

WEBVTT

NOTE duration:"00:16:00.5400000"

NOTE language:en-us

NOTE Confidence: 0.843998

00:00:00.000 --> 00:00:01.785 It's my pleasure to introduce

NOTE Confidence: 0.843998

00:00:01.785 --> 00:00:03.910 our final speaker of the day.

NOTE Confidence: 0.843998

00:00:03.910 --> 00:00:04.544 Doctor Muenkel,

NOTE Confidence: 0.843998

00:00:04.544 --> 00:00:06.446 like he is an assistant professor

NOTE Confidence: 0.843998

00:00:06.446 --> 00:00:08.450 in the Department of Genetics.

NOTE Confidence: 0.843998

00:00:08.450 --> 00:00:10.249 He got his PhD at the University

NOTE Confidence: 0.843998

00:00:10.249 --> 00:00:11.896 of Sydney and did postdoctoral

NOTE Confidence: 0.843998

00:00:11.896 --> 00:00:13.528 training at Massachusetts General

NOTE Confidence: 0.843998

00:00:13.528 --> 00:00:15.780 Hospital and the Broad Institute.

NOTE Confidence: 0.843998

00:00:15.780 --> 00:00:18.104 His lab here at Yale is focused

NOTE Confidence: 0.843998

00:00:18.104 --> 00:00:20.308 on the genetics of rare disease,

NOTE Confidence: 0.843998

00:00:20.310 --> 00:00:21.710 an understanding genetic mechanisms

NOTE Confidence: 0.843998

00:00:21.710 --> 00:00:22.760 of neuromuscular diseases,

NOTE Confidence: 0.843998

00:00:22.760 --> 00:00:24.848 and as a patient with a

NOTE Confidence: 0.843998

00:00:24.848 --> 00:00:25.892 rare muscular dystrophy,

NOTE Confidence: 0.843998

00:00:25.900 --> 00:00:28.000 Dr Lake is very passionate about

NOTE Confidence: 0.843998

00:00:28.000 --> 00:00:29.050 translating genetic discoveries

NOTE Confidence: 0.843998

00:00:29.050 --> 00:00:30.419 into targeted therapies.

NOTE Confidence: 0.843998

00:00:30.420 --> 00:00:31.440 Thank you Doctor

NOTE Confidence: 0.826636

00:00:31.440 --> 00:00:33.992 like thank you and thank you for the

NOTE Confidence: 0.826636

00:00:33.992 --> 00:00:36.552 kind introduction and thank you for the

NOTE Confidence: 0.826636

00:00:36.552 --> 00:00:38.412 organizers for putting together wonderful

NOTE Confidence: 0.826636

00:00:38.477 --> 00:00:41.029 workshop and also give me the honor and

NOTE Confidence: 0.826636

00:00:41.029 --> 00:00:43.365 opportunity to present on some of the

NOTE Confidence: 0.826636

00:00:43.365 --> 00:00:46.108 fantastic work that my lab is working on.

NOTE Confidence: 0.826636

00:00:46.110 --> 00:00:49.064 So I'll be talking about translating genetic

NOTE Confidence: 0.826636

00:00:49.064 --> 00:00:51.000 discoveries into targeted therapies.

NOTE Confidence: 0.826636

00:00:51.000 --> 00:00:53.944 I have no conflicts of interest to disclose,

NOTE Confidence: 0.826636

00:00:53.950 --> 00:00:57.262 so my lab works on the full patient journey.

NOTE Confidence: 0.826636

00:00:57.270 --> 00:00:59.524 So from the diagnostic Odyssey that the
NOTE Confidence: 0.826636

00:00:59.524 --> 00:01:02.824 young we had touched upon all the way to
NOTE Confidence: 0.826636

00:01:02.824 --> 00:01:05.020 the development of individualized therapies,
NOTE Confidence: 0.826636

00:01:05.020 --> 00:01:07.240 and I can reflect upon this,
NOTE Confidence: 0.826636

00:01:07.240 --> 00:01:08.712 this whole patient journey
NOTE Confidence: 0.826636

00:01:08.712 --> 00:01:10.552 through reflecting on my own.
NOTE Confidence: 0.826636

00:01:10.560 --> 00:01:13.136 So I went on a diagnostic Odyssey,
NOTE Confidence: 0.826636

00:01:13.140 --> 00:01:16.356 took over 10 years to actually find the
NOTE Confidence: 0.826636

00:01:16.356 --> 00:01:18.607 gene underlying my ran your muscular
NOTE Confidence: 0.826636

00:01:18.607 --> 00:01:21.329 disease and you can see on the key.
NOTE Confidence: 0.826636

00:01:21.330 --> 00:01:23.508 The gene, the mutation, they fight,
NOTE Confidence: 0.826636

00:01:23.510 --> 00:01:26.044 found finally found an and this an.
NOTE Confidence: 0.826636

00:01:26.050 --> 00:01:28.246 Nowadays we can actually rapidly shorten
NOTE Confidence: 0.826636

00:01:28.246 --> 00:01:30.468 this diagnostic Odyssey time by using
NOTE Confidence: 0.826636

00:01:30.468 --> 00:01:32.574 genomic technologies such as xom sequencing,
NOTE Confidence: 0.826636

00:01:32.580 --> 00:01:34.400 genome sequencing and RNA sequencing,

NOTE Confidence: 0.826636

00:01:34.400 --> 00:01:37.296 and also developing methods to get at that.

NOTE Confidence: 0.826636

00:01:37.300 --> 00:01:40.124 And that's one of the things that my

NOTE Confidence: 0.826636

00:01:40.124 --> 00:01:42.485 lab focuses on and but still there's

NOTE Confidence: 0.826636

00:01:42.485 --> 00:01:45.355 so much work to be done because the

NOTE Confidence: 0.826636

00:01:45.355 --> 00:01:47.779 diagnosis rate is still only about

NOTE Confidence: 0.826636

00:01:47.779 --> 00:01:50.368 50% for a lot of rare diseases,

NOTE Confidence: 0.826636

00:01:50.370 --> 00:01:51.922 and when patients actually.

NOTE Confidence: 0.826636

00:01:51.922 --> 00:01:53.862 Find out their genetic diagnosis.

NOTE Confidence: 0.826636

00:01:53.870 --> 00:01:56.198 They can work on specific disease

NOTE Confidence: 0.826636

00:01:56.198 --> 00:01:57.750 management such as stretching.

NOTE Confidence: 0.826636

00:01:57.750 --> 00:02:01.233 As you can see here in this example here,

NOTE Confidence: 0.826636

00:02:01.240 --> 00:02:03.928 but one of the most exciting things

NOTE Confidence: 0.826636

00:02:03.928 --> 00:02:06.262 that has only become possible in

NOTE Confidence: 0.826636

00:02:06.262 --> 00:02:09.390 the last five years or so is there.

NOTE Confidence: 0.826636

00:02:09.390 --> 00:02:11.330 The concept of development of

NOTE Confidence: 0.826636

00:02:11.330 --> 00:02:12.106 individualized therapies.
NOTE Confidence: 0.826636

00:02:12.110 --> 00:02:14.721 I had the good fortune that my
NOTE Confidence: 0.826636

00:02:14.721 --> 00:02:16.728 collaborators and colleagues at the
NOTE Confidence: 0.826636

00:02:16.728 --> 00:02:18.372 University of Massachusetts Medical
NOTE Confidence: 0.826636

00:02:18.372 --> 00:02:21.029 School one dashly work on my mutation,
NOTE Confidence: 0.826636

00:02:21.030 --> 00:02:22.578 which was which was.
NOTE Confidence: 0.7905694

00:02:26.110 --> 00:02:31.198 Oh sorry. Which was working on
NOTE Confidence: 0.7905694

00:02:31.198 --> 00:02:33.926 the 8 basepair duplication.
NOTE Confidence: 0.7905694

00:02:33.930 --> 00:02:36.114 And designing a custom crispr target
NOTE Confidence: 0.7905694

00:02:36.114 --> 00:02:39.258 that and to to cleanly remove one copy
NOTE Confidence: 0.7905694

00:02:39.258 --> 00:02:41.688 of the eight base pair duplication.
NOTE Confidence: 0.7905694

00:02:41.690 --> 00:02:44.794 So and they actually perform this on a
NOTE Confidence: 0.7905694

00:02:44.794 --> 00:02:47.508 skin biopsy I gave and they created.
NOTE Confidence: 0.7905694

00:02:47.510 --> 00:02:49.078 I PS cell line.
NOTE Confidence: 0.7905694

00:02:49.078 --> 00:02:51.780 It was able to achieve nearly 80%
NOTE Confidence: 0.7905694

00:02:51.780 --> 00:02:54.174 correction of my cells and this

NOTE Confidence: 0.7905694

00:02:54.174 --> 00:02:56.429 was published in Nature in 2019.

NOTE Confidence: 0.7905694

00:02:56.430 --> 00:02:59.922 So we were inspired by this effort and story.

NOTE Confidence: 0.7905694

00:02:59.930 --> 00:03:02.618 And we wanted Ashley replicate some of

NOTE Confidence: 0.7905694

00:03:02.618 --> 00:03:06.109 this idea of the individualized therapy so.

NOTE Confidence: 0.7905694

00:03:06.110 --> 00:03:08.891 So I'm going to talk for the rest of

NOTE Confidence: 0.7905694

00:03:08.891 --> 00:03:11.410 touch upon on the rest of the talk.

NOTE Confidence: 0.7905694

00:03:11.410 --> 00:03:12.970 A story of the patient.

NOTE Confidence: 0.7905694

00:03:12.970 --> 00:03:14.848 So the patient here is Terry,

NOTE Confidence: 0.7905694

00:03:14.850 --> 00:03:17.027 who has a rare form of Duchene,

NOTE Confidence: 0.7905694

00:03:17.030 --> 00:03:18.896 muscular dystrophy and his brother here,

NOTE Confidence: 0.7905694

00:03:18.900 --> 00:03:20.796 Richt, who created our foundation to

NOTE Confidence: 0.7905694

00:03:20.796 --> 00:03:22.959 actually find a therapy for his brother.

NOTE Confidence: 0.7905694

00:03:22.960 --> 00:03:25.120 And you can read a little bit more

NOTE Confidence: 0.7905694

00:03:25.120 --> 00:03:27.172 about their story on a Harvard

NOTE Confidence: 0.7905694

00:03:27.172 --> 00:03:28.255 Business School article.

NOTE Confidence: 0.7905694

00:03:28.260 --> 00:03:30.556 And so we set up on this project
NOTE Confidence: 0.7905694

00:03:30.556 --> 00:03:32.320 in the summer of 2018.
NOTE Confidence: 0.7905694

00:03:32.320 --> 00:03:34.567 So this was shortly after I saw
NOTE Confidence: 0.7905694

00:03:34.567 --> 00:03:36.190 the lab here at Yale.
NOTE Confidence: 0.7905694

00:03:36.190 --> 00:03:38.218 And this is the genetic report,
NOTE Confidence: 0.7905694

00:03:38.220 --> 00:03:40.548 and it's very typical for genetic report of
NOTE Confidence: 0.7905694

00:03:40.548 --> 00:03:42.950 a patient with Duchene muscular dystrophy.
NOTE Confidence: 0.7905694

00:03:42.950 --> 00:03:45.983 So this patient has an X on one deletion,
NOTE Confidence: 0.7905694

00:03:45.990 --> 00:03:48.265 so it's it's quite a large deletion
NOTE Confidence: 0.7905694

00:03:48.265 --> 00:03:50.692 that takes out Exxon one and the
NOTE Confidence: 0.7905694

00:03:50.692 --> 00:03:53.090 promoter region of the muscle Exxon one.
NOTE Confidence: 0.7905694

00:03:53.090 --> 00:03:54.875 So the first thing we wanted to
NOTE Confidence: 0.7905694

00:03:54.875 --> 00:03:57.149 do is a full characterization.
NOTE Confidence: 0.7905694

00:03:57.150 --> 00:03:59.166 So we performed whole genome sequencing.
NOTE Confidence: 0.7905694

00:03:59.170 --> 00:04:01.198 So for those that aren't familiar,
NOTE Confidence: 0.7905694

00:04:01.200 --> 00:04:02.865 follow genome sequencing,

NOTE Confidence: 0.7905694

00:04:02.865 --> 00:04:04.530 these reads represent.

NOTE Confidence: 0.7905694

00:04:04.530 --> 00:04:06.525 Next generation sequencing reads and

NOTE Confidence: 0.7905694

00:04:06.525 --> 00:04:09.490 where they map to the human genome.

NOTE Confidence: 0.7905694

00:04:09.490 --> 00:04:13.538 And you can see here in this particular

NOTE Confidence: 0.7905694

00:04:13.538 --> 00:04:16.718 case this is the X chromosome.

NOTE Confidence: 0.7905694

00:04:16.720 --> 00:04:18.628 And in particular the DMD gene.

NOTE Confidence: 0.7905694

00:04:18.630 --> 00:04:21.166 And keeping in mind the DMD gene goes,

NOTE Confidence: 0.7905694

00:04:21.170 --> 00:04:22.760 is on the negative strand,

NOTE Confidence: 0.7905694

00:04:22.760 --> 00:04:24.668 so goes from right to left.

NOTE Confidence: 0.7905694

00:04:24.670 --> 00:04:26.896 And here is muscle X on one,

NOTE Confidence: 0.7905694

00:04:26.900 --> 00:04:29.084 so in the patient we see no

NOTE Confidence: 0.7905694

00:04:29.084 --> 00:04:30.390 sequencing reads mapping here,

NOTE Confidence: 0.7905694

00:04:30.390 --> 00:04:32.496 and this indicates that there is

NOTE Confidence: 0.7905694

00:04:32.496 --> 00:04:35.014 a large deletion and if you look

NOTE Confidence: 0.7905694

00:04:35.014 --> 00:04:36.744 carefully here at the mother.

NOTE Confidence: 0.7905694

00:04:36.750 --> 00:04:39.690 You can see the histogram here that
NOTE Confidence: 0.7905694

00:04:39.690 --> 00:04:42.049 represents coverage that we see this
NOTE Confidence: 0.7905694

00:04:42.049 --> 00:04:44.450 dip and this dip is approximately 50%
NOTE Confidence: 0.7905694

00:04:44.450 --> 00:04:47.607 and this represents that the mother is
NOTE Confidence: 0.7905694

00:04:47.607 --> 00:04:50.298 a heterozygous carrier of this deletion.
NOTE Confidence: 0.7905694

00:04:50.300 --> 00:04:52.592 We also performed RNA sequencing on
NOTE Confidence: 0.7905694

00:04:52.592 --> 00:04:55.520 the patient and so for those are not
NOTE Confidence: 0.7905694

00:04:55.520 --> 00:04:58.648 familiar this is what we call a sashimi plot.
NOTE Confidence: 0.7905694

00:04:58.650 --> 00:05:00.545 These arcs represent next generation
NOTE Confidence: 0.7905694

00:05:00.545 --> 00:05:02.440 sequencing reads that span from
NOTE Confidence: 0.7905694

00:05:02.502 --> 00:05:03.730 1X onto another Exon.
NOTE Confidence: 0.7905694

00:05:03.730 --> 00:05:04.762 So not surprisingly,
NOTE Confidence: 0.7905694

00:05:04.762 --> 00:05:07.720 we saw no read support for the muscle,
NOTE Confidence: 0.7905694

00:05:07.720 --> 00:05:10.987 the muscle isoform, and this is X on one.
NOTE Confidence: 0.7905694

00:05:10.990 --> 00:05:12.805 But surprisingly we we sorry
NOTE Confidence: 0.7905694

00:05:12.805 --> 00:05:14.620 support for the cortical isoforms,

NOTE Confidence: 0.7905694

00:05:14.620 --> 00:05:18.250 so you can see in our that goes from Exxon,

NOTE Confidence: 0.7905694

00:05:18.250 --> 00:05:20.548 one from the cortical ice form.

NOTE Confidence: 0.7905694

00:05:20.550 --> 00:05:22.428 *** on two and so forth,

NOTE Confidence: 0.7905694

00:05:22.430 --> 00:05:24.015 and the the difference between

NOTE Confidence: 0.7905694

00:05:24.015 --> 00:05:25.955 the cortical eyes from the muscle

NOTE Confidence: 0.7905694

00:05:25.955 --> 00:05:27.435 isoform is only exon one.

NOTE Confidence: 0.7905694

00:05:27.440 --> 00:05:29.000 And we've been Exxon one.

NOTE Confidence: 0.7905694

00:05:29.000 --> 00:05:31.184 Most of it is the untranslated region.

NOTE Confidence: 0.793868

00:05:31.190 --> 00:05:34.007 So the coding starts later on next on one.

NOTE Confidence: 0.793868

00:05:34.010 --> 00:05:36.187 So there's only a few amino acids

NOTE Confidence: 0.793868

00:05:36.187 --> 00:05:37.795 difference between the muscle and

NOTE Confidence: 0.793868

00:05:37.795 --> 00:05:39.959 the cortical ice form, and this is

NOTE Confidence: 0.793868

00:05:39.959 --> 00:05:42.150 important for the rest of the talk.

NOTE Confidence: 0.793868

00:05:42.150 --> 00:05:44.103 But one of the things we hypothesize

NOTE Confidence: 0.793868

00:05:44.103 --> 00:05:45.690 is could the upregulation in

NOTE Confidence: 0.793868

00:05:45.690 --> 00:05:48.084 switching on the cortical or ice form,
NOTE Confidence: 0.793868

00:05:48.090 --> 00:05:49.398 which you typically don't
NOTE Confidence: 0.793868

00:05:49.398 --> 00:05:50.706 see in skeletal muscle.
NOTE Confidence: 0.793868

00:05:50.710 --> 00:05:52.835 Can this actually be contributing
NOTE Confidence: 0.793868

00:05:52.835 --> 00:05:55.562 to some of the delayed progression
NOTE Confidence: 0.793868

00:05:55.562 --> 00:05:57.630 of the muscle disease?
NOTE Confidence: 0.793868

00:05:57.630 --> 00:06:00.368 We actually seen the patient, so.
NOTE Confidence: 0.793868

00:06:00.368 --> 00:06:03.434 When we look at the protein levels,
NOTE Confidence: 0.793868

00:06:03.440 --> 00:06:05.520 when you do Western blot you can see
NOTE Confidence: 0.793868

00:06:05.520 --> 00:06:07.441 what our collaborators at Boston
NOTE Confidence: 0.793868

00:06:07.441 --> 00:06:09.225 Children's Hospital has shown.
NOTE Confidence: 0.793868

00:06:09.230 --> 00:06:11.358 Is that the patient does have residual
NOTE Confidence: 0.793868

00:06:11.358 --> 00:06:13.329 level of dystrophin protein expression,
NOTE Confidence: 0.793868

00:06:13.330 --> 00:06:14.350 and they approximate.
NOTE Confidence: 0.793868

00:06:14.350 --> 00:06:16.700 This is about 3% of normal levels,
NOTE Confidence: 0.793868

00:06:16.700 --> 00:06:19.010 and when you look at the muscle

NOTE Confidence: 0.793868
00:06:19.080 --> 00:06:21.168 biopsy and the Histology of it,
NOTE Confidence: 0.793868
00:06:21.170 --> 00:06:23.786 you can see that in control you can
NOTE Confidence: 0.793868
00:06:23.786 --> 00:06:26.607 see this nice staining of dystrophin.
NOTE Confidence: 0.793868
00:06:26.610 --> 00:06:28.460 Represented by green at the
NOTE Confidence: 0.793868
00:06:28.460 --> 00:06:30.310 muscle membrane of these fibers.
NOTE Confidence: 0.793868
00:06:30.310 --> 00:06:32.998 While in the patient you do see some
NOTE Confidence: 0.793868
00:06:32.998 --> 00:06:35.119 dystrophin but it's patchy standing.
NOTE Confidence: 0.793868
00:06:35.120 --> 00:06:37.460 So there's some patching mosaic standing
NOTE Confidence: 0.793868
00:06:37.460 --> 00:06:39.754 and this represents what we believe
NOTE Confidence: 0.793868
00:06:39.754 --> 00:06:41.776 to be a stochastic random process.
NOTE Confidence: 0.793868
00:06:41.780 --> 00:06:44.370 So this is a snapshot in time,
NOTE Confidence: 0.793868
00:06:44.370 --> 00:06:46.464 and sometimes you do get the
NOTE Confidence: 0.793868
00:06:46.464 --> 00:06:48.440 expression of the cortical isoform,
NOTE Confidence: 0.793868
00:06:48.440 --> 00:06:52.140 but you know it's not as strong as a control,
NOTE Confidence: 0.793868
00:06:52.140 --> 00:06:56.892 and it's not in each one of the fibers.
NOTE Confidence: 0.793868

00:06:56.900 --> 00:06:57.588 And so,
NOTE Confidence: 0.793868

00:06:57.588 --> 00:06:59.996 so this shows that the patient is
NOTE Confidence: 0.793868

00:06:59.996 --> 00:07:02.126 expressing a quarter Kreisel and when
NOTE Confidence: 0.793868

00:07:02.126 --> 00:07:04.638 we look in the literature there are
NOTE Confidence: 0.793868

00:07:04.638 --> 00:07:06.816 other patients that were reported in
NOTE Confidence: 0.793868

00:07:06.816 --> 00:07:09.866 the late 80s and early 90s that Ashley
NOTE Confidence: 0.793868

00:07:09.866 --> 00:07:12.520 have a muscle Exon one deletion.
NOTE Confidence: 0.793868

00:07:12.520 --> 00:07:12.884 However,
NOTE Confidence: 0.793868

00:07:12.884 --> 00:07:14.704 the interesting thing about these
NOTE Confidence: 0.793868

00:07:14.704 --> 00:07:16.999 patients is that they don't actually
NOTE Confidence: 0.793868

00:07:16.999 --> 00:07:18.949 have a skeletal muscle phenotype,
NOTE Confidence: 0.793868

00:07:18.950 --> 00:07:21.449 so they don't have dish in muscular
NOTE Confidence: 0.793868

00:07:21.449 --> 00:07:24.239 dystrophy or any muscle phenotype at all.
NOTE Confidence: 0.793868

00:07:24.240 --> 00:07:26.184 But they still have a cardiac
NOTE Confidence: 0.793868

00:07:26.184 --> 00:07:28.343 phenotype and what we believe is
NOTE Confidence: 0.793868

00:07:28.343 --> 00:07:30.713 happening here is there's this complex

NOTE Confidence: 0.793868

00:07:30.713 --> 00:07:32.368 interplay of enhancers surrounding

NOTE Confidence: 0.793868

00:07:32.368 --> 00:07:35.175 the muscle Exxon one and also the

NOTE Confidence: 0.793868

00:07:35.175 --> 00:07:37.092 cortical Exxon which they've called

NOTE Confidence: 0.793868

00:07:37.092 --> 00:07:39.360 the brain isoform in this paper,

NOTE Confidence: 0.793868

00:07:39.360 --> 00:07:41.664 and that when you get the

NOTE Confidence: 0.793868

00:07:41.664 --> 00:07:43.520 deletion of the muscle X1,

NOTE Confidence: 0.793868

00:07:43.520 --> 00:07:45.470 the other enhancers switches on.

NOTE Confidence: 0.793868

00:07:45.470 --> 00:07:47.710 And actually turn on the cortical ice

NOTE Confidence: 0.793868

00:07:47.710 --> 00:07:50.368 form and they turn it on high enough

NOTE Confidence: 0.793868

00:07:50.368 --> 00:07:52.482 in skeletal muscle that these patients

NOTE Confidence: 0.793868

00:07:52.482 --> 00:07:54.960 don't actually have a muscle disease.

NOTE Confidence: 0.793868

00:07:54.960 --> 00:07:55.329 However,

NOTE Confidence: 0.793868

00:07:55.329 --> 00:07:58.281 what we think is happening with the patient

NOTE Confidence: 0.793868

00:07:58.281 --> 00:08:01.203 is that he his deletion is a lot larger,

NOTE Confidence: 0.793868

00:08:01.210 --> 00:08:03.674 takes out some of these enhancers so

NOTE Confidence: 0.793868

00:08:03.674 --> 00:08:06.058 the cortical ice form can switch on,
NOTE Confidence: 0.793868

00:08:06.060 --> 00:08:08.384 but not at the levels that is
NOTE Confidence: 0.793868

00:08:08.384 --> 00:08:10.230 actually happening in the patient,
NOTE Confidence: 0.793868

00:08:10.230 --> 00:08:12.750 so I'm going to stop and pause you
NOTE Confidence: 0.793868

00:08:12.750 --> 00:08:15.430 so we have a very good example.
NOTE Confidence: 0.793868

00:08:15.430 --> 00:08:17.670 A human example that in the absence
NOTE Confidence: 0.793868

00:08:17.670 --> 00:08:19.250 of a muscle isoform,
NOTE Confidence: 0.793868

00:08:19.250 --> 00:08:21.420 if you can switch on the cortical
NOTE Confidence: 0.793868

00:08:21.420 --> 00:08:23.593 isoform that this could actually perform
NOTE Confidence: 0.793868

00:08:23.593 --> 00:08:25.945 a genetic rescue and actually save.
NOTE Confidence: 0.793868

00:08:25.950 --> 00:08:27.905 From having a muscle disease
NOTE Confidence: 0.793868

00:08:27.905 --> 00:08:30.244 phenotype and this is motivated us
NOTE Confidence: 0.793868

00:08:30.244 --> 00:08:32.463 to actually do this for the patient,
NOTE Confidence: 0.793868

00:08:32.470 --> 00:08:34.504 will develop a therapy for the
NOTE Confidence: 0.793868

00:08:34.504 --> 00:08:35.860 patient so so the
NOTE Confidence: 0.8324777

00:08:35.937 --> 00:08:38.611 ability to actually switch on and robustly

NOTE Confidence: 0.8324777

00:08:38.611 --> 00:08:41.880 turn on the cortical ice form in one way.

NOTE Confidence: 0.8324777

00:08:41.880 --> 00:08:44.768 You can do this is by using CRISPR,

NOTE Confidence: 0.8324777

00:08:44.770 --> 00:08:47.666 so in this case a dead cast line,

NOTE Confidence: 0.8324777

00:08:47.670 --> 00:08:50.542 so the ability to home into a particular

NOTE Confidence: 0.8324777

00:08:50.542 --> 00:08:53.099 place in the genome and bind and.

NOTE Confidence: 0.8324777

00:08:53.100 --> 00:08:55.996 But in this case not cause a double

NOTE Confidence: 0.8324777

00:08:55.996 --> 00:08:58.064 stranded break, but bring in.

NOTE Confidence: 0.8324777

00:08:58.064 --> 00:08:59.856 Inscription activating these transcription

NOTE Confidence: 0.8324777

00:08:59.856 --> 00:09:02.012 activator allows for the expression

NOTE Confidence: 0.8324777

00:09:02.012 --> 00:09:04.250 so it's not the best transcription

NOTE Confidence: 0.8324777

00:09:04.250 --> 00:09:05.870 activated that you could use,

NOTE Confidence: 0.8324777

00:09:05.870 --> 00:09:08.186 but we had translation in mind.

NOTE Confidence: 0.8324777

00:09:08.190 --> 00:09:11.601 It was small enough to package into a Navy

NOTE Confidence: 0.8324777

00:09:11.601 --> 00:09:14.744 which has only a 4.7 KB packaging limit,

NOTE Confidence: 0.8324777

00:09:14.750 --> 00:09:17.487 so everything we've done has been designed

NOTE Confidence: 0.8324777

00:09:17.487 --> 00:09:19.770 with translation mine and not rescuing.

NOTE Confidence: 0.8324777

00:09:19.770 --> 00:09:21.310 Save particularly the cells

NOTE Confidence: 0.8324777

00:09:21.310 --> 00:09:22.850 or the mouse itself.

NOTE Confidence: 0.8324777

00:09:22.850 --> 00:09:25.112 But how can this actually translate

NOTE Confidence: 0.8324777

00:09:25.112 --> 00:09:27.100 to a human clinical trial?

NOTE Confidence: 0.8324777

00:09:27.100 --> 00:09:28.632 So we also picked.

NOTE Confidence: 0.8324777

00:09:28.632 --> 00:09:30.930 Skeletal muscle promoter called CCA D

NOTE Confidence: 0.8324777

00:09:31.003 --> 00:09:33.547 This is their truncated small synthetic

NOTE Confidence: 0.8324777

00:09:33.547 --> 00:09:36.616 promoter that has also been used in

NOTE Confidence: 0.8324777

00:09:36.616 --> 00:09:39.214 clinical trials already for mini dystrophin,

NOTE Confidence: 0.8324777

00:09:39.220 --> 00:09:43.119 so it has a good safety profile.

NOTE Confidence: 0.8324777

00:09:43.120 --> 00:09:45.424 So next thing we wanted to do is

NOTE Confidence: 0.8324777

00:09:45.424 --> 00:09:47.737 going over all the possible guides

NOTE Confidence: 0.8324777

00:09:47.737 --> 00:09:49.817 so upstream of the transcription

NOTE Confidence: 0.8324777

00:09:49.817 --> 00:09:51.941 start site or the code acquires

NOTE Confidence: 0.8324777

00:09:51.941 --> 00:09:54.286 form that we could bind and possibly

NOTE Confidence: 0.8324777

00:09:54.286 --> 00:09:56.374 switch on the cortical eyes form.

NOTE Confidence: 0.8324777

00:09:56.380 --> 00:10:00.030 So this was done in human cells and we found

NOTE Confidence: 0.8324777

00:10:00.119 --> 00:10:03.359 that the C7 one of the cortical guides.

NOTE Confidence: 0.8324777

00:10:03.360 --> 00:10:05.810 One of the guides targeting the cortical

NOTE Confidence: 0.8324777

00:10:05.810 --> 00:10:08.577 ice form seems to have good upregulation,

NOTE Confidence: 0.8324777

00:10:08.580 --> 00:10:11.562 so the next thing we wanted

NOTE Confidence: 0.8324777

00:10:11.562 --> 00:10:14.160 to do is then ask.

NOTE Confidence: 0.8324777

00:10:14.160 --> 00:10:16.696 How the performance would look like in vivo.

NOTE Confidence: 0.8324777

00:10:16.700 --> 00:10:18.280 So we're very fortunate 44.

NOTE Confidence: 0.8324777

00:10:18.280 --> 00:10:20.800 DMD research that there is a range of

NOTE Confidence: 0.8324777

00:10:20.800 --> 00:10:23.036 great mouse models that you can use,

NOTE Confidence: 0.8324777

00:10:23.040 --> 00:10:27.837 but the best mouse model you can use for.

NOTE Confidence: 0.8324777

00:10:27.840 --> 00:10:29.645 Very specific genetic therapies such

NOTE Confidence: 0.8324777

00:10:29.645 --> 00:10:32.239 as this is a transgenic mouse model,

NOTE Confidence: 0.8324777

00:10:32.240 --> 00:10:34.075 so this transgenic mouse has

NOTE Confidence: 0.8324777

00:10:34.075 --> 00:10:35.543 the whole DMD gene,
NOTE Confidence: 0.8324777

00:10:35.550 --> 00:10:37.310 so a 2.6 megabass fragmente
NOTE Confidence: 0.8324777

00:10:37.310 --> 00:10:39.580 that not only has the axons,
NOTE Confidence: 0.8324777

00:10:39.580 --> 00:10:42.016 but also has the introns and also
NOTE Confidence: 0.8324777

00:10:42.016 --> 00:10:43.620 the flanking intergenic regions.
NOTE Confidence: 0.8324777

00:10:43.620 --> 00:10:45.460 So guides that we've designed,
NOTE Confidence: 0.8324777

00:10:45.460 --> 00:10:49.051 and we've proven to work in the human cell
NOTE Confidence: 0.8324777

00:10:49.051 --> 00:10:52.367 model can also work in this mouse model.
NOTE Confidence: 0.8324777

00:10:52.370 --> 00:10:54.170 And in an in addition,
NOTE Confidence: 0.8324777

00:10:54.170 --> 00:10:56.690 this mouse more has the MTX mutation.
NOTE Confidence: 0.8324777

00:10:56.690 --> 00:10:58.866 So at the mouse locus for the DMD
NOTE Confidence: 0.8324777

00:10:58.866 --> 00:11:01.684 gene has a nonsense mutation and can't
NOTE Confidence: 0.8324777

00:11:01.684 --> 00:11:03.889 actually express the mouse dystrophin.
NOTE Confidence: 0.8324777

00:11:03.890 --> 00:11:05.846 So the only dystrophin this mouse
NOTE Confidence: 0.8324777

00:11:05.846 --> 00:11:07.850 expresses is the human dystrophin.
NOTE Confidence: 0.8324777

00:11:07.850 --> 00:11:09.929 And this was this was given to

NOTE Confidence: 0.8324777

00:11:09.929 --> 00:11:12.528 us by our collaborators at UCLA.

NOTE Confidence: 0.8324777

00:11:12.530 --> 00:11:15.421 So the mouse study we ran with

NOTE Confidence: 0.8324777

00:11:15.421 --> 00:11:18.939 was a low dose and a high dose.

NOTE Confidence: 0.8324777

00:11:18.940 --> 00:11:21.600 At four weeks in eight weeks looking

NOTE Confidence: 0.8324777

00:11:21.600 --> 00:11:24.815 for the UP regulation as what we saw

NOTE Confidence: 0.8324777

00:11:24.815 --> 00:11:27.998 in the human cell model and the reason

NOTE Confidence: 0.8324777

00:11:27.998 --> 00:11:31.022 why we picked this dose is this dose.

NOTE Confidence: 0.8324777

00:11:31.022 --> 00:11:33.082 Ashley correlates to a human

NOTE Confidence: 0.8324777

00:11:33.082 --> 00:11:36.054 clinical trial of 3×10 to the

NOTE Confidence: 0.8324777

00:11:36.054 --> 00:11:38.382 3rd 14 vector genomes per kilo.

NOTE Confidence: 0.8324777

00:11:38.390 --> 00:11:40.054 Kilogram is the largest.

NOTE Confidence: 0.8324777

00:11:40.054 --> 00:11:42.550 The highest dose that has ever

NOTE Confidence: 0.8324777

00:11:42.635 --> 00:11:44.755 been used safely in delivering

NOTE Confidence: 0.8324777

00:11:44.755 --> 00:11:47.755 effectors such as this in in a

NOTE Confidence: 0.8324777

00:11:47.755 --> 00:11:49.755 systemic way to skeletal muscle.

NOTE Confidence: 0.82745665

00:11:49.760 --> 00:11:52.856 So the results I'm going to show you are
NOTE Confidence: 0.82745665

00:11:52.856 --> 00:11:55.667 the results from the high dose eight
NOTE Confidence: 0.82745665

00:11:55.667 --> 00:11:58.587 week mice where we have two untreated
NOTE Confidence: 0.82745665

00:11:58.587 --> 00:12:01.881 mice and four treated mice and you can
NOTE Confidence: 0.82745665

00:12:01.881 --> 00:12:05.400 see that we do get on an RNA level.
NOTE Confidence: 0.82745665

00:12:05.400 --> 00:12:07.724 Looking at qPCR, we do see upregulation
NOTE Confidence: 0.82745665

00:12:07.724 --> 00:12:10.479 in in cardiac tissue and disappointingly,
NOTE Confidence: 0.82745665

00:12:10.480 --> 00:12:12.988 the UP regulations in other skeletal
NOTE Confidence: 0.82745665

00:12:12.988 --> 00:12:15.777 muscles are not just not as high
NOTE Confidence: 0.82745665

00:12:15.777 --> 00:12:17.517 and in non muscle tissues.
NOTE Confidence: 0.82745665

00:12:17.520 --> 00:12:19.956 We don't see any regulation or.
NOTE Confidence: 0.82745665

00:12:19.960 --> 00:12:24.040 And and this was expected because we news.
NOTE Confidence: 0.82745665

00:12:24.040 --> 00:12:26.476 A skeletal muscle specific ice form.
NOTE Confidence: 0.82583815

00:12:29.530 --> 00:12:31.994 So what are the lies is so
NOTE Confidence: 0.82583815

00:12:31.994 --> 00:12:33.510 like preference for harm?
NOTE Confidence: 0.82583815

00:12:33.510 --> 00:12:35.766 Is that the AAV 9 that will be

NOTE Confidence: 0.82583815

00:12:35.766 --> 00:12:38.220 using to deliver that therapeutic?

NOTE Confidence: 0.82583815

00:12:38.220 --> 00:12:39.588 Well? Not surprisingly,

NOTE Confidence: 0.82583815

00:12:39.588 --> 00:12:43.615 most of that goes to the liver when we

NOTE Confidence: 0.82583815

00:12:43.615 --> 00:12:46.930 look when we isolate the DNA an look at.

NOTE Confidence: 0.82583815

00:12:46.930 --> 00:12:48.202 The viral copy numbers,

NOTE Confidence: 0.82583815

00:12:48.202 --> 00:12:51.017 but also you can see that a lot of

NOTE Confidence: 0.82583815

00:12:51.017 --> 00:12:53.467 it goes to the cardiac tissue with a

NOTE Confidence: 0.82583815

00:12:53.467 --> 00:12:56.029 preference toward versus to skeletal muscle,

NOTE Confidence: 0.82583815

00:12:56.030 --> 00:12:59.414 and this is something that the field has

NOTE Confidence: 0.82583815

00:12:59.414 --> 00:13:03.086 found that AAV 9 has a preference for heart.

NOTE Confidence: 0.82583815

00:13:03.090 --> 00:13:05.568 So using this data and other

NOTE Confidence: 0.82583815

00:13:05.568 --> 00:13:07.220 data that we've generated,

NOTE Confidence: 0.82583815

00:13:07.220 --> 00:13:09.726 but I don't have time to present

NOTE Confidence: 0.82583815

00:13:09.726 --> 00:13:11.844 and will very well successful

NOTE Confidence: 0.82583815

00:13:11.844 --> 00:13:14.239 with a pre Ind application.

NOTE Confidence: 0.82583815

00:13:14.240 --> 00:13:16.490 And since then we've replicated this
NOTE Confidence: 0.82583815

00:13:16.490 --> 00:13:18.904 data on multiple other mouse cohorts
NOTE Confidence: 0.82583815

00:13:18.904 --> 00:13:21.454 and also at other AAV production
NOTE Confidence: 0.82583815

00:13:21.454 --> 00:13:23.329 facilities producing robust results.
NOTE Confidence: 0.82583815

00:13:23.330 --> 00:13:26.597 But the future work we have to do after
NOTE Confidence: 0.82583815

00:13:26.597 --> 00:13:29.860 talking to the FDA is we'd like to
NOTE Confidence: 0.82583815

00:13:29.860 --> 00:13:32.000 measure increased protein expression.
NOTE Confidence: 0.82583815

00:13:32.000 --> 00:13:34.140 So what I've shown you?
NOTE Confidence: 0.82583815

00:13:34.140 --> 00:13:36.000 Is increased in RNA expression,
NOTE Confidence: 0.82583815

00:13:36.000 --> 00:13:38.646 but we'd also like to show an
NOTE Confidence: 0.82583815

00:13:38.646 --> 00:13:40.710 increase in protein expression.
NOTE Confidence: 0.82583815

00:13:40.710 --> 00:13:42.978 One of the challenges is that
NOTE Confidence: 0.82583815

00:13:42.978 --> 00:13:44.490 the cortical muscle isoforms
NOTE Confidence: 0.82583815

00:13:44.558 --> 00:13:46.580 are very similar to each other.
NOTE Confidence: 0.82583815

00:13:46.580 --> 00:13:48.834 It's only a few amino acids at
NOTE Confidence: 0.82583815

00:13:48.834 --> 00:13:50.990 the intern that Steve different.

NOTE Confidence: 0.82583815

00:13:50.990 --> 00:13:53.559 We're trying to create also an exon,

NOTE Confidence: 0.82583815

00:13:53.560 --> 00:13:55.028 one knockout mouse model,

NOTE Confidence: 0.82583815

00:13:55.028 --> 00:13:56.129 but there's difficulties

NOTE Confidence: 0.82583815

00:13:56.129 --> 00:13:57.230 naturally creating that.

NOTE Confidence: 0.82583815

00:13:57.230 --> 00:13:59.652 So what we're working towards is just

NOTE Confidence: 0.82583815

00:13:59.652 --> 00:14:02.051 showing an increase in overall protein

NOTE Confidence: 0.82583815

00:14:02.051 --> 00:14:03.775 expression and therefore inferring

NOTE Confidence: 0.82583815

00:14:03.775 --> 00:14:06.378 that the increase is actually due to

NOTE Confidence: 0.82583815

00:14:06.378 --> 00:14:08.628 the increase in the cortical ice form.

NOTE Confidence: 0.82583815

00:14:08.628 --> 00:14:11.732 The other thing we're working on is the

NOTE Confidence: 0.82583815

00:14:11.732 --> 00:14:14.295 development of a robust patient cell model.

NOTE Confidence: 0.82583815

00:14:14.300 --> 00:14:16.185 And this is necessarily necessary

NOTE Confidence: 0.82583815

00:14:16.185 --> 00:14:18.070 to determine off target effects.

NOTE Confidence: 0.82583815

00:14:18.070 --> 00:14:20.709 So what are the genes weight would?

NOTE Confidence: 0.82583815

00:14:20.710 --> 00:14:23.032 We may be switching on other

NOTE Confidence: 0.82583815

00:14:23.032 --> 00:14:24.860 than the dystrophin isoform so,
NOTE Confidence: 0.82583815

00:14:24.860 --> 00:14:27.092 but we believe that there probably
NOTE Confidence: 0.82583815

00:14:27.092 --> 00:14:29.343 shouldn't be any off target effects
NOTE Confidence: 0.82583815

00:14:29.343 --> 00:14:32.103 given that a lot of these off target
NOTE Confidence: 0.82583815

00:14:32.174 --> 00:14:34.568 sites are actually into Jenny and
NOTE Confidence: 0.82583815

00:14:34.568 --> 00:14:36.507 shouldn't be switching on genes.
NOTE Confidence: 0.82583815

00:14:36.507 --> 00:14:38.529 And the last is functional assays
NOTE Confidence: 0.82583815

00:14:38.529 --> 00:14:40.405 that show the restoration of
NOTE Confidence: 0.82583815

00:14:40.405 --> 00:14:42.799 dystrophin at the cell membrane in
NOTE Confidence: 0.82583815

00:14:42.799 --> 00:14:45.248 these patients else comes along with
NOTE Confidence: 0.82583815

00:14:45.248 --> 00:14:47.223 the restoration of other complexes.
NOTE Confidence: 0.82583815

00:14:47.230 --> 00:14:49.744 Such as the dystrophin complex
NOTE Confidence: 0.82583815

00:14:49.744 --> 00:14:52.740 and also the dystrophin complex.
NOTE Confidence: 0.82583815

00:14:52.740 --> 00:14:55.708 So our lab is very excited that
NOTE Confidence: 0.82583815

00:14:55.708 --> 00:14:57.830 we're in this new era.
NOTE Confidence: 0.82583815

00:14:57.830 --> 00:15:00.315 This shift from personalized medicine

NOTE Confidence: 0.82583815
00:15:00.315 --> 00:15:02.303 to individualized medicine and
NOTE Confidence: 0.82583815
00:15:02.303 --> 00:15:04.707 the the contrast is an explained
NOTE Confidence: 0.82583815
00:15:04.707 --> 00:15:07.398 well by Peter Marks from the FDA
NOTE Confidence: 0.82583815
00:15:07.398 --> 00:15:09.368 is that personalized medicine is
NOTE Confidence: 0.82583815
00:15:09.368 --> 00:15:11.726 when you profile patient and use
NOTE Confidence: 0.82583815
00:15:11.726 --> 00:15:14.078 off the shelf drugs and therapies
NOTE Confidence: 0.82583815
00:15:14.078 --> 00:15:16.458 personalized based on their profile
NOTE Confidence: 0.82583815
00:15:16.458 --> 00:15:17.895 while individualized medicine.
NOTE Confidence: 0.82583815
00:15:17.900 --> 00:15:19.590 Is looking at a patient's
NOTE Confidence: 0.82583815
00:15:19.590 --> 00:15:21.280 mutation and developing a therapy
NOTE Confidence: 0.82583815
00:15:21.340 --> 00:15:22.828 specific to that mutation,
NOTE Confidence: 0.82583815
00:15:22.830 --> 00:15:25.126 and so there are two exciting clinical
NOTE Confidence: 0.82583815
00:15:25.126 --> 00:15:27.294 trials that are going on right
NOTE Confidence: 0.82583815
00:15:27.294 --> 00:15:29.154 now and using CRISPR technologies.
NOTE Confidence: 0.82583815
00:15:29.160 --> 00:15:31.272 One is sickle cell which is
NOTE Confidence: 0.82583815

00:15:31.272 --> 00:15:32.680 using ex vivo approach.
NOTE Confidence: 0.82583815

00:15:32.680 --> 00:15:35.046 The other one is delivery to the
NOTE Confidence: 0.82583815

00:15:35.046 --> 00:15:36.794 eye for inherited blindness and
NOTE Confidence: 0.82583815

00:15:36.794 --> 00:15:39.111 with that I would like to thank
NOTE Confidence: 0.82583815

00:15:39.111 --> 00:15:41.005 the audience for their attention
NOTE Confidence: 0.82583815

00:15:41.005 --> 00:15:43.237 and a huge thanks to Karen
NOTE Confidence: 0.8503214

00:15:43.240 --> 00:15:45.000 K You from my lab.
NOTE Confidence: 0.8503214

00:15:45.000 --> 00:15:47.904 Have driven a lot of the research that.
NOTE Confidence: 0.8503214

00:15:47.910 --> 00:15:50.320 I've presented today our funders,
NOTE Confidence: 0.8503214

00:15:50.320 --> 00:15:51.718 cure a disease,
NOTE Confidence: 0.8503214

00:15:51.718 --> 00:15:54.048 and also a wonderful collaborators
NOTE Confidence: 0.8503214

00:15:54.048 --> 00:15:56.090 academic and also industry,
NOTE Confidence: 0.8503214

00:15:56.090 --> 00:15:58.495 particularly Charles River and our
NOTE Confidence: 0.8503214

00:15:58.495 --> 00:16:00.540 collaborators at UMass, thanks.