WEBVTT

NOTE duration:"00:38:11.1760000"

NOTE language:en-us

NOTE Confidence: 0.8013401

00:00:00.000 --> 00:00:02.520 OK, so welcome. I'm going to be

NOTE Confidence: 0.8013401

00:00:02.520 --> 00:00:04.529 talking about hematopoietic stem and

NOTE Confidence: 0.8013401

00:00:04.529 --> 00:00:06.261 progenitor cell fate specification

NOTE Confidence: 0.8013401

00:00:06.261 --> 00:00:08.465 in health predominantly and disease

NOTE Confidence: 0.8013401

00:00:08.465 --> 00:00:10.500 and I just loved megakaryocytes,

NOTE Confidence: 0.8013401

00:00:10.500 --> 00:00:13.332 so I have a picture of a gorgeous

NOTE Confidence: 0.8013401

00:00:13.332 --> 00:00:16.028 megakaryocyte here you can see all of

NOTE Confidence: 0.8013401

00:00:16.028 --> 00:00:17.938 the nuclei inside this megakaryocyte.

NOTE Confidence: 0.8013401

00:00:17.940 --> 00:00:19.424 This megakaryocyte became polyploid

NOTE Confidence: 0.8013401

00:00:19.424 --> 00:00:21.286 in vitro, so the nuclei,

NOTE Confidence: 0.8013401

00:00:21.286 --> 00:00:23.518 or separate and abstained the cytoskeleton.

NOTE Confidence: 0.8013401

00:00:23.520 --> 00:00:26.194 The actin and tubulin and you can

NOTE Confidence: 0.8013401

00:00:26.194 --> 00:00:29.510 just see how gorgeous these cells are.

NOTE Confidence: 0.8013401

00:00:29.510 --> 00:00:32.759 So I have to be able to go forward.

NOTE Confidence: 0.8013401

00:00:32.760 --> 00:00:33.831 There we go.

NOTE Confidence: 0.8013401

00:00:33.831 --> 00:00:36.330 So this is just the hematopoietic tree

NOTE Confidence: 0.8013401

00:00:36.404 --> 00:00:39.113 for anybody who is new to him out of

NOTE Confidence: 0.8013401

00:00:39.113 --> 00:00:42.145 police is these are hematopoietic stem cells.

NOTE Confidence: 0.8013401

00:00:42.150 --> 00:00:43.694 They divide and differentiate,

NOTE Confidence: 0.8013401

00:00:43.694 --> 00:00:45.624 become hematopoetic progenitor cells and

NOTE Confidence: 0.8013401

00:00:45.624 --> 00:00:47.867 then the classic tree shows that they

NOTE Confidence: 0.8013401

00:00:47.867 --> 00:00:49.730 split into common lymphoid progenitors,

NOTE Confidence: 0.8013401

00:00:49.730 --> 00:00:52.376 which will make the B&T cells and NK cells

NOTE Confidence: 0.8013401

00:00:52.376 --> 00:00:54.778 and Common myeloid progenitor cells,

NOTE Confidence: 0.8013401

00:00:54.780 --> 00:00:56.664 amongst which then you get me

NOTE Confidence: 0.8013401

00:00:56.664 --> 00:00:58.750 P Mega Rich Royd Projectors,

NOTE Confidence: 0.8013401

00:00:58.750 --> 00:01:01.766 which is what I'll be focusing on today.

NOTE Confidence: 0.8013401

00:01:01.770 --> 00:01:03.114 Since it's bright orange

NOTE Confidence: 0.8013401

00:01:03.114 --> 00:01:04.458 granulocyte monocyte for janitors,

NOTE Confidence: 0.8013401

00:01:04.460 --> 00:01:06.560 the maker annulus sites and monocytes

NOTE Confidence: 0.8013401

00:01:06.560 --> 00:01:09.134 and these mega rich right for janitors

NOTE Confidence: 0.8013401

00:01:09.134 --> 00:01:10.979 that make the megakaryocytes and

NOTE Confidence: 0.8013401

00:01:10.979 --> 00:01:13.084 are it's red cells and we produce

NOTE Confidence: 0.8013401

00:01:13.084 --> 00:01:15.158 about 2 * 10 to the 11th.

NOTE Confidence: 0.8013401

00:01:15.158 --> 00:01:17.560 New platelets and red blood cells daily,

NOTE Confidence: 0.8013401

00:01:17.560 --> 00:01:19.814 and if you calculate that down it's

NOTE Confidence: 0.8013401

00:01:19.814 --> 00:01:21.882 about 2 million platelets in 2

NOTE Confidence: 0.8013401

00:01:21.882 --> 00:01:23.946 million red cells at every second.

NOTE Confidence: 0.8013401

00:01:23.950 --> 00:01:25.630 So these MVP are very,

NOTE Confidence: 0.8013401

00:01:25.630 --> 00:01:28.108 very busy and we'd like to better

NOTE Confidence: 0.8013401

00:01:28.108 --> 00:01:30.216 understand how the MVP Self Renew

NOTE Confidence: 0.8013401

00:01:30.216 --> 00:01:32.166 and choose a fate to become.

NOTE Confidence: 0.8013401

00:01:32.170 --> 00:01:35.010 With Droid Versus Megakaryocytic.

NOTE Confidence: 0.8013401

00:01:35.010 --> 00:01:36.678 This image just shows that there

NOTE Confidence: 0.8013401

00:01:36.678 --> 00:01:38.394 are over 14 million platelet

NOTE Confidence: 0.8013401

00:01:38.394 --> 00:01:40.038 transfusions worldwide annually,

NOTE Confidence: 0.8013401

00:01:40.040 --> 00:01:42.420 which we would like to be able

NOTE Confidence: 0.8013401

00:01:42.420 --> 00:01:44.699 to decrease the need for donors.

NOTE Confidence: 0.8013401

00:01:44.700 --> 00:01:47.129 An if there were ways of making

NOTE Confidence: 0.8013401

00:01:47.129 --> 00:01:49.264 platelets in vitro that would be

NOTE Confidence: 0.8013401

00:01:49.264 --> 00:01:51.644 great and this is from an excellent

NOTE Confidence: 0.8013401

00:01:51.718 --> 00:01:53.968 video that might all talk will

NOTE Confidence: 0.8013401

00:01:53.968 --> 00:01:55.833 be available on YouTube later.

NOTE Confidence: 0.8013401

00:01:55.833 --> 00:01:58.374 Jonathan has a video of Platelet formation

NOTE Confidence: 0.8013401

00:01:58.374 --> 00:02:00.858 and this these images are from that.

NOTE Confidence: 0.8013401

00:02:00.860 --> 00:02:01.576 It's spectacular.

NOTE Confidence: 0.8013401

00:02:01.576 --> 00:02:03.008 I highly recommend it.

NOTE Confidence: 0.8013401

00:02:03.010 --> 00:02:03.378 OK,

NOTE Confidence: 0.8013401

00:02:03.378 --> 00:02:05.954 so why do we study the megakaryocyte?

NOTE Confidence: 0.8013401

00:02:05.960 --> 00:02:08.040 Erythroid progenitor or me P,

NOTE Confidence: 0.8013401

00:02:08.040 --> 00:02:10.764 which makes Mega Mega Carey site

NOTE Confidence: 0.8013401

00:02:10.764 --> 00:02:12.580 progenitors which make megakaryocytes

NOTE Confidence: 0.8013401

00:02:12.652 --> 00:02:14.364 and platelets and erythroid

NOTE Confidence: 0.8013401

00:02:14.364 --> 00:02:16.932 progenitors that make red blood cells.

NOTE Confidence: 0.8013401

00:02:16.940 --> 00:02:18.816 So we studied this for lots of

NOTE Confidence: 0.8013401

00:02:18.816 --> 00:02:20.728 reasons and you guys could probably

NOTE Confidence: 0.8013401

00:02:20.728 --> 00:02:22.822 come up with additional good ones.

NOTE Confidence: 0.8013401

00:02:22.830 --> 00:02:24.951 But first of all it just by

NOTE Confidence: 0.8013401

00:02:24.951 --> 00:02:27.170 learning how the MVP makes it date,

NOTE Confidence: 0.8013401

00:02:27.170 --> 00:02:27.800 it's fate.

NOTE Confidence: 0.8013401

00:02:27.800 --> 00:02:29.375 Decisions were learning really about

NOTE Confidence: 0.8013401

00:02:29.375 --> 00:02:30.890 cell fate decisions in general,

NOTE Confidence: 0.8013401

00:02:30.890 --> 00:02:32.440 basics of stem cell biology,

NOTE Confidence: 0.8013401

00:02:32.440 --> 00:02:34.300 what makes us Ella stem cell?

NOTE Confidence: 0.8013401

00:02:34.300 --> 00:02:36.470 What makes a cell at progenitor cell,

NOTE Confidence: 0.8013401

00:02:36.470 --> 00:02:38.640 and how does it make its fate?

NOTE Confidence: 0.8013401

00:02:38.640 --> 00:02:40.190 Decisions for regenerative medicine purposes,

NOTE Confidence: 0.8013401

00:02:40.190 --> 00:02:42.050 it's good to understand how this

NOTE Confidence: 0.8013401

00:02:42.050 --> 00:02:43.290 actually occurs in vivo,

NOTE Confidence: 0.8013401

00:02:43.290 --> 00:02:45.318 so that we can improve our

NOTE Confidence: 0.8013401

00:02:45.318 --> 00:02:46.670 approaches for making platelets

NOTE Confidence: 0.8013401

00:02:46.729 --> 00:02:48.415 and red blood cells in vitro.

NOTE Confidence: 0.8013401

00:02:48.420 --> 00:02:49.424 For patients,

NOTE Confidence: 0.8013401

00:02:49.424 --> 00:02:51.934 and also because understanding this

NOTE Confidence: 0.8013401

00:02:51.934 --> 00:02:54.779 entire lineages and how it's regulated

NOTE Confidence: 0.8013401

00:02:54.779 --> 00:02:57.733 will lead to the discovery of potential

NOTE Confidence: 0.8013401

00:02:57.806 --> 00:03:00.278 targets for disorders of the Mega.

NOTE Confidence: 0.8013401

00:03:00.280 --> 00:03:01.800 Carey's itinerary thread lineages.

NOTE Confidence: 0.8013401

00:03:01.800 --> 00:03:04.080 So how can one distinguish an

NOTE Confidence: 0.8016149

00:03:04.142 --> 00:03:05.806 Emmy PA megathread progenitor

NOTE Confidence: 0.8016149

00:03:05.806 --> 00:03:07.470 cell functionally in FINA?

NOTE Confidence: 0.8016149

00:03:07.470 --> 00:03:09.648 Typically this is a challenge that

NOTE Confidence: 0.8016149

00:03:09.648 --> 00:03:11.975 was initially taken up by Ched Sonata

NOTE Confidence: 0.8016149

00:03:11.975 --> 00:03:14.170 when he was a postdoc in my lab,

NOTE Confidence: 0.8016149

00:03:14.170 --> 00:03:15.760 and then followed up by

NOTE Confidence: 0.8016149

00:03:15.760 --> 00:03:16.714 Juliana Javie Ferruccio,

NOTE Confidence: 0.8016149

00:03:16.720 --> 00:03:18.841 who then finish the work and what

NOTE Confidence: 0.8016149

00:03:18.841 --> 00:03:21.340 they did is they figured out a way

NOTE Confidence: 0.8016149

00:03:21.340 --> 00:03:23.740 where you start with a single cell.

NOTE Confidence: 0.8016149

00:03:23.740 --> 00:03:26.284 You hope it's a mega rich Rd progenitor,

NOTE Confidence: 0.8016149

00:03:26.290 --> 00:03:28.530 and if you keep that cell in

NOTE Confidence: 0.8016149

00:03:28.530 --> 00:03:30.119 culture for about 2 weeks,

NOTE Confidence: 0.8016149

00:03:30.120 --> 00:03:32.584 it will form a colony of cells where

NOTE Confidence: 0.8016149

00:03:32.584 --> 00:03:34.707 the only cells in that colony.

NOTE Confidence: 0.8016149

00:03:34.710 --> 00:03:36.708 Are megakaryocytes and red blood cells,

NOTE Confidence: 0.8016149

00:03:36.710 --> 00:03:39.830 so this is this was the goal and this is

NOTE Confidence: 0.8016149

00:03:39.914 --> 00:03:43.037 what Chad and you figured out how to do.

NOTE Confidence: 0.8016149

00:03:43.040 --> 00:03:44.710 So basically what they would

NOTE Confidence: 0.8016149

00:03:44.710 --> 00:03:46.700 do is fax sort candidate MPs,

NOTE Confidence: 0.8016149

00:03:46.700 --> 00:03:48.032 megathread progenitor cells and

NOTE Confidence: 0.8016149

00:03:48.032 --> 00:03:49.697 put them into semisolid media.

NOTE Confidence: 0.8016149

00:03:49.700 --> 00:03:50.813 Very, very seriously.

NOTE Confidence: 0.8016149

00:03:50.813 --> 00:03:53.410 So none of the two colonies would

NOTE Confidence: 0.8016149

00:03:53.480 --> 00:03:55.544 be bumping up against each other

NOTE Confidence: 0.8016149

00:03:55.544 --> 00:03:58.070 and then waited 13 to 15 days and

NOTE Confidence: 0.8016149

00:03:58.070 --> 00:04:00.016 at the end of that period they

NOTE Confidence: 0.8016149

00:04:00.016 --> 00:04:00.994 stained with antibodies.

NOTE Confidence: 0.8016149

00:04:00.994 --> 00:04:02.624 They staying with an antibody

NOTE Confidence: 0.8016149

00:04:02.624 --> 00:04:03.679 against Glycophorin A,

NOTE Confidence: 0.8016149

00:04:03.680 --> 00:04:06.214 which is a red blood cell antigen.

NOTE Confidence: 0.8016149

00:04:06.220 --> 00:04:07.141 And again CD41A,

NOTE Confidence: 0.8016149

00:04:07.141 --> 00:04:09.290 which is on Maggie Carey sites and

NOTE Confidence: 0.8016149

00:04:09.360 --> 00:04:11.558 you can see we have some colonies

NOTE Confidence: 0.8016149

00:04:11.558 --> 00:04:13.502 that are erythroid only and some

NOTE Confidence: 0.8016149

00:04:13.502 --> 00:04:15.386 colonies that are Mega Carey site

NOTE Confidence: 0.8016149

00:04:15.386 --> 00:04:17.668 only and then we have some colonies

NOTE Confidence: 0.8016149

00:04:17.668 --> 00:04:19.941 that have both colors in them and

NOTE Confidence: 0.8016149

00:04:19.941 --> 00:04:22.020 these are colonies that we can see.

NOTE Confidence: 0.8016149

00:04:22.020 --> 00:04:24.722 If you mke because they have both

NOTE Confidence: 0.8016149

00:04:24.722 --> 00:04:25.880 megakaryocytes and erythrocytes.

NOTE Confidence: 0.8016149

00:04:25.880 --> 00:04:27.905 So using this functional readout

NOTE Confidence: 0.8016149

00:04:27.905 --> 00:04:30.784 for a by potent progenitor chat and

NOTE Confidence: 0.8016149

00:04:30.784 --> 00:04:33.130 then shoe went through Facs sorting

NOTE Confidence: 0.8016149

00:04:33.130 --> 00:04:35.976 protocols to try to identify the best

NOTE Confidence: 0.8016149

00:04:35.976 --> 00:04:37.966 enrichment they could for primary

NOTE Confidence: 0.8016149

00:04:37.970 --> 00:04:39.630 human megathread progenitors and

NOTE Confidence: 0.8016149

00:04:39.630 --> 00:04:42.567 what they discovered is if you gate

NOTE Confidence: 0.8016149

00:04:42.567 --> 00:04:45.220 within the 34 positive Lynn negative 135.

NOTE Confidence: 0.8016149

00:04:45.220 --> 00:04:47.638 That's all three receptor negative CD,

NOTE Confidence: 0.8016149

00:04:47.640 --> 00:04:49.252 45 RA negative population.

NOTE Confidence: 0.8016149

00:04:49.252 --> 00:04:51.267 If you look for Maple,

NOTE Confidence: 0.8016149

00:04:51.270 --> 00:04:53.490 which is the receptor for thrombo

NOTE Confidence: 0.8016149

00:04:53.490 --> 00:04:55.939 poet and the vast majority of

NOTE Confidence: 0.8016149

00:04:55.939 --> 00:04:58.169 these cells are Maple positive.

NOTE Confidence: 0.8016149

00:04:58.170 --> 00:04:59.900 It's not limited to megakaryocytes,

NOTE Confidence: 0.8016149

00:04:59.900 --> 00:05:01.976 we know he mad about extending

NOTE Confidence: 0.8016149

00:05:01.976 --> 00:05:03.360 projector cells expressed nipple,

NOTE Confidence: 0.8016149

00:05:03.360 --> 00:05:05.936 but then there was also this nipple

NOTE Confidence: 0.8016149

00:05:05.936 --> 00:05:08.616 low population and if we look at that

NOTE Confidence: 0.8016149

00:05:08.616 --> 00:05:11.254 and compared to excuse me CD 38 which

NOTE Confidence: 0.8016149

00:05:11.254 --> 00:05:13.739 I'm sorry that should be labeled better.

NOTE Confidence: 0.8016149

00:05:13.740 --> 00:05:16.218 What we can find is that the

NOTE Confidence: 0.8016149

00:05:16.218 --> 00:05:17.864 erythroid progenitors tend to have

NOTE Confidence: 0.8016149

00:05:17.864 --> 00:05:19.663 less nipple and more CD 38 so

NOTE Confidence: 0.8016149

00:05:19.663 --> 00:05:22.054 if we enrich for this population

NOTE Confidence: 0.8016149

00:05:22.054 --> 00:05:24.114 they grow erythroid only colonies.

NOTE Confidence: 0.8016149

00:05:24.120 --> 00:05:26.514 If you then take this population which

NOTE Confidence: 0.8016149

00:05:26.514 --> 00:05:28.960 is really in the middle for 3038,

NOTE Confidence: 0.8016149

00:05:28.960 --> 00:05:30.952 it's not the most negative and

NOTE Confidence: 0.8016149

00:05:30.952 --> 00:05:32.980 it's not the most positive.

NOTE Confidence: 0.8016149

00:05:32.980 --> 00:05:35.360 And then you get on CD 41.

NOTE Confidence: 0.8016149

00:05:35.360 --> 00:05:37.677 You can find CD 41 positive Meg

NOTE Confidence: 0.8016149

00:05:37.677 --> 00:05:39.418 progenitors that make Mega Carey

NOTE Confidence: 0.8016149

00:05:39.418 --> 00:05:41.476 site only colonies and these MVP,

NOTE Confidence: 0.8016149

00:05:41.480 --> 00:05:43.950 which made colonies that have

NOTE Confidence: 0.8016149

00:05:43.950 --> 00:05:46.420 both Meg Anorith Royd cells.

NOTE Confidence: 0.8016149

00:05:46.420 --> 00:05:48.448 When they enriched for these populations,

NOTE Confidence: 0.8016149

00:05:48.450 --> 00:05:50.809 this is what the colonies look like,

NOTE Confidence: 0.8016149

00:05:50.810 --> 00:05:53.085 and you'll see a lot of these

NOTE Confidence: 0.8016149

00:05:53.085 --> 00:05:54.530 graphs in my talk,

NOTE Confidence: 0.8016149

00:05:54.530 --> 00:05:57.234 so I'm going to go through this slowly.

NOTE Confidence: 0.8016149

00:05:57.240 --> 00:05:58.998 What I'm showing you here on

NOTE Confidence: 0.8016149

00:05:58.998 --> 00:06:00.170 the Y axis is

NOTE Confidence: 0.86811244

00:06:00.245 --> 00:06:03.317 the number of colonies per 100 cells plated,

NOTE Confidence: 0.86811244

00:06:03.320 --> 00:06:05.786 and what I'm showing you in blue is the

NOTE Confidence: 0.86811244

00:06:05.786 --> 00:06:08.482 number of those colonies that had both

NOTE Confidence: 0.86811244

00:06:08.482 --> 00:06:10.420 megakaryocytes and erythrocytes in them,

NOTE Confidence: 0.86811244

00:06:10.420 --> 00:06:13.255 so a by potent colony and you can see

NOTE Confidence: 0.86811244

00:06:13.255 --> 00:06:15.915 about half of the colonies that grow.

NOTE Confidence: 0.86811244

00:06:15.920 --> 00:06:18.146 Have megakaryocytes and erythrocytes and E.

NOTE Confidence: 0.86811244

00:06:18.150 --> 00:06:21.126 Only colony has just erythrocytes and a Meg.

NOTE Confidence: 0.86811244

00:06:21.130 --> 00:06:22.990 Only colony has just megakaryocytes.

NOTE Confidence: 0.86811244

00:06:22.990 --> 00:06:25.958 We also enriched as I said for Meg

NOTE Confidence: 0.86811244

00:06:25.958 --> 00:06:28.995 Progenitors which are in the CD 41

NOTE Confidence: 0.86811244

00:06:28.995 --> 00:06:30.767 positive population and erythroid

NOTE Confidence: 0.86811244

00:06:30.767 --> 00:06:32.500 progenitors which are in the.

NOTE Confidence: 0.86811244

00:06:32.500 --> 00:06:36.204 Nipple low population that has more CD 38.

NOTE Confidence: 0.86811244

00:06:36.210 --> 00:06:39.180 So we were able to enrich for these cells.

NOTE Confidence: 0.86811244

00:06:39.180 --> 00:06:41.820 We always get some erythroid, only an MB.

NOTE Confidence: 0.86811244

00:06:41.820 --> 00:06:44.130 Only colonies in our cultures of MVP.

NOTE Confidence: 0.86811244

00:06:44.130 --> 00:06:46.440 So one of our questions is really,

NOTE Confidence: 0.86811244

00:06:46.440 --> 00:06:48.736 is this a uniform population where by

NOTE Confidence: 0.86811244

00:06:48.736 --> 00:06:51.098 chance some of the colonies will be

NOTE Confidence: 0.86811244

00:06:51.098 --> 00:06:53.700 erythroid only and some will be Meg only?

NOTE Confidence: 0.86811244

00:06:53.700 --> 00:06:55.305 Or we really contaminated here

NOTE Confidence: 0.86811244

00:06:55.305 --> 00:06:56.910 with some erythroid progenitors and

NOTE Confidence: 0.86811244

00:06:56.966 --> 00:06:57.989 some Meg progenitors.

NOTE Confidence: 0.86811244

00:06:57.990 --> 00:07:00.246 So what we next did is to better

NOTE Confidence: 0.86811244

00:07:00.246 --> 00:07:02.018 understand this Anne how fake

NOTE Confidence: 0.86811244

00:07:02.018 --> 00:07:03.928 decisions actually occur is performed.

NOTE Confidence: 0.86811244

00:07:03.930 --> 00:07:05.946 Single cell RNA seq on the

NOTE Confidence: 0.86811244

00:07:05.946 --> 00:07:07.660 different populations in this work.

NOTE Confidence: 0.86811244

00:07:07.660 --> 00:07:09.248 Was done by Yishun,

NOTE Confidence: 0.86811244

00:07:09.248 --> 00:07:11.630 or will Lou in my laboratory.

NOTE Confidence: 0.86811244

00:07:11.630 --> 00:07:14.980 So what he did is he fax sorted out the

NOTE Confidence: 0.86811244

00:07:15.072 --> 00:07:18.167 candidate Mega Rich Ride Projectors.

NOTE Confidence: 0.86811244

00:07:18.170 --> 00:07:19.795 The Meg Progenitors the erythroid

NOTE Confidence: 0.86811244

00:07:19.795 --> 00:07:21.420 progenitors and then the upstream

NOTE Confidence: 0.86811244

00:07:21.477 --> 00:07:22.788 common myeloid progenitors,

NOTE Confidence: 0.86811244

00:07:22.790 --> 00:07:25.798 and this was done with single cell seq

NOTE Confidence: 0.86811244

00:07:25.798 --> 00:07:28.842 that was run by Amazong in the Yale

NOTE Confidence: 0.86811244

00:07:28.842 --> 00:07:31.519 stem cell center or genomics core.

NOTE Confidence: 0.86811244

00:07:31.520 --> 00:07:33.062 The data were then analyzed with

NOTE Confidence: 0.86811244

00:07:33.062 --> 00:07:34.542 the assistance of Nathan Salamone's

NOTE Confidence: 0.86811244

00:07:34.542 --> 00:07:35.760 at University Cincinnati,

NOTE Confidence: 0.86811244

00:07:35.760 --> 00:07:38.128 and I want to take a minute to

NOTE Confidence: 0.86811244

00:07:38.128 --> 00:07:39.999 really look at this heat map,

NOTE Confidence: 0.86811244

00:07:40.000 --> 00:07:42.424 'cause it gave us a lot of information.

NOTE Confidence: 0.86811244

00:07:42.430 --> 00:07:44.544 That was some of it quite surprising.

NOTE Confidence: 0.86811244

00:07:44.550 --> 00:07:46.863 So what you can see here is these are

NOTE Confidence: 0.86811244

00:07:46.863 --> 00:07:49.195 single cells from top to bottom that are

NOTE Confidence: 0.86811244

00:07:49.195 --> 00:07:51.518 from the common myeloid progenitor gate.

NOTE Confidence: 0.86811244

00:07:51.520 --> 00:07:53.320 These are single cells from the

NOTE Confidence: 0.86811244

00:07:53.320 --> 00:07:54.850 Mega Rich Royd Projector Gate.

NOTE Confidence: 0.86811244

00:07:54.850 --> 00:07:56.686 These are cells from the megakaryocyte

NOTE Confidence: 0.86811244

00:07:56.686 --> 00:07:58.231 progenitor gate and the erythroid

NOTE Confidence: 0.86811244

00:07:58.231 --> 00:07:59.857 progenitor gate and what you can

NOTE Confidence: 0.86811244

00:07:59.857 --> 00:08:01.723 see is there are distinct gene

NOTE Confidence: 0.86811244

00:08:01.723 --> 00:08:03.388 expression patterns that are unique.

NOTE Confidence: 0.86811244

00:08:03.390 --> 00:08:05.020 To arrest rate for janitors.

NOTE Confidence: 0.86811244

00:08:05.020 --> 00:08:07.288 Jeans are from left to right here.

NOTE Confidence: 0.86811244

00:08:07.290 --> 00:08:09.341 So these are genes that are expressed

NOTE Confidence: 0.86811244

00:08:09.341 --> 00:08:11.190 uniquely in Eryth Roid Progenitors.

NOTE Confidence: 0.86811244

00:08:11.190 --> 00:08:13.140 These are genes that are expressed

NOTE Confidence: 0.86811244

00:08:13.140 --> 00:08:14.440 uniquely in Meg progenitors.

NOTE Confidence: 0.86811244

00:08:14.440 --> 00:08:16.370 Mazer jeans that are expressed

NOTE Confidence: 0.86811244

00:08:16.370 --> 00:08:18.300 predominantly in common myeloid progenitors.

NOTE Confidence: 0.86811244

00:08:18.300 --> 00:08:20.344 But there's no such group of jeans

NOTE Confidence: 0.86811244

00:08:20.344 --> 00:08:22.380 for the Mega Mega Carey Cider

NOTE Confidence: 0.86811244

00:08:22.380 --> 00:08:23.836 Ridge Rd for generators.

NOTE Confidence: 0.86811244

00:08:23.840 --> 00:08:25.796 They really seem to be a

NOTE Confidence: 0.86811244

00:08:25.796 --> 00:08:26.448 transitional population.

NOTE Confidence: 0.86811244

00:08:26.450 --> 00:08:28.436 That's between the CMP where some

NOTE Confidence: 0.86811244

00:08:28.436 --> 00:08:30.690 of the CMP jeans are still on,

NOTE Confidence: 0.86811244

00:08:30.690 --> 00:08:32.520 but they're all going to turn

NOTE Confidence: 0.86811244

00:08:32.520 --> 00:08:34.270 off when they be picked.

NOTE Confidence: 0.86811244

00:08:34.270 --> 00:08:36.226 A fate to be Mega Rich,

NOTE Confidence: 0.86811244

00:08:36.230 --> 00:08:36.558 Royd,

NOTE Confidence: 0.86811244

00:08:36.558 --> 00:08:38.854 and then you see some that are

NOTE Confidence: 0.86811244

00:08:38.854 --> 00:08:40.469 me P an MB only,

NOTE Confidence: 0.86811244

00:08:40.470 --> 00:08:43.395 and some that are me P Anorith Droid only,

NOTE Confidence: 0.86811244

00:08:43.400 --> 00:08:45.437 and what that taught us is that

NOTE Confidence: 0.86811244

00:08:45.437 --> 00:08:47.640 MVP really are a unique population.

NOTE Confidence: 0.86811244

00:08:47.640 --> 00:08:49.000 They are not contaminated

NOTE Confidence: 0.86811244

00:08:49.000 --> 00:08:50.020 with Meg Progenitors.

NOTE Confidence: 0.86811244

00:08:50.020 --> 00:08:51.156 'cause these jeans are

NOTE Confidence: 0.86811244

00:08:51.156 --> 00:08:52.860 not on yet and they're not

NOTE Confidence: 0.8134995

00:08:52.922 --> 00:08:55.358 contaminated with the rich Roy Projectors.

NOTE Confidence: 0.8134995

00:08:55.360 --> 00:08:57.490 They really are at their own

NOTE Confidence: 0.8134995

00:08:57.490 --> 00:08:59.270 unique population that has a

NOTE Confidence: 0.8134995

00:08:59.270 --> 00:09:01.040 little bit of expression of E.

NOTE Confidence: 0.8134995

00:09:01.040 --> 00:09:02.710 Only jeans, MK only jeans,

NOTE Confidence: 0.8134995

00:09:02.710 --> 00:09:04.375 and some leftover from this

NOTE Confidence: 0.8134995

00:09:04.375 --> 00:09:05.374 common myeloid progenitor.

NOTE Confidence: 0.8134995

00:09:05.380 --> 00:09:07.788 So true transitional population.

NOTE Confidence: 0.8134995

00:09:07.790 --> 00:09:10.102 And we wanted to then look at the

NOTE Confidence: 0.8134995

00:09:10.102 --> 00:09:12.632 specific genes that turn on and off and

NOTE Confidence: 0.8134995

00:09:12.632 --> 00:09:14.304 understand whether the fate decisions

NOTE Confidence: 0.8134995

00:09:14.304 --> 00:09:16.796 are made according to what had been

NOTE Confidence: 0.8134995

00:09:16.796 --> 00:09:18.551 classic dogma in the literature.

NOTE Confidence: 0.8134995

00:09:18.551 --> 00:09:20.980 So the classic dug in the literature

NOTE Confidence: 0.8134995

00:09:21.045 --> 00:09:23.013 is than any pee pics to be a

NOTE Confidence: 0.8134995

00:09:23.013 --> 00:09:25.033 mega Carey Cider Inner it's roid

NOTE Confidence: 0.8134995

00:09:25.033 --> 00:09:27.229 commitment by expression of KLF one,

NOTE Confidence: 0.8134995

00:09:27.230 --> 00:09:29.526 or flee once OK LF one is also

NOTE Confidence: 0.8134995

00:09:29.526 --> 00:09:31.645 known as a rich Rd krupa like

NOTE Confidence: 0.8134995

00:09:31.645 --> 00:09:34.695 factor or E KLF and flea one is a

NOTE Confidence: 0.8134995

00:09:34.695 --> 00:09:36.570 transcription factor that is known

NOTE Confidence: 0.8134995

00:09:36.570 --> 00:09:38.710 to be necessary for Meg progenitors.

NOTE Confidence: 0.8134995

00:09:38.710 --> 00:09:40.528 To differentiate into megakaryocytes so when

NOTE Confidence: 0.8134995

00:09:40.528 --> 00:09:43.040 we looked at the gene expression of these,

NOTE Confidence: 0.8134995

00:09:43.040 --> 00:09:45.007 we thought we would find is that

NOTE Confidence: 0.8134995

00:09:45.007 --> 00:09:46.740 these cells are uniquely flea.

NOTE Confidence: 0.8134995

00:09:46.740 --> 00:09:48.708 One positive these cells are uniquely

NOTE Confidence: 0.8134995

00:09:48.708 --> 00:09:50.625 KLF one positive and these cells

NOTE Confidence: 0.8134995

00:09:50.625 --> 00:09:52.616 pick one or the other, or neither.

NOTE Confidence: 0.8134995

00:09:52.616 --> 00:09:54.464 But that's not what we got.

NOTE Confidence: 0.8134995

00:09:54.470 --> 00:09:56.318 Oh, and it's already been shown.

NOTE Confidence: 0.8134995

00:09:56.320 --> 00:09:58.228 I'm sorry in the dogma that

NOTE Confidence: 0.8134995

00:09:58.228 --> 00:10:00.109 fully one turns off KLF one.

NOTE Confidence: 0.8134995

00:10:00.110 --> 00:10:02.526 Tail F1 turns awfully one in cell lines,

NOTE Confidence: 0.8134995

00:10:02.530 --> 00:10:04.810 but when we looked at the gene expression

NOTE Confidence: 0.8134995

00:10:04.810 --> 00:10:07.057 patterns of all the various gene patterns,

NOTE Confidence: 0.8134995

00:10:07.060 --> 00:10:09.244 we could have jeans that are on and

NOTE Confidence: 0.8134995

00:10:09.244 --> 00:10:11.277 then off and the other lineages,

NOTE Confidence: 0.8134995

00:10:11.280 --> 00:10:13.394 or often CMP in them on in

NOTE Confidence: 0.8134995

00:10:13.394 --> 00:10:14.300 the other lineages,

NOTE Confidence: 0.8134995

00:10:14.300 --> 00:10:16.638 we found that there were very few

NOTE Confidence: 0.8134995

00:10:16.638 --> 00:10:17.941 transcription factors that really

NOTE Confidence: 0.8134995

00:10:17.941 --> 00:10:19.853 told us the story to as to how

NOTE Confidence: 0.8134995

00:10:19.853 --> 00:10:21.548 these state decisions are made.

NOTE Confidence: 0.8134995

00:10:21.550 --> 00:10:23.990 So just looking at flea one and KLF

NOTE Confidence: 0.8134995

00:10:23.990 --> 00:10:26.387 one what you can see is now what

NOTE Confidence: 0.8134995

00:10:26.387 --> 00:10:28.499 I've done is from the heat map.

NOTE Confidence: 0.8134995

00:10:28.500 --> 00:10:29.406 We still have.

NOTE Confidence: 0.8134995

00:10:29.406 --> 00:10:31.218 These are individual CMP's individual MPs,

NOTE Confidence: 0.8134995

00:10:31.220 --> 00:10:32.480 individual Meg progenitors and

NOTE Confidence: 0.8134995

00:10:32.480 --> 00:10:33.740 individual rich Rd projectors.

NOTE Confidence: 0.8134995

00:10:33.740 --> 00:10:36.516 And these are comb plots of individual genes,

NOTE Confidence: 0.8134995

00:10:36.520 --> 00:10:37.564 so KLF one.

NOTE Confidence: 0.8134995

00:10:37.564 --> 00:10:38.956 As you can see,

NOTE Confidence: 0.8134995

00:10:38.960 --> 00:10:41.396 is expressed in some me P in

NOTE Confidence: 0.8134995

00:10:41.396 --> 00:10:42.440 some Meg progenitors.

NOTE Confidence: 0.8134995

00:10:42.440 --> 00:10:43.484 It's predominantly expressed

NOTE Confidence: 0.8134995

00:10:43.484 --> 00:10:44.876 in the erythroid progenitors,

NOTE Confidence: 0.8134995

00:10:44.880 --> 00:10:47.808 but it's really not showing you that it's

NOTE Confidence: 0.8134995

00:10:47.808 --> 00:10:50.438 clearly making a fate decision of any sort.

NOTE Confidence: 0.8134995

00:10:50.440 --> 00:10:50.798 Flea.

NOTE Confidence: 0.8134995

00:10:50.798 --> 00:10:52.946 One is also expressed in almost

NOTE Confidence: 0.8134995

00:10:52.946 --> 00:10:54.619 every Lenny Edge at well.

NOTE Confidence: 0.8134995

00:10:54.620 --> 00:10:54.967 Yes,

NOTE Confidence: 0.8134995

00:10:54.967 --> 00:10:57.396 it is expressed strongly in Meg Progenitors.

NOTE Confidence: 0.8134995

00:10:57.400 --> 00:10:59.140 It's also expressed throughout MVP,

NOTE Confidence: 0.8134995

00:10:59.140 --> 00:11:01.709 and if you look for Co expression

NOTE Confidence: 0.8134995

00:11:01.709 --> 00:11:04.448 of flea one and KLF one in MVP.

NOTE Confidence: 0.8134995

00:11:04.450 --> 00:11:05.830 Sometimes it is coexpressed in,

NOTE Confidence: 0.8134995

00:11:05.830 --> 00:11:06.655 sometimes it's not,

NOTE Confidence: 0.8134995

00:11:06.655 --> 00:11:08.305 so it wasn't quite so simple.

NOTE Confidence: 0.8134995

00:11:08.310 --> 00:11:09.899 We also looked at Gotta One and

NOTE Confidence: 0.8134995

00:11:09.899 --> 00:11:11.596 got it two which are known to

NOTE Confidence: 0.8134995

00:11:11.596 --> 00:11:13.610 be critical for a rich Rd in

NOTE Confidence: 0.8134995

00:11:13.610 --> 00:11:14.658 megakaryocytic maturation.

NOTE Confidence: 0.8134995

00:11:14.660 --> 00:11:16.524 And what you can see is that gotta

NOTE Confidence: 0.8134995

00:11:16.524 --> 00:11:18.687 one comes on when the cells commit

NOTE Confidence: 0.8134995

00:11:18.687 --> 00:11:19.999 to the megathread progenitor

NOTE Confidence: 0.8134995

00:11:19.999 --> 00:11:21.288 lineages and then stay on.

NOTE Confidence: 0.8134995

00:11:21.290 --> 00:11:23.775 In both lineages got it too is

NOTE Confidence: 0.8134995

00:11:23.775 --> 00:11:25.569 expressed throughout as his NFE 2.

NOTE Confidence: 0.8134995

00:11:25.570 --> 00:11:27.600 So what really is going on and

NOTE Confidence: 0.8134995

00:11:27.600 --> 00:11:29.400 what's making these fate decisions?

NOTE Confidence: 0.8134995

00:11:29.400 --> 00:11:31.591 We looked at all of the different

NOTE Confidence: 0.8134995

00:11:31.591 --> 00:11:32.903 gene expression patterns and

NOTE Confidence: 0.8134995

00:11:32.903 --> 00:11:34.155 found some pretty interesting

NOTE Confidence: 0.8134995

00:11:34.155 --> 00:11:35.720 genes and genes that are

NOTE Confidence: 0.8618278

00:11:35.780 --> 00:11:37.375 somewhat me specific that we're

NOTE Confidence: 0.8618278

00:11:37.375 --> 00:11:38.970 pursuing further in my lab.

NOTE Confidence: 0.8618278

00:11:38.970 --> 00:11:40.926 But really, what the data ended

NOTE Confidence: 0.8618278

00:11:40.926 --> 00:11:43.236 up showing is that it's not one

NOTE Confidence: 0.8618278

00:11:43.236 --> 00:11:45.114 specific group of jeans I wanted

NOTE Confidence: 0.8618278

00:11:45.114 --> 00:11:47.579 to make sure to stop and say again,

NOTE Confidence: 0.8618278

00:11:47.580 --> 00:11:50.390 this is going to be.

NOTE Confidence: 0.8618278

00:11:50.390 --> 00:11:51.885 Publicly available on YouTube at

NOTE Confidence: 0.8618278

00:11:51.885 --> 00:11:54.320 this URL that I have down here and

NOTE Confidence: 0.8618278

00:11:54.320 --> 00:11:56.084 it's also available in the paper,

NOTE Confidence: 0.8618278

00:11:56.090 --> 00:11:57.902 you can actually put any gene

NOTE Confidence: 0.8618278

00:11:57.902 --> 00:12:00.197 of interest in an get the comb

NOTE Confidence: 0.8618278

00:12:00.197 --> 00:12:01.897 plots that you're interested in

NOTE Confidence: 0.8618278

00:12:01.897 --> 00:12:03.998 from the are single cell data.

NOTE Confidence: 0.8618278

00:12:04.000 --> 00:12:06.030 OK, So what ended up happening is

NOTE Confidence: 0.8618278

00:12:06.030 --> 00:12:08.334 when we analyze these data using gene

NOTE Confidence: 0.8618278

00:12:08.334 --> 00:12:10.049 ontogeny and other approaches is

NOTE Confidence: 0.8618278

00:12:10.049 --> 00:12:12.161 the cell cycle was amongst the most

NOTE Confidence: 0.8618278

00:12:12.161 --> 00:12:14.230 the genes of the cell cycle where

NOTE Confidence: 0.8618278

00:12:14.230 --> 00:12:15.470 the most differentially expressed.

NOTE Confidence: 0.8618278

00:12:15.470 --> 00:12:17.020 When you compared me, PETA,

NOTE Confidence: 0.8618278

00:12:17.020 --> 00:12:18.570 Meg progenitors and me Peter

NOTE Confidence: 0.8618278

00:12:18.570 --> 00:12:19.500 it's red projectors.

NOTE Confidence: 0.8618278

00:12:19.500 --> 00:12:21.348 You can see regulation of the

NOTE Confidence: 0.8618278

00:12:21.348 --> 00:12:23.530 cell cycle comes up here and then.

NOTE Confidence: 0.8618278

00:12:23.530 --> 00:12:25.205 All of these different differences

NOTE Confidence: 0.8618278

00:12:25.205 --> 00:12:27.492 between MVP and MKP and this actually

NOTE Confidence: 0.8618278

00:12:27.492 --> 00:12:29.708 kind of hit a nerve with us 'cause

NOTE Confidence: 0.8618278

00:12:29.769 --> 00:12:31.605 we already had a finding that

NOTE Confidence: 0.8618278

00:12:31.605 --> 00:12:33.640 suggested this might be the case.

NOTE Confidence: 0.8618278

00:12:33.640 --> 00:12:35.260 What we already knew.

NOTE Confidence: 0.8618278

00:12:35.260 --> 00:12:36.700 We had tried some candidate

NOTE Confidence: 0.8618278

00:12:36.700 --> 00:12:38.580 drugs to see if we could.

NOTE Confidence: 0.8618278

00:12:38.580 --> 00:12:40.449 If they affect the Mega Rich Rd

NOTE Confidence: 0.8618278

00:12:40.449 --> 00:12:42.421 fate decision and we had added for

NOTE Confidence: 0.8618278

00:12:42.421 --> 00:12:43.811 example all trans retinoic acid

NOTE Confidence: 0.8618278

00:12:43.811 --> 00:12:45.892 at low and high concentrations and

NOTE Confidence: 0.8618278

00:12:45.892 --> 00:12:47.936 seeing a dose dependent increase in

NOTE Confidence: 0.8618278

00:12:47.936 --> 00:12:48.824 megakaryocyte fate specification.

NOTE Confidence: 0.8618278

00:12:48.824 --> 00:12:50.952 Similarly with Rappo Mison with a dose

NOTE Confidence: 0.8618278

00:12:50.952 --> 00:12:52.764 dependent increase in Meg fate specification.

NOTE Confidence: 0.8618278

00:12:52.770 --> 00:12:55.146 And this was true for a lot of other

NOTE Confidence: 0.8618278

00:12:55.146 --> 00:12:57.551 drugs that we had tried that were

NOTE Confidence: 0.8618278

00:12:57.551 --> 00:12:59.282 known kinase inhibitors and what

NOTE Confidence: 0.8618278

00:12:59.282 --> 00:13:01.263 became clear to us is the thing

NOTE Confidence: 0.8618278

00:13:01.263 --> 00:13:02.990 that all of these inhibitors had.

NOTE Confidence: 0.8618278

00:13:02.990 --> 00:13:05.810 All of these drugs head is that they inhibit.

NOTE Confidence: 0.8618278

00:13:05.810 --> 00:13:07.480 Or slowed the cell cycle.

NOTE Confidence: 0.8618278

00:13:07.480 --> 00:13:09.478 So we decided to test that.

NOTE Confidence: 0.8618278

00:13:09.480 --> 00:13:11.608 So what we've done here is just

NOTE Confidence: 0.8618278

00:13:11.608 --> 00:13:13.829 treating with all trans retinoic acid,

NOTE Confidence: 0.8618278

00:13:13.830 --> 00:13:14.853 the 50 nanomolar,

NOTE Confidence: 0.8618278

00:13:14.853 --> 00:13:17.240 which is the same thing is down

NOTE Confidence: 0.8618278

00:13:17.309 --> 00:13:18.839 here you take any pee,

NOTE Confidence: 0.8618278

00:13:18.840 --> 00:13:21.010 stain them with CFC and then as

NOTE Confidence: 0.8618278

00:13:21.010 --> 00:13:23.066 the cells are in culture for

NOTE Confidence: 0.8618278

00:13:23.066 --> 00:13:25.178 72 hours that CFC gets diluted.

NOTE Confidence: 0.8618278

00:13:25.180 --> 00:13:27.532 So from as cells become lower in

NOTE Confidence: 0.8618278

00:13:27.532 --> 00:13:29.645 their fluorescence for CFC that means

NOTE Confidence: 0.8618278

00:13:29.645 --> 00:13:31.385 that they have proliferated more

NOTE Confidence: 0.8618278

00:13:31.385 --> 00:13:33.975 times and what you can see is the

NOTE Confidence: 0.8618278

00:13:33.975 --> 00:13:35.956 control population or blue population here.

NOTE Confidence: 0.8618278

00:13:35.956 --> 00:13:37.846 Has proliferated more times than

NOTE Confidence: 0.8618278

00:13:37.846 --> 00:13:39.552 the population that was treated

NOTE Confidence: 0.8618278

00:13:39.552 --> 00:13:41.351 with Aptra and which gave you a

NOTE Confidence: 0.8618278

00:13:41.351 --> 00:13:42.986 Meg lineages bias suggesting that

NOTE Confidence: 0.8618278

00:13:42.986 --> 00:13:44.960 this lower cell cycle might be

NOTE Confidence: 0.8618278

00:13:44.960 --> 00:13:46.560 associated with the Meg bias.

NOTE Confidence: 0.8618278

00:13:46.560 --> 00:13:48.730 Similarly with Rappa Mice and we see

NOTE Confidence: 0.8618278

00:13:48.730 --> 00:13:51.035 a slower cell cycle that's shown here

NOTE Confidence: 0.8618278

00:13:51.035 --> 00:13:53.630 in red is associated with the Meg bias.

NOTE Confidence: 0.8618278

00:13:53.630 --> 00:13:56.086 So what we did is we tested just

NOTE Confidence: 0.8618278

00:13:56.086 --> 00:13:57.737 inhibiting the cell cycle with

NOTE Confidence: 0.8618278

00:13:57.737 --> 00:13:59.984 the CD K46 Inhibitor and here you

NOTE Confidence: 0.8618278

00:14:00.050 --> 00:14:02.297 can see that the cells that were

NOTE Confidence: 0.8618278

00:14:02.297 --> 00:14:03.900 treated with the inhibitor or

NOTE Confidence: 0.8618278

00:14:03.900 --> 00:14:05.500 practically not dividing at all,

NOTE Confidence: 0.8618278

00:14:05.500 --> 00:14:07.666 whereas the control population is Dividing.

NOTE Confidence: 0.8618278

00:14:07.670 --> 00:14:10.400 And again we see a dose.

NOTE Confidence: 0.8618278

00:14:10.400 --> 00:14:12.130 Specific increase in the Meg

NOTE Confidence: 0.8618278

00:14:12.130 --> 00:14:13.514 progenitor cell fate decision.

NOTE Confidence: 0.8618278

00:14:13.520 --> 00:14:16.000 So the next thing we had to do

NOTE Confidence: 0.8618278

00:14:16.000 --> 00:14:18.599 is figure out a way to increase

NOTE Confidence: 0.8618278

00:14:18.599 --> 00:14:20.115 this cell cycle speed,

NOTE Confidence: 0.80757487

00:14:20.120 --> 00:14:22.759 and for this we ended up trying

NOTE Confidence: 0.80757487

00:14:22.759 --> 00:14:25.318 to knock down CD 21 and CD 57,

NOTE Confidence: 0.80757487

00:14:25.320 --> 00:14:27.749 but those things didn't work very well.

NOTE Confidence: 0.80757487

00:14:27.750 --> 00:14:29.916 They killed the cells so we

NOTE Confidence: 0.80757487

00:14:29.916 --> 00:14:32.073 tried something else and what we

NOTE Confidence: 0.80757487

00:14:32.073 --> 00:14:33.999 did is from the Vascular lab.

NOTE Confidence: 0.80757487

00:14:34.000 --> 00:14:35.730 We got two different constructs.

NOTE Confidence: 0.80757487

00:14:35.730 --> 00:14:37.806 One is C DK2 cycle independent,

NOTE Confidence: 0.80757487

00:14:37.810 --> 00:14:39.270 two cyclin dependent kinase

NOTE Confidence: 0.80757487

00:14:39.270 --> 00:14:40.365 to driving cycling.

NOTE Confidence: 0.80757487

00:14:40.370 --> 00:14:42.140 E phosphorylation and cycling depending

NOTE Confidence: 0.80757487

00:14:42.140 --> 00:14:43.910 kinase for which promotes cycling.

NOTE Confidence: 0.80757487

00:14:43.910 --> 00:14:44.842 D phosphorylation,

NOTE Confidence: 0.80757487

00:14:44.842 --> 00:14:48.104 and we coexpressed either CD K2 and

NOTE Confidence: 0.80757487

00:14:48.104 --> 00:14:50.339 Cyclin E or C DK foreign cyclin

NOTE Confidence: 0.80757487

00:14:50.339 --> 00:14:52.742 D in our MVP and what you can

NOTE Confidence: 0.80757487

00:14:52.742 --> 00:14:55.236 see up here is for the site CD.

NOTE Confidence: 0.80757487

00:14:55.236 --> 00:14:58.086 K for Cyclin D, which we call for.

NOTE Confidence: 0.80757487

00:14:58.086 --> 00:15:01.003 D actually promotes G one of the cell

NOTE Confidence: 0.80757487

00:15:01.003 --> 00:15:03.747 cycle and CK2 Cyclin E which we call

NOTE Confidence: 0.80757487

00:15:03.832 --> 00:15:07.262 2 E promotes the G1 to S transition and down.

NOTE Confidence: 0.80757487

00:15:07.270 --> 00:15:10.555 Here we can see the data the two E.

NOTE Confidence: 0.80757487

00:15:10.560 --> 00:15:11.744 And four deconstructs gave

NOTE Confidence: 0.80757487

00:15:11.744 --> 00:15:12.928 us Anorith Royd Bias,

NOTE Confidence: 0.80757487

00:15:12.930 --> 00:15:14.995 which is what we were looking for.

NOTE Confidence: 0.80757487

00:15:15.000 --> 00:15:17.240 It's the opposite of the Meg bias that

NOTE Confidence: 0.80757487

00:15:17.240 --> 00:15:19.735 we get when we slowed the cell cycle.

NOTE Confidence: 0.80757487

00:15:19.740 --> 00:15:22.080 When you just had to Ian 40 to Meg

NOTE Confidence: 0.80757487

00:15:22.080 --> 00:15:23.877 progenitors in Eryth Roid Progenitors,

NOTE Confidence: 0.80757487

00:15:23.880 --> 00:15:25.656 you don't see any specific change

NOTE Confidence: 0.80757487

00:15:25.656 --> 00:15:26.840 in their fate specification.

NOTE Confidence: 0.80757487

00:15:26.840 --> 00:15:27.863 And up here,

NOTE Confidence: 0.80757487

00:15:27.863 --> 00:15:29.909 we're just showing that when you

NOTE Confidence: 0.80757487

00:15:29.909 --> 00:15:31.867 overexpress the 2E or 4D in the cells,

NOTE Confidence: 0.80757487

00:15:31.870 --> 00:15:33.646 you get a faster cell cycle,

NOTE Confidence: 0.80757487

00:15:33.650 --> 00:15:35.420 or the CFC is more diluted.

NOTE Confidence: 0.80757487

00:15:35.420 --> 00:15:37.130 So this really suggested to us

NOTE Confidence: 0.80757487

00:15:37.130 --> 00:15:38.954 that the faster cell cycle is

NOTE Confidence: 0.80757487

00:15:38.954 --> 00:15:40.838 associated with the River police is.

NOTE Confidence: 0.80757487

00:15:40.840 --> 00:15:42.952 Slower cell cycle with Mega Carey

NOTE Confidence: 0.80757487

00:15:42.952 --> 00:15:45.626 side of Louise is so can we assess

NOTE Confidence: 0.80757487

00:15:45.626 --> 00:15:48.229 cell cycle in vivo and now I'm bout

NOTE Confidence: 0.80757487

00:15:48.229 --> 00:15:50.467 to present to you some unpublished

NOTE Confidence: 0.80757487

00:15:50.467 --> 00:15:52.600 work using and now recently published

NOTE Confidence: 0.80757487

00:15:52.600 --> 00:15:53.635 fluorescent Reporter mouse.

NOTE Confidence: 0.80757487

00:15:53.640 --> 00:15:56.328 They shun ching glow at Yale University

NOTE Confidence: 0.80757487

00:15:56.328 --> 00:15:58.813 has developed and what she did is

NOTE Confidence: 0.80757487

00:15:58.813 --> 00:16:00.709 she made a mouse that basically

NOTE Confidence: 0.80757487

00:16:00.785 --> 00:16:03.046 tells you the cell cycle speed of

NOTE Confidence: 0.80757487

00:16:03.046 --> 00:16:05.256 any given cell that you look at.

NOTE Confidence: 0.80757487

00:16:05.256 --> 00:16:07.511 She did this by overexpressing a cell

NOTE Confidence: 0.80757487

00:16:07.511 --> 00:16:09.515 cycle timer protein that starts out

NOTE Confidence: 0.80757487

00:16:09.515 --> 00:16:12.497 blue and then gradually becomes red overtime.

NOTE Confidence: 0.80757487

00:16:12.500 --> 00:16:14.516 And this has been described previously.

NOTE Confidence: 0.80757487

00:16:14.520 --> 00:16:16.205 She fused it with Histone

NOTE Confidence: 0.80757487

00:16:16.205 --> 00:16:17.890 H2B so it was nuclear.

NOTE Confidence: 0.80757487

00:16:17.890 --> 00:16:20.302 And what you can see in the math is

NOTE Confidence: 0.80757487

00:16:20.302 --> 00:16:22.473 all in this paper that's available

NOTE Confidence: 0.80757487

00:16:22.473 --> 00:16:25.118 in bio archive soon to be out

NOTE Confidence: 0.80757487

00:16:25.118 --> 00:16:26.988 in a peer reviewed publication.

NOTE Confidence: 0.80757487

00:16:26.990 --> 00:16:29.144 Basically what happens is the blue

NOTE Confidence: 0.80757487

00:16:29.144 --> 00:16:31.242 because it is expressed for just

NOTE Confidence: 0.80757487

00:16:31.242 --> 00:16:33.024 a short time after the protein

NOTE Confidence: 0.80757487

00:16:33.024 --> 00:16:34.739 comes on is always steady,

NOTE Confidence: 0.80757487

00:16:34.740 --> 00:16:37.356 whereas the red gets brighter and

NOTE Confidence: 0.80757487

00:16:37.356 --> 00:16:40.080 brighter and brighter as as the cell.

NOTE Confidence: 0.80757487

00:16:40.080 --> 00:16:42.075 Proliferates So what you can see is

NOTE Confidence: 0.80757487

00:16:42.075 --> 00:16:44.202 if you look at the blue red ratio

NOTE Confidence: 0.80757487

00:16:44.202 --> 00:16:46.260 you get a sense of how quickly

NOTE Confidence: 0.80757487

00:16:46.260 --> 00:16:48.210 that cell has been proliferating.

NOTE Confidence: 0.80757487

00:16:48.210 --> 00:16:50.730 So when we did this and I think the

NOTE Confidence: 0.80757487

00:16:50.730 --> 00:16:53.320 main thing here is to look down here,

NOTE Confidence: 0.80757487

00:16:53.320 --> 00:16:55.126 I'll tell you what we did.

NOTE Confidence: 0.80757487

00:16:55.130 --> 00:16:57.686 What we did is we took urine cells from

NOTE Confidence: 0.80757487

00:16:57.686 --> 00:17:00.250 these Reporter Mice and we gated on them.

NOTE Confidence: 0.80757487

00:17:00.250 --> 00:17:01.750 You Ring me pee Meg.

NOTE Confidence: 0.80757487

00:17:01.750 --> 00:17:03.730 Progenitor eryth roid for genitor

NOTE Confidence: 0.80757487

00:17:03.730 --> 00:17:05.710 or fully differentiated cells that

NOTE Confidence: 0.80757487

00:17:05.775 --> 00:17:07.593 were Lynn positive and then for

NOTE Confidence: 0.80757487

00:17:07.593 --> 00:17:08.805 every cell we looked

NOTE Confidence: 0.799561560000001

00:17:08.867 --> 00:17:10.498 at the ratio of blue to red.

NOTE Confidence: 0.799561560000001

00:17:10.500 --> 00:17:12.740 If you have a lot of red,

NOTE Confidence: 0.799561560000001

00:17:12.740 --> 00:17:14.980 so a high red to blue ratio,

NOTE Confidence: 0.799561560000001

00:17:14.980 --> 00:17:16.580 that means you're proliferating much,

NOTE Confidence: 0.799561560000001

00:17:16.580 --> 00:17:18.960 much faster and what you can see is that the

NOTE Confidence: 0.799561560000001

00:17:19.017 --> 00:17:20.802 erythroid cells the erythroid progenitors

NOTE Confidence: 0.799561560000001

00:17:20.802 --> 00:17:23.619 are the fastest cells in the proliferation.

NOTE Confidence: 0.799561560000001

00:17:23.620 --> 00:17:26.469 The Meg progenitors were in the middle.

NOTE Confidence: 0.799561560000001

00:17:26.470 --> 00:17:27.394 And the MVP,

NOTE Confidence: 0.799561560000001

00:17:27.394 --> 00:17:28.934 the by potent progenitors upstream,

NOTE Confidence: 0.799561560000001

00:17:28.940 --> 00:17:29.867 where the slowest.

NOTE Confidence: 0.799561560000001

00:17:29.867 --> 00:17:32.030 So this was new information to us,

NOTE Confidence: 0.799561560000001

00:17:32.030 --> 00:17:33.490 although it was consistent with

NOTE Confidence: 0.799561560000001

00:17:33.490 --> 00:17:35.556 what we've seen in vitro for human

NOTE Confidence: 0.799561560000001

00:17:35.556 --> 00:17:37.188 cells is that MVP are probably

NOTE Confidence: 0.799561560000001

00:17:37.188 --> 00:17:38.830 the slowest proliferating cells,

NOTE Confidence: 0.799561560000001

00:17:38.830 --> 00:17:40.380 then the Meg progenitor cells,

NOTE Confidence: 0.799561560000001

00:17:40.380 --> 00:17:41.920 then the erythroid progenitor cells.

NOTE Confidence: 0.799561560000001

00:17:41.920 --> 00:17:43.901 So our model right now is that

NOTE Confidence: 0.799561560000001

00:17:43.901 --> 00:17:45.630 me PR is quite slow.

NOTE Confidence: 0.799561560000001

00:17:45.630 --> 00:17:47.376 They do self renew and then

NOTE Confidence: 0.799561560000001

00:17:47.376 --> 00:17:49.648 if you pick up the cell cycle,

NOTE Confidence: 0.799561560000001

00:17:49.650 --> 00:17:51.806 so it's a little faster than that,

NOTE Confidence: 0.799561560000001

00:17:51.810 --> 00:17:53.966 you're a Meg progenitor and a faster

NOTE Confidence: 0.799561560000001

00:17:53.966 --> 00:17:55.670 cell cycle, Anorith raid progenitor.

NOTE Confidence: 0.799561560000001

00:17:55.670 --> 00:17:56.750 And we haven't.

NOTE Confidence: 0.799561560000001

00:17:56.750 --> 00:17:58.174 Still answered the question

NOTE Confidence: 0.799561560000001

00:17:58.174 --> 00:17:59.954 as to how this happens.

NOTE Confidence: 0.799561560000001

00:17:59.960 --> 00:18:01.034 That's a really,

NOTE Confidence: 0.799561560000001

00:18:01.034 --> 00:18:01.750 really exciting,

NOTE Confidence: 0.799561560000001

00:18:01.750 --> 00:18:02.434 important question.

NOTE Confidence: 0.799561560000001

00:18:02.434 --> 00:18:04.828 We think there are lots of things

NOTE Confidence: 0.799561560000001

00:18:04.828 --> 00:18:07.058 going on in addition to maybe

NOTE Confidence: 0.799561560000001

00:18:07.058 --> 00:18:08.883 epigenetic things with DNA methylation.

NOTE Confidence: 0.799561560000001

00:18:08.890 --> 00:18:11.425 There are probably also changes

NOTE Confidence: 0.799561560000001

00:18:11.425 --> 00:18:13.453 in phosphorylation of critical

NOTE Confidence: 0.799561560000001

00:18:13.453 --> 00:18:15.076 transcription factors when you

NOTE Confidence: 0.799561560000001

00:18:15.076 --> 00:18:17.164 have a slower fast cell cycle.

NOTE Confidence: 0.799561560000001

00:18:17.170 --> 00:18:19.708 So I'm going to move on to now is

NOTE Confidence: 0.799561560000001

00:18:19.708 --> 00:18:21.714 whether there are clinical scenarios

NOTE Confidence: 0.799561560000001

00:18:21.714 --> 00:18:24.192 in patients where MP fate might

NOTE Confidence: 0.799561560000001

00:18:24.262 --> 00:18:26.158 play a critical role and this

NOTE Confidence: 0.799561560000001

00:18:26.158 --> 00:18:28.417 is work that was done again bij

NOTE Confidence: 0.799561560000001

00:18:28.417 --> 00:18:30.391 Juliana Javie Ferruccio and now in

NOTE Confidence: 0.799561560000001

00:18:30.391 --> 00:18:31.879 collaboration with Vanessa Scanlon,

NOTE Confidence: 0.799561560000001

00:18:31.880 --> 00:18:33.842 an instructor in my laboratory and

NOTE Confidence: 0.799561560000001

00:18:33.842 --> 00:18:36.320 what we're looking at is iron deficiency.

NOTE Confidence: 0.799561560000001

00:18:36.320 --> 00:18:38.366 So it's long been known anecdotally,

NOTE Confidence: 0.799561560000001

00:18:38.370 --> 00:18:40.422 the patients who become iron deficient

NOTE Confidence: 0.799561560000001

00:18:40.422 --> 00:18:41.790 have elevated platelet counts,

NOTE Confidence: 0.799561560000001

00:18:41.790 --> 00:18:43.824 and we decided we really wanted

NOTE Confidence: 0.799561560000001

00:18:43.824 --> 00:18:46.313 to look at that and see whether

NOTE Confidence: 0.799561560000001

00:18:46.313 --> 00:18:48.784 it might be an MVP fake decision.

NOTE Confidence: 0.799561560000001

00:18:48.790 --> 00:18:51.212 So what you're seeing here is just

NOTE Confidence: 0.799561560000001

00:18:51.212 --> 00:18:53.221 data from published accounts of

NOTE Confidence: 0.799561560000001

00:18:53.221 --> 00:18:55.466 patients with iron deficiency anemia,

NOTE Confidence: 0.799561560000001

00:18:55.470 --> 00:18:57.290 specifically in.

NOTE Confidence: 0.799561560000001

00:18:57.290 --> 00:19:00.104 Um Irida, which I'll tell you bout,

NOTE Confidence: 0.799561560000001

00:19:00.110 --> 00:19:02.924 which is a temporal 6 Mutation population,

NOTE Confidence: 0.799561560000001

00:19:02.930 --> 00:19:06.236 but they're highly iron deficient and.

NOTE Confidence: 0.799561560000001

00:19:06.240 --> 00:19:08.418 Refractory to when you add iron.

NOTE Confidence: 0.799561560000001

00:19:08.420 --> 00:19:10.195 So iron refractory iron deficiency

NOTE Confidence: 0.799561560000001

00:19:10.195 --> 00:19:12.776 anemia and what you can see is

NOTE Confidence: 0.799561560000001

00:19:12.776 --> 00:19:13.880 in individual patients.

NOTE Confidence: 0.799561560000001

00:19:13.880 --> 00:19:16.070 These are 11 of individual patients.

NOTE Confidence: 0.799561560000001

00:19:16.070 --> 00:19:17.890 As the hemoglobin goes down,

NOTE Confidence: 0.799561560000001

00:19:17.890 --> 00:19:20.310 the platelet count goes up.

NOTE Confidence: 0.799561560000001

00:19:20.310 --> 00:19:22.186 So if we could look at the

NOTE Confidence: 0.799561560000001

00:19:22.186 --> 00:19:23.680 MVP in these patients,

NOTE Confidence: 0.799561560000001

00:19:23.680 --> 00:19:25.210 that would be really cool.

NOTE Confidence: 0.799561560000001

00:19:25.210 --> 00:19:26.780 'cause we could determine whether

NOTE Confidence: 0.799561560000001

00:19:26.780 --> 00:19:28.350 or not there are megakaryocytes

NOTE Confidence: 0.799561560000001

00:19:28.405 --> 00:19:30.097 biased in the iron deficient state,

NOTE Confidence: 0.799561560000001

00:19:30.100 --> 00:19:31.936 given that we couldn't get bone

NOTE Confidence: 0.799561560000001

00:19:31.936 --> 00:19:33.160 marrow from these patients,

NOTE Confidence: 0.799561560000001

00:19:33.160 --> 00:19:34.996 we did the next best thing,

NOTE Confidence: 0.799561560000001

00:19:35.000 --> 00:19:36.836 which is to look at Immuring

NOTE Confidence: 0.799561560000001

00:19:36.836 --> 00:19:38.060 model of this disease.

NOTE Confidence: 0.799561560000001

00:19:38.060 --> 00:19:40.083 So the mooring model of the diseases

NOTE Confidence: 0.799561560000001

00:19:40.083 --> 00:19:42.042 that Empress 6 knockout mouse an

NOTE Confidence: 0.799561560000001

00:19:42.042 --> 00:19:44.112 we were very fortunate that current

NOTE Confidence: 0.799561560000001

00:19:44.112 --> 00:19:46.152 finberg is here at Yale and she

NOTE Confidence: 0.799561560000001

00:19:46.152 --> 00:19:47.553 really helped define the temper

NOTE Confidence: 0.799561560000001

00:19:47.553 --> 00:19:49.431 6 knockout in patients and she

NOTE Confidence: 0.799561560000001

00:19:49.431 --> 00:19:50.750 also had the mice.

NOTE Confidence: 0.799561560000001

00:19:50.750 --> 00:19:52.670 So Long story short,

NOTE Confidence: 0.799561560000001

00:19:52.670 --> 00:19:55.960 temper 6 normally keeps hepcidin levels low.

NOTE Confidence: 0.799561560000001

00:19:55.960 --> 00:20:00.200 Low hepcidin levels allow you to absorb iron.

NOTE Confidence: 0.799561560000001

00:20:00.200 --> 00:20:01.855 If you have high hepcidin

NOTE Confidence: 0.799561560000001

00:20:01.855 --> 00:20:03.510 then you don't absorb any

NOTE Confidence: 0.80322874

00:20:03.585 --> 00:20:05.318 iron, so Long story short,

NOTE Confidence: 0.80322874

00:20:05.318 --> 00:20:08.323 our temporal 6 knockout mice do not absorb

NOTE Confidence: 0.80322874

00:20:08.323 --> 00:20:11.214 iron because their hepcidin levels are high.

NOTE Confidence: 0.80322874

00:20:11.220 --> 00:20:13.510 And So what we did is we got the temporal

NOTE Confidence: 0.80322874

00:20:13.569 --> 00:20:15.985 6 nicean we looked to see whether they

NOTE Confidence: 0.80322874

00:20:15.985 --> 00:20:18.098 have microcytic anemia and thrombocytosis,

NOTE Confidence: 0.80322874

00:20:18.100 --> 00:20:19.894 and they do so with what

NOTE Confidence: 0.80322874

00:20:19.894 --> 00:20:21.090 I'm showing you here.

NOTE Confidence: 0.80322874

00:20:21.090 --> 00:20:22.580 Here's the temper 6 knockout.

NOTE Confidence: 0.80322874

00:20:22.580 --> 00:20:23.772 Here's the wild type.

NOTE Confidence: 0.80322874

00:20:23.772 --> 00:20:26.178 The hemoglobin is low, the hematocrit is low.

NOTE Confidence: 0.80322874

00:20:26.178 --> 00:20:28.260 The MCD mean corpuscular volume is low.

NOTE Confidence: 0.80322874

00:20:28.260 --> 00:20:30.213 So that's why their microcytic their small

NOTE Confidence: 0.80322874

00:20:30.213 --> 00:20:32.357 red blood cells and the platelet counts

NOTE Confidence: 0.80322874

00:20:32.357 --> 00:20:34.235 are significantly elevated in these mice.

NOTE Confidence: 0.80322874

00:20:34.240 --> 00:20:36.448 So the next thing we could do is

NOTE Confidence: 0.80322874

00:20:36.448 --> 00:20:38.727 look at these mice at their MPs.

NOTE Confidence: 0.80322874

00:20:38.730 --> 00:20:40.626 So what we did is we fax sorted

NOTE Confidence: 0.80322874

00:20:40.626 --> 00:20:42.500 out murin megathread progenitors.

NOTE Confidence: 0.80322874

00:20:42.500 --> 00:20:44.635 And we grew colonies and what you

NOTE Confidence: 0.80322874

00:20:44.635 --> 00:20:47.539 can see is that the temper 6 knockout

NOTE Confidence: 0.80322874

00:20:47.539 --> 00:20:49.870 me peas have a megakaryocyte bias,

NOTE Confidence: 0.80322874

00:20:49.870 --> 00:20:51.976 which is exactly what we predicted.

NOTE Confidence: 0.80322874

00:20:51.980 --> 00:20:54.451 So this was super exciting to us

NOTE Confidence: 0.80322874

00:20:54.451 --> 00:20:57.236 and the next thing we did is well,

NOTE Confidence: 0.80322874

00:20:57.240 --> 00:20:58.644 do they proliferate slower?

NOTE Confidence: 0.80322874

00:20:58.644 --> 00:21:00.399 And the answer is yes.

NOTE Confidence: 0.80322874

00:21:00.400 --> 00:21:01.234 So again,

NOTE Confidence: 0.80322874

00:21:01.234 --> 00:21:03.319 it's consistent with our previous

NOTE Confidence: 0.80322874

00:21:03.319 --> 00:21:06.392 data that slower cell cycle goes is

NOTE Confidence: 0.80322874

00:21:06.392 --> 00:21:09.020 consistent with a higher megakaryocyte bias.

NOTE Confidence: 0.80322874

00:21:09.020 --> 00:21:11.428 We wanted to also study this in human

NOTE Confidence: 0.80322874

00:21:11.428 --> 00:21:13.807 cells an what we did initially didn't

NOTE Confidence: 0.80322874

00:21:13.807 --> 00:21:16.430 workout and what we did initially is.

NOTE Confidence: 0.80322874

00:21:16.430 --> 00:21:18.410 We just tried growing the cells

NOTE Confidence: 0.80322874

00:21:18.410 --> 00:21:20.526 in the presence of iron chelators

NOTE Confidence: 0.80322874

00:21:20.526 --> 00:21:23.067 hoping to get a low iron environment

NOTE Confidence: 0.80322874

00:21:23.067 --> 00:21:25.020 for the cells that would allow

NOTE Confidence: 0.80322874

00:21:25.020 --> 00:21:26.939 us to see the human cells.

NOTE Confidence: 0.80322874

00:21:26.939 --> 00:21:28.535 Reiterate these Mooring Dataware

NOTE Confidence: 0.80322874

00:21:28.535 --> 00:21:31.384 with human me pee when you grew them

NOTE Confidence: 0.80322874

00:21:31.384 --> 00:21:33.620 in low iron you'd get a Meg bias.

NOTE Confidence: 0.80322874

00:21:33.620 --> 00:21:35.495 However that didn't workout because

NOTE Confidence: 0.80322874

00:21:35.495 --> 00:21:37.732 whenever we lowered the iron in

NOTE Confidence: 0.80322874

00:21:37.732 --> 00:21:39.526 vitro the cells didn't grow colonies

NOTE Confidence: 0.80322874

00:21:39.526 --> 00:21:41.479 so we ended up instead using.

NOTE Confidence: 0.80322874

00:21:41.480 --> 00:21:43.116 A more biochemical approach,

NOTE Confidence: 0.80322874

00:21:43.116 --> 00:21:46.239 which was to knock down TF are two,

NOTE Confidence: 0.80322874

00:21:46.240 --> 00:21:49.380 so TF are let me just go through back that

NOTE Confidence: 0.80322874

00:21:49.457 --> 00:21:52.595 there are two different cvars transfusion.

NOTE Confidence: 0.80322874

00:21:52.600 --> 00:21:54.765 I mean transferrin receptors, there's

NOTE Confidence: 0.80322874

00:21:54.765 --> 00:21:57.360 transparent Receptor One which is CD 71,

NOTE Confidence: 0.80322874

00:21:57.360 --> 00:22:00.536 and it's an all proliferating cells and very,

NOTE Confidence: 0.80322874

00:22:00.540 --> 00:22:03.319 very highly expressed in Eryth roid cells.

NOTE Confidence: 0.80322874

00:22:03.320 --> 00:22:05.300 And there's transparent receptor 2,

NOTE Confidence: 0.80322874

00:22:05.300 --> 00:22:06.960 which is entirely different.

NOTE Confidence: 0.80322874

00:22:06.960 --> 00:22:08.205 Transferrin receptor two

NOTE Confidence: 0.80322874

00:22:08.205 --> 00:22:10.458 acts more as an iron sensor,

NOTE Confidence: 0.80322874

00:22:10.460 --> 00:22:12.968 but not as an iron transporter.

NOTE Confidence: 0.80322874

00:22:12.970 --> 00:22:15.644 And what it does is it binds

NOTE Confidence: 0.80322874

00:22:15.644 --> 00:22:16.790 to Jolo transparent.

NOTE Confidence: 0.80322874

00:22:16.790 --> 00:22:17.554 That's transparent,

NOTE Confidence: 0.80322874

00:22:17.554 --> 00:22:19.846 that has iron bound to it,

NOTE Confidence: 0.80322874

00:22:19.850 --> 00:22:23.846 and when it's bound to that.

NOTE Confidence: 0.80322874

00:22:23.850 --> 00:22:25.390 Transparent it activates Erk 1

NOTE Confidence: 0.80322874

00:22:25.390 --> 00:22:27.580 two and P38 map kinase signaling,

NOTE Confidence: 0.80322874

00:22:27.580 --> 00:22:29.608 so it's kind of a baseline.

NOTE Confidence: 0.80322874

00:22:29.610 --> 00:22:31.305 The cells are always expressing

NOTE Confidence: 0.80322874

00:22:31.305 --> 00:22:33.000 TF R2 they have iron,

NOTE Confidence: 0.80322874

00:22:33.000 --> 00:22:34.700 they're saying to the MVP.

NOTE Confidence: 0.80322874

00:22:34.700 --> 00:22:36.390 We got plenty of iron.

NOTE Confidence: 0.80322874

00:22:36.390 --> 00:22:38.750 Do what you need to do if you

NOTE Confidence: 0.80322874

00:22:38.750 --> 00:22:40.569 actually have no iron present

NOTE Confidence: 0.80322874

00:22:40.569 --> 00:22:42.499 so that you have transparent,

NOTE Confidence: 0.80322874

00:22:42.500 --> 00:22:44.924 but it's not bound to iron then that

NOTE Confidence: 0.80322874

00:22:44.924 --> 00:22:47.579 Fr two comes off the cell membrane.

NOTE Confidence: 0.80322874

00:22:47.580 --> 00:22:49.782 It gets internalised and you lose

NOTE Confidence: 0.80322874

00:22:49.782 --> 00:22:52.672 your Erk 12 signal so it's really an

NOTE Confidence: 0.80322874

00:22:52.672 --> 00:22:54.990 iron sensor in the presence of it.

NOTE Confidence: 0.80322874

00:22:54.990 --> 00:22:56.380 It's saying to the cell.

NOTE Confidence: 0.80322874

00:22:56.380 --> 00:22:57.760 There's plenty of iron available.

NOTE Confidence: 0.80322874

00:22:57.760 --> 00:22:59.890 Do what you need to do in the absence is

NOTE Confidence: 0.81601065

00:22:59.948 --> 00:23:02.186 saying wait no eryth roid differentiation.

NOTE Confidence: 0.81601065

00:23:02.190 --> 00:23:03.575 I'll show you the data

NOTE Confidence: 0.81601065

00:23:03.575 --> 00:23:04.960 'cause we're low on iron.

NOTE Confidence: 0.81601065

00:23:04.960 --> 00:23:08.010 So what we do is we knocked down TF are two.

NOTE Confidence: 0.81601065

00:23:08.010 --> 00:23:09.882 This is just showing you the M RNA

NOTE Confidence: 0.81601065

00:23:09.882 --> 00:23:11.494 levels are decreased and true enough

NOTE Confidence: 0.81601065

00:23:11.494 --> 00:23:13.853 when you knock down TF are to the

NOTE Confidence: 0.81601065

00:23:13.853 --> 00:23:15.209 cells have slower proliferation.

NOTE Confidence: 0.81601065

00:23:15.210 --> 00:23:17.388 So that told us we might end up seeing

NOTE Confidence: 0.81601065

00:23:17.388 --> 00:23:19.636 a Meg Bias which is what happened.

NOTE Confidence: 0.81601065

00:23:19.640 --> 00:23:21.578 So what you're looking at here

NOTE Confidence: 0.81601065

00:23:21.578 --> 00:23:23.280 is the colony distribution and

NOTE Confidence: 0.81601065

00:23:23.280 --> 00:23:25.025 non transduced primary human MVP.

NOTE Confidence: 0.81601065

00:23:25.030 --> 00:23:27.250 Me P transduced with the scrambled

NOTE Confidence: 0.81601065

00:23:27.250 --> 00:23:29.991 SH RNA and transduced with an SH RNA

NOTE Confidence: 0.81601065

00:23:29.991 --> 00:23:31.980 against TF are one against TFR 2.

NOTE Confidence: 0.81601065

00:23:31.980 --> 00:23:33.654 Two different SH RNA's against TF

NOTE Confidence: 0.81601065

00:23:33.654 --> 00:23:35.920 R2 and in both cases we're seeing

NOTE Confidence: 0.81601065

00:23:35.920 --> 00:23:38.086 a megakaryocyte bias so this was

NOTE Confidence: 0.81601065

00:23:38.086 --> 00:23:40.256 consistent with the low iron environment.

NOTE Confidence: 0.81601065

00:23:40.260 --> 00:23:42.171 Both the mouse and human leading to

NOTE Confidence: 0.81601065

00:23:42.171 --> 00:23:44.216 a Meg bias Anna slower proliferation

NOTE Confidence: 0.81601065

00:23:44.216 --> 00:23:46.550 but what's the mechanism of this?

NOTE Confidence: 0.81601065

00:23:46.550 --> 00:23:48.895 So in order to assess the mechanism

NOTE Confidence: 0.81601065

00:23:48.895 --> 00:23:51.551 what we did is we went back to

NOTE Confidence: 0.81601065

00:23:51.551 --> 00:23:53.931 the Meyssan we fax sorted out the

NOTE Confidence: 0.81601065

00:23:53.931 --> 00:23:55.766 Mega Rich ride for janitors.

NOTE Confidence: 0.81601065

00:23:55.770 --> 00:23:57.494 And looked at weather.

NOTE Confidence: 0.81601065

00:23:57.494 --> 00:24:00.080 Looked at the gene expression patterns

NOTE Confidence: 0.81601065

00:24:00.157 --> 00:24:02.365 between wild type and temper 6

NOTE Confidence: 0.81601065

00:24:02.365 --> 00:24:05.075 knockout MVP and this work was done

NOTE Confidence: 0.81601065

00:24:05.075 --> 00:24:07.020 in collaboration with Tomata Baldy

NOTE Confidence: 0.81601065

00:24:07.020 --> 00:24:09.610 who is does hit our bioinformatics and

NOTE Confidence: 0.81601065

00:24:09.610 --> 00:24:12.620 what he showed is that there are many

NOTE Confidence: 0.81601065

00:24:12.620 --> 00:24:14.876 genes that are upregulated when you

NOTE Confidence: 0.81601065

00:24:14.876 --> 00:24:17.395 knock out temper 6 in these empiezan.

NOTE Confidence: 0.81601065

00:24:17.400 --> 00:24:20.344 Those tend to be targets of veg F,

NOTE Confidence: 0.81601065

00:24:20.350 --> 00:24:22.620 whereas there's downregulation of Erk

NOTE Confidence: 0.81601065

00:24:22.620 --> 00:24:25.309 target genes again consistent with the

NOTE Confidence: 0.81601065

00:24:25.309 --> 00:24:27.427 idea that you're losing Erk signaling

NOTE Confidence: 0.81601065

00:24:27.427 --> 00:24:30.129 because the TF R2 would be internalised.

NOTE Confidence: 0.81601065

00:24:30.130 --> 00:24:32.314 So what we did is we looked in

NOTE Confidence: 0.81601065

00:24:32.314 --> 00:24:34.789 those MVP's to see if their fasfa

NOTE Confidence: 0.81601065

00:24:34.789 --> 00:24:36.644 lurk levels were actually decreased

NOTE Confidence: 0.81601065

00:24:36.717 --> 00:24:39.063 and they were consistent with the

NOTE Confidence: 0.81601065

00:24:39.063 --> 00:24:40.980 decreased Erk target genes being

NOTE Confidence: 0.81601065

00:24:40.980 --> 00:24:42.380 phosphorylated and being expressed.

NOTE Confidence: 0.81601065

00:24:42.380 --> 00:24:44.448 We actually have decreased

NOTE Confidence: 0.81601065

00:24:44.448 --> 00:24:46.516 or toss work levels.

NOTE Confidence: 0.81601065

00:24:46.520 --> 00:24:49.048 Similarly, so that was in the mooring system.

NOTE Confidence: 0.81601065

00:24:49.050 --> 00:24:50.630 Similarly in the human system,

NOTE Confidence: 0.81601065

00:24:50.630 --> 00:24:52.835 when you knock down the TF R2,

NOTE Confidence: 0.81601065

00:24:52.840 --> 00:24:55.510 you have decreased phospho, Erk levels.

NOTE Confidence: 0.81601065

00:24:55.510 --> 00:24:58.190 So the model that we have for what's

NOTE Confidence: 0.81601065

00:24:58.190 --> 00:25:01.530 going on in this fake decision is that

NOTE Confidence: 0.81601065

00:25:01.530 --> 00:25:04.109 under normal iron conditions you have

NOTE Confidence: 0.81601065

00:25:04.109 --> 00:25:06.818 the cells making their decisions to go

NOTE Confidence: 0.81601065

00:25:06.818 --> 00:25:08.898 down the megakaryocytic versus areth

NOTE Confidence: 0.81601065

00:25:08.898 --> 00:25:11.430 ride progenitor cell lineages and in

NOTE Confidence: 0.81601065

00:25:11.497 --> 00:25:14.225 the presence in the case of iron deficiency,

NOTE Confidence: 0.81601065

00:25:14.230 --> 00:25:17.156 you get anemia at least in part

NOTE Confidence: 0.81601065

00:25:17.156 --> 00:25:19.899 because you have decreased TF R2.

NOTE Confidence: 0.81601065

00:25:19.900 --> 00:25:22.784 Which leads to decreased phospho Erk levels,

NOTE Confidence: 0.81601065

00:25:22.790 --> 00:25:24.450 decreased proliferation and a

NOTE Confidence: 0.81601065

00:25:24.450 --> 00:25:26.525 megakaryocyte bias which leads you

NOTE Confidence: 0.81601065

00:25:26.525 --> 00:25:28.563 to have elevated platelet count

NOTE Confidence: 0.81601065

00:25:28.563 --> 00:25:30.903 and a decrease erythroid count and

NOTE Confidence: 0.81601065

00:25:30.967 --> 00:25:33.072 the microcytic microcytosis is the

NOTE Confidence: 0.81601065

00:25:33.072 --> 00:25:35.177 downstream effect of lacking iron

NOTE Confidence: 0.81601065

00:25:35.180 --> 00:25:37.658 as the erythroid cells are maturing.

NOTE Confidence: 0.90009916

00:25:39.800 --> 00:25:42.278 So there are a lot of

NOTE Confidence: 0.90009916

00:25:42.278 --> 00:25:43.930 unanswered questions here and.

NOTE Confidence: 0.90009916

00:25:43.930 --> 00:25:46.186 Some of them are listed here.

NOTE Confidence: 0.90009916

00:25:46.190 --> 00:25:49.190 To what extent do me P self renew?

NOTE Confidence: 0.90009916

00:25:49.190 --> 00:25:52.158 I showed you that me PR this

NOTE Confidence: 0.90009916

00:25:52.158 --> 00:25:54.044 unique transitional state can

NOTE Confidence: 0.90009916

00:25:54.044 --> 00:25:56.180 me pee themselves proliferate.

NOTE Confidence: 0.90009916

00:25:56.180 --> 00:25:57.595 Does cell cycle speed actually

NOTE Confidence: 0.90009916

00:25:57.595 --> 00:25:59.431 change as cells are about to

NOTE Confidence: 0.90009916

00:25:59.431 --> 00:26:00.887 undergo their fate specification?

NOTE Confidence: 0.90009916

00:26:00.890 --> 00:26:02.934 Could we see cells starting to slow

NOTE Confidence: 0.90009916

00:26:02.934 --> 00:26:05.245 their cell cycle speed in that predicts

NOTE Confidence: 0.90009916

00:26:05.245 --> 00:26:07.303 there about to pick the megakaryocytic

NOTE Confidence: 0.90009916

00:26:07.362 --> 00:26:09.048 and speed up their their fate,

NOTE Confidence: 0.90009916

00:26:09.050 --> 00:26:10.940 their speed up their cell cycle,

NOTE Confidence: 0.90009916

00:26:10.940 --> 00:26:13.768 and start to pick the erythritol image?

NOTE Confidence: 0.90009916

00:26:13.770 --> 00:26:16.460 Are there cell characteristics that

NOTE Confidence: 0.90009916

00:26:16.460 --> 00:26:19.150 predict fate decisions like the

NOTE Confidence: 0.90009916

00:26:19.231 --> 00:26:21.715 cell size or the cell motility?

NOTE Confidence: 0.90009916

00:26:21.720 --> 00:26:24.268 What about if we changed the cytokines?

NOTE Confidence: 0.90009916

00:26:24.270 --> 00:26:26.090 Does that affect fate decisions?

NOTE Confidence: 0.90009916

00:26:26.090 --> 00:26:28.268 How did cells in the micro

NOTE Confidence: 0.90009916

00:26:28.268 --> 00:26:29.357 environment affect decisions?

NOTE Confidence: 0.90009916

00:26:29.360 --> 00:26:31.544 These are all unanswered questions that

NOTE Confidence: 0.90009916

00:26:31.544 --> 00:26:34.098 we're addressing in our laboratory now in.

NOTE Confidence: 0.90009916

00:26:34.100 --> 00:26:36.422 This brings me to the beautiful

NOTE Confidence: 0.90009916

00:26:36.422 --> 00:26:38.465 dynamic system that Vanessa Scanlon

NOTE Confidence: 0.90009916

00:26:38.465 --> 00:26:40.595 in my laboratory has developed.

NOTE Confidence: 0.90009916

00:26:40.600 --> 00:26:43.858 So what Vanessa is doing is she is live

NOTE Confidence: 0.90009916

00:26:43.858 --> 00:26:46.719 image Ng me P as they form colonies.

NOTE Confidence: 0.90009916

00:26:46.720 --> 00:26:49.321 So she fax or it's out the MVP and

NOTE Confidence: 0.90009916

00:26:49.321 --> 00:26:52.116 she puts them into semisolid medium,

NOTE Confidence: 0.90009916

00:26:52.120 --> 00:26:54.082 the same semisolid medium that we

NOTE Confidence: 0.90009916

00:26:54.082 --> 00:26:56.440 use for a colony forming assays.

NOTE Confidence: 0.90009916

00:26:56.440 --> 00:26:58.240 It's a collagen based medium.

NOTE Confidence: 0.90009916

00:26:58.240 --> 00:27:00.040 It's basically mega cult from

NOTE Confidence: 0.90009916

00:27:00.040 --> 00:27:01.120 stem cell technologies,

NOTE Confidence: 0.90009916

00:27:01.120 --> 00:27:02.920 and we've added Orris Republican,

NOTE Confidence: 0.90009916

00:27:02.920 --> 00:27:05.080 and she puts these into an

NOTE Confidence: 0.90009916

00:27:05.080 --> 00:27:06.520 Olympus Viva view system.

NOTE Confidence: 0.90009916

00:27:06.520 --> 00:27:08.680 And what this system is is.

NOTE Confidence: 0.90009916

00:27:08.680 --> 00:27:10.216 It's basically an incubator.

NOTE Confidence: 0.90009916

00:27:10.216 --> 00:27:12.520 And inside the incubator are these

NOTE Confidence: 0.90009916

00:27:12.585 --> 00:27:14.720 spots where you can put the dishes.

NOTE Confidence: 0.90009916

00:27:14.720 --> 00:27:17.123 You can put eight dishes in its hooked up

NOTE Confidence: 0.90009916

00:27:17.123 --> 00:27:19.816 to a computer an underneath the incubator.

NOTE Confidence: 0.90009916

00:27:19.820 --> 00:27:21.776 There is a fluorescence camera so

NOTE Confidence: 0.90009916

00:27:21.776 --> 00:27:23.421 you tell the fluorescence camera

NOTE Confidence: 0.90009916

00:27:23.421 --> 00:27:25.113 where to take pictures and how

NOTE Confidence: 0.90009916

00:27:25.113 --> 00:27:26.690 often to take pictures overtime

NOTE Confidence: 0.90009916

00:27:26.690 --> 00:27:29.049 and you since this is actually an

NOTE Confidence: 0.90009916

00:27:29.049 --> 00:27:31.226 incubator you can keep them in the

NOTE Confidence: 0.90009916

00:27:31.226 --> 00:27:33.220 incubator for up to two weeks,

NOTE Confidence: 0.90009916

00:27:33.220 --> 00:27:34.810 three weeks and really watch

NOTE Confidence: 0.90009916

00:27:34.810 --> 00:27:36.082 the individual colonies form.

NOTE Confidence: 0.90009916

00:27:36.090 --> 00:27:39.336 I should mention also that when.

NOTE Confidence: 0.90009916

00:27:39.340 --> 00:27:41.086 Vanessa was putting this system together.

NOTE Confidence: 0.90009916

00:27:41.090 --> 00:27:42.861 What she realizes she had to have

NOTE Confidence: 0.90009916

00:27:42.861 --> 00:27:44.945 some way of flattening the colonies so

NOTE Confidence: 0.90009916

00:27:44.945 --> 00:27:47.197 that we could watch the cells overtime

NOTE Confidence: 0.90009916

00:27:47.197 --> 00:27:49.269 and they didn't form a big stick.

NOTE Confidence: 0.90009916

00:27:49.270 --> 00:27:50.730 Big, thick 3 dimensional colony.

NOTE Confidence: 0.90009916

00:27:50.730 --> 00:27:53.010 So when she puts the cells in the

NOTE Confidence: 0.90009916

00:27:53.010 --> 00:27:55.474 first thing she does is she puts him

NOTE Confidence: 0.90009916

00:27:55.474 --> 00:27:57.306 in about 15 microliters and then

NOTE Confidence: 0.90009916

00:27:57.306 --> 00:27:59.490 put the cover slip on top of that.

NOTE Confidence: 0.90009916

00:27:59.490 --> 00:28:00.990 It doesn't affect the distribution

NOTE Confidence: 0.90009916

00:28:00.990 --> 00:28:02.818 of colony subtypes and it allows

NOTE Confidence: 0.90009916

00:28:02.818 --> 00:28:04.450 us to see the individual cells.

NOTE Confidence: 0.90009916

00:28:04.450 --> 00:28:06.202 So what we're seeing here is

NOTE Confidence: 0.90009916

00:28:06.202 --> 00:28:07.370 a colony with megakaryocytes.

NOTE Confidence: 0.90009916

00:28:07.370 --> 00:28:09.589 That's the green and erythrocytes in red.

NOTE Confidence: 0.90009916

00:28:09.590 --> 00:28:11.060 Here's a megakaryocyte only colony,

NOTE Confidence: 0.90009916

00:28:11.060 --> 00:28:12.806 and here's an Areth Royd Colony.

NOTE Confidence: 0.90009916

00:28:12.810 --> 00:28:13.647 I left out.

NOTE Confidence: 0.90009916

00:28:13.647 --> 00:28:15.321 The fact I'm sorry that towards

NOTE Confidence: 0.90009916

00:28:15.321 --> 00:28:17.209 the end of the culture period,

NOTE Confidence: 0.90009916

00:28:17.210 --> 00:28:18.968 she adds antibodies against 235 a.

NOTE Confidence: 0.90009916

00:28:18.970 --> 00:28:20.826 That's like a four and A and CD

NOTE Confidence: 0.90009916

00:28:20.826 --> 00:28:23.154 41 so that we can identify the

NOTE Confidence: 0.90009916

00:28:23.154 --> 00:28:24.234 different cell types.

NOTE Confidence: 0.90009916

00:28:24.240 --> 00:28:25.705 If you add these antibodies

NOTE Confidence: 0.90009916

00:28:25.705 --> 00:28:27.170 too early in the culture,

NOTE Confidence: 0.90009916

00:28:27.170 --> 00:28:28.640 then the cells will die.

NOTE Confidence: 0.90009916

00:28:28.640 --> 00:28:29.812 So there's some phototoxicity

NOTE Confidence: 0.90009916

00:28:29.812 --> 00:28:31.570 that we really needed to address,

NOTE Confidence: 0.8274063

00:28:31.570 --> 00:28:32.446 but we're still.

NOTE Confidence: 0.8274063

00:28:32.446 --> 00:28:33.614 We're still optimizing that,

NOTE Confidence: 0.8274063

00:28:33.620 --> 00:28:36.630 so we can add the antibodies sooner.

NOTE Confidence: 0.8274063

00:28:36.630 --> 00:28:38.849 So what I'm going to show you

NOTE Confidence: 0.8274063

00:28:38.849 --> 00:28:41.589 here is one of the initial images.

NOTE Confidence: 0.8274063

00:28:41.590 --> 00:28:43.290 Stacked images or movies that

NOTE Confidence: 0.8274063

00:28:43.290 --> 00:28:45.427 Vanessa was able to get when

NOTE Confidence: 0.8274063

00:28:45.427 --> 00:28:47.247 she sorted primary human MVP,

NOTE Confidence: 0.8274063

00:28:47.250 --> 00:28:48.715 put them in culture and

NOTE Confidence: 0.8274063

00:28:48.715 --> 00:28:50.704 watch them form a colony of

NOTE Confidence: 0.8274063

00:28:50.704 --> 00:28:52.556 megakaryocytes and erythroid cells.

NOTE Confidence: 0.92436194

00:28:56.960 --> 00:28:58.787 I'm still here, I just want you to watch.

NOTE Confidence: 0.8286442

00:29:01.470 --> 00:29:03.606 When you start to see the green color,

NOTE Confidence: 0.8286442

00:29:03.610 --> 00:29:04.674 that's when she's added

NOTE Confidence: 0.8286442

00:29:04.674 --> 00:29:06.004 the antibody against E 41,

NOTE Confidence: 0.8286442

00:29:06.010 --> 00:29:07.606 so those are the megakaryocytes an,

NOTE Confidence: 0.8286442

00:29:07.610 --> 00:29:09.479 then the pink cells are the ones

NOTE Confidence: 0.8286442

00:29:09.479 --> 00:29:10.820 that are standing with two,

NOTE Confidence: 0.8286442

00:29:10.820 --> 00:29:13.780 35 A and those are the erythroid cells.

NOTE Confidence: 0.8286442

00:29:13.780 --> 00:29:15.490 So the wonderful thing about

NOTE Confidence: 0.8286442

00:29:15.490 --> 00:29:17.507 having these time lapse images is

NOTE Confidence: 0.8286442

00:29:17.507 --> 00:29:19.250 that then you can play it back

NOTE Confidence: 0.8286442

00:29:19.250 --> 00:29:21.152 and forth and actually figure out

NOTE Confidence: 0.8286442

00:29:21.152 --> 00:29:23.168 which cell divided to Weikum which

NOTE Confidence: 0.8286442

00:29:23.170 --> 00:29:25.354 other cell type and make a tree.

NOTE Confidence: 0.8286442

00:29:25.360 --> 00:29:26.436 So here is it.

NOTE Confidence: 0.8286442

00:29:26.436 --> 00:29:28.483 Tree or lineages tree from a single

NOTE Confidence: 0.8286442

00:29:28.483 --> 00:29:30.328 by potent megathread for Genitor

NOTE Confidence: 0.8286442

00:29:30.328 --> 00:29:32.446 and what we've done with these

NOTE Confidence: 0.8286442

00:29:32.446 --> 00:29:34.462 trees is any cell that was green

NOTE Confidence: 0.8286442

00:29:34.462 --> 00:29:36.906 at the end or Meg committed at the

NOTE Confidence: 0.8286442

00:29:36.906 --> 00:29:39.447 end became a green cell at the end.

NOTE Confidence: 0.8286442

00:29:39.450 --> 00:29:41.666 Any cell at the end? That's red.

NOTE Confidence: 0.8286442

00:29:41.666 --> 00:29:43.962 That was a rich Rd committed if.

NOTE Confidence: 0.8286442

00:29:43.970 --> 00:29:46.354 There's a cell that has downstream of it,

NOTE Confidence: 0.8286442

00:29:46.360 --> 00:29:48.460 some green cells and some red cells.

NOTE Confidence: 0.8286442

00:29:48.460 --> 00:29:51.910 We call data by Potence Elan, it's blue.

NOTE Confidence: 0.8286442

00:29:51.910 --> 00:29:53.974 So one of the first things you can

NOTE Confidence: 0.8286442

00:29:53.974 --> 00:29:56.062 see here is the blue cells can self

NOTE Confidence: 0.8286442

00:29:56.062 --> 00:29:58.378 renew that me P can self renew so

NOTE Confidence: 0.8286442

00:29:58.378 --> 00:30:00.250 that had not previously been show.

NOTE Confidence: 0.8286442

00:30:00.250 --> 00:30:02.274 Phone and now we have that in every

NOTE Confidence: 0.8286442

00:30:02.274 --> 00:30:04.506 video we have with by potent colonies,

NOTE Confidence: 0.8286442

00:30:04.510 --> 00:30:06.792 we can see that the MVP themselves

NOTE Confidence: 0.8286442

00:30:06.792 --> 00:30:08.250 self renew in vitro.

NOTE Confidence: 0.8286442

00:30:08.250 --> 00:30:10.035 What you can also see is that

NOTE Confidence: 0.8286442

00:30:10.035 --> 00:30:11.330 there are different patterns.

NOTE Confidence: 0.8286442

00:30:11.330 --> 00:30:13.466 Sometimes we have a cell that commits to

NOTE Confidence: 0.8286442

00:30:13.466 --> 00:30:15.247 the erythroid lineages quite early on,

NOTE Confidence: 0.8286442

00:30:15.250 --> 00:30:16.650 and sometimes it takes much,

NOTE Confidence: 0.8286442

00:30:16.650 --> 00:30:18.050 much longer to commit to

NOTE Confidence: 0.8286442

00:30:18.050 --> 00:30:18.890 the erythroid lineage.

NOTE Confidence: 0.8286442

00:30:18.890 --> 00:30:22.070 Similarly with the megakaryocyte Lenny edge.

NOTE Confidence: 0.8286442

00:30:22.070 --> 00:30:23.440 We also have colonies that

NOTE Confidence: 0.8286442

00:30:23.440 --> 00:30:24.536 are eryth roid only.

NOTE Confidence: 0.8286442

00:30:24.540 --> 00:30:26.451 That came from our sorted me pee

NOTE Confidence: 0.8286442

00:30:26.451 --> 00:30:28.098 and colonies that are mega only.

NOTE Confidence: 0.8286442

00:30:28.100 --> 00:30:30.284 They come from our original MVP and

NOTE Confidence: 0.8286442

00:30:30.284 --> 00:30:32.660 what you can see is initially the.

NOTE Confidence: 0.8286442

00:30:32.660 --> 00:30:34.530 Cell cycle is relatively slow,

NOTE Confidence: 0.8286442

00:30:34.530 --> 00:30:36.959 but when it's a erythroid only colony

NOTE Confidence: 0.8286442

00:30:36.959 --> 00:30:39.368 they get very very fast overtime,

NOTE Confidence: 0.8286442

00:30:39.370 --> 00:30:40.862 whereas the megakaryocytes or

NOTE Confidence: 0.8286442

00:30:40.862 --> 00:30:42.727 have a slower colony formation.

NOTE Confidence: 0.9027187

00:30:44.990 --> 00:30:47.496 So this gave us the opportunity to

NOTE Confidence: 0.9027187

00:30:47.496 --> 00:30:50.053 address to start to address some of

NOTE Confidence: 0.9027187

00:30:50.053 --> 00:30:52.590 the many many questions that we have,

NOTE Confidence: 0.9027187

00:30:52.590 --> 00:30:54.756 and the analysis is on going.

NOTE Confidence: 0.9027187

00:30:54.760 --> 00:30:57.559 I'll just give you a glimpse as to some

NOTE Confidence: 0.9027187

00:30:57.559 --> 00:31:00.480 one of the stories that has become more

NOTE Confidence: 0.9027187

00:31:00.480 --> 00:31:03.230 clear now that we have this dynamic

NOTE Confidence: 0.9027187

00:31:03.230 --> 00:31:05.618 system of looking at the colonies.

NOTE Confidence: 0.9027187

00:31:05.620 --> 00:31:08.770 So the dogma that many people believe.

NOTE Confidence: 0.9027187

00:31:08.770 --> 00:31:09.492 Maybe not.

NOTE Confidence: 0.9027187

00:31:09.492 --> 00:31:12.019 The folks on this conference on this

NOTE Confidence: 0.9027187

00:31:12.019 --> 00:31:14.609 web and R is that an Emmy P might

NOTE Confidence: 0.9027187

00:31:14.609 --> 00:31:17.063 make its fate decision by in the

NOTE Confidence: 0.9027187

00:31:17.063 --> 00:31:19.173 presence of Thrombo poet and picking

NOTE Confidence: 0.9027187

00:31:19.173 --> 00:31:20.488 the megakaryocyte Lenny edge and

NOTE Confidence: 0.9027187

00:31:20.488 --> 00:31:21.950 in the presence of erythropoetin

NOTE Confidence: 0.9027187

00:31:21.950 --> 00:31:23.735 picking theorist Freud Lenny Edge.

NOTE Confidence: 0.9027187

00:31:23.740 --> 00:31:25.844 So just to blow that up here that

NOTE Confidence: 0.9027187

00:31:25.844 --> 00:31:28.430 if we were to grow the cells in

NOTE Confidence: 0.9027187

00:31:28.430 --> 00:31:30.419 the presence of Thrombo poet and

NOTE Confidence: 0.9027187

00:31:30.419 --> 00:31:32.795 in the absence of Ipoh we would get

NOTE Confidence: 0.9027187

00:31:32.800 --> 00:31:34.310 colonies that are only megakaryocyte.

NOTE Confidence: 0.9027187

00:31:34.310 --> 00:31:36.718 And if we left out the thrombi poet,

NOTE Confidence: 0.9027187

00:31:36.720 --> 00:31:39.378 and we'd get colonies that are

NOTE Confidence: 0.9027187

00:31:39.378 --> 00:31:40.707 just eryth roid.

NOTE Confidence: 0.9027187

00:31:40.710 --> 00:31:42.873 Those of you who know the literature

NOTE Confidence: 0.9027187

00:31:42.873 --> 00:31:44.859 more deeply might not predict that

NOTE Confidence: 0.9027187

00:31:44.859 --> 00:31:46.845 something just didn't show up here,

NOTE Confidence: 0.9027187

00:31:46.850 --> 00:31:48.782 which is the background on this

NOTE Confidence: 0.9027187

00:31:48.782 --> 00:31:49.748 information background here.

NOTE Confidence: 0.9027187

00:31:49.750 --> 00:31:52.495 So what I wanted to make sure to tell

NOTE Confidence: 0.9027187

00:31:52.495 --> 00:31:55.564 you is some of the background as to why.

NOTE Confidence: 0.9027187

00:31:55.570 --> 00:31:57.656 If you culture the cells in the

NOTE Confidence: 0.9027187

00:31:57.656 --> 00:31:59.768 presence of minus Devitte Bowen tipo,

NOTE Confidence: 0.9027187

00:31:59.770 --> 00:32:01.654 you might not predict that this

NOTE Confidence: 0.9027187

00:32:01.654 --> 00:32:03.640 is what that you would get.

NOTE Confidence: 0.9027187

00:32:03.640 --> 00:32:06.856 The only in MK only colonies.

NOTE Confidence: 0.9027187

00:32:06.860 --> 00:32:09.324 So initially what we did is we just

NOTE Confidence: 0.9027187

00:32:09.324 --> 00:32:11.061 perform static colony forming unit

NOTE Confidence: 0.9027187

00:32:11.061 --> 00:32:13.518 assays in the presence and absence of

NOTE Confidence: 0.9027187

00:32:13.589 --> 00:32:15.619 vivo en tipo and the background here

NOTE Confidence: 0.9027187

00:32:15.619 --> 00:32:18.095 is that we knew that if you don't

NOTE Confidence: 0.9027187

00:32:18.095 --> 00:32:19.379 have the Erythropoetin Receptor,

NOTE Confidence: 0.9027187

00:32:19.380 --> 00:32:21.627 you don't form normal Aris Freud Colonies,

NOTE Confidence: 0.9027187

00:32:21.630 --> 00:32:23.230 but you have some colonies,

NOTE Confidence: 0.9027187

00:32:23.230 --> 00:32:25.477 so it's not that they can't pick

NOTE Confidence: 0.9027187

00:32:25.477 --> 00:32:26.440 the fate decision.

NOTE Confidence: 0.9027187

00:32:26.440 --> 00:32:28.596 It really sounds like it really appears

NOTE Confidence: 0.9027187

00:32:28.596 --> 00:32:30.608 more that it's a survival signal,

NOTE Confidence: 0.9027187

00:32:30.610 --> 00:32:32.215 and similarly if you overexpress

NOTE Confidence: 0.9027187

00:32:32.215 --> 00:32:33.178 the thrombopoietin receptor,

NOTE Confidence: 0.9027187

00:32:33.180 --> 00:32:36.204 it doesn't cause the cells to just become

NOTE Confidence: 0.9027187

00:32:36.204 --> 00:32:38.409 megakaryocytes and not a rich way itself.

NOTE Confidence: 0.9027187

00:32:38.410 --> 00:32:40.042 So we thought there would probably

NOTE Confidence: 0.9027187

00:32:40.042 --> 00:32:42.387 be more to it and we decided to

NOTE Confidence: 0.9027187

00:32:42.387 --> 00:32:44.187 test that using this dynamic model.

NOTE Confidence: 0.9027187

00:32:44.190 --> 00:32:46.310 So the first thing that we did and

NOTE Confidence: 0.9027187

00:32:46.310 --> 00:32:48.527 this was work that was done by an

NOTE Confidence: 0.9027187

00:32:48.527 --> 00:32:50.316 undergrad in my lab several years

NOTE Confidence: 0.9027187

00:32:50.316 --> 00:32:52.563 ago is when we grew the colonies.

NOTE Confidence: 0.9027187

00:32:52.570 --> 00:32:54.020 In the absence of tipo,

NOTE Confidence: 0.9027187

00:32:54.020 --> 00:32:55.748 we saw no difference in the

NOTE Confidence: 0.9027187

00:32:55.748 --> 00:32:56.324 fate specification,

NOTE Confidence: 0.9027187

00:32:56.330 --> 00:32:57.946 just fewer colonies suggesting

NOTE Confidence: 0.9027187

00:32:57.946 --> 00:32:59.158 a survival detect.

NOTE Confidence: 0.9027187

00:32:59.160 --> 00:32:59.880 In contrast,

NOTE Confidence: 0.9027187

00:32:59.880 --> 00:33:01.680 when we leave out erythropoetin,

NOTE Confidence: 0.9027187

00:33:01.680 --> 00:33:03.840 we got absolutely no erythroid colonies,

NOTE Confidence: 0.9027187

00:33:03.840 --> 00:33:06.000 suggesting maybe that it was that

NOTE Confidence: 0.9027187

00:33:06.000 --> 00:33:07.850 are important is necessary for

NOTE Confidence: 0.9027187

00:33:07.850 --> 00:33:09.600 the erythroid colonies to grow,

NOTE Confidence: 0.9027187

00:33:09.600 --> 00:33:12.480 or it was necessary for the fate decision,

NOTE Confidence: 0.9027187

00:33:12.480 --> 00:33:14.640 but we couldn't tell the difference,

NOTE Confidence: 0.9027187

00:33:14.640 --> 00:33:17.520 but we've gotten a little smarter since then,

NOTE Confidence: 0.9027187

00:33:17.520 --> 00:33:20.688 and what we did is instead of just using

NOTE Confidence: 0.9027187

00:33:20.688 --> 00:33:23.640 CD-235-A as our marker for with Tripoli Sis,

NOTE Confidence: 0.9027187

00:33:23.640 --> 00:33:25.080 we're using an earlier

NOTE Confidence: 0.9027187

00:33:25.080 --> 00:33:27.240 marker which is CD71 CD 71,

NOTE Confidence: 0.9027187

00:33:27.240 --> 00:33:28.365 which is transparent.

NOTE Confidence: 0.9027187

00:33:28.365 --> 00:33:29.865 Receptor one goes up.

NOTE Confidence: 0.9027187

00:33:29.870 --> 00:33:31.282 Logarithmically as cells commit

NOTE Confidence: 0.9027187

00:33:31.282 --> 00:33:33.400 to the erythroid lineage and we

NOTE Confidence: 0.7891538

00:33:33.465 --> 00:33:35.145 found that if we staying for

NOTE Confidence: 0.7891538

00:33:35.145 --> 00:33:36.779 CD71 and CD41 instead of 2:35,

NOTE Confidence: 0.7891538

00:33:36.780 --> 00:33:39.036 but we could actually see that

NOTE Confidence: 0.7891538

00:33:39.036 --> 00:33:40.924 the cells were committing to

NOTE Confidence: 0.7891538

00:33:40.924 --> 00:33:43.087 be a Richard Lenny Edge even in

NOTE Confidence: 0.7891538

00:33:43.087 --> 00:33:44.920 the absence of a riffle poet.

NOTE Confidence: 0.7891538

00:33:44.920 --> 00:33:46.810 So these are now the data

NOTE Confidence: 0.7891538

00:33:46.810 --> 00:33:48.070 with this new marker,

NOTE Confidence: 0.7891538

00:33:48.070 --> 00:33:50.905 the CD 235 a what you're looking at is.

NOTE Confidence: 0.7891538

00:33:50.910 --> 00:33:53.430 This is a colony that formed both Mega.

NOTE Confidence: 0.7891538

00:33:53.430 --> 00:33:55.326 These are three different colonies that

NOTE Confidence: 0.7891538

00:33:55.326 --> 00:33:56.890 formed megakaryocytes and erythroid cells.

NOTE Confidence: 0.7891538

00:33:56.890 --> 00:33:57.520 Control colony.

NOTE Confidence: 0.7891538

00:33:57.520 --> 00:33:59.725 Here's one in the absence of Thrombopoietin,

NOTE Confidence: 0.7891538

00:33:59.730 --> 00:34:02.331 and here's one in the absence of A with

NOTE Confidence: 0.7891538

00:34:02.331 --> 00:34:05.075 report and see the colonies are much smaller,

NOTE Confidence: 0.7891538

00:34:05.080 --> 00:34:06.264 but they're still here.

NOTE Confidence: 0.7891538

00:34:06.264 --> 00:34:08.550 This is the and MK only colony,

NOTE Confidence: 0.7891538

00:34:08.550 --> 00:34:09.806 and here's a risk.

NOTE Confidence: 0.7891538

00:34:09.806 --> 00:34:12.010 Weighed only colony, so they all form.

NOTE Confidence: 0.7891538

00:34:12.010 --> 00:34:12.955 They're just much,

NOTE Confidence: 0.7891538

00:34:12.960 --> 00:34:14.928 much smaller in the absence of.

NOTE Confidence: 0.7891538

00:34:14.930 --> 00:34:15.289 Otherwise,

NOTE Confidence: 0.7891538

00:34:15.289 --> 00:34:17.443 report when we think in retrospect

NOTE Confidence: 0.7891538

00:34:17.443 --> 00:34:19.596 that this is that the standing

NOTE Confidence: 0.7891538

00:34:19.596 --> 00:34:21.618 for 235 was really the problem,

NOTE Confidence: 0.7891538

00:34:21.620 --> 00:34:23.882 but that neither equal nor tipo

NOTE Confidence: 0.7891538

00:34:23.882 --> 00:34:26.190 affects the fate decision of the MVP,

NOTE Confidence: 0.7891538

00:34:26.190 --> 00:34:28.702 so we wanted to look at to improve

NOTE Confidence: 0.7891538

00:34:28.702 --> 00:34:31.334 this further by making a video of

NOTE Confidence: 0.7891538

00:34:31.334 --> 00:34:33.269 timeless microscopy of colony forming

NOTE Confidence: 0.7891538

00:34:33.344 --> 00:34:35.348 in the absence of humble poet.

NOTE Confidence: 0.7891538

00:34:35.350 --> 00:34:37.681 And so this is a colony growing

NOTE Confidence: 0.7891538

00:34:37.681 --> 00:34:39.920 in the absence of crumble poet,

NOTE Confidence: 0.7891538

00:34:39.920 --> 00:34:42.279 and you saw that other colony earlier

NOTE Confidence: 0.7891538

00:34:42.279 --> 00:34:45.087 and it just kept growing and growing.

NOTE Confidence: 0.7891538

00:34:45.090 --> 00:34:47.637 And what you'll see here is as the cells.

NOTE Confidence: 0.7891538

00:34:47.640 --> 00:34:51.736 So start to proliferate and form a colony.

NOTE Confidence: 0.7891538

00:34:51.740 --> 00:34:53.539 Still start to die so you can

NOTE Confidence: 0.7891538

00:34:53.539 --> 00:34:54.820 see that we picked.

NOTE Confidence: 0.7891538

00:34:54.820 --> 00:34:56.220 Those red ones are the

NOTE Confidence: 0.7891538

00:34:56.220 --> 00:34:57.060 erythroid limited sells.

NOTE Confidence: 0.7891538

00:34:57.060 --> 00:34:59.300 The green ones are the Meg committed cells,

NOTE Confidence: 0.7891538

00:34:59.300 --> 00:35:01.358 but you're not seeing that same

NOTE Confidence: 0.7891538

00:35:01.358 --> 00:35:02.730 log Arhythmic expansion itself

NOTE Confidence: 0.7891538

00:35:02.788 --> 00:35:04.558 because they're starting to die.

NOTE Confidence: 0.7891538

00:35:04.560 --> 00:35:06.373 And what this is showing us is

NOTE Confidence: 0.7891538

00:35:06.373 --> 00:35:08.191 that are smaller colonies are not

NOTE Confidence: 0.7891538

00:35:08.191 --> 00:35:09.821 just due to decreased proliferation

NOTE Confidence: 0.7891538

00:35:09.821 --> 00:35:11.908 in the absence of from a potent,

NOTE Confidence: 0.7891538

00:35:11.910 --> 00:35:13.695 but actually do to cell death in

NOTE Confidence: 0.7891538

00:35:13.695 --> 00:35:15.439 the absence of Fraud Department,

NOTE Confidence: 0.7891538

00:35:15.440 --> 00:35:16.910 which we wouldn't want otherwise

NOTE Confidence: 0.7891538

00:35:16.910 --> 00:35:18.380 have been able to see.

NOTE Confidence: 0.79554796

00:35:20.500 --> 00:35:22.135 Mrs. Just showing you these

NOTE Confidence: 0.79554796

00:35:22.135 --> 00:35:22.789 colonies side-by-side,

NOTE Confidence: 0.79554796

00:35:22.790 --> 00:35:24.425 here's a normal colony with

NOTE Confidence: 0.79554796

00:35:24.425 --> 00:35:25.406 megakaryocytes in erythrocytes,

NOTE Confidence: 0.79554796

00:35:25.410 --> 00:35:28.050 and here's a colony with that has both

NOTE Confidence: 0.79554796

00:35:28.050 --> 00:35:29.660 megakaryocytes in a richer sites,

NOTE Confidence: 0.79554796

00:35:29.660 --> 00:35:32.225 but the vast majority of cells died so that

NOTE Confidence: 0.79554796

00:35:32.225 --> 00:35:34.889 you didn't get this huge colony expansion.

NOTE Confidence: 0.79554796

00:35:34.890 --> 00:35:37.144 So our data really have shown quite

NOTE Confidence: 0.79554796

00:35:37.144 --> 00:35:39.153 nicely that tipo versus EPO do

NOTE Confidence: 0.79554796

00:35:39.153 --> 00:35:41.097 not affect the MVP fake decision,

NOTE Confidence: 0.79554796

00:35:41.100 --> 00:35:42.520 so it's negative data,

NOTE Confidence: 0.79554796

00:35:42.520 --> 00:35:44.650 but it's negative data where we're

NOTE Confidence: 0.79554796

00:35:44.719 --> 00:35:46.815 starting to get a clue as to the

NOTE Confidence: 0.79554796

00:35:46.815 --> 00:35:49.206 fact that tipo is necessary for cell

NOTE Confidence: 0.79554796

00:35:49.206 --> 00:35:51.011 survival and Ipoh is necessary.

NOTE Confidence: 0.79554796

00:35:51.020 --> 00:35:53.768 For with Droid maturation.

NOTE Confidence: 0.79554796

00:35:53.770 --> 00:35:56.083 So of course we have a lot of on

NOTE Confidence: 0.79554796

00:35:56.083 --> 00:35:58.506 going studies looking at whether MVP,

NOTE Confidence: 0.79554796

00:35:58.510 --> 00:36:00.589 self renew and weather cell cycle speed

NOTE Confidence: 0.79554796

00:36:00.589 --> 00:36:01.990 predicts subsequent fate decisions,

NOTE Confidence: 0.79554796

00:36:01.990 --> 00:36:03.805 whether there are other characteristics

NOTE Confidence: 0.79554796

00:36:03.805 --> 00:36:05.964 that affect cell motility and really

NOTE Confidence: 0.79554796

00:36:05.964 --> 00:36:07.698 exciting new data that Vanessa is

NOTE Confidence: 0.79554796

00:36:07.698 --> 00:36:09.570 getting was on the role of cells,

NOTE Confidence: 0.79554796

00:36:09.570 --> 00:36:11.466 other cells within the micro environment,

NOTE Confidence: 0.79554796

00:36:11.470 --> 00:36:13.050 and how they affect MVP,

NOTE Confidence: 0.79554796

00:36:13.050 --> 00:36:14.955 fate specification 'cause all of

NOTE Confidence: 0.79554796

00:36:14.955 --> 00:36:17.205 our videos to Dayton colonies or

NOTE Confidence: 0.79554796

00:36:17.205 --> 00:36:19.144 with pure MVP and not with the

NOTE Confidence: 0.79554796

00:36:19.144 --> 00:36:21.145 other cells that they would be next

NOTE Confidence: 0.79554796

00:36:21.145 --> 00:36:24.120 to in the bone marrow environment.

NOTE Confidence: 0.79554796

00:36:24.120 --> 00:36:27.400 So to summarize what I told you today,

NOTE Confidence: 0.79554796

00:36:27.400 --> 00:36:29.992 single cell RNA seq reveals that

NOTE Confidence: 0.79554796

00:36:29.992 --> 00:36:31.720 MVP represented unique transitional

NOTE Confidence: 0.79554796

00:36:31.786 --> 00:36:34.072 state in both primary human cells

NOTE Confidence: 0.79554796

00:36:34.072 --> 00:36:35.596 and primary mooring cells.

NOTE Confidence: 0.79554796

00:36:35.600 --> 00:36:38.880 That these MVP are capable of self renewal.

NOTE Confidence: 0.79554796

00:36:38.880 --> 00:36:41.640 The single cell RNA seq also gave us

NOTE Confidence: 0.79554796

00:36:41.640 --> 00:36:44.540 a clue that cell cycle differences

NOTE Confidence: 0.79554796

00:36:44.540 --> 00:36:47.190 between MVP Meg progenitors inner

NOTE Confidence: 0.79554796

00:36:47.190 --> 00:36:49.658 it's right for janitors are.

NOTE Confidence: 0.79554796

00:36:49.660 --> 00:36:51.376 Probably playing a role in that

NOTE Confidence: 0.79554796

00:36:51.376 --> 00:36:53.292 fake decision where the slower cell

NOTE Confidence: 0.79554796

00:36:53.292 --> 00:36:55.077 cycle promotes a megakaryocyte fate,

NOTE Confidence: 0.79554796

00:36:55.080 --> 00:36:57.624 whereas a faster cell cycle promotes

NOTE Confidence: 0.79554796

00:36:57.624 --> 00:36:59.320 in Eryth Roid Fate.

NOTE Confidence: 0.79554796

00:36:59.320 --> 00:37:01.336 The in vivo timer mice supported

NOTE Confidence: 0.79554796

00:37:01.336 --> 00:37:03.930 the fact that the MVP or slower

NOTE Confidence: 0.79554796

00:37:03.930 --> 00:37:05.106 than Meg Progenitors,

NOTE Confidence: 0.79554796

00:37:05.110 --> 00:37:08.236 which are slower than originally projected.

NOTE Confidence: 0.79554796

00:37:08.240 --> 00:37:10.571 I showed you data that the iron

NOTE Confidence: 0.79554796

00:37:10.571 --> 00:37:12.737 content in MVP toggles the MK

NOTE Confidence: 0.79554796

00:37:12.737 --> 00:37:15.271 versus E fate decision via veg FERK

NOTE Confidence: 0.79554796

00:37:15.353 --> 00:37:17.831 or signaling and the time lapse

NOTE Confidence: 0.79554796

00:37:17.831 --> 00:37:20.576 imaging reveals that tipo Niko do

NOTE Confidence: 0.79554796

00:37:20.576 --> 00:37:23.834 not affect fate decisions per say.

NOTE Confidence: 0.79554796

00:37:23.840 --> 00:37:25.600 So I've not acknowledge people

NOTE Confidence: 0.79554796

00:37:25.600 --> 00:37:27.008 as we've gone along,

NOTE Confidence: 0.79554796

00:37:27.010 --> 00:37:29.618 so I hope that I called on everybody

NOTE Confidence: 0.79554796

00:37:29.618 --> 00:37:32.216 who was played a role in this work

NOTE Confidence: 0.79554796

00:37:32.216 --> 00:37:35.098 on may not have mentioned Lee Grimes,

NOTE Confidence: 0.79554796

00:37:35.100 --> 00:37:36.588 who works with Nathan,

NOTE Confidence: 0.79554796

00:37:36.588 --> 00:37:38.820 and Lee has also been incredibly

NOTE Confidence: 0.79554796

00:37:38.891 --> 00:37:40.376 helpful in us with us.

NOTE Confidence: 0.79554796

00:37:40.380 --> 00:37:42.160 Analyzing the time lapse images

NOTE Confidence: 0.79554796

00:37:42.160 --> 00:37:43.940 'cause he asks tremendously important

NOTE Confidence: 0.79554796

00:37:43.995 --> 00:37:45.690 questions that we're now beginning

NOTE Confidence: 0.79554796

00:37:45.690 --> 00:37:46.368 to address,

NOTE Confidence: 0.79554796

00:37:46.370 --> 00:37:48.908 which is how likely is it that an MVP

NOTE Confidence: 0.79554796

00:37:48.908 --> 00:37:51.464 will self renew versus undergoing mag

NOTE Confidence: 0.79554796

00:37:51.464 --> 00:37:53.818 versus inner. If word fate decision.

NOTE Confidence: 0.79554796

00:37:53.818 --> 00:37:54.919 Overtime in culture.

NOTE Confidence: 0.79554796

00:37:54.920 --> 00:37:57.265 I want to mention that my lab

NOTE Confidence: 0.79554796

00:37:57.265 --> 00:37:59.339 is looking for new postdocs,

NOTE Confidence: 0.79554796

00:37:59.340 --> 00:38:01.145 so please consider applying and

NOTE Confidence: 0.79554796

00:38:01.145 --> 00:38:03.476 that this work was also supported

NOTE Confidence: 0.79554796

00:38:03.476 --> 00:38:05.741 by the Yale Cooperative Center

NOTE Confidence: 0.79554796

00:38:05.741 --> 00:38:07.553 of excellence in Hematology.

NOTE Confidence: 0.79554796

00:38:07.560 --> 00:38:08.300 Thank you very much.