing of
- NOTE Confidence: 0.675399618333333
- $00{:}14{:}29{.}710 \dashrightarrow 00{:}14{:}31{.}985$ isoform 2 between independent HR and
- NOTE Confidence: 0.675399618333333
- $00{:}14{:}31{.}985 \dashrightarrow 00{:}14{:}34{.}848$ I had a very significant decrease in
- NOTE Confidence: 0.675399618333333
- $00{:}14{:}34{.}848 \dashrightarrow 00{:}14{:}36{.}938$ metastatic potential in this model,
- NOTE Confidence: 0.675399618333333
- $00:14:36.940 \longrightarrow 00:14:39.136$ which is quantified here on the
- NOTE Confidence: 0.675399618333333

00:14:39.136 - 00:14:40.882 right by bioluminescence as well

NOTE Confidence: 0.675399618333333

 $00:14:40.882 \longrightarrow 00:14:42.522$ as fluorescence intensity when

NOTE Confidence: 0.675399618333333

 $00:14:42.522 \longrightarrow 00:14:44.418$ we extract the organs.

NOTE Confidence: 0.675399618333333

 $00:14:44.420 \rightarrow 00:14:47.864$ So it seems like isoform 2 silencing

NOTE Confidence: 0.675399618333333

 $00{:}14{:}47.864 \dashrightarrow 00{:}14{:}49.910$ suppresses metastasis in these.

NOTE Confidence: 0.675399618333333

00:14:49.910 --> 00:14:52.444 A model and now we move into

NOTE Confidence: 0.675399618333333

 $00{:}14{:}52{.}444 \dashrightarrow 00{:}14{:}54{.}079$ over expression systems in which

NOTE Confidence: 0.675399618333333

 $00:14:54.079 \rightarrow 00:14:56.489$ ectopic expression of isoform 2IN

NOTE Confidence: 0.675399618333333

 $00:14:56.489 \rightarrow 00:14:58.844$ cells that have hypermethylation has

NOTE Confidence: 0.675399618333333

 $00:14:58.844 \rightarrow 00:15:00.968$ no effect again into the culture,

NOTE Confidence: 0.675399618333333

 $00:15:00.970 \longrightarrow 00:15:03.553$ but it has a significant ability to

NOTE Confidence: 0.675399618333333

 $00{:}15{:}03.553 \dashrightarrow 00{:}15{:}05.841$ increase the number of colonies and

NOTE Confidence: 0.675399618333333

 $00:15:05.841 \rightarrow 00:15:08.424$ soft tagar as well as sphere formation.

NOTE Confidence: 0.675399618333333

 $00:15:08.430 \longrightarrow 00:15:10.164$ In vivo we injected these cells

NOTE Confidence: 0.675399618333333

 $00:15:10.164 \longrightarrow 00:15:12.677$ in the flank of the mice and then

NOTE Confidence: 0.675399618333333

00:15:12.677 - 00:15:13.969 we did survival surgery.

 $00{:}15{:}13.970 \dashrightarrow 00{:}15{:}15.826$ You can see that we cover two more

NOTE Confidence: 0.675399618333333

 $00:15:15.826 \rightarrow 00:15:17.506$ area because there is always a lot

NOTE Confidence: 0.675399618333333

 $00{:}15{:}17.506 \dashrightarrow 00{:}15{:}19.019$ of signal that comes even after

NOTE Confidence: 0.675399618333333

 $00{:}15{:}19{.}019 \dashrightarrow 00{:}15{:}19{.}910$ you have resected.

NOTE Confidence: 0.675399618333333

 $00:15:19.910 \longrightarrow 00:15:22.788$ The majority of the tumor is the

NOTE Confidence: 0.675399618333333

 $00{:}15{:}22.788 \dashrightarrow 00{:}15{:}25.212$ subcutaneous tumor and you can see

NOTE Confidence: 0.675399618333333

 $00:15:25.212 \rightarrow 00:15:27.565$ that isotonic expression enhances

NOTE Confidence: 0.675399618333333

 $00:15:27.565 \rightarrow 00:15:29.605$ metastasis both by bioluminescence

NOTE Confidence: 0.675399618333333

 $00:15:29.605 \rightarrow 00:15:32.215$ as well as by histological analysis.

NOTE Confidence: 0.675399618333333

 $00{:}15{:}32{.}220 \dashrightarrow 00{:}15{:}34{.}182$ We did this also by intracardiac

NOTE Confidence: 0.675399618333333

00:15:34.182 --> 00:15:35.932 injection and again we observed

NOTE Confidence: 0.675399618333333

 $00{:}15{:}35{.}932 \dashrightarrow 00{:}15{:}38{.}212$ the same effect with an increase

NOTE Confidence: 0.675399618333333

00:15:38.212 --> 00:15:39.352 in metastasis overall.

NOTE Confidence: 0.675399618333333

00:15:39.360 --> 00:15:41.496 So it seems like ISO two,

NOTE Confidence: 0.675399618333333

 $00{:}15{:}41.500 \dashrightarrow 00{:}15{:}43.770$ this particular truncated isoform of

 $00{:}15{:}43.770 \dashrightarrow 00{:}15{:}46.558$ the orphan nuclear receptor and R2F2

NOTE Confidence: 0.675399618333333

 $00{:}15{:}46{.}558 \dashrightarrow 00{:}15{:}50{.}184$ is able to promote metastasis and is.

NOTE Confidence: 0.675399618333333

 $00:15:50.190 \longrightarrow 00:15:51.726$ There's to be required in some

NOTE Confidence: 0.675399618333333

 $00:15:51.726 \longrightarrow 00:15:53.477$ models to in most of the models

NOTE Confidence: 0.675399618333333

00:15:53.477 - 00:15:55.420 that we tried in lab and the paper,

NOTE Confidence: 0.675399618333333

 $00:15:55.420 \longrightarrow 00:15:57.716$ we have three or four different models.

NOTE Confidence: 0.675399618333333

 $00{:}15{:}57{.}720 \dashrightarrow 00{:}16{:}00{.}552$ You can see a decrease in metastasis when

NOTE Confidence: 0.675399618333333

 $00:16:00.552 \rightarrow 00:16:02.839$ you're silence or knockout this change.

NOTE Confidence: 0.675399618333333

 $00{:}16{:}02{.}840 \dashrightarrow 00{:}16{:}04{.}802$ So now what are the programs

NOTE Confidence: 0.675399618333333

 $00:16:04.802 \rightarrow 00:16:06.840$ that are modulated by isoform 2?

NOTE Confidence: 0.675399618333333

00:16:06.840 --> 00:16:09.536 We did RNA sequencing to get to this

NOTE Confidence: 0.675399618333333

 $00:16:09.536 \rightarrow 00:16:11.928$ question and one of the pathways

NOTE Confidence: 0.675399618333333

 $00{:}16{:}11.928 \dashrightarrow 00{:}16{:}13.988$ that was significantly modulated was

NOTE Confidence: 0.675399618333333

 $00:16:13.988 \rightarrow 00:16:16.540$ EMT DPL to mesenchymal transition.

NOTE Confidence: 0.675399618333333

 $00{:}16{:}16{.}540 \dashrightarrow 00{:}16{:}18{.}794$ Now we went on to demonstrate that

NOTE Confidence: 0.675399618333333

 $00:16:18.794 \longrightarrow 00:16:21.724$ a lot of the typical genes involved

- NOTE Confidence: 0.675399618333333
- $00:16:21.724 \rightarrow 00:16:24.092$ in the epithelial transition where
- NOTE Confidence: 0.675399618333333
- $00{:}16{:}24.092 \dashrightarrow 00{:}16{:}26.472$ silence in different cell lines
- NOTE Confidence: 0.675399618333333
- $00:16:26.472 \longrightarrow 00:16:28.852$ when we deplete isoform 2.
- NOTE Confidence: 0.675399618333333
- $00:16:28.852 \rightarrow 00:16:32.170$ And here you can see a validation
- NOTE Confidence: 0.675399618333333
- $00{:}16{:}32.268 \dashrightarrow 00{:}16{:}34.766$ for snail which is reduced both
- NOTE Confidence: 0.675399618333333
- $00{:}16{:}34.766 \dashrightarrow 00{:}16{:}36.861$ transcriptionally and at the protein
- NOTE Confidence: 0.675399618333333
- $00:16:36.861 \rightarrow 00:16:39.120$ level when we silence ice form 2.
- NOTE Confidence: 0.791361322727273
- 00:16:41.430 --> 00:16:43.040 Importantly, I as I mentioned
- NOTE Confidence: 0.791361322727273
- $00:16:43.040 \longrightarrow 00:16:45.030$ at the beginning of my talk,
- NOTE Confidence: 0.791361322727273
- $00:16:45.030 \rightarrow 00:16:47.658$ now we have an understanding of
- NOTE Confidence: 0.791361322727273
- 00:16:47.658 --> 00:16:48.972 intratumoral heterogeneity Melanoma
- NOTE Confidence: 0.791361322727273
- $00{:}16{:}48{.}972 \dashrightarrow 00{:}16{:}51{.}326$ and we have seen that most melanomas
- NOTE Confidence: 0.791361322727273
- $00:16:51.326 \longrightarrow 00:16:53.575$ both in mouse and human display
- NOTE Confidence: 0.791361322727273
- $00{:}16{:}53{.}575 \dashrightarrow 00{:}16{:}55{.}416$ different transcriptional states,
- NOTE Confidence: 0.791361322727273
- $00:16:55.416 \longrightarrow 00:16:57.465$ intermediate neural Crest
- NOTE Confidence: 0.791361322727273

00:16:57.465 --> 00:17:00.880 like proliferation EMT or more

NOTE Confidence: 0.791361322727273

 $00{:}17{:}00{.}968 \dashrightarrow 00{:}17{:}03{.}530$ melanocytic or differentiated.

NOTE Confidence: 0.791361322727273

 $00:17:03.530 \longrightarrow 00:17:05.644$ So we were curious to see whether

NOTE Confidence: 0.791361322727273

 $00:17:05.644 \rightarrow 00:17:08.204$ NRF 2 and particularly the signature

NOTE Confidence: 0.791361322727273

 $00{:}17{:}08{.}204 \dashrightarrow 00{:}17{:}10{.}839$ of genes modulated by isophorone.

NOTE Confidence: 0.791361322727273

 $00{:}17{:}10.840$ --> $00{:}17{:}13.336$ Who were particularly enriched in any NOTE Confidence: 0.791361322727273

 $00{:}17{:}13.336 \dashrightarrow 00{:}17{:}15.542$ of these transcriptional States and as

NOTE Confidence: 0.791361322727273

 $00:17:15.542 \rightarrow 00:17:18.252$ you can see in this heat map, the EMT,

NOTE Confidence: 0.791361322727273

00:17:18.252 --> 00:17:20.296 in particular the transcriptional

NOTE Confidence: 0.791361322727273

 $00:17:20.296 \longrightarrow 00:17:23.022$ state that corresponds to an

NOTE Confidence: 0.791361322727273

 $00{:}17{:}23.022 \dashrightarrow 00{:}17{:}25.234$ epithelial to mesenchymal transition

NOTE Confidence: 0.791361322727273

 $00{:}17{:}25{.}234 \dashrightarrow 00{:}17{:}28{.}553$ seems to be the one that is more

NOTE Confidence: 0.791361322727273

 $00{:}17{:}28.553 \dashrightarrow 00{:}17{:}30.734$ enriched in these two signature

NOTE Confidence: 0.791361322727273

 $00{:}17{:}30{.}734 \dashrightarrow 00{:}17{:}33{.}174$ together with the neural signature.

NOTE Confidence: 0.791361322727273

 $00:17:33.180 \longrightarrow 00:17:36.099$ So this goes together with our findings

NOTE Confidence: 0.791361322727273

 $00:17:36.099 \rightarrow 00:17:40.762$ that if two could be regulating the EMT.

 $00{:}17{:}40.762 \dashrightarrow 00{:}17{:}43.198$ Um estate and in this additional

NOTE Confidence: 0.791361322727273

 $00:17:43.198 \rightarrow 00:17:46.267$ analysis in which we used scenic

NOTE Confidence: 0.791361322727273

 $00{:}17{:}46.267 \dashrightarrow 00{:}17{:}48.447$ to understand the regulations the

NOTE Confidence: 0.791361322727273

 $00:17:48.531 \longrightarrow 00:17:51.361$ the basically the epigenetic and

NOTE Confidence: 0.791361322727273

 $00{:}17{:}51{.}361 \dashrightarrow 00{:}17{:}54{.}191$ transcriptional factors that control each

NOTE Confidence: 0.791361322727273

 $00{:}17{:}54.200 \dashrightarrow 00{:}17{:}56.475$ of these states melanocytic intermediate

NOTE Confidence: 0.791361322727273

 $00:17:56.475 \rightarrow 00:17:59.060$ proliferative neural Crest like an EMT.

NOTE Confidence: 0.791361322727273

00:17:59.060 --> 00:18:01.388 You can see an I think it's more

NOTE Confidence: 0.791361322727273

 $00{:}18{:}01{.}388 \dashrightarrow 00{:}18{:}03{.}970$ visible in the next slide that the

NOTE Confidence: 0.791361322727273

 $00:18:03.970 \longrightarrow 00:18:05.900$ top regulon controlling the event

NOTE Confidence: 0.791361322727273

 $00:18:05.968 \rightarrow 00:18:08.446$ signature and this is a completely

NOTE Confidence: 0.791361322727273

 $00{:}18{:}08{.}446 \dashrightarrow 00{:}18{:}11{.}024$ different analysis done in our mouse.

NOTE Confidence: 0.791361322727273

00:18:11.024 --> 00:18:14.258 Levels of Melanoma you can see that

NOTE Confidence: 0.791361322727273

 $00{:}18{:}14.258 \dashrightarrow 00{:}18{:}17.915$ inner 2F2 is at the top is a top regulon.

NOTE Confidence: 0.791361322727273

 $00{:}18{:}17{.}920 \dashrightarrow 00{:}18{:}19{.}784$ Controlling the EMT signature.

 $00{:}18{:}19{.}784 \dashrightarrow 00{:}18{:}23{.}086$ So it seems like these transcriptional state

NOTE Confidence: 0.791361322727273

 $00{:}18{:}23.086 \dashrightarrow 00{:}18{:}25.246$ of epithelium to mesenchymal transition

NOTE Confidence: 0.791361322727273

 $00:18:25.246 \rightarrow 00:18:29.186$ seems to be mostly controlled by NF2 now.

NOTE Confidence: 0.791361322727273

 $00:18:29.186 \longrightarrow 00:18:31.070$ How this happens?

NOTE Confidence: 0.791361322727273

00:18:31.070 --> 00:18:34.141 Like I told you at the beginning that N2I2,

NOTE Confidence: 0.791361322727273

 $00{:}18{:}34{.}141 \dashrightarrow 00{:}18{:}38{.}069$ another two ISO two is a truncated isoform.

NOTE Confidence: 0.791361322727273

 $00{:}18{:}38{.}070 \dashrightarrow 00{:}18{:}40{.}326$ It lacks the DNA binding domain.

NOTE Confidence: 0.791361322727273

 $00{:}18{:}40{.}330 \dashrightarrow 00{:}18{:}43{.}147$ So how is it possible that it has this

NOTE Confidence: 0.791361322727273

 $00:18:43.147 \rightarrow 00:18:45.330$ capacity to control gene expression?

NOTE Confidence: 0.791361322727273

 $00:18:45.330 \rightarrow 00:18:47.913$ Now when we started to work in this isoform,

NOTE Confidence: 0.791361322727273

 $00{:}18{:}47{.}920 \dashrightarrow 00{:}18{:}51{.}928$ there were only two other papers that were

NOTE Confidence: 0.791361322727273

 $00:18:51.928 \rightarrow 00:18:54.288$ studying alternative isoforms of two,

NOTE Confidence: 0.791361322727273

 $00{:}18{:}54{.}290 \dashrightarrow 00{:}18{:}58{.}124$ all the papers that have been put out there.

NOTE Confidence: 0.791361322727273

 $00:18:58.130 \longrightarrow 00:19:00.559$ And study only the full length isoform.

NOTE Confidence: 0.791361322727273

 $00:19:00.560 \longrightarrow 00:19:02.900$ So there were two papers.

NOTE Confidence: 0.791361322727273

 $00:19:02.900 \longrightarrow 00:19:06.740$ One was suggesting that I2 acted as a

 $00{:}19{:}06{.}740 \dashrightarrow 00{:}19{:}09{.}957$ dominant negative and was not removing

NOTE Confidence: 0.791361322727273

 $00{:}19{:}09{.}957 \dashrightarrow 00{:}19{:}13.203$ or displacing isoform one from chromatin.

NOTE Confidence: 0.791361322727273

 $00:19:13.210 \rightarrow 00:19:15.317$ With us another paper suggested the opposite

NOTE Confidence: 0.791361322727273

 $00:19:15.317 \rightarrow 00:19:17.818$ that I saw two contributed to bind ISO,

NOTE Confidence: 0.791361322727273

 $00{:}19{:}17.820 \dashrightarrow 00{:}19{:}20.466$ one to the chromatin and also what

NOTE Confidence: 0.791361322727273

 $00:19:20.466 \longrightarrow 00:19:22.059$ complicates more this interpretation

NOTE Confidence: 0.791361322727273

 $00:19:22.059 \longrightarrow 00:19:24.796$ of the results is the fact that

NOTE Confidence: 0.791361322727273

 $00{:}19{:}24.796 \dashrightarrow 00{:}19{:}27.594$ isoform one has been or inner 2F2 has

NOTE Confidence: 0.791361322727273

 $00{:}19{:}27{.}594 \dashrightarrow 00{:}19{:}29{.}576$ been described both as a receptor

NOTE Confidence: 0.791361322727273

 $00:19:29.576 \longrightarrow 00:19:31.634$ as a repressor of transcription as

NOTE Confidence: 0.791361322727273

 $00:19:31.634 \longrightarrow 00:19:33.230$ well as an activator.

NOTE Confidence: 0.791361322727273

00:19:33.230 --> 00:19:36.110 So we.

NOTE Confidence: 0.791361322727273

00:19:36.110 $\operatorname{-->}$ 00:19:37.976 It tested and the possibility that

NOTE Confidence: 0.791361322727273

00:19:37.976 --> 00:19:40.591 NRF 2 ISO two was interacting with

NOTE Confidence: 0.791361322727273

 $00{:}19{:}40.591 \dashrightarrow 00{:}19{:}42.761$ isoform one and somehow modulating

 $00:19:42.761 \longrightarrow 00:19:45.681$ the ability of an artist to to bind

NOTE Confidence: 0.791361322727273

 $00:19:45.681 \rightarrow 00:19:47.359$ chromatin and regulate gene expression.

NOTE Confidence: 0.791361322727273

 $00{:}19{:}47.359 \dashrightarrow 00{:}19{:}50.151$ But for this to be true also from

NOTE Confidence: 0.791361322727273

 $00:19:50.151 \longrightarrow 00:19:52.586$ two had to have the capacity to

NOTE Confidence: 0.791361322727273

 $00:19:52.586 \longrightarrow 00:19:55.068$ bind to isoform one in the nucleus,

NOTE Confidence: 0.791361322727273

 $00{:}19{:}55{.}070 \dashrightarrow 00{:}19{:}59{.}370$ and isoform 2 lacks the nuclear

NOTE Confidence: 0.791361322727273

 $00:19:59.370 \rightarrow 00:20:01.770$ localization signal present in isoform one.

NOTE Confidence: 0.791361322727273

 $00{:}20{:}01.770 \dashrightarrow 00{:}20{:}03.918$ It has an alternative nuclear localization

NOTE Confidence: 0.791361322727273

 $00{:}20{:}03{.}918 \dashrightarrow 00{:}20{:}06{.}110$ signal that is much less potent.

NOTE Confidence: 0.791361322727273

 $00:20:06.110 \longrightarrow 00:20:11.297$ Thought we first show in a A

NOTE Confidence: 0.791361322727273

 $00{:}20{:}11{.}300 \dashrightarrow 00{:}20{:}13{.}180$ fragmentation analysis that isoform

NOTE Confidence: 0.791361322727273

 $00:20:13.180 \longrightarrow 00:20:16.000$ 2 is able to reach the

NOTE Confidence: 0.803509978181818

 $00{:}20{:}16.082 \dashrightarrow 00{:}20{:}18.635$ nucleus. You can see there not

NOTE Confidence: 0.803509978181818

 $00:20:18.635 \rightarrow 00:20:20.266$ only the cytoplasm, right?

NOTE Confidence: 0.803509978181818

 $00:20:20.266 \rightarrow 00:20:23.182$ So this suggests the possibility that

NOTE Confidence: 0.803509978181818

 $00{:}20{:}23.182 \dashrightarrow 00{:}20{:}26.019$ it can interact with iPhone one.

 $00:20:26.020 \longrightarrow 00:20:27.754$ Then we also did IP analysis

NOTE Confidence: 0.803509978181818

 $00{:}20{:}27.754 \dashrightarrow 00{:}20{:}29.692$ where we show that isoform one

NOTE Confidence: 0.803509978181818

 $00{:}20{:}29{.}692 \dashrightarrow 00{:}20{:}32{.}191$ can pull down isoform two with two

NOTE Confidence: 0.803509978181818

 $00:20:32.191 \longrightarrow 00:20:33.694$ independent antibodies and the

NOTE Confidence: 0.803509978181818

 $00:20:33.694 \rightarrow 00:20:36.295$ opposite is also true, we can use.

NOTE Confidence: 0.803509978181818

00:20:36.295 --> 00:20:38.605 Mile a GFP pulled down because

NOTE Confidence: 0.803509978181818

 $00:20:38.605 \rightarrow 00:20:40.633$ they unfortunately the endogenous

NOTE Confidence: 0.803509978181818

 $00:20:40.633 \longrightarrow 00:20:43.747$ IP for I2 doesn't work well.

NOTE Confidence: 0.803509978181818

 $00{:}20{:}43.750 \dashrightarrow 00{:}20{:}45.760$ So we use the exogenous construct

NOTE Confidence: 0.803509978181818

 $00:20:45.760 \longrightarrow 00:20:48.547$ that has a GP fusion and so we

NOTE Confidence: 0.803509978181818

 $00{:}20{:}48.547 \dashrightarrow 00{:}20{:}50.599$ can pull down again isoform too

NOTE Confidence: 0.803509978181818

 $00{:}20{:}50.679 \dashrightarrow 00{:}20{:}52.594$ with the isoform 1 suggesting

NOTE Confidence: 0.803509978181818

 $00{:}20{:}52{.}594 \dashrightarrow 00{:}20{:}54{.}740$ that it's two isoforms interact.

NOTE Confidence: 0.803509978181818

 $00{:}20{:}54.740 \dashrightarrow 00{:}20{:}58.700$ And when we look now at Chief of

NOTE Confidence: 0.803509978181818

 $00{:}20{:}58.801 \dashrightarrow 00{:}21{:}02.588$ isoform or chromatin IP of isoform one,

 $00:21:02.590 \longrightarrow 00:21:05.014$ we can see that indeed when we silence

NOTE Confidence: 0.803509978181818

 $00{:}21{:}05{.}014 \dashrightarrow 00{:}21{:}07{.}325$ isoform 2 there is a group of peaks.

NOTE Confidence: 0.803509978181818

 $00:21:07.330 \longrightarrow 00:21:09.640$ Of isoform one that now lose

NOTE Confidence: 0.803509978181818

00:21:09.640 --> 00:21:11.180 binding of isoform one,

NOTE Confidence: 0.803509978181818

 $00{:}21{:}11{.}180 \dashrightarrow 00{:}21{:}13{.}791$ and this is more dramatic in this

NOTE Confidence: 0.803509978181818

00:21:13.791 --> 00:21:16.179 group of targets is a little bit

NOTE Confidence: 0.803509978181818

 $00{:}21{:}16.179 \dashrightarrow 00{:}21{:}18.898$ less prevalent in this subset of of

NOTE Confidence: 0.803509978181818

 $00:21:18.898 \rightarrow 00:21:21.800$ pigs that are bound by isoform one.

NOTE Confidence: 0.803509978181818

 $00{:}21{:}21{.}800 \dashrightarrow 00{:}21{:}23.620$ But we can also observe the opposite.

NOTE Confidence: 0.803509978181818

 $00:21:23.620 \rightarrow 00:21:26.020$ We can see that in a group of peaks there

NOTE Confidence: 0.803509978181818

 $00{:}21{:}26.088 \dashrightarrow 00{:}21{:}28.377$ is an increased binding of isoform one.

NOTE Confidence: 0.803509978181818

 $00:21:28.380 \longrightarrow 00:21:29.840$ When I do is gone.

NOTE Confidence: 0.803509978181818

 $00:21:29.840 \longrightarrow 00:21:32.130$ So definitely the interaction between

NOTE Confidence: 0.803509978181818

 $00:21:32.130 \rightarrow 00:21:34.420$ these two isoforms is complex.

NOTE Confidence: 0.803509978181818

 $00:21:34.420 \longrightarrow 00:21:37.759$ We know that the chip works because.

NOTE Confidence: 0.803509978181818

 $00:21:37.760 \longrightarrow 00:21:40.952$ Um and active two is their most
$00:21:40.952 \rightarrow 00:21:42.320$ significantly enriched transcription

NOTE Confidence: 0.803509978181818

 $00:21:42.397 \longrightarrow 00:21:45.097$ factor in these pics together with

NOTE Confidence: 0.803509978181818

 $00{:}21{:}45.097 \dashrightarrow 00{:}21{:}46.897$ some other transcription factors

NOTE Confidence: 0.803509978181818

 $00:21:46.966 \longrightarrow 00:21:48.711$ which are we're also interested

NOTE Confidence: 0.803509978181818

 $00:21:48.711 \longrightarrow 00:21:50.830$ in in looking at the potential

NOTE Confidence: 0.803509978181818

 $00{:}21{:}50.830 \dashrightarrow 00{:}21{:}52.680$ interaction of MR2 with those.

NOTE Confidence: 0.803509978181818

 $00{:}21{:}52.680 \dashrightarrow 00{:}21{:}56.632$ So we can see that N22ISO2 modulates

NOTE Confidence: 0.803509978181818

 $00:21:56.632 \rightarrow 00:21:59.302$ the binding capacity of the chromatin,

NOTE Confidence: 0.803509978181818

 $00{:}21{:}59{.}302 \dashrightarrow 00{:}22{:}02{.}675$ but it not only in a way seems to be

NOTE Confidence: 0.803509978181818

 $00{:}22{:}02{.}675 \dashrightarrow 00{:}22{:}05{.}212$ in a in a gene specific manner and

NOTE Confidence: 0.803509978181818

 $00:22:05.212 \longrightarrow 00:22:07.810$ when we integrate the cheap data.

NOTE Confidence: 0.803509978181818

 $00:22:07.810 \longrightarrow 00:22:10.510$ Our transcriptomic sequencing of cells

NOTE Confidence: 0.803509978181818

 $00{:}22{:}10.510 \dashrightarrow 00{:}22{:}13.969$ in which we have depleted ISO two,

NOTE Confidence: 0.803509978181818

 $00{:}22{:}13.970 \dashrightarrow 00{:}22{:}16.904$ we found that the majority of the genes are.

NOTE Confidence: 0.803509978181818

 $00{:}22{:}16{.}910 \dashrightarrow 00{:}22{:}19{.}150$ A significant portion of the genes that

 $00:22:19.150 \longrightarrow 00:22:20.988$ are downregulated when we sell in size.

NOTE Confidence: 0.803509978181818

 $00{:}22{:}20{.}990 \dashrightarrow 00{:}22{:}24{.}094$ Or two that are targets for an R2F2

NOTE Confidence: 0.803509978181818

 $00:22:24.094 \rightarrow 00:22:27.466$ are genes that are involved in the EMT,

NOTE Confidence: 0.803509978181818

00:22:27.470 --> 00:22:30.410 snail, twist, Peter two, etc.

NOTE Confidence: 0.803509978181818

 $00{:}22{:}30{.}410 \dashrightarrow 00{:}22{:}34{.}478$ So it seems like a lot of the NR2ISO1

NOTE Confidence: 0.803509978181818

 $00{:}22{:}34{.}478 \dashrightarrow 00{:}22{:}37{.}766$ targets are actually in the direct.

NOTE Confidence: 0.803509978181818

00:22:37.770 --> 00:22:38.626 Same teachings.

NOTE Confidence: 0.803509978181818

 $00:22:38.626 \rightarrow 00:22:41.622$ The opposite is that when we see

NOTE Confidence: 0.803509978181818

 $00:22:41.622 \longrightarrow 00:22:44.608$ look at the genes that are now

NOTE Confidence: 0.803509978181818

 $00:22:44.608 \rightarrow 00:22:46.256$ upregulated by isoform 2,

NOTE Confidence: 0.803509978181818

 $00:22:46.260 \longrightarrow 00:22:48.774$ the majority of those genes are

NOTE Confidence: 0.803509978181818

00:22:48.774 --> 00:22:50.031 involving differentiation and

NOTE Confidence: 0.803509978181818

 $00:22:50.031 \rightarrow 00:22:51.590$ pigmentation such as tyrosinase,

NOTE Confidence: 0.803509978181818

 $00:22:51.590 \rightarrow 00:22:52.646$ DCT, etcetera.

NOTE Confidence: 0.803509978181818

 $00{:}22{:}52.646 \dashrightarrow 00{:}22{:}55.286$ So with all of these,

NOTE Confidence: 0.803509978181818

 $00:22:55.290 \rightarrow 00:22:57.558$ and still with many questions still open,

 $00:22:57.560 \longrightarrow 00:22:59.590$ we propose this modeling which

NOTE Confidence: 0.803509978181818

 $00:22:59.590 \longrightarrow 00:23:02.232$ in primary tumors isoform 2 is

NOTE Confidence: 0.803509978181818

 $00:23:02.232 \rightarrow 00:23:03.858$ hypermethylated and silence.

NOTE Confidence: 0.803509978181818

 $00:23:03.860 \rightarrow 00:23:07.124$ So the dimers of isoform 1 prevail over

NOTE Confidence: 0.803509978181818

 $00:23:07.124 \rightarrow 00:23:09.975$ the heterodimers of isoform 2 and isoform 1,

NOTE Confidence: 0.803509978181818

 $00{:}23{:}09{.}980 \dashrightarrow 00{:}23{:}13{.}179$ whereas in metastasis we see a displacement

NOTE Confidence: 0.803509978181818

00:23:13.179 - 00:23:15.880 of the equilibrium towards the home,

NOTE Confidence: 0.803509978181818

 $00:23:15.880 \longrightarrow 00:23:16.602$ the heterodimer.

NOTE Confidence: 0.803509978181818

00:23:16.602 --> 00:23:16.963 Sorry,

NOTE Confidence: 0.803509978181818

00:23:16.963 - > 00:23:19.129 because now isoform 2 is present

NOTE Confidence: 0.803509978181818

 $00{:}23{:}19{.}129 \dashrightarrow 00{:}23{:}21{.}424$ and is this heterodimer that allows

NOTE Confidence: 0.803509978181818

 $00{:}23{:}21{.}424 \dashrightarrow 00{:}23{:}23{.}600$ the expression of neural Crest and.

NOTE Confidence: 0.803509978181818

 $00:23:23.600 \rightarrow 00:23:26.680$ Empty genes such as twisted slag etcetera.

NOTE Confidence: 0.803509978181818

 $00{:}23{:}26.680 \dashrightarrow 00{:}23{:}29.808$ So what we are now in the process

NOTE Confidence: 0.803509978181818

 $00:23:29.808 \longrightarrow 00:23:31.913$ of understanding is how these

00:23:31.913 --> 00:23:33.983 heterodimer is able to activate

NOTE Confidence: 0.803509978181818

 $00{:}23{:}33{.}983 \dashrightarrow 00{:}23{:}36{.}617$ this EMT genes and we think that

NOTE Confidence: 0.803509978181818

 $00:23:36.617 \longrightarrow 00:23:38.337$ in part could be due

NOTE Confidence: 0.78339154368421

 $00:23:38.340 \longrightarrow 00:23:41.220$ to the interaction with third parties

NOTE Confidence: 0.78339154368421

 $00:23:41.220 \longrightarrow 00:23:43.140$ like additional transcription factors

NOTE Confidence: 0.78339154368421

 $00{:}23{:}43{.}207 \dashrightarrow 00{:}23{:}44{.}915$ that are specifically attracted

NOTE Confidence: 0.78339154368421

 $00:23:44.915 \longrightarrow 00:23:47.050$ to the complex like isoform.

NOTE Confidence: 0.78339154368421

 $00:23:47.050 \rightarrow 00:23:49.466$ So we are doing our time and other

NOTE Confidence: 0.78339154368421

 $00{:}23{:}49{.}466$ --> $00{:}23{:}51{.}793$ techniques to understand what are the NOTE Confidence: 0.78339154368421

 $00{:}23{:}51{.}793$ --> $00{:}23{:}53{.}840$ complexes that are attracted in bound NOTE Confidence: 0.78339154368421

 $00{:}23{:}53{.}840 \dashrightarrow 00{:}23{:}56{.}442$ to the chromatin where we have the

NOTE Confidence: 0.78339154368421

 $00:23:56.442 \rightarrow 00:23:58.250$ homodimers versus the heterodimers.

NOTE Confidence: 0.78339154368421

 $00:23:58.250 \longrightarrow 00:24:00.638$ Now moving on into the second

NOTE Confidence: 0.78339154368421

00:24:00.638 --> 00:24:02.230 part of my talk,

NOTE Confidence: 0.78339154368421

 $00{:}24{:}02{.}230 \dashrightarrow 00{:}24{:}04{.}478$ I don't think I have to repeat the

NOTE Confidence: 0.78339154368421

 $00:24:04.478 \rightarrow 00:24:05.736$ conclusions because that's basically

- NOTE Confidence: 0.78339154368421
- $00:24:05.736 \longrightarrow 00:24:07.962$ the summary that I just have provided.
- NOTE Confidence: 0.78339154368421
- $00:24:07.970 \longrightarrow 00:24:09.950$ So in the interest of time,
- NOTE Confidence: 0.78339154368421
- $00:24:09.950 \rightarrow 00:24:12.620$ I will move to the second part of the story
- NOTE Confidence: 0.78339154368421
- $00:24:12.689 \rightarrow 00:24:15.572$ in which we have focused on brain metastasis.
- NOTE Confidence: 0.78339154368421
- $00{:}24{:}15{.}572 \dashrightarrow 00{:}24{:}19{.}779$ This was the work mostly done by.
- NOTE Confidence: 0.78339154368421
- 00:24:19.780 --> 00:24:21.475 I actually incorrectly stated PhD
- NOTE Confidence: 0.78339154368421
- 00:24:21.475 --> 00:24:24.239 is an MD PhD from our laboratory,
- NOTE Confidence: 0.78339154368421
- $00:24:24.240 \longrightarrow 00:24:28.140$ Kevin Kleffman that is now a.
- NOTE Confidence: 0.78339154368421
- $00:24:28.140 \longrightarrow 00:24:31.332$ Accident at mass general.
- NOTE Confidence: 0.78339154368421
- $00:24:31.332 \longrightarrow 00:24:34.218$ So we of course are interested in
- NOTE Confidence: 0.78339154368421
- 00:24:34.218 --> 00:24:36.007 brain metastasis because it's an
- NOTE Confidence: 0.78339154368421
- $00{:}24{:}36{.}007 \dashrightarrow 00{:}24{:}37{.}697$ important and met clinical need.
- NOTE Confidence: 0.78339154368421
- $00{:}24{:}37.700 \dashrightarrow 00{:}24{:}40.715$ And although these tumors can
- NOTE Confidence: 0.78339154368421
- $00{:}24{:}40.715 \dashrightarrow 00{:}24{:}42.524$ respond to immunotherapy,
- NOTE Confidence: 0.78339154368421
- $00{:}24{:}42.530 \dashrightarrow 00{:}24{:}44.714$ we know that these responses are mostly
- NOTE Confidence: 0.78339154368421

 $00:24:44.714 \longrightarrow 00:24:46.828$ seen in patients that are asymptomatic

NOTE Confidence: 0.78339154368421

 $00:24:46.828 \longrightarrow 00:24:49.066$ and in the symptomatic patients the

NOTE Confidence: 0.78339154368421

 $00:24:49.066 \rightarrow 00:24:51.086$ responses are much poorer and these

NOTE Confidence: 0.78339154368421

 $00:24:51.086 \rightarrow 00:24:53.236$ patients have overall very poor survival.

NOTE Confidence: 0.78339154368421

 $00{:}24{:}53{.}236 \dashrightarrow 00{:}24{:}56{.}016$ So this remains very important

NOTE Confidence: 0.78339154368421

 $00{:}24{:}56.016 \dashrightarrow 00{:}24{:}58.240$ clinical question and definitely.

NOTE Confidence: 0.78339154368421

00:24:58.240 --> 00:25:00.100 Fascinating biological, um.

NOTE Confidence: 0.78339154368421

00:25:00.100 --> 00:25:00.720 Uh,

NOTE Confidence: 0.78339154368421

 $00{:}25{:}00{.}720 \dashrightarrow 00{:}25{:}01{.}340$ question.

NOTE Confidence: 0.78339154368421

 $00:25:01.340 \longrightarrow 00:25:03.746$ How Melanoma cells adapt to the

NOTE Confidence: 0.78339154368421

 $00{:}25{:}03.746 \dashrightarrow 00{:}25{:}05.350$ brain microenvironment and why

NOTE Confidence: 0.78339154368421

 $00{:}25{:}05{.}420 \dashrightarrow 00{:}25{:}07{.}358$ they have such a profound tropism

NOTE Confidence: 0.78339154368421

 $00{:}25{:}07{.}358 \dashrightarrow 00{:}25{:}09{.}646$ for the brain is still something

NOTE Confidence: 0.78339154368421

 $00:25:09.646 \rightarrow 00:25:11.956$ that we don't entirely understand.

NOTE Confidence: 0.78339154368421

 $00:25:11.960 \rightarrow 00:25:14.634$ So uh in collaboration with the management,

NOTE Confidence: 0.78339154368421

 $00:25:14.640 \rightarrow 00:25:17.160$ the director of the Melanoma program at NYU,

- NOTE Confidence: 0.78339154368421
- $00{:}25{:}17.160 \dashrightarrow 00{:}25{:}19.448$ we develop Melanoma short-term
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}19{.}448 \dashrightarrow 00{:}25{:}21{.}580$ cultures that in some cases are
- NOTE Confidence: 0.78339154368421
- $00:25:21.580 \longrightarrow 00:25:23.140$ derived from the same patients.
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}23.140 \dashrightarrow 00{:}25{:}25.327$ So we can in some of the cases we
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}25{.}327 \dashrightarrow 00{:}25{:}27{.}463$ were able to obtain a short-term
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}27.463 \dashrightarrow 00{:}25{:}29.288$ cultures derived from a brain
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}29{.}353 \dashrightarrow 00{:}25{:}31{.}305$ metastasis and extracranial metastasis
- NOTE Confidence: 0.78339154368421
- $00:25:31.305 \longrightarrow 00:25:33.257$ from the same patient.
- NOTE Confidence: 0.78339154368421
- $00:25:33.260 \longrightarrow 00:25:35.294$ And again this is a very
- NOTE Confidence: 0.78339154368421
- 00:25:35.294 --> 00:25:36.650 difficult comparison to make,
- NOTE Confidence: 0.78339154368421
- $00:25:36.650 \rightarrow 00:25:38.690$ but we think it's very useful
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}38.690 \dashrightarrow 00{:}25{:}41.019$ because it reduces some of the
- NOTE Confidence: 0.78339154368421
- 00:25:41.019 --> 00:25:42.366 inter tumoral heterogeneity.
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}42{.}370 \dashrightarrow 00{:}25{:}44{.}465$ As we observe what genetically
- NOTE Confidence: 0.78339154368421
- $00{:}25{:}44{.}465 \dashrightarrow 00{:}25{:}46{.}560$ and transcriptionally and what was
- NOTE Confidence: 0.78339154368421

 $00:25:46.630 \rightarrow 00:25:48.933$ really exciting to us is that when

NOTE Confidence: 0.78339154368421

 $00:25:48.933 \rightarrow 00:25:51.069$ we labeled these cells with GFP

NOTE Confidence: 0.78339154368421

00:25:51.069 --> 00:25:53.235 luciferase and inject them back into NOTE Confidence: 0.78339154368421

 $00:25:53.235 \rightarrow 00:25:55.445$ mice with intracardiac injections,

NOTE Confidence: 0.78339154368421

 $00:25:55.445 \longrightarrow 00:25:58.902$ we observed that that metastatic the NOTE Confidence: 0.78339154368421

 $00:25:58.902 \longrightarrow 00:26:01.194$ the the short-term culture that has NOTE Confidence: 0.78339154368421

 $00{:}26{:}01{.}194 \dashrightarrow 00{:}26{:}04{.}652$ been derived from the brain has in

NOTE Confidence: 0.78339154368421

 $00:26:04.652 \rightarrow 00:26:06.700$ general more metastatic potential

NOTE Confidence: 0.78339154368421

 $00{:}26{:}06{.}700 \dashrightarrow 00{:}26{:}08{.}488$ than the one that was derived

NOTE Confidence: 0.78339154368421

 $00{:}26{:}08{.}488 \dashrightarrow 00{:}26{:}09{.}680$ from an extra cranial metastasis.

NOTE Confidence: 0.78339154368421

 $00:26:09.680 \rightarrow 00:26:11.619$ This is represented here on the right,

NOTE Confidence: 0.78339154368421

 $00:26:11.620 \longrightarrow 00:26:12.829$ but more specifically.

NOTE Confidence: 0.78339154368421

 $00{:}26{:}12.829 \dashrightarrow 00{:}26{:}15.650$ The one that derives from the brain

NOTE Confidence: 0.78339154368421

00:26:15.727 --> 00:26:18.310 has more ability to metastasize to the

NOTE Confidence: 0.78339154368421

 $00{:}26{:}18.310 \dashrightarrow 00{:}26{:}21.017$ brain and this is measured here as a

NOTE Confidence: 0.78339154368421

 $00:26:21.017 \rightarrow 00:26:22.834$ ratio of brain to body luminescence.

- NOTE Confidence: 0.78339154368421
- $00{:}26{:}22{.}834 \dashrightarrow 00{:}26{:}25{.}390$ So it seems like this is a short term.
- NOTE Confidence: 0.78339154368421
- $00:26:25.390 \rightarrow 00:26:28.570$ Cultures retain some of the properties,
- NOTE Confidence: 0.78339154368421
- $00:26:28.570 \longrightarrow 00:26:30.970$ some of the ability that they
- NOTE Confidence: 0.78339154368421
- $00:26:30.970 \longrightarrow 00:26:33.270$ had gained in in people,
- NOTE Confidence: 0.78339154368421
- $00:26:33.270 \longrightarrow 00:26:35.979$ in the patients of colonizing the brain
- NOTE Confidence: 0.78339154368421
- $00{:}26{:}35{.}979 \dashrightarrow 00{:}26{:}38{.}261$ and therefore could be a good model
- NOTE Confidence: 0.78339154368421
- $00:26:38.261 \rightarrow 00:26:40.482$ to study brain specific adaptations.
- NOTE Confidence: 0.78339154368421
- $00:26:40.482 \longrightarrow 00:26:43.866$ So we went on to conduct.
- NOTE Confidence: 0.78339154368421
- $00{:}26{:}43.870 \dashrightarrow 00{:}26{:}46.922$ For the Omega analysis of these short-term
- NOTE Confidence: 0.78339154368421
- $00:26:46.922 \rightarrow 00:26:49.490$ cultures, in total we profile 25,
- NOTE Confidence: 0.78339154368421
- 00:26:49.490 --> 00:26:52.080 approximately 12 and 13 brain
- NOTE Confidence: 0.78339154368421
- $00{:}26{:}52.080 \dashrightarrow 00{:}26{:}54.152$ metastasis versus second metastasis.
- NOTE Confidence: 0.78339154368421
- $00:26:54.160 \longrightarrow 00:26:55.700$ Only a few of them, of course,
- NOTE Confidence: 0.78339154368421
- $00:26:55.700 \longrightarrow 00:26:56.200$ were pair,
- NOTE Confidence: 0.78339154368421
- $00{:}26{:}56{.}200 \dashrightarrow 00{:}26{:}57{.}700$ the rest were unfair and the
- NOTE Confidence: 0.752316441666667

 $00:26:57.754 \rightarrow 00:27:00.194$ idea was to try to identify proteins that

NOTE Confidence: 0.752316441666667

 $00:27:00.194 \rightarrow 00:27:02.010$ were differentially expressed in the brain.

NOTE Confidence: 0.752316441666667

 $00:27:02.010 \longrightarrow 00:27:04.615$ Metastasis input could be potential

NOTE Confidence: 0.752316441666667

00:27:04.615 --> 00:27:06.699 drivers of the adaptation.

NOTE Confidence: 0.752316441666667

00:27:06.700 --> 00:27:08.796 The first reply is that we found when

NOTE Confidence: 0.752316441666667

 $00{:}27{:}08.796 \dashrightarrow 00{:}27{:}10.963$ analyzing the data is that the majority

NOTE Confidence: 0.752316441666667

 $00{:}27{:}10{.}963 \dashrightarrow 00{:}27{:}12{.}553$ of the proteins found differentially

NOTE Confidence: 0.752316441666667

 $00:27:12.612 \longrightarrow 00:27:14.360$ expressed where proteins involved.

NOTE Confidence: 0.752316441666667

 $00{:}27{:}14.360 \dashrightarrow 00{:}27{:}15.770$ Being neurodegenerative disorders

NOTE Confidence: 0.7523164416666667

00:27:15.770 --> 00:27:17.811 such as Parkinson's, Alzheimer's,

NOTE Confidence: 0.752316441666667

00:27:17.811 --> 00:27:21.597 Oxfords and this was rewarding because

NOTE Confidence: 0.752316441666667

 $00{:}27{:}21.597 \dashrightarrow 00{:}27{:}25.269$ our collaborator in at in the Anderson,

NOTE Confidence: 0.752316441666667

 $00{:}27{:}25{.}270 \dashrightarrow 00{:}27{:}27{.}496$ Mike Davis had previously found that

NOTE Confidence: 0.752316441666667

 $00{:}27{:}27{.}496 \dashrightarrow 00{:}27{:}30{.}924$ a lot of the proteins involved in

NOTE Confidence: 0.752316441666667

00:27:30.924 --> 00:27:33.408 different study transcript transcriptional

NOTE Confidence: 0.752316441666667

 $00{:}27{:}33{.}408 \dashrightarrow 00{:}27{:}36{.}308$ profiling of brain metastases wouldn't

 $00{:}27{:}36.308 \dashrightarrow 00{:}27{:}39.170$ reach in Oxford or proteins involved

NOTE Confidence: 0.752316441666667

 $00{:}27{:}39{.}170 \dashrightarrow 00{:}27{:}41{.}447$ in the respiratory chain mitochondria

NOTE Confidence: 0.752316441666667

 $00:27:41.447 \longrightarrow 00:27:44.520$ and so this was confirmed in our.

NOTE Confidence: 0.752316441666667

 $00:27:44.520 \longrightarrow 00:27:44.958$ Plans.

NOTE Confidence: 0.7523164416666667

 $00{:}27{:}44.958 \dashrightarrow 00{:}27{:}47.586$ We found that short-term cultures of

NOTE Confidence: 0.752316441666667

00:27:47.586 --> 00:27:50.550 brain metastasis had elongated mitochondria,

NOTE Confidence: 0.752316441666667

 $00{:}27{:}50{.}550 \dashrightarrow 00{:}27{:}53{.}532$ and they also had increased oxygen

NOTE Confidence: 0.752316441666667

 $00:27:53.532 \rightarrow 00:27:57.130$ consumption rate in this seahorse analysis.

NOTE Confidence: 0.752316441666667

 $00:27:57.130 \longrightarrow 00:28:00.433$ But what we focus on was in the differential

NOTE Confidence: 0.752316441666667

00:28:00.433 --> 00:28:02.880 expression of proteins involved in

NOTE Confidence: 0.752316441666667

 $00{:}28{:}02{.}880 \dashrightarrow 00{:}28{:}04{.}884$ Alzheimer's and Parkinson's disease.

NOTE Confidence: 0.752316441666667

00:28:04.890 --> 00:28:05.716 In particular,

NOTE Confidence: 0.752316441666667

 $00:28:05.716 \longrightarrow 00:28:08.607$ we landed for this study on AP.

NOTE Confidence: 0.752316441666667

 $00{:}28{:}08{.}610 \dashrightarrow 00{:}28{:}11{.}315$ The amyloid processing protein there

NOTE Confidence: 0.752316441666667

 $00{:}28{:}11{.}315 \dashrightarrow 00{:}28{:}14{.}540$ is a precursor for amyloid beta.

 $00:28:14.540 \longrightarrow 00:28:17.012$ It was induced in pre metastasis

NOTE Confidence: 0.752316441666667

 $00{:}28{:}17{.}012 \dashrightarrow 00{:}28{:}18{.}246$ compared to metastasis,

NOTE Confidence: 0.752316441666667

00:28:18.246 --> 00:28:20.276 but not only AP itself,

NOTE Confidence: 0.752316441666667

 $00:28:20.280 \rightarrow 00:28:23.262$ but the proteins that leave AP into

NOTE Confidence: 0.752316441666667

 $00{:}28{:}23{.}262 \dashrightarrow 00{:}28{:}25{.}782$ amyloid beta like beta secretase

NOTE Confidence: 0.752316441666667

 $00{:}28{:}25.782 \dashrightarrow 00{:}28{:}27.519$ or present presently.

NOTE Confidence: 0.752316441666667

 $00:28:27.520 \longrightarrow 00:28:29.389$ So we decided to modulate the loss

NOTE Confidence: 0.752316441666667

 $00:28:29.389 \longrightarrow 00:28:32.404$ of a P to see if it had an effect

NOTE Confidence: 0.752316441666667

 $00{:}28{:}32{.}404 \dashrightarrow 00{:}28{:}33{.}322$ on brain metastasis.

NOTE Confidence: 0.752316441666667

 $00:28:33.330 \longrightarrow 00:28:36.700$ And initially we found that

NOTE Confidence: 0.752316441666667

 $00:28:36.700 \longrightarrow 00:28:39.064$ sorry went too fast.

NOTE Confidence: 0.752316441666667

 $00:28:39.064 \longrightarrow 00:28:41.388$ We found that supernatants

NOTE Confidence: 0.752316441666667

 $00:28:41.388 \longrightarrow 00:28:43.580$ of brain metastasis, Dr.

NOTE Confidence: 0.752316441666667

 $00:28:43.580 \rightarrow 00:28:44.400$ short-term cultures,

NOTE Confidence: 0.752316441666667

 $00:28:44.400 \rightarrow 00:28:46.860$ had higher secretion of family beta

NOTE Confidence: 0.752316441666667

 $00:28:46.860 \rightarrow 00:28:48.824$ compared to the extracranial brain

00:28:48.824 --> 00:28:51.351 metastasis not only in our own hands,

NOTE Confidence: 0.752316441666667

 $00{:}28{:}51{.}360 \dashrightarrow 00{:}28{:}54{.}587$ but also in short term cultures obtained

NOTE Confidence: 0.752316441666667

 $00{:}28{:}54{.}587 \dashrightarrow 00{:}28{:}57{.}410$ from collaborators that we study institute.

NOTE Confidence: 0.752316441666667

 $00{:}28{:}57{.}410 \dashrightarrow 00{:}28{:}57{.}803$ Silence.

NOTE Confidence: 0.752316441666667

00:28:57.803 --> 00:29:00.161 AP Again, we found no effect

NOTE Confidence: 0.752316441666667

00:29:00.161 -> 00:29:01.960 in proliferation in culture,

NOTE Confidence: 0.752316441666667

 $00:29:01.960 \longrightarrow 00:29:04.678$ but when we inject these cells

NOTE Confidence: 0.752316441666667

 $00:29:04.678 \rightarrow 00:29:06.490$ intracardiac in immunodeficient mice,

NOTE Confidence: 0.752316441666667

 $00{:}29{:}06{.}490 \dashrightarrow 00{:}29{:}08{.}520$ we observed this reduction of

NOTE Confidence: 0.752316441666667

 $00:29:08.520 \longrightarrow 00:29:10.550$ the brain to body ratio,

NOTE Confidence: 0.752316441666667

 $00:29:10.550 \rightarrow 00:29:13.042$ suggesting that the loss of AP was

NOTE Confidence: 0.752316441666667

 $00{:}29{:}13.042 \dashrightarrow 00{:}29{:}14.790$ particularly affecting brain metastasis.

NOTE Confidence: 0.752316441666667

00:29:14.790 --> 00:29:17.610 This was confirmed by histological analysis.

NOTE Confidence: 0.752316441666667

 $00{:}29{:}17.610 \dashrightarrow 00{:}29{:}19.810$ This is entertaining of NUMA,

NOTE Confidence: 0.752316441666667

 $00:29:19.810 \longrightarrow 00:29:21.520$ which is a human marker,

 $00:29:21.520 \longrightarrow 00:29:24.650$ and therefore it can perfectly

NOTE Confidence: 0.752316441666667

 $00{:}29{:}24.650 \dashrightarrow 00{:}29{:}27.420$ mark the cells that are.

NOTE Confidence: 0.752316441666667

 $00:29:27.420 \rightarrow 00:29:29.534$ The deriving from the from the scenography,

NOTE Confidence: 0.752316441666667

 $00:29:29.540 \longrightarrow 00:29:32.396$ from the implant and you can see that

NOTE Confidence: 0.752316441666667

 $00:29:32.396 \rightarrow 00:29:35.016$ there was a very significant reduction

NOTE Confidence: 0.752316441666667

 $00{:}29{:}35{.}016 \dashrightarrow 00{:}29{:}38{.}669$ of brain and one positive cells but no

NOTE Confidence: 0.752316441666667

 $00{:}29{:}38.669 \dashrightarrow 00{:}29{:}41.177$ effect on kidney or liver metastasis.

NOTE Confidence: 0.752316441666667

 $00{:}29{:}41.180 \dashrightarrow 00{:}29{:}45.948$ We did ex vivo imaging MRI to conduct

NOTE Confidence: 0.752316441666667

 $00{:}29{:}45{.}948 \dashrightarrow 00{:}29{:}49{.}163$ volumetric analysis that show us that

NOTE Confidence: 0.752316441666667

 $00:29:49.163 \rightarrow 00:29:51.950$ we're not only less perimeter studies

NOTE Confidence: 0.752316441666667

 $00{:}29{:}51{.}950 \dashrightarrow 00{:}29{:}54{.}650$ but also smaller brain metastasis and

NOTE Confidence: 0.752316441666667

 $00{:}29{:}54.650 \dashrightarrow 00{:}29{:}57.459$ of course we show that this effect.

NOTE Confidence: 0.752316441666667

00:29:57.460 --> 00:29:58.570 Happens, you know other models,

NOTE Confidence: 0.752316441666667

 $00{:}29{:}58{.}570 \dashrightarrow 00{:}30{:}01{.}146$ this is not a brain Tropic Melanoma cell

NOTE Confidence: 0.752316441666667

 $00:30:01.146 \rightarrow 00:30:03.985$ line type one and we use national approach,

NOTE Confidence: 0.752316441666667

 $00:30:03.990 \longrightarrow 00:30:06.125$ in this case a crisper cast 9,

- NOTE Confidence: 0.752316441666667
- $00:30:06.130 \longrightarrow 00:30:08.890$ to show again a reduction in

 $00:30:08.890 \longrightarrow 00:30:09.810$ brain metastasis.

NOTE Confidence: 0.752316441666667

 $00:30:09.810 \rightarrow 00:30:12.294$ But this of course open a lot of questions.

NOTE Confidence: 0.752316441666667

 $00:30:12.300 \rightarrow 00:30:15.807$ The first one is which step offering

NOTE Confidence: 0.752316441666667

 $00{:}30{:}15.807 \dashrightarrow 00{:}30{:}17.680$ metastasis is the one in which

NOTE Confidence: 0.752316441666667

 $00:30:17.680 \longrightarrow 00:30:19.775$ is particularly required for the

NOTE Confidence: 0.752316441666667

 $00{:}30{:}19.775 \dashrightarrow 00{:}30{:}21.870$ adaptation and the the arrival

NOTE Confidence: 0.752316441666667

 $00:30:21.944 \longrightarrow 00:30:24.008$ of Melanoma cells to the brain.

NOTE Confidence: 0.752316441666667

 $00:30:24.010 \longrightarrow 00:30:26.439$ So as you know brain metas is

NOTE Confidence: 0.752316441666667

 $00:30:26.439 \rightarrow 00:30:27.480$ a complex process.

NOTE Confidence: 0.752316441666667

 $00:30:27.480 \longrightarrow 00:30:30.704$ It involves multiple steps,

NOTE Confidence: 0.752316441666667

 $00{:}30{:}30{.}704 \dashrightarrow 00{:}30{:}33{.}460$ the intravasation from the tumor

NOTE Confidence: 0.752316441666667

 $00:30:33.460 \longrightarrow 00:30:35.740$ into the first into the stroma,

NOTE Confidence: 0.679529493333333

 $00{:}30{:}35{.}740 \dashrightarrow 00{:}30{:}37{.}960$ then the intravasation into the vasculature,

NOTE Confidence: 0.679529493333333

00:30:37.960 --> 00:30:38.782 survival in circulation,

 $00:30:38.782 \rightarrow 00:30:41.330$ and when the cells arrive to the distal site,

NOTE Confidence: 0.679529493333333

 $00{:}30{:}41{.}330 \dashrightarrow 00{:}30{:}42{.}480$ in this case the brain,

NOTE Confidence: 0.679529493333333

 $00{:}30{:}42{.}480 \dashrightarrow 00{:}30{:}44{.}340$ they have to again extravasate.

NOTE Confidence: 0.679529493333333

 $00:30:44.340 \rightarrow 00:30:46.182$ Many of these cells will undergo

NOTE Confidence: 0.679529493333333

00:30:46.182 --> 00:30:47.940 cell death or become dormant,

NOTE Confidence: 0.679529493333333

 $00{:}30{:}47{.}940 \dashrightarrow 00{:}30{:}51{.}425$ but those that are able to proliferate

NOTE Confidence: 0.679529493333333

 $00{:}30{:}51{.}425 \dashrightarrow 00{:}30{:}53{.}340$ and survive in this environment will

NOTE Confidence: 0.679529493333333

 $00:30:53.340 \longrightarrow 00:30:54.925$ form micro and macro metastasis.

NOTE Confidence: 0.679529493333333

 $00:30:54.930 \longrightarrow 00:30:58.276$ So when was abeta required for this?

NOTE Confidence: 0.679529493333333

 $00:30:58.280 \longrightarrow 00:31:00.098$ For this process,

NOTE Confidence: 0.679529493333333

 $00{:}31{:}00{.}098 \dashrightarrow 00{:}31{:}03{.}046$ so Kevin embark himself in really

NOTE Confidence: 0.679529493333333

 $00{:}31{:}03.046 \dashrightarrow 00{:}31{:}05.650$ a difficult task of monitoring the

NOTE Confidence: 0.679529493333333

 $00:31:05.723 \rightarrow 00:31:08.213$ kinetics of cancer cells injected

NOTE Confidence: 0.679529493333333

 $00{:}31{:}08{.}213 \dashrightarrow 00{:}31{:}10{.}205$ intracardiac in these mice.

NOTE Confidence: 0.679529493333333

 $00{:}31{:}10{.}210 \dashrightarrow 00{:}31{:}13{.}318$ So he did brain slice immunofluorescence

NOTE Confidence: 0.679529493333333

 $00:31:13.320 \dashrightarrow 00:31:15.480$ and a lot of confocal microscopy

- NOTE Confidence: 0.679529493333333
- $00:31:15.480 \rightarrow 00:31:17.689$ and was tracking this GFP positive
- NOTE Confidence: 0.679529493333333
- $00:31:17.689 \longrightarrow 00:31:19.789$ cells in the brain over days.
- NOTE Confidence: 0.679529493333333
- $00:31:19.790 \longrightarrow 00:31:24.155$ So you can see that just one day after.
- NOTE Confidence: 0.679529493333333
- $00:31:24.160 \longrightarrow 00:31:25.184$ In the cardiac injection,
- NOTE Confidence: 0.679529493333333
- $00{:}31{:}25{.}184 \dashrightarrow 00{:}31{:}27{.}340$ these cells are stuck in the vasculature.
- NOTE Confidence: 0.679529493333333
- $00:31:27.340 \longrightarrow 00:31:28.395$ They even have the shape
- NOTE Confidence: 0.679529493333333
- $00:31:28.395 \longrightarrow 00:31:29.239$ of the blood vessels.
- NOTE Confidence: 0.679529493333333
- 00:31:29.240 --> 00:31:30.940 This is a tomato, tomato,
- NOTE Confidence: 0.679529493333333
- $00:31:30.940 \longrightarrow 00:31:33.210$ lectin marking the blood
- NOTE Confidence: 0.679529493333333
- $00:31:33.210 \longrightarrow 00:31:35.480$ vessels you can see there.
- NOTE Confidence: 0.679529493333333
- $00:31:35.480 \longrightarrow 00:31:38.360$ At day three they start extravasation,
- NOTE Confidence: 0.679529493333333
- $00:31:38.360 \rightarrow 00:31:41.258$ they start getting out of the vasculature.
- NOTE Confidence: 0.679529493333333
- $00{:}31{:}41{.}260 \dashrightarrow 00{:}31{:}42{.}500$ Some of these cells die.
- NOTE Confidence: 0.679529493333333
- 00:31:42.500 --> 00:31:44.844 A lot of these cells die in the
- NOTE Confidence: 0.679529493333333
- $00{:}31{:}44{.}844 \dashrightarrow 00{:}31{:}46{.}983$ blood vessels or outside when they
- NOTE Confidence: 0.679529493333333

 $00:31:46.983 \dashrightarrow 00:31:49.209$ are able to extravasate as sustained

NOTE Confidence: 0.679529493333333

00:31:49.280 --> 00:31:51.268 by Clifton Space Stream and you

NOTE Confidence: 0.679529493333333

 $00:31:51.268 \longrightarrow 00:31:52.976$ can see that later on they start

NOTE Confidence: 0.679529493333333

 $00:31:52.976 \rightarrow 00:31:54.569$ crawling through the blood vessels.

NOTE Confidence: 0.679529493333333

 $00:31:54.570 \rightarrow 00:31:57.510$ In these process called Vascular Co option,

NOTE Confidence: 0.679529493333333

 $00{:}31{:}57{.}510$ --> $00{:}31{:}59{.}856$ they can later on form micrometastasis

NOTE Confidence: 0.679529493333333

 $00:31:59.856 \rightarrow 00:32:03.230$ that day 14 and finally micrometastasis.

NOTE Confidence: 0.679529493333333

 $00:32:03.230 \rightarrow 00:32:05.660$ So when you compare the kinetics

NOTE Confidence: 0.679529493333333

 $00:32:05.660 \longrightarrow 00:32:08.045$ of control cells here in the

NOTE Confidence: 0.679529493333333

 $00:32:08.045 \rightarrow 00:32:10.439$ black line to those that lack app,

NOTE Confidence: 0.679529493333333

 $00:32:10.440 \longrightarrow 00:32:12.526$ you can see that the first steps

NOTE Confidence: 0.679529493333333

 $00:32:12.526 \dashrightarrow 00:32:14.670$ of the kinetics are really similar.

NOTE Confidence: 0.679529493333333

 $00:32:14.670 \longrightarrow 00:32:17.310$ There is this big crisis where most of

NOTE Confidence: 0.679529493333333

 $00{:}32{:}17{.}310 \dashrightarrow 00{:}32{:}19{.}898$ the cells that are able to extravasate

NOTE Confidence: 0.679529493333333

 $00:32:19.898 \longrightarrow 00:32:22.609$ die either in the vessels or right

NOTE Confidence: 0.679529493333333

 $00:32:22.609 \rightarrow 00:32:25.339$ after extravasation but then after they 7.

 $00{:}32{:}25{.}340 \dashrightarrow 00{:}32{:}27{.}828$ When the control cells are able to start

NOTE Confidence: 0.679529493333333

 $00{:}32{:}27.828 \dashrightarrow 00{:}32{:}29.763$ expanding and proliferating happily,

NOTE Confidence: 0.679529493333333

 $00{:}32{:}29{.}763 \dashrightarrow 00{:}32{:}33{.}516$ the ones that lack APP can no longer

NOTE Confidence: 0.679529493333333

 $00{:}32{:}33{.}516 \dashrightarrow 00{:}32{:}36{.}400$ grow after the first or second division

NOTE Confidence: 0.679529493333333

 $00:32:36.482 \rightarrow 00:32:38.413$ and they eventually disappear.

NOTE Confidence: 0.679529493333333

 $00:32:38.413 \longrightarrow 00:32:40.678$ They are they are dead.

NOTE Confidence: 0.679529493333333

 $00:32:40.680 \rightarrow 00:32:44.558$ So we wonder which effects were required

NOTE Confidence: 0.679529493333333

 $00{:}32{:}44.558 \dashrightarrow 00{:}32{:}49.159$ for for this role of ebata in the brain.

NOTE Confidence: 0.679529493333333

 $00{:}32{:}49{.}160 \dashrightarrow 00{:}32{:}51{.}379$ And remember that this is a very

NOTE Confidence: 0.679529493333333

 $00:32:51.379 \rightarrow 00:32:52.940$ complex environment where there are,

NOTE Confidence: 0.679529493333333

 $00:32:52.940 \longrightarrow 00:32:54.638$ you know, the resident myeloid cells,

NOTE Confidence: 0.679529493333333

 $00{:}32{:}54{.}640 \dashrightarrow 00{:}32{:}55{.}410$ the microglia.

NOTE Confidence: 0.679529493333333

 $00:32:55.410 \longrightarrow 00:32:57.335$ Macrophages in some cases will

NOTE Confidence: 0.679529493333333

 $00{:}32{:}57{.}335 \dashrightarrow 00{:}32{:}58{.}854$ matter derived macrophages that

NOTE Confidence: 0.679529493333333

 $00{:}32{:}58{.}854 \dashrightarrow 00{:}33{:}00{.}989$ attracted to the tumor and it excels

 $00:33:00.989 \rightarrow 00:33:02.780$ interfiling for sites and astrocytes.

NOTE Confidence: 0.679529493333333

 $00{:}33{:}02{.}780 \dashrightarrow 00{:}33{:}05{.}060$ So we first look at a strocytes

NOTE Confidence: 0.679529493333333

 $00:33:05.060 \longrightarrow 00:33:06.200$ as a potential.

NOTE Confidence: 0.833985785833333

 $00:33:08.620 \rightarrow 00:33:11.290$ Still of interest, because of the

NOTE Confidence: 0.833985785833333

 $00:33:11.290 \longrightarrow 00:33:13.714$ literature that had shown previously

NOTE Confidence: 0.833985785833333

00:33:13.714 --> 00:33:15.835 that activated astrocytes can be Co

NOTE Confidence: 0.833985785833333

 $00{:}33{:}15.835 \dashrightarrow 00{:}33{:}18.100$ opted by the cancer cells to support

NOTE Confidence: 0.833985785833333

 $00:33:18.100 \dashrightarrow 00:33:20.056$ the growth in the brain environment.

NOTE Confidence: 0.833985785833333

 $00:33:20.060 \longrightarrow 00:33:21.740$ And this is indeed the case.

NOTE Confidence: 0.833985785833333

 $00{:}33{:}21.740 \dashrightarrow 00{:}33{:}24.420$ Also in our models where we see that

NOTE Confidence: 0.833985785833333

 $00:33:24.420 \longrightarrow 00:33:27.039$ if you look at the left panels,

NOTE Confidence: 0.833985785833333

 $00{:}33{:}27{.}040 \dashrightarrow 00{:}33{:}28{.}816$ you can see that over time,

NOTE Confidence: 0.833985785833333

 $00:33:28.820 \rightarrow 00:33:30.549$ as the muscles arrive to the brain,

NOTE Confidence: 0.833985785833333

 $00:33:30.550 \rightarrow 00:33:32.040$ these are the control cells,

NOTE Confidence: 0.833985785833333

 $00{:}33{:}32{.}040 \dashrightarrow 00{:}33{:}34{.}674$ you can see an increased presence

NOTE Confidence: 0.833985785833333

00:33:34.674 --> 00:33:36.430 of GFP positive astrocytes.

- NOTE Confidence: 0.833985785833333
- $00:33:36.430 \longrightarrow 00:33:37.690$ So there is.
- NOTE Confidence: 0.833985785833333
- $00:33:37.690 \rightarrow 00:33:39.790$ Some equipment of activated astrocytes
- NOTE Confidence: 0.833985785833333
- $00:33:39.790 \longrightarrow 00:33:42.337$ we cannot distinguish if it's
- NOTE Confidence: 0.833985785833333
- $00:33:42.337 \longrightarrow 00:33:44.513$ recruitment versus activation of
- NOTE Confidence: 0.833985785833333
- $00{:}33{:}44{.}513 \dashrightarrow 00{:}33{:}46{.}910$ the surrounding astrocytes to the
- NOTE Confidence: 0.833985785833333
- $00:33:46.910 \rightarrow 00:33:48.974$ point that they form this network
- NOTE Confidence: 0.833985785833333
- $00:33:48.974 \rightarrow 00:33:52.982$ of active astrocytes that is called
- NOTE Confidence: 0.833985785833333
- 00:33:52.982 --> 00:33:56.819 active Astro cytosis supporting
- NOTE Confidence: 0.833985785833333
- $00{:}33{:}56{.}819 \dashrightarrow 00{:}33{:}59{.}828$ the Melanoma micrometastasis.
- NOTE Confidence: 0.833985785833333
- $00:33:59.830 \longrightarrow 00:34:02.686$ So what we observe is that cells
- NOTE Confidence: 0.833985785833333
- $00:34:02.686 \rightarrow 00:34:05.639$ that lack APP are unable to
- NOTE Confidence: 0.833985785833333
- $00:34:05.639 \longrightarrow 00:34:07.280$ trigger these reactive.
- NOTE Confidence: 0.833985785833333
- $00{:}34{:}07{.}280 \dashrightarrow 00{:}34{:}08{.}360$ Cytosis around them.
- NOTE Confidence: 0.833985785833333
- $00:34:08.360 \longrightarrow 00:34:10.520$ When they arrive to the brain,
- NOTE Confidence: 0.833985785833333
- $00:34:10.520 \longrightarrow 00:34:13.185$ there is a significant reduction
- NOTE Confidence: 0.833985785833333

00:34:13.185 --> 00:34:15.600 of positive cells of GFP.

NOTE Confidence: 0.833985785833333

 $00{:}34{:}15{.}600 \dashrightarrow 00{:}34{:}17{.}950$ Positive cells around the cells

NOTE Confidence: 0.833985785833333

00:34:17.950 --> 00:34:20.199 demand muscles that lack APP,

NOTE Confidence: 0.833985785833333

 $00:34:20.200 \rightarrow 00:34:23.329$ suggesting that perhaps a beta is important

NOTE Confidence: 0.833985785833333

 $00:34:23.329 \rightarrow 00:34:25.740$ in triggering these Astro cytosis.

NOTE Confidence: 0.833985785833333

00:34:25.740 --> 00:34:27.624 Now, in data that I don't

NOTE Confidence: 0.833985785833333

 $00:34:27.624 \longrightarrow 00:34:29.560$ have the time to explain,

NOTE Confidence: 0.833985785833333

 $00{:}34{:}29{.}560 \dashrightarrow 00{:}34{:}33{.}070$ we also show that Amelia better not only has

NOTE Confidence: 0.833985785833333

 $00:34:33.070 \rightarrow 00:34:36.479$ the capacity to activate the astrocytes,

NOTE Confidence: 0.833985785833333

 $00{:}34{:}36{.}480 \dashrightarrow 00{:}34{:}37{.}528$ but also.

NOTE Confidence: 0.833985785833333

 $00{:}34{:}37{.}528 \dashrightarrow 00{:}34{:}39{.}624$ Can suppress the phagocytosis

NOTE Confidence: 0.833985785833333

 $00{:}34{:}39{.}624 \dashrightarrow 00{:}34{:}41{.}720$ coming from the microglia.

NOTE Confidence: 0.833985785833333

 $00{:}34{:}41.720 \dashrightarrow 00{:}34{:}43.864$ So we can see that there is a

NOTE Confidence: 0.833985785833333

 $00{:}34{:}43.864 \dashrightarrow 00{:}34{:}45.283$ reduction of neural inflammation

NOTE Confidence: 0.833985785833333

 $00:34:45.283 \rightarrow 00:34:47.725$ in the presence of family beta.

NOTE Confidence: 0.833985785833333

 $00{:}34{:}47{.}730 \dashrightarrow 00{:}34{:}49{.}767$ So we think that Ali beta secreted

- NOTE Confidence: 0.833985785833333
- 00:34:49.767 -> 00:34:51.788 by cancer cells can have multiple
- NOTE Confidence: 0.833985785833333
- $00:34:51.788 \longrightarrow 00:34:53.603$ effects in the brain metastasis,
- NOTE Confidence: 0.833985785833333
- $00:34:53.610 \rightarrow 00:34:55.910$ macular vironment, particularly on
- NOTE Confidence: 0.833985785833333
- $00:34:55.910 \rightarrow 00:34:58.785$ the astrocytes and the microglia,
- NOTE Confidence: 0.833985785833333
- $00:34:58.790 \longrightarrow 00:35:01.070$ but also it can influence,
- NOTE Confidence: 0.833985785833333
- $00:35:01.070 \dashrightarrow 00:35:03.464$ as it has been reported in Alzheimer's,
- NOTE Confidence: 0.833985785833333
- $00:35:03.470 \longrightarrow 00:35:06.428$ the interaction with the endothelial cells.
- NOTE Confidence: 0.833985785833333
- $00:35:06.430 \longrightarrow 00:35:08.650$ So of course these open,
- NOTE Confidence: 0.833985785833333
- $00:35:08.650 \longrightarrow 00:35:11.406$ these findings open some
- NOTE Confidence: 0.833985785833333
- 00:35:11.406 00:35:13.473 possibilities and therapeutic
- NOTE Confidence: 0.833985785833333
- $00:35:13.473 \rightarrow 00:35:16.330$ opportunities because of all the.
- NOTE Confidence: 0.833985785833333
- $00:35:16.330 \longrightarrow 00:35:19.570$ Armamentarium of drugs that have
- NOTE Confidence: 0.833985785833333
- $00:35:19.570 \dashrightarrow 00:35:22.426$ been developed against America beta,
- NOTE Confidence: 0.833985785833333
- $00{:}35{:}22.426 \dashrightarrow 00{:}35{:}25.597$ some of them beta secret ase inhibitors and
- NOTE Confidence: 0.833985785833333
- $00{:}35{:}25{.}597 \dashrightarrow 00{:}35{:}28{.}649$ more recently anti American antibodies,
- NOTE Confidence: 0.833985785833333

 $00:35:28.650 \rightarrow 00:35:30.996$ some of which have been developed

NOTE Confidence: 0.833985785833333

 $00:35:30.996 \longrightarrow 00:35:33.346$ for clinical use and in some

NOTE Confidence: 0.833985785833333

00:35:33.346 - > 00:35:35.464 cases even approved by the FDA.

NOTE Confidence: 0.833985785833333

 $00:35:35.470 \longrightarrow 00:35:37.564$ So these open the possibility of

NOTE Confidence: 0.833985785833333

 $00:35:37.564 \rightarrow 00:35:39.733$ repurposing some of these drugs which

NOTE Confidence: 0.833985785833333

00:35:39.733 --> 00:35:41.863 are generally safe for brain metastasis

NOTE Confidence: 0.833985785833333

 $00{:}35{:}41{.}863 \dashrightarrow 00{:}35{:}44{.}128$ and for proof of principle we've

NOTE Confidence: 0.833985785833333

 $00:35:44.128 \rightarrow 00:35:46.366$ been testing some of these compounds.

NOTE Confidence: 0.833985785833333

00:35:46.370 --> 00:35:48.400 In collaboration with Eli Lilly,

NOTE Confidence: 0.833985785833333

 $00{:}35{:}48{.}400 \dashrightarrow 00{:}35{:}50{.}985$ so we obtain beta secret ase

NOTE Confidence: 0.833985785833333

 $00:35:50.985 \longrightarrow 00:35:54.088$ inhibitors in the diet of the mice.

NOTE Confidence: 0.833985785833333

 $00:35:54.088 \rightarrow 00:35:56.868$ So initially we injected a cancer cells

NOTE Confidence: 0.833985785833333

 $00{:}35{:}56.868 \dashrightarrow 00{:}35{:}59.521$ Melanoma cells in these mice and gave

NOTE Confidence: 0.833985785833333

 $00{:}35{:}59{.}521 \dashrightarrow 00{:}36{:}02{.}183$ them a better second base inhibitor

NOTE Confidence: 0.833985785833333

 $00:36:02.183 \longrightarrow 00:36:04.913$ in the food or controlled diet.

NOTE Confidence: 0.833985785833333

 $00:36:04.920 \longrightarrow 00:36:07.448$ And you can see how this reduces the

 $00:36:07.448 \longrightarrow 00:36:10.286$ number of brain metastasis in this model.

NOTE Confidence: 0.833985785833333

 $00{:}36{:}10.290 \dashrightarrow 00{:}36{:}12.355$ This is a short term culture but

NOTE Confidence: 0.833985785833333

 $00{:}36{:}12.355 \dashrightarrow 00{:}36{:}14.918$ also in the five one Melanoma cells.

NOTE Confidence: 0.833985785833333

 $00:36:14.920 \rightarrow 00:36:16.736$ Now of course this is more a prophylactic.

NOTE Confidence: 0.833985785833333

 $00:36:16.740 \longrightarrow 00:36:18.092$ Model because treatment starts

NOTE Confidence: 0.833985785833333

 $00:36:18.092 \longrightarrow 00:36:19.782$ at the time of injection,

NOTE Confidence: 0.833985785833333

 $00{:}36{:}19.790 \dashrightarrow 00{:}36{:}22.510$ so we raise the bar a little bit

NOTE Confidence: 0.833985785833333

 $00:36:22.510 \rightarrow 00:36:25.040$ by allowing the cells to establish

NOTE Confidence: 0.833985785833333

 $00{:}36{:}25{.}040 \dashrightarrow 00{:}36{:}27{.}686$ metastasis first and then after 21

NOTE Confidence: 0.833985785833333

 $00:36:27.769 \rightarrow 00:36:30.480$ days we gave doxycycline to the food

NOTE Confidence: 0.833985785833333

 $00:36:30.480 \longrightarrow 00:36:32.960$ and the in the water of the mice

NOTE Confidence: 0.833985785833333

 $00{:}36{:}32{.}960 \dashrightarrow 00{:}36{:}35{.}747$ to activate docs inducible SH RNA.

NOTE Confidence: 0.833985785833333

 $00{:}36{:}35{.}750 \dashrightarrow 00{:}36{:}37{.}726$ And again we saw that in this context,

NOTE Confidence: 0.894519268333333

 $00:36:37.730 \longrightarrow 00:36:39.446$ even when the treatment is initiated,

NOTE Confidence: 0.894519268333333

 $00:36:39.450 \rightarrow 00:36:41.370$ once metastasis have been formed,

 $00{:}36{:}41{.}370 \dashrightarrow 00{:}36{:}43{.}330$ we can see a reduction in the number

NOTE Confidence: 0.894519268333333

 $00{:}36{:}43{.}330 \dashrightarrow 00{:}36{:}45{.}158$ of brain metastases and if we do

NOTE Confidence: 0.894519268333333

 $00:36:45.158 \dashrightarrow 00:36:47.040$ the same thing with the Secretary.

NOTE Confidence: 0.894519268333333

 $00:36:47.040 \rightarrow 00:36:50.480$ Keep in touch. We can also see again

NOTE Confidence: 0.894519268333333

 $00:36:50.480 \dashrightarrow 00:36:52.008$ after initiating the treatment.

NOTE Confidence: 0.894519268333333

 $00{:}36{:}52{.}008 \dashrightarrow 00{:}36{:}54{.}570$ Once the micrometer studies have been formed,

NOTE Confidence: 0.894519268333333

 $00{:}36{:}54{.}570 \dashrightarrow 00{:}36{:}57{.}020$ we can see a reduction in brain

NOTE Confidence: 0.894519268333333

 $00:36:57.020 \rightarrow 00:36:59.580$ metastasis we are currently trying to.

NOTE Confidence: 0.780478655

 $00{:}37{:}01{.}720 \dashrightarrow 00{:}37{:}05{.}200$ Moving to well before I go into that,

NOTE Confidence: 0.780478655

 $00:37:05.200 \longrightarrow 00:37:09.770$ we are now testing the antibodies against

NOTE Confidence: 0.780478655

00:37:09.770 $\operatorname{-->}$ 00:37:12.140 Emily Beta either as a monotherapy

NOTE Confidence: 0.780478655

 $00:37:12.140 \longrightarrow 00:37:14.292$ or in combination with immunotherapy

NOTE Confidence: 0.780478655

 $00:37:14.292 \longrightarrow 00:37:16.866$ to see if we can recapitulate

NOTE Confidence: 0.780478655

 $00{:}37{:}16.866 \dashrightarrow 00{:}37{:}19.106$ the same effects of surf here.

NOTE Confidence: 0.780478655

 $00:37:19.110 \longrightarrow 00:37:21.190$ So to summarize this part of the talk,

NOTE Confidence: 0.780478655

 $00:37:21.190 \rightarrow 00:37:24.004$ we have shown the proteomic studies

- NOTE Confidence: 0.780478655
- $00{:}37{:}24.004 \dashrightarrow 00{:}37{:}26.708$ have revealed a novel connection

 $00{:}37{:}26.708 \dashrightarrow 00{:}37{:}29.010$ between brain metastasis and

NOTE Confidence: 0.780478655

 $00:37:29.010 \rightarrow 00:37:30.050$ neurodegenerative pathologies.

NOTE Confidence: 0.780478655

 $00:37:30.050 \rightarrow 00:37:32.234$ This has now been confirmed by other studies.

NOTE Confidence: 0.780478655

 $00:37:32.240 \longrightarrow 00:37:33.724$ We collaborated with a group of men,

NOTE Confidence: 0.780478655

 $00:37:33.730 \longrightarrow 00:37:35.239$ iser, last year.

NOTE Confidence: 0.780478655

 $00:37:35.239 \rightarrow 00:37:39.290$ We got a study done in single cell

NOTE Confidence: 0.780478655

 $00:37:39.290 \rightarrow 00:37:42.642$ analysis of primate testis that also

NOTE Confidence: 0.780478655

 $00{:}37{:}42.642 \dashrightarrow 00{:}37{:}45.276$ recapitulated these these finding

NOTE Confidence: 0.780478655

 $00{:}37{:}45{.}276$ --> $00{:}37{:}49{.}026$ that Melanoma cells mimic the.

NOTE Confidence: 0.780478655

 $00{:}37{:}49.026 \dashrightarrow 00{:}37{:}53.085$ A neuronal pathways in another of the

NOTE Confidence: 0.780478655

 $00{:}37{:}53.085 \dashrightarrow 00{:}37{:}54.980$ alterations that are seen in you know,

NOTE Confidence: 0.780478655

 $00:37:54.980 \longrightarrow 00:37:56.435$ degenerative disorders once

NOTE Confidence: 0.780478655

 $00{:}37{:}56{.}435 \dashrightarrow 00{:}37{:}58{.}375$ they reach the brain.

NOTE Confidence: 0.780478655

 $00:37:58.380 \longrightarrow 00:38:00.865$ We see that Amelia Beta is particularly

00:38:00.865 --> 00:38:02.758 required for pre metastasis and

NOTE Confidence: 0.780478655

 $00{:}38{:}02.758 \dashrightarrow 00{:}38{:}05.038$ not other sites of metastasis is

NOTE Confidence: 0.780478655

 $00:38:05.038 \dashrightarrow 00:38:07.467$ acquired for steps that happen after

NOTE Confidence: 0.780478655

 $00{:}38{:}07{.}467 \dashrightarrow 00{:}38{:}09{.}063$ extravasation and early survival

NOTE Confidence: 0.780478655

 $00{:}38{:}09{.}063 \dashrightarrow 00{:}38{:}11.766$ in the brain parenchyma and among

NOTE Confidence: 0.780478655

 $00{:}38{:}11.766 \dashrightarrow 00{:}38{:}14.136$ the multiple functions of family.

NOTE Confidence: 0.780478655

00:38:14.140 --> 00:38:16.363 But in this context we have seen that it

NOTE Confidence: 0.780478655

00:38:16.363 --> 00:38:17.853 triggers an anti-inflammatory response

NOTE Confidence: 0.780478655

 $00{:}38{:}17.853 \dashrightarrow 00{:}38{:}19.838$ in the astrocytes and suppresses.

NOTE Confidence: 0.780478655

00:38:19.840 --> 00:38:22.800 Your inflammation, as I mentioned,

NOTE Confidence: 0.780478655

 $00:38:22.800 \dashrightarrow 00:38:25.884$ we are also studying now whether

NOTE Confidence: 0.780478655

 $00:38:25.884 \dashrightarrow 00:38:27.675$ these effects of family beta can

NOTE Confidence: 0.780478655

 $00:38:27.675 \longrightarrow 00:38:29.380$ be seen also in other models.

NOTE Confidence: 0.780478655

 $00:38:29.380 \longrightarrow 00:38:31.102$ So we have done a spatial

NOTE Confidence: 0.780478655

00:38:31.102 --> 00:38:31.676 transcriptomic analysis.

NOTE Confidence: 0.780478655

 $00:38:31.680 \longrightarrow 00:38:32.505$ In this case,

- NOTE Confidence: 0.780478655
- 00:38:32.505 --> 00:38:33.880 it's not a Melanoma model,
- NOTE Confidence: 0.780478655
- $00:38:33.880 \longrightarrow 00:38:36.656$ is the 41 model which is a breast
- NOTE Confidence: 0.780478655
- 00:38:36.656 --> 00:38:38.523 cancer triple negative model
- NOTE Confidence: 0.780478655
- $00:38:38.523 \longrightarrow 00:38:41.258$ that also colonizes the brain
- NOTE Confidence: 0.780478655
- $00:38:41.258 \rightarrow 00:38:42.899$ after intracardiac injection.
- NOTE Confidence: 0.780478655
- $00:38:42.900 \longrightarrow 00:38:46.640$ These are on the top is a is a brain,
- NOTE Confidence: 0.780478655
- $00{:}38{:}46.640 \dashrightarrow 00{:}38{:}48.928$ is half of a brain of a sham
- NOTE Confidence: 0.780478655
- $00{:}38{:}48{.}928 \dashrightarrow 00{:}38{:}50{.}479$ mouse and this is the.
- NOTE Confidence: 0.780478655
- $00:38:50.480 \longrightarrow 00:38:50.802$ Mouse,
- NOTE Confidence: 0.780478655
- $00:38:50.802 \rightarrow 00:38:53.056$ this is a mouse that was injected
- NOTE Confidence: 0.780478655
- $00:38:53.056 \rightarrow 00:38:55.394$ with the 41 cells and what I want
- NOTE Confidence: 0.780478655
- $00:38:55.394 \rightarrow 00:38:57.659$ to bring to your attention is that
- NOTE Confidence: 0.780478655
- $00{:}38{:}57.660 \dashrightarrow 00{:}39{:}01.286$ in this the next vision analysis we
- NOTE Confidence: 0.780478655
- $00{:}39{:}01{.}286 \dashrightarrow 00{:}39{:}04{.}212$ can see that the gfap positive cells
- NOTE Confidence: 0.780478655
- $00:39:04.212 \rightarrow 00:39:06.580$ around the areas where the tumors are,
- NOTE Confidence: 0.780478655

 $00:39{:}06.580 \dashrightarrow 00{:}39{:}08.924$ you have to probably take me you know

NOTE Confidence: 0.780478655

 $00{:}39{:}08{.}924 \dashrightarrow 00{:}39{:}11{.}459$ take my my word these are the areas

NOTE Confidence: 0.780478655

 $00{:}39{:}11{.}459 \dashrightarrow 00{:}39{:}13{.}507$ where the tumors are circle here

NOTE Confidence: 0.780478655

 $00{:}39{:}13.507 \dashrightarrow 00{:}39{:}16.062$ and we see a special expression or

NOTE Confidence: 0.780478655

00:39:16.062 --> 00:39:17.882 increased expression of GFP around

NOTE Confidence: 0.780478655

 $00{:}39{:}17.882 \dashrightarrow 00{:}39{:}20.060$ those tumor cells we also see.

NOTE Confidence: 0.780478655

 $00:39:20.060 \longrightarrow 00:39:22.388$ This one is 100 which has been seen

NOTE Confidence: 0.780478655

 $00{:}39{:}22{.}388 \dashrightarrow 00{:}39{:}25{.}283$ it used in tumor cells in the brain

NOTE Confidence: 0.780478655

00:39:25.283 --> 00:39:27.250 environment in other cancer types.

NOTE Confidence: 0.780478655

 $00:39:27.250 \rightarrow 00:39:29.440$ And interestingly we find these

NOTE Confidence: 0.780478655

 $00{:}39{:}29{.}440 \dashrightarrow 00{:}39{:}32{.}231$ signature that we have that has

NOTE Confidence: 0.780478655

 $00:39:32.231 \dashrightarrow 00:39:35.393$ been previously reported as the an NOTE Confidence: 0.780478655

 $00:39:35.393 \rightarrow 00:39:36.974$ Alzheimer's associated microglia

NOTE Confidence: 0.780478655

 $00:39:37.050 \dashrightarrow 00:39:38.980$ signature that is a combination

NOTE Confidence: 0.780478655

 $00{:}39{:}38{.}980 \dashrightarrow 00{:}39{:}42{.}616$ of 10 markers and is again seen

NOTE Confidence: 0.780478655

 $00:39:42.616 \rightarrow 00:39:46.028$ particularly activated around the

- NOTE Confidence: 0.780478655
- $00:39:46.030 \longrightarrow 00:39:48.740$ brain metastasis here in here.

 $00{:}39{:}48.740 \dashrightarrow 00{:}39{:}51.547$ So it seems like perhaps these Alzheimer

NOTE Confidence: 0.780478655

 $00:39:51.547 \rightarrow 00:39:54.092$ like response in the microglia genes

NOTE Confidence: 0.780478655

 $00:39:54.092 \rightarrow 00:39:55.771$ around the brain metastasis cells

NOTE Confidence: 0.780478655

 $00{:}39{:}55{.}771 \dashrightarrow 00{:}39{:}57{.}710$ could be a more general finding and

NOTE Confidence: 0.780478655

 $00:39:57.760 \dashrightarrow 00:39:59.480$ not only characteristic of Melanoma.

NOTE Confidence: 0.780478655

00:39:59.480 --> 00:40:00.419 As I mentioned,

NOTE Confidence: 0.780478655

 $00:40:00.419 \longrightarrow 00:40:02.898$ we are now very excited by studying

NOTE Confidence: 0.780478655

 $00{:}40{:}02.898 \dashrightarrow 00{:}40{:}05.888$ whether app genetic and pharmacological

NOTE Confidence: 0.780478655

 $00{:}40{:}05{.}888 \dashrightarrow 00{:}40{:}08{.}905$ inhibition extends to other cancer

NOTE Confidence: 0.780478655

 $00{:}40{:}08{.}905 \dashrightarrow 00{:}40{:}12{.}163$ types and whether the combination of

NOTE Confidence: 0.780478655

 $00{:}40{:}12{.}163 \dashrightarrow 00{:}40{:}14{.}491$ beta secret ase inhibitors or antibodies

NOTE Confidence: 0.780478655

 $00:40:14.491 \longrightarrow 00:40:17.257$ can work alone or in combination

NOTE Confidence: 0.780478655

 $00{:}40{:}17.257 \dashrightarrow 00{:}40{:}19.170$ with checkpoint checkpoint locate.

NOTE Confidence: 0.780478655

00:40:19.170 --> 00:40:20.484 In immunocompetent models,

 $00:40:20.484 \longrightarrow 00:40:24.028$ I was hoping to tell you about a

NOTE Confidence: 0.780478655

 $00:40:24.028 \rightarrow 00:40:26.786$ another very exciting story in the lab,

NOTE Confidence: 0.780478655

 $00{:}40{:}26.790$ --> $00{:}40{:}29.220$ but I I see the clock and we are reaching NOTE Confidence: 0.831005876153846

 $00:40:29.279 \rightarrow 00:40:34.748$ the 1:00 PM. So I will stop here and.

NOTE Confidence: 0.831005876153846

 $00{:}40{:}34.750 \dashrightarrow 00{:}40{:}37.654$ This is this data I wanted to present

NOTE Confidence: 0.831005876153846

 $00{:}40{:}37.654 \dashrightarrow 00{:}40{:}39.982$ but perhaps at the second occasion

NOTE Confidence: 0.831005876153846

 $00:40:39.982 \longrightarrow 00:40:42.850$ and just wanna thank all the members

NOTE Confidence: 0.831005876153846

 $00{:}40{:}42.850 \dashrightarrow 00{:}40{:}46.270$ of the lab for their contributions.

NOTE Confidence: 0.831005876153846

 $00:40:46.270 \longrightarrow 00:40:48.112$ This work that I presented was

NOTE Confidence: 0.831005876153846

 $00:40:48.112 \longrightarrow 00:40:50.099$ mostly done by the first part,

NOTE Confidence: 0.831005876153846

 $00{:}40{:}50{.}100 \dashrightarrow 00{:}40{:}52{.}962$ which are Veronica and Claudia in

NOTE Confidence: 0.831005876153846

00:40:52.962 --> 00:40:57.008 NRF 2 and Ali and Maya in CDP one.

NOTE Confidence: 0.831005876153846

 $00{:}40{:}57{.}010 \dashrightarrow 00{:}41{:}01{.}564$ And Kevin Kleffman led the I'm a

NOTE Confidence: 0.831005876153846

 $00{:}41{:}01{.}564 \dashrightarrow 00{:}41{:}03{.}860$ little better story and I want to thank

NOTE Confidence: 0.831005876153846

 $00:41:03.928 \rightarrow 00:41:06.100$ of course all our funding sources.

NOTE Confidence: 0.831005876153846

 $00:41:06.100 \rightarrow 00:41:08.858$ And thank you all for your attention

 $00:41:08.858 \rightarrow 00:41:11.036$ and your patience with the technical

NOTE Confidence: 0.831005876153846

 $00:41:11.036 \longrightarrow 00:41:12.380$ issues at the beginning.

NOTE Confidence: 0.831005876153846

 $00:41:12.380 \rightarrow 00:41:13.815$ I'll stop here and take any questions.

NOTE Confidence: 0.831005876153846

00:41:13.820 --> 00:41:14.180 Thank you.

NOTE Confidence: 0.821179502

 $00{:}41{:}16{.}510 \dashrightarrow 00{:}41{:}18{.}593$ Alright, thank you so much, Eva.

NOTE Confidence: 0.821179502

00:41:18.593 --> 00:41:20.651 I'm going to ask folks to

NOTE Confidence: 0.821179502

 $00:41:20.651 \longrightarrow 00:41:22.409$ put questions in the chat.

NOTE Confidence: 0.821179502

 $00:41:22.410 \longrightarrow 00:41:24.370$ We only got a couple of minutes,

NOTE Confidence: 0.821179502

 $00{:}41{:}24.370 \dashrightarrow 00{:}41{:}26.225$ but while people type in the questions,

NOTE Confidence: 0.821179502

 $00:41:26.230 \longrightarrow 00:41:27.742$ maybe I don't know if Marcus

NOTE Confidence: 0.821179502

00:41:27.742 --> 00:41:29.550 has any or I can ask one.

NOTE Confidence: 0.821179502

00:41:29.550 --> 00:41:33.600 Go ahead Marcus, I've got plenty.

NOTE Confidence: 0.821179502

 $00{:}41{:}33.600 \dashrightarrow 00{:}41{:}36.869$ So either really Congrats also on the,

NOTE Confidence: 0.821179502

 $00{:}41{:}36.870 \dashrightarrow 00{:}41{:}39.240$ you know the nature communications paper.

NOTE Confidence: 0.821179502

00:41:39.240 --> 00:41:42.140 And I was wondering with the NR2F2 story,

00:41:42.140 --> 00:41:44.265 you're probably aware of Chris,

NOTE Confidence: 0.821179502

 $00{:}41{:}44{.}270 \dashrightarrow 00{:}41{:}49{.}184$ Marines work in Nature last fall on TCF 4.

NOTE Confidence: 0.821179502

00:41:49.190 --> 00:41:52.598 And I'm kind of wondering if you think

NOTE Confidence: 0.821179502

00:41:52.598 --> 00:41:55.102 your NR 2F2 is upstream of TCF four if

NOTE Confidence: 0.821179502

 $00{:}41{:}55{.}102 \dashrightarrow 00{:}41{:}57{.}102$ you've seen any role there and there's

NOTE Confidence: 0.821179502

 $00{:}41{:}57.102 \dashrightarrow 00{:}41{:}59.027$ probably going to be a subsequent

NOTE Confidence: 0.821179502

00:41:59.027 --> 00:42:01.287 story about resistance to therapies,

NOTE Confidence: 0.821179502

 $00:42:01.290 \longrightarrow 00:42:02.230$ so there's.

NOTE Confidence: 0.821179502

 $00{:}42{:}02{.}230 \dashrightarrow 00{:}42{:}04{.}580$ Multiple parts to this question.

NOTE Confidence: 0.821179502

 $00:42:04.580 \rightarrow 00:42:06.260$ A, the upstream downstream part,

NOTE Confidence: 0.821179502

 $00:42:06.260 \longrightarrow 00:42:08.055$ but then also the heterogeneity

NOTE Confidence: 0.821179502

 $00:42:08.055 \rightarrow 00:42:09.850$ because you're talking about these

NOTE Confidence: 0.821179502

 $00{:}42{:}09{.}906 \dashrightarrow 00{:}42{:}11{.}806$ things happening sort of uniformly.

NOTE Confidence: 0.821179502

 $00{:}42{:}11.810 \dashrightarrow 00{:}42{:}13.670$ But I'm imagining that the epigenetic

NOTE Confidence: 0.821179502

00:42:13.670 -> 00:42:15.648 regulation is kind of cell by cell

NOTE Confidence: 0.821179502

 $00:42:15.648 \longrightarrow 00:42:17.230$ and that there's going to be a

- NOTE Confidence: 0.821179502
- $00:42:17.287 \rightarrow 00:42:18.865$ population of cells that have more
- NOTE Confidence: 0.821179502
- $00:42:18.865 \longrightarrow 00:42:22.458$ or less of that along the way.
- NOTE Confidence: 0.821179502
- $00:42:22.460 \longrightarrow 00:42:23.540$ All excellent questions and and
- NOTE Confidence: 0.821179502
- $00:42:23.540 \longrightarrow 00:42:24.840$ these are all the questions that
- NOTE Confidence: 0.821179502
- $00:42:24.840 \longrightarrow 00:42:25.908$ we are trying to address now.
- NOTE Confidence: 0.821179502
- 00:42:25.910 --> 00:42:26.346 So,
- NOTE Confidence: 0.821179502
- $00:42:26.346 \longrightarrow 00:42:29.398$ so we didn't see a total overlap
- NOTE Confidence: 0.821179502
- $00{:}42{:}29{.}398 \dashrightarrow 00{:}42{:}32{.}290$ between our population of N2F2
- NOTE Confidence: 0.821179502
- $00{:}42{:}32{.}290 \dashrightarrow 00{:}42{:}35{.}250$ with Chris Marines population.
- NOTE Confidence: 0.821179502
- $00:42:35.250 \rightarrow 00:42:38.694$ It seems like his population is smaller
- NOTE Confidence: 0.821179502
- $00:42:38.694 \rightarrow 00:42:42.767$ one inner two seems to be more broadly.
- NOTE Confidence: 0.821179502
- $00{:}42{:}42{.}770 \dashrightarrow 00{:}42{:}47{.}190$ I expressed in both EMT and also partially
- NOTE Confidence: 0.821179502
- $00{:}42{:}47.190 \dashrightarrow 00{:}42{:}49.020$ in the neural Crest like population.
- NOTE Confidence: 0.821179502
- $00{:}42{:}49{.}020 \dashrightarrow 00{:}42{:}51{.}180$ So his population seems to be a more,
- NOTE Confidence: 0.821179502
- 00:42:51.180 --> 00:42:52.436 I wouldn't say minority,
- NOTE Confidence: 0.821179502

 $00:42:52.436 \longrightarrow 00:42:54.006$ but it's a smaller population

NOTE Confidence: 0.821179502

 $00:42:54.006 \rightarrow 00:42:55.739$ and not not directly overlap.

NOTE Confidence: 0.840730844782609

 $00{:}42{:}58{.}330 \dashrightarrow 00{:}43{:}00{.}563$ You also ask the other challenge that

NOTE Confidence: 0.840730844782609

 $00:43:00.563 \longrightarrow 00:43:03.503$ we have is that a lot of the single

NOTE Confidence: 0.840730844782609

 $00{:}43{:}03{.}503 \dashrightarrow 00{:}43{:}05{.}779$ cell analysis that have allowed us to.

NOTE Confidence: 0.840730844782609

 $00:43:05.780 \longrightarrow 00:43:07.110$ Distinguish this.

NOTE Confidence: 0.840730844782609

00:43:07.110 --> 00:43:09.770 Transcriptionist states don't have,

NOTE Confidence: 0.840730844782609

 $00:43:09.770 \longrightarrow 00:43:11.877$ don't allow us, don't don't have the

NOTE Confidence: 0.840730844782609

 $00{:}43{:}11.877 \dashrightarrow 00{:}43{:}14.058$ death to look into isoforms, right?

NOTE Confidence: 0.840730844782609

 $00{:}43{:}14.058 \dashrightarrow 00{:}43{:}17.002$ So you really have to have a different

NOTE Confidence: 0.840730844782609

 $00:43:17.002 \rightarrow 00:43:19.275$ library preparation and pipeline to

NOTE Confidence: 0.840730844782609

 $00:43:19.275 \rightarrow 00:43:22.023$ identify the different types of forms.

NOTE Confidence: 0.840730844782609

 $00:43:22.030 \longrightarrow 00:43:23.836$ So when people look at R2,

NOTE Confidence: 0.840730844782609

 $00{:}43{:}23{.}840 \dashrightarrow 00{:}43{:}26{.}808$ they are just examining an R2 ISO one,

NOTE Confidence: 0.840730844782609

 $00:43:26.810 \longrightarrow 00:43:29.477$ the full length and the one that

NOTE Confidence: 0.840730844782609

 $00:43:29.477 \longrightarrow 00:43:31.509$ is really switching from the
- NOTE Confidence: 0.840730844782609
- $00:43:31.509 \rightarrow 00:43:33.549$ the primary to the metastatic,
- NOTE Confidence: 0.840730844782609
- $00:43:33.550 \rightarrow 00:43:36.054$ the one that is really triggering the EMT.
- NOTE Confidence: 0.840730844782609
- 00:43:36.060 --> 00:43:38.610 Like program is only isoform
- NOTE Confidence: 0.840730844782609
- $00:43:38.610 \longrightarrow 00:43:40.650$ 2 because of this.
- NOTE Confidence: 0.840730844782609
- $00{:}43{:}40.650 \dashrightarrow 00{:}43{:}42.450$ Balance between home and headliners.
- NOTE Confidence: 0.793306258125
- $00:43:44.690 \longrightarrow 00:43:46.060$ Yeah. So we we're definitely
- NOTE Confidence: 0.793306258125
- $00:43:46.060 \rightarrow 00:43:47.783$ trying to understand what is the
- NOTE Confidence: 0.793306258125
- $00:43:47.783 \rightarrow 00:43:49.268$ error key between these pathways,
- NOTE Confidence: 0.793306258125
- $00:43:49.270 \longrightarrow 00:43:51.377$ where is upstream, it seems to be
- NOTE Confidence: 0.793306258125
- 00:43:51.377 00:43:53.730 one of the critical regulators.
- NOTE Confidence: 0.793306258125
- $00:43:53.730 \longrightarrow 00:43:56.856$ So again when we do this.
- NOTE Confidence: 0.793306258125
- 00:43:56.860 --> 00:43:59.040 Um, analysis is sending analysis,
- NOTE Confidence: 0.793306258125
- $00:43:59.040 \longrightarrow 00:44:00.336$ but still we don't know which
- NOTE Confidence: 0.793306258125
- $00:44:00.336 \rightarrow 00:44:01.822$ one is upstream of which, right?
- NOTE Confidence: 0.793306258125
- $00:44:01.822 \rightarrow 00:44:03.634$ Yeah, that's a very good question.
- NOTE Confidence: 0.793306258125

 $00:44:03.640 \longrightarrow 00:44:05.068$ And then you ask.

NOTE Confidence: 0.793306258125

 $00{:}44{:}05{.}068 \dashrightarrow 00{:}44{:}06{.}853$ The dynamics right of this

NOTE Confidence: 0.793306258125

 $00:44:06.853 \rightarrow 00:44:08.510$ of this population.

NOTE Confidence: 0.793306258125

 $00:44:08.510 \longrightarrow 00:44:10.792$ So we are now in the process

NOTE Confidence: 0.793306258125

 $00{:}44{:}10.792 \dashrightarrow 00{:}44{:}12.124$ of labeling endogenously this

NOTE Confidence: 0.793306258125

 $00{:}44{:}12{.}124 \dashrightarrow 00{:}44{:}14{.}168$ isoform to be able to trace them.

NOTE Confidence: 0.793306258125

 $00{:}44{:}14{.}170 \dashrightarrow 00{:}44{:}17{.}195$ We want to understand whether

NOTE Confidence: 0.793306258125

00:44:17.195 --> 00:44:19.787 these isoforms are, you know,

NOTE Confidence: 0.793306258125

00:44:19.787 --> 00:44:21.623 dynamically regulated during the

NOTE Confidence: 0.793306258125

 $00:44:21.623 \longrightarrow 00:44:23.795$ metastatic process and we have

NOTE Confidence: 0.793306258125

 $00{:}44{:}23.795 \dashrightarrow 00{:}44{:}25.239$ different labeling systems now

NOTE Confidence: 0.793306258125

 $00{:}44{:}25{.}239 \dashrightarrow 00{:}44{:}27{.}682$ that allow us to sell to monitor

NOTE Confidence: 0.793306258125

 $00{:}44{:}27.682 \dashrightarrow 00{:}44{:}29.570$ single cells during metastasis.

NOTE Confidence: 0.793306258125

 $00:44:29.570 \longrightarrow 00:44:31.282$ So I hope we will have soon an

NOTE Confidence: 0.793306258125

 $00:44:31.282 \rightarrow 00:44:33.134$ answer for that, but we don't know.

NOTE Confidence: 0.793306258125

 $00:44:33.134 \rightarrow 00:44:35.650$ I I also dissipate that it would change.

- NOTE Confidence: 0.793306258125
- $00:44:35.650 \longrightarrow 00:44:37.846$ From the primary tumor as the
- NOTE Confidence: 0.793306258125
- $00:44:37.850 \longrightarrow 00:44:39.685$ cells switch from the more
- NOTE Confidence: 0.793306258125
- 00:44:39.685 00:44:41.520 proliferative to the more invasive,
- NOTE Confidence: 0.793306258125
- $00:44:41.520 \longrightarrow 00:44:43.122$ and then back when they arrive
- NOTE Confidence: 0.793306258125
- $00:44:43.122 \longrightarrow 00:44:44.970$ to them at the static side.
- NOTE Confidence: 0.793306258125
- 00:44:44.970 --> 00:44:46.385 But still we don't have
- NOTE Confidence: 0.793306258125
- $00:44:46.385 \longrightarrow 00:44:47.517$ full evidence for that.
- NOTE Confidence: 0.854771814210526
- $00:44:50.660 \longrightarrow 00:44:52.697$ So, but there were a couple of
- NOTE Confidence: 0.854771814210526
- $00{:}44{:}52.697 \dashrightarrow 00{:}44{:}55.367$ questions in the chat both relating to
- NOTE Confidence: 0.854771814210526
- $00{:}44{:}55{.}367 \dashrightarrow 00{:}44{:}57{.}532$ Alzheimer's disease and brain metastases.
- NOTE Confidence: 0.854771814210526
- $00:44:57.540 \longrightarrow 00:45:00.144$ One is there an increased incidence
- NOTE Confidence: 0.854771814210526
- $00{:}45{:}00{.}144 \dashrightarrow 00{:}45{:}02{.}370$ in brain metastases amount Alzheimer's
- NOTE Confidence: 0.854771814210526
- $00{:}45{:}02{.}370 \dashrightarrow 00{:}45{:}05{.}310$ disease patients and the other one was
- NOTE Confidence: 0.854771814210526
- $00{:}45{:}05{.}310 \dashrightarrow 00{:}45{:}08{.}208$ whether a postmortem you see Alzheimer's
- NOTE Confidence: 0.854771814210526
- $00{:}45{:}08{.}208 \dashrightarrow 00{:}45{:}11{.}190$ clogs in patients with brain metastases.
- NOTE Confidence: 0.854771814210526

 $00:45:11.190 \longrightarrow 00:45:12.254$ Good questions.

NOTE Confidence: 0.854771814210526

 $00:45:12.254 \rightarrow 00:45:15.446$ So we haven't seen brain metastasis,

NOTE Confidence: 0.854771814210526

 $00:45:15.450 \rightarrow 00:45:17.788$ we haven't seen plaques in brain metastasis.

NOTE Confidence: 0.854771814210526

 $00:45:17.790 \longrightarrow 00:45:19.410$ We look for for them,

NOTE Confidence: 0.854771814210526

 $00:45:19.410 \longrightarrow 00:45:22.170$ we don't think that they get to accumulate,

NOTE Confidence: 0.854771814210526

 $00{:}45{:}22.170 \dashrightarrow 00{:}45{:}25.082$ so we don't, we don't think that the

NOTE Confidence: 0.854771814210526

 $00:45:25.082 \rightarrow 00:45:27.860$ processing is wrong is to simply induced.

NOTE Confidence: 0.854771814210526

 $00:45:27.860 \rightarrow 00:45:30.708$ So we see more soluble abeta being produced,

NOTE Confidence: 0.854771814210526

 $00:45:30.710 \longrightarrow 00:45:32.742$ but we don't see oligomers and we don't

NOTE Confidence: 0.854771814210526

 $00:45:32.742 \longrightarrow 00:45:34.586$ see plaques. We did that staining.

NOTE Confidence: 0.854771814210526

 $00{:}45{:}34{.}586 \dashrightarrow 00{:}45{:}36{.}590$ That conversion is done by pathologists

NOTE Confidence: 0.854771814210526

 $00{:}45{:}36.647 \dashrightarrow 00{:}45{:}38.861$ to to look at the plaques and we couldn't

NOTE Confidence: 0.854771814210526

 $00{:}45{:}38.861 \dashrightarrow 00{:}45{:}41.029$ see either in our models nor in human.

NOTE Confidence: 0.854771814210526

 $00{:}45{:}41.030 \dashrightarrow 00{:}45{:}44.556$ Examples and that's the reason why we

NOTE Confidence: 0.854771814210526

 $00{:}45{:}44.556 \dashrightarrow 00{:}45{:}46.724$ think that the antibodies that we wanna

NOTE Confidence: 0.854771814210526

 $00:45:46.724 \rightarrow 00:45:48.566$ try are antibodies that design against

NOTE Confidence: 0.854771814210526

 $00:45:48.566 \rightarrow 00:45:50.815$ the soluble and libetta and not against

NOTE Confidence: 0.854771814210526

 $00{:}45{:}50{.}815 \dashrightarrow 00{:}45{:}52{.}913$ the plaques which are the ones that

NOTE Confidence: 0.854771814210526

 $00:45:52.913 \rightarrow 00:45:54.880$ have been now or the oligomers which

NOTE Confidence: 0.854771814210526

 $00:45:54.944 \rightarrow 00:45:56.855$ have been now approved by the FDA.

NOTE Confidence: 0.854771814210526

00:45:56.860 - 00:45:59.278 Now you ask the other question,

NOTE Confidence: 0.854771814210526

 $00:45:59.280 \longrightarrow 00:46:01.350$ the incidence in in Alzheimer's.

NOTE Confidence: 0.854771814210526

 $00{:}46{:}01{.}350 \dashrightarrow 00{:}46{:}05{.}855$ So we'll look into that and we didn't see

NOTE Confidence: 0.854771814210526

 $00{:}46{:}05{.}855 \dashrightarrow 00{:}46{:}09{.}380$ a epidemiological studies and association

NOTE Confidence: 0.854771814210526

 $00{:}46{:}09{.}380 \dashrightarrow 00{:}46{:}12.870$ between Alzheimer's and brain metastasis.

NOTE Confidence: 0.854771814210526

 $00:46:12.870 \longrightarrow 00:46:15.040$ But normally remember that in the majority

NOTE Confidence: 0.854771814210526

 $00:46:15.040 \rightarrow 00:46:16.490$ of the neurodegenerative disorders,

NOTE Confidence: 0.854771814210526

 $00{:}46{:}16{.}490 \dashrightarrow 00{:}46{:}19{.}000$ there is an inverse correlation

NOTE Confidence: 0.854771814210526

 $00{:}46{:}19.000 \dashrightarrow 00{:}46{:}21.008$ between cancer incidence and

NOTE Confidence: 0.854771814210526

 $00{:}46{:}21.010 \dashrightarrow 00{:}46{:}22.316$ neurodegenerative disorders,

NOTE Confidence: 0.854771814210526

 $00{:}46{:}22.316$ --> $00{:}46{:}24.928$ particularly Alzheimer's and Parkinson's.

NOTE Confidence: 0.854771814210526

 $00:46:24.930 \longrightarrow 00:46:26.155$ And even though there is

NOTE Confidence: 0.854771814210526

 $00{:}46{:}26.155 \dashrightarrow 00{:}46{:}27.135$ not a positive correlation,

NOTE Confidence: 0.854771814210526

 $00{:}46{:}27.140 \dashrightarrow 00{:}46{:}29.430$ there is no negative association.

NOTE Confidence: 0.854771814210526

 $00:46:29.430 \longrightarrow 00:46:32.328$ Now there is a reported association

NOTE Confidence: 0.854771814210526

 $00:46:32.328 \longrightarrow 00:46:34.260$ between Parkinson's and and

NOTE Confidence: 0.854771814210526

00:46:34.337 --> 00:46:36.329 Melanoma brain metastases.

NOTE Confidence: 0.854771814210526

00:46:36.330 --> 00:46:39.266 And I mean I think Harry,

NOTE Confidence: 0.854771814210526

 $00:46:39.266 \rightarrow 00:46:42.653$ this is skeptical or not, I see you.

NOTE Confidence: 0.854771814210526

 $00{:}46{:}42.653 \dashrightarrow 00{:}46{:}44.658$ No, there's a slight increase,

NOTE Confidence: 0.854771814210526

00:46:44.660 --> 00:46:47.316 but anyway, I don't know that it's brain

NOTE Confidence: 0.854771814210526

 $00{:}46{:}47{.}316 \dashrightarrow 00{:}46{:}49{.}820$ metastases specifically, it's just.

NOTE Confidence: 0.854771814210526

 $00:46:49.820 \longrightarrow 00:46:50.660$ Come on. I'm sorry. Sorry.

NOTE Confidence: 0.854771814210526

00:46:50.660 --> 00:46:51.850 Yeah, yeah, yeah. I misspoke.

NOTE Confidence: 0.854771814210526

00:46:51.850 --> 00:46:53.266 I I meant that. I know my kid.

NOTE Confidence: 0.854771814210526

 $00:46:53.270 \longrightarrow 00:46:54.120$ Right.

NOTE Confidence: 0.854771814210526

 $00:46:54.120 \longrightarrow 00:46:54.970$ Yeah.

- NOTE Confidence: 0.854771814210526
- 00:46:54.970 --> 00:46:55.820 Yeah.
- NOTE Confidence: 0.854771814210526
- 00:46:55.820 --> 00:46:56.195 No,
- NOTE Confidence: 0.854771814210526
- $00:46:56.195 \rightarrow 00:46:58.070$ but there's a really interesting
- NOTE Confidence: 0.854771814210526
- $00{:}46{:}58.070 \dashrightarrow 00{:}46{:}58.820$ observation there.
- NOTE Confidence: 0.854771814210526
- $00{:}46{:}58{.}820 \dashrightarrow 00{:}47{:}00{.}636$ So thank you and thank you so much
- NOTE Confidence: 0.854771814210526
- $00{:}47{:}00{.}636 \dashrightarrow 00{:}47{:}02{.}259$ for this amazing presentation.
- NOTE Confidence: 0.854771814210526
- $00:47:02.260 \longrightarrow 00:47:03.500$ I have more questions,
- NOTE Confidence: 0.854771814210526
- $00:47:03.500 \rightarrow 00:47:05.850$ but I'm going to e-mail them to you.
- NOTE Confidence: 0.854771814210526
- $00:47:05.850 \dashrightarrow 00:47:07.570$ I don't believe there anymore in the chat.
- NOTE Confidence: 0.854771814210526
- $00:47:07.570 \longrightarrow 00:47:09.340$ We appreciate your patience with all
- NOTE Confidence: 0.854771814210526
- $00:47:09.340 \longrightarrow 00:47:10.874$ the technical challenges and thanks
- NOTE Confidence: 0.854771814210526
- 00:47:10.874 --> 00:47:12.389 for virtually visiting next time.
- NOTE Confidence: 0.854771814210526
- $00:47:12.390 \longrightarrow 00:47:13.902$ It'll be in person.
- NOTE Confidence: 0.854771814210526
- $00{:}47{:}13.902 \dashrightarrow 00{:}47{:}15.792$ Thanks for wonderful talk to
- NOTE Confidence: 0.854771814210526
- $00:47:15.792 \longrightarrow 00:47:17.409$ really fascinating work.
- NOTE Confidence: 0.854771814210526

00:47:17.410 --> 00:47:18.058 Thank you,

NOTE Confidence: 0.854771814210526

00:47:18.058 --> 00:47:18.382 Harriet.

NOTE Confidence: 0.854771814210526

 $00:47:18.382 \longrightarrow 00:47:20.002$ Looking forward to see you

NOTE Confidence: 0.854771814210526

 $00:47:20.002 \longrightarrow 00:47:21.709$ soon and thanks everybody.

NOTE Confidence: 0.854771814210526

 $00:47:21.710 \longrightarrow 00:47:22.160$ Bye.