

WEBVTT

NOTE duration:"00:55:33"

NOTE recognizability:0.927

NOTE language:en-us

NOTE Confidence: 0.94125994

00:00:00.000 --> 00:00:02.424 Hi, everyone. Thank you so much

NOTE Confidence: 0.94125994

00:00:02.424 --> 00:00:04.606 for joining us today. I'm Tiffany,

NOTE Confidence: 0.94125994

00:00:04.606 --> 00:00:06.587 I'm a fourth year PhD student and

NOTE Confidence: 0.94125994

00:00:06.587 --> 00:00:08.351 the Pathology Grand Round Student

NOTE Confidence: 0.94125994

00:00:08.351 --> 00:00:10.720 Committee is very excited to have Dr.

NOTE Confidence: 0.94125994

00:00:10.720 --> 00:00:14.116 Jennifer Jones here with us today.

NOTE Confidence: 0.94125994

00:00:14.120 --> 00:00:15.877 It's not. Maybe I'll just use the.

NOTE Confidence: 0.950317

00:00:18.200 --> 00:00:18.640 Hello.

NOTE Confidence: 0.950317

00:00:22.680 --> 00:00:27.144 Hello. Great. Okay, great.

NOTE Confidence: 0.950317

00:00:27.144 --> 00:00:29.783 Thank you again for joining us today.

NOTE Confidence: 0.950317

00:00:29.790 --> 00:00:31.944 The Pathology Grand Rand Student Committee

NOTE Confidence: 0.950317

00:00:31.944 --> 00:00:34.469 is really excited to host Doctor Jones.

NOTE Confidence: 0.950317

00:00:34.470 --> 00:00:36.518 Doctor Jones received her

NOTE Confidence: 0.950317

00:00:36.518 --> 00:00:38.566 Bachelor's at Princeton University  
NOTE Confidence: 0.950317

00:00:38.566 --> 00:00:40.562 and her MD&PHD at Stanford.  
NOTE Confidence: 0.950317

00:00:40.562 --> 00:00:43.234 She also spends a few years at Harvard  
NOTE Confidence: 0.950317

00:00:43.234 --> 00:00:46.202 and is currently at the National Center,  
NOTE Confidence: 0.950317

00:00:46.202 --> 00:00:50.074 the NCI and the CCR at NIH.  
NOTE Confidence: 0.950317

00:00:50.074 --> 00:00:53.510 And she is also the head of the  
NOTE Confidence: 0.950317

00:00:53.510 --> 00:00:56.849 Translational Nanobiology section.  
NOTE Confidence: 0.950317

00:00:56.850 --> 00:00:59.135 Currently, her lab does wonderful  
NOTE Confidence: 0.950317

00:00:59.135 --> 00:01:01.415 work on extracellular vesicles and  
NOTE Confidence: 0.950317

00:01:01.415 --> 00:01:03.195 identifying the different types  
NOTE Confidence: 0.950317

00:01:03.195 --> 00:01:05.420 of vesicles secreted by distinct  
NOTE Confidence: 0.950317

00:01:05.490 --> 00:01:08.202 tumor types and analyzing how they  
NOTE Confidence: 0.950317

00:01:08.202 --> 00:01:10.010 affect downstream immune pathways.  
NOTE Confidence: 0.950317

00:01:10.010 --> 00:01:12.056 She is also working on the  
NOTE Confidence: 0.950317

00:01:12.056 --> 00:01:13.420 development of the characterization  
NOTE Confidence: 0.950317

00:01:13.483 --> 00:01:15.133 and analysis of these extracellular

NOTE Confidence: 0.950317

00:01:15.133 --> 00:01:17.475 vesicles and we are very excited to

NOTE Confidence: 0.950317

00:01:17.475 --> 00:01:19.050 hear more about her research.

NOTE Confidence: 0.950317

00:01:19.050 --> 00:01:21.570 So please join me in welcoming Doctor Jones.

NOTE Confidence: 0.914721334

00:01:26.430 --> 00:01:28.950 So thank you very much.

NOTE Confidence: 0.914721334

00:01:28.950 --> 00:01:31.636 In one of the chats that I

NOTE Confidence: 0.914721334

00:01:31.636 --> 00:01:33.088 had with one of your faculty

NOTE Confidence: 0.914721334

00:01:33.088 --> 00:01:34.430 members earlier this morning,

NOTE Confidence: 0.914721334

00:01:34.430 --> 00:01:35.974 the comment was made.

NOTE Confidence: 0.914721334

00:01:35.974 --> 00:01:38.909 This all sounds kind of like a mess,

NOTE Confidence: 0.914721334

00:01:38.910 --> 00:01:40.214 and actually it is.

NOTE Confidence: 0.914721334

00:01:40.214 --> 00:01:42.871 And what I'm going to show you is

NOTE Confidence: 0.914721334

00:01:42.871 --> 00:01:45.055 what we're going to try to make

NOTE Confidence: 0.914721334

00:01:45.055 --> 00:01:47.632 sense of the mess that is all of

NOTE Confidence: 0.914721334

00:01:47.632 --> 00:01:49.316 the extracellular structures and

NOTE Confidence: 0.914721334

00:01:49.316 --> 00:01:51.869 complexes and what they can tell us.

NOTE Confidence: 0.914721334

00:01:51.870 --> 00:01:56.870 So there is a book called The Commotion

NOTE Confidence: 0.914721334

00:01:56.870 --> 00:02:02.106 in the Blood that is a bit of a back

NOTE Confidence: 0.914721334

00:02:02.106 --> 00:02:05.069 story of of how I got started with this.

NOTE Confidence: 0.914721334

00:02:05.070 --> 00:02:06.990 First, these are the the folks

NOTE Confidence: 0.914721334

00:02:06.990 --> 00:02:09.046 who have done the the legwork,

NOTE Confidence: 0.914721334

00:02:09.046 --> 00:02:10.108 the hard work,

NOTE Confidence: 0.914721334

00:02:10.110 --> 00:02:11.735 the experiments behind these things

NOTE Confidence: 0.914721334

00:02:11.735 --> 00:02:13.971 that I'm going to show you and.

NOTE Confidence: 0.914721334

00:02:13.971 --> 00:02:16.257 So they really deserve all the

NOTE Confidence: 0.914721334

00:02:16.257 --> 00:02:18.859 credit for for getting this done.

NOTE Confidence: 0.914721334

00:02:18.860 --> 00:02:20.528 And I hope I'll show you

NOTE Confidence: 0.914721334

00:02:20.528 --> 00:02:21.860 collectively where this ends up.

NOTE Confidence: 0.941371687777778

00:02:24.100 --> 00:02:25.876 I'm a radiation oncologist,

NOTE Confidence: 0.941371687777778

00:02:25.876 --> 00:02:29.060 so I just wanted to loop you in

NOTE Confidence: 0.941371687777778

00:02:29.060 --> 00:02:30.980 on my perspective and interest on

NOTE Confidence: 0.91493164

00:02:33.500 --> 00:02:34.564 these problems.

NOTE Confidence: 0.91493164

00:02:34.564 --> 00:02:37.756 There's a a phenomenon called abscopal

NOTE Confidence: 0.91493164

00:02:37.756 --> 00:02:40.500 immune responses where you radiate one

NOTE Confidence: 0.91493164

00:02:40.500 --> 00:02:44.100 side of a tumor and the immune system.

NOTE Confidence: 0.91493164

00:02:44.100 --> 00:02:46.500 Causes other sites to regress.

NOTE Confidence: 0.91493164

00:02:46.500 --> 00:02:49.804 We have had some clinical trials at at

NOTE Confidence: 0.91493164

00:02:49.804 --> 00:02:52.900 NIH looking at this and as you know with

NOTE Confidence: 0.91493164

00:02:52.900 --> 00:02:54.500 immunotherapy it doesn't always work.

NOTE Confidence: 0.91493164

00:02:54.500 --> 00:02:55.958 In fact, it often doesn't work

NOTE Confidence: 0.91493164

00:02:55.958 --> 00:02:57.857 as well as you might want it to

NOTE Confidence: 0.91493164

00:02:57.860 --> 00:02:59.580 and the abscopal will response.

NOTE Confidence: 0.91493164

00:02:59.580 --> 00:03:01.340 The combination of radiation

NOTE Confidence: 0.91493164

00:03:01.340 --> 00:03:02.832 and immunotherapy works even

NOTE Confidence: 0.91493164

00:03:02.832 --> 00:03:04.056 less often than that.

NOTE Confidence: 0.938576366666667

00:03:06.180 --> 00:03:08.728 So I want to find tools that

NOTE Confidence: 0.938576366666667

00:03:08.728 --> 00:03:11.179 help me unpack and understand.

NOTE Confidence: 0.938576366666667

00:03:11.180 --> 00:03:12.688 What's happening when those  
NOTE Confidence: 0.938576366666667

00:03:12.688 --> 00:03:14.573 immune responses are going right,  
NOTE Confidence: 0.938576366666667

00:03:14.580 --> 00:03:16.155 what we need to do to drive  
NOTE Confidence: 0.938576366666667

00:03:16.155 --> 00:03:17.820 them in the right direction.  
NOTE Confidence: 0.938576366666667

00:03:17.820 --> 00:03:19.374 This is the structure of the talk.  
NOTE Confidence: 0.938576366666667

00:03:19.380 --> 00:03:21.300 I'm going to talk about not just the  
NOTE Confidence: 0.938576366666667

00:03:21.300 --> 00:03:22.500 motivations that they just went through,  
NOTE Confidence: 0.938576366666667

00:03:22.500 --> 00:03:24.810 but also the basics about extracellular  
NOTE Confidence: 0.938576366666667

00:03:24.810 --> 00:03:26.780 vesicles and other extracellular things.  
NOTE Confidence: 0.938576366666667

00:03:26.780 --> 00:03:28.092 The technology development we've  
NOTE Confidence: 0.938576366666667

00:03:28.092 --> 00:03:30.060 been doing to crack the nut.  
NOTE Confidence: 0.938576366666667

00:03:30.060 --> 00:03:31.560 Some basics discoveries that we've  
NOTE Confidence: 0.938576366666667

00:03:31.560 --> 00:03:33.407 made as we're beginning to actually  
NOTE Confidence: 0.938576366666667

00:03:33.407 --> 00:03:35.177 start to leverage these tools now  
NOTE Confidence: 0.938576366666667

00:03:35.180 --> 00:03:36.650 and some conclusions that really  
NOTE Confidence: 0.938576366666667

00:03:36.650 --> 00:03:38.660 are not really what we expected,

NOTE Confidence: 0.938576366666667  
00:03:38.660 --> 00:03:39.756 but where we're going.  
NOTE Confidence: 0.938576366666667  
00:03:39.756 --> 00:03:42.419 So for those of you who haven't seen it,  
NOTE Confidence: 0.938576366666667  
00:03:42.420 --> 00:03:44.004 you may or may not have a book  
NOTE Confidence: 0.938576366666667  
00:03:44.004 --> 00:03:46.041 or two in your past that sort of  
NOTE Confidence: 0.938576366666667  
00:03:46.041 --> 00:03:47.686 stoked your interest in the field  
NOTE Confidence: 0.938576366666667  
00:03:47.686 --> 00:03:49.294 that you went into in science.  
NOTE Confidence: 0.938576366666667  
00:03:49.300 --> 00:03:52.340 This was a book in the 90s about  
NOTE Confidence: 0.938576366666667  
00:03:52.340 --> 00:03:54.608 tumor immunology in the early days and  
NOTE Confidence: 0.938576366666667  
00:03:54.608 --> 00:03:56.817 Cole's Toxin and all of these things.  
NOTE Confidence: 0.938576366666667  
00:03:56.820 --> 00:03:59.804 And that really led me to want to  
NOTE Confidence: 0.938576366666667  
00:03:59.804 --> 00:04:01.416 follow a path of studying these things.  
NOTE Confidence: 0.938576366666667  
00:04:01.420 --> 00:04:03.660 Overall,  
NOTE Confidence: 0.938576366666667  
00:04:03.660 --> 00:04:05.585 it's all about cells in the immune  
NOTE Confidence: 0.938576366666667  
00:04:05.585 --> 00:04:07.845 system of the cells in the immune system  
NOTE Confidence: 0.938576366666667  
00:04:07.845 --> 00:04:10.138 and the way that that book frames it.  
NOTE Confidence: 0.938576366666667

00:04:10.140 --> 00:04:11.736 So there's more in our blood  
NOTE Confidence: 0.938576366666667

00:04:11.740 --> 00:04:13.340 than just the cells.  
NOTE Confidence: 0.938576366666667

00:04:13.340 --> 00:04:15.740 There is an assortment of vesicles,  
NOTE Confidence: 0.938576366666667

00:04:15.740 --> 00:04:17.400 things with lipid bilayers and  
NOTE Confidence: 0.938576366666667

00:04:17.400 --> 00:04:19.060 various cargo released in different  
NOTE Confidence: 0.938576366666667

00:04:19.112 --> 00:04:20.420 ways from different cells.  
NOTE Confidence: 0.938576366666667

00:04:20.420 --> 00:04:22.400 They're also obviously lipoproteins.  
NOTE Confidence: 0.938576366666667

00:04:22.400 --> 00:04:25.025 They're also ribonuclear proteins complexes.  
NOTE Confidence: 0.938576366666667

00:04:25.025 --> 00:04:28.250 They're also classes of extracellular  
NOTE Confidence: 0.938576366666667

00:04:28.250 --> 00:04:30.830 things called exomeres and  
NOTE Confidence: 0.938576366666667

00:04:30.917 --> 00:04:33.461 supermeres isolated in different  
NOTE Confidence: 0.938576366666667

00:04:33.461 --> 00:04:37.300 ways from these broadly speaking,  
NOTE Confidence: 0.938576366666667

00:04:37.300 --> 00:04:39.200 I'm going to focus on.  
NOTE Confidence: 0.938576366666667

00:04:39.200 --> 00:04:40.082 The vesicles,  
NOTE Confidence: 0.938576366666667

00:04:40.082 --> 00:04:44.171 with a caveat that I can't guarantee to you  
NOTE Confidence: 0.938576366666667

00:04:44.171 --> 00:04:47.517 with the technology that anybody is using,



NOTE Confidence: 0.938576366666667

00:04:47.520 --> 00:04:49.746 that every single vesicle is actually

NOTE Confidence: 0.938576366666667

00:04:49.746 --> 00:04:52.344 vesicle and not just a particle that

NOTE Confidence: 0.938576366666667

00:04:52.344 --> 00:04:54.432 happens to have the same density,

NOTE Confidence: 0.938576366666667

00:04:54.440 --> 00:04:57.480 size, or other properties, but.

NOTE Confidence: 0.938576366666667

00:04:57.480 --> 00:04:59.880 This is this is where we are with the field.

NOTE Confidence: 0.938576366666667

00:04:59.880 --> 00:05:02.971 So Ev's are heterogeneous exosomes.

NOTE Confidence: 0.938576366666667

00:05:02.971 --> 00:05:06.696 Perche tends to be less than 150 nanometers.

NOTE Confidence: 0.938576366666667

00:05:06.696 --> 00:05:09.236 They have protein surface markers.

NOTE Confidence: 0.938576366666667

00:05:09.240 --> 00:05:11.400 They have nucleic acid cargo

NOTE Confidence: 0.938576366666667

00:05:11.400 --> 00:05:13.280 inside and sometimes outside,

NOTE Confidence: 0.938576366666667

00:05:13.280 --> 00:05:15.160 stuck to the surface,

NOTE Confidence: 0.938576366666667

00:05:15.160 --> 00:05:17.434 and there's widespread interest in them

NOTE Confidence: 0.938576366666667

00:05:17.434 --> 00:05:19.950 from tumors and other types of cells.

NOTE Confidence: 0.938576366666667

00:05:19.950 --> 00:05:22.236 People are interested in them because

NOTE Confidence: 0.938576366666667

00:05:22.236 --> 00:05:24.510 everybody who's interested in some tissue,

NOTE Confidence: 0.938576366666667

00:05:24.510 --> 00:05:25.198 some disease,  
NOTE Confidence: 0.938576366666667

00:05:25.198 --> 00:05:27.606 some something wants to look at the  
NOTE Confidence: 0.938576366666667

00:05:27.606 --> 00:05:29.348 vesicles coming from that tissue,  
NOTE Confidence: 0.938576366666667

00:05:29.348 --> 00:05:30.026 that disease.  
NOTE Confidence: 0.938576366666667

00:05:30.026 --> 00:05:32.622 And so it's really a great framework  
NOTE Confidence: 0.938576366666667

00:05:32.622 --> 00:05:34.550 for potentially doing systematic  
NOTE Confidence: 0.938576366666667

00:05:34.550 --> 00:05:37.190 systems biology if we could be  
NOTE Confidence: 0.938576366666667

00:05:37.190 --> 00:05:39.309 organized and structured about this.  
NOTE Confidence: 0.945742663636364

00:05:41.590 --> 00:05:43.767 So I want you to remember this  
NOTE Confidence: 0.945742663636364

00:05:43.767 --> 00:05:45.390 figure because it's important.  
NOTE Confidence: 0.945742663636364

00:05:45.390 --> 00:05:47.642 Strictly speaking exosomes come  
NOTE Confidence: 0.945742663636364

00:05:47.642 --> 00:05:49.894 from multi vesicular bodies.  
NOTE Confidence: 0.945742663636364

00:05:49.900 --> 00:05:53.320 In the cell and released into  
NOTE Confidence: 0.945742663636364

00:05:53.320 --> 00:05:55.060 the extracellular space,  
NOTE Confidence: 0.945742663636364

00:05:55.060 --> 00:05:56.698 other vessels are set from the surface.  
NOTE Confidence: 0.945742663636364

00:05:56.700 --> 00:05:58.325 Those might be called micro

NOTE Confidence: 0.945742663636364

00:05:58.325 --> 00:05:59.300 vessels or microparticles.

NOTE Confidence: 0.945742663636364

00:05:59.300 --> 00:06:01.856 But exosomes imply a certain Biogenesis

NOTE Confidence: 0.95232978

00:06:04.700 --> 00:06:06.580 in the liquid biopsy community.

NOTE Confidence: 0.95232978

00:06:06.580 --> 00:06:07.900 So most of my clinical colleagues,

NOTE Confidence: 0.95232978

00:06:07.900 --> 00:06:10.396 when they do exosome studies or

NOTE Confidence: 0.95232978

00:06:10.396 --> 00:06:12.060 liquid biopsies for biomarkers,

NOTE Confidence: 0.95232978

00:06:12.060 --> 00:06:13.532 they'll take a biofluid,

NOTE Confidence: 0.95232978

00:06:13.532 --> 00:06:15.740 they'll isolate the exosomes often with

NOTE Confidence: 0.95232978

00:06:15.800 --> 00:06:18.186 some kit and then they'll do some other.

NOTE Confidence: 0.95232978

00:06:18.190 --> 00:06:20.230 Cargo assay RNA or DNA,

NOTE Confidence: 0.95232978

00:06:20.230 --> 00:06:22.426 some sequencing to identify some biomarkers.

NOTE Confidence: 0.948487223636364

00:06:25.390 --> 00:06:28.264 The reason there's so much huge

NOTE Confidence: 0.948487223636364

00:06:28.264 --> 00:06:30.870 excitement about this is because

NOTE Confidence: 0.948487223636364

00:06:30.870 --> 00:06:34.790 robust consistency in the protocols

NOTE Confidence: 0.948487223636364

00:06:34.790 --> 00:06:39.886 and a useful payload in the readouts

NOTE Confidence: 0.948487223636364

00:06:39.886 --> 00:06:42.426 has had some big successes.  
NOTE Confidence: 0.948487223636364

00:06:42.430 --> 00:06:45.094 So Exosome diagnostics has  
NOTE Confidence: 0.948487223636364

00:06:45.094 --> 00:06:47.194 prostate cancer assays.  
NOTE Confidence: 0.948487223636364

00:06:47.194 --> 00:06:52.844 Which can help predict the the aggressiveness  
NOTE Confidence: 0.948487223636364

00:06:52.844 --> 00:06:55.229 and discriminate between high grade  
NOTE Confidence: 0.948487223636364

00:06:55.229 --> 00:06:57.907 and low grade prostate cancer and  
NOTE Confidence: 0.948487223636364

00:06:57.907 --> 00:07:01.010 indicate to a patient based on their PS:.  
NOTE Confidence: 0.948487223636364

00:07:01.010 --> 00:07:03.523 A and this test whether or not  
NOTE Confidence: 0.948487223636364

00:07:03.523 --> 00:07:05.770 they need to get a biopsy.  
NOTE Confidence: 0.948487223636364

00:07:05.770 --> 00:07:08.994 This was the result of one of the  
NOTE Confidence: 0.948487223636364

00:07:08.994 --> 00:07:11.127 randomized control trials where they  
NOTE Confidence: 0.948487223636364

00:07:11.127 --> 00:07:14.610 looked at this and showed the benefit.  
NOTE Confidence: 0.948487223636364

00:07:14.610 --> 00:07:18.809 Of combining this test with standard of care.  
NOTE Confidence: 0.948487223636364

00:07:18.810 --> 00:07:21.150 There have been since 3 randomized  
NOTE Confidence: 0.948487223636364

00:07:21.150 --> 00:07:23.415 control trials and so I'm not saying  
NOTE Confidence: 0.948487223636364

00:07:23.415 --> 00:07:25.890 that this kind of assay is not useful.

NOTE Confidence: 0.948487223636364

00:07:25.890 --> 00:07:28.578 But if you talk to Johan Skog who

NOTE Confidence: 0.948487223636364

00:07:28.578 --> 00:07:30.280 developed these assays you'll be

NOTE Confidence: 0.948487223636364

00:07:30.280 --> 00:07:33.100 the first to tell you yes 3 Two of

NOTE Confidence: 0.948487223636364

00:07:33.100 --> 00:07:35.440 the three Rna's that they isolated

NOTE Confidence: 0.948487223636364

00:07:35.440 --> 00:07:38.170 in the first versions of this test

NOTE Confidence: 0.948487223636364

00:07:38.170 --> 00:07:40.606 were not actually in vesicles at all.

NOTE Confidence: 0.948487223636364

00:07:40.610 --> 00:07:43.976 They were Co isolated with those.

NOTE Confidence: 0.948487223636364

00:07:43.980 --> 00:07:44.460 And so

NOTE Confidence: 0.949725035294118

00:07:47.060 --> 00:07:48.440 remember I showed you the figure

NOTE Confidence: 0.949725035294118

00:07:48.440 --> 00:07:49.985 where it really means something very

NOTE Confidence: 0.949725035294118

00:07:49.985 --> 00:07:51.420 specific in terms of Biogenesis.

NOTE Confidence: 0.949725035294118

00:07:51.420 --> 00:07:54.076 We have another part of the field that's

NOTE Confidence: 0.949725035294118

00:07:54.076 --> 00:07:55.564 really approaching this strictly as

NOTE Confidence: 0.949725035294118

00:07:55.564 --> 00:07:57.380 a I don't know about the Biogenesis,

NOTE Confidence: 0.949725035294118

00:07:57.380 --> 00:07:58.996 I don't really care.

NOTE Confidence: 0.949725035294118

00:07:58.996 --> 00:08:01.944 I'm taking a blood sample and and  
NOTE Confidence: 0.949725035294118

00:08:01.944 --> 00:08:04.254 doing a procedural based thing.  
NOTE Confidence: 0.949725035294118

00:08:04.260 --> 00:08:06.708 So there is an ontology initiative  
NOTE Confidence: 0.949725035294118

00:08:06.708 --> 00:08:09.003 that we've started because if you  
NOTE Confidence: 0.949725035294118

00:08:09.003 --> 00:08:10.767 want to create atlases you have  
NOTE Confidence: 0.949725035294118

00:08:10.767 --> 00:08:13.239 to be speaking the same language.  
NOTE Confidence: 0.949725035294118

00:08:13.240 --> 00:08:14.768 And I'll get to that at the very  
NOTE Confidence: 0.949725035294118

00:08:14.768 --> 00:08:16.253 end because I know ontology talks  
NOTE Confidence: 0.949725035294118

00:08:16.253 --> 00:08:17.313 make everybody fall asleep.  
NOTE Confidence: 0.949725035294118

00:08:17.320 --> 00:08:19.800 So I just got a couple of slides to to  
NOTE Confidence: 0.949725035294118

00:08:19.865 --> 00:08:22.240 share with you and thank those of you  
NOTE Confidence: 0.949725035294118

00:08:22.240 --> 00:08:26.040 who feel about the the survey for us.  
NOTE Confidence: 0.949725035294118

00:08:26.040 --> 00:08:27.480 But back to the original idea,  
NOTE Confidence: 0.949725035294118

00:08:27.480 --> 00:08:30.679 So how do we do this systematically  
NOTE Confidence: 0.949725035294118

00:08:30.679 --> 00:08:31.593 and correctly?  
NOTE Confidence: 0.949725035294118

00:08:31.600 --> 00:08:35.648 What we want to do is to sort

NOTE Confidence: 0.949725035294118

00:08:35.648 --> 00:08:37.465 subsets of the vesicles and then

NOTE Confidence: 0.949725035294118

00:08:37.465 --> 00:08:39.494 look at the the cargo and the

NOTE Confidence: 0.949725035294118

00:08:39.494 --> 00:08:41.154 messages that those sets contain.

NOTE Confidence: 0.941371867777778

00:08:43.470 --> 00:08:45.982 So the state of the field is taking

NOTE Confidence: 0.941371867777778

00:08:45.982 --> 00:08:48.650 the whole mess of vesicles from all

NOTE Confidence: 0.941371867777778

00:08:48.650 --> 00:08:51.226 the different cells and we want to

NOTE Confidence: 0.941371867777778

00:08:51.226 --> 00:08:53.320 parse them out into vesicles from

NOTE Confidence: 0.941371867777778

00:08:53.399 --> 00:08:57.790 particular sources and and study those.

NOTE Confidence: 0.941371867777778

00:08:57.790 --> 00:09:01.489 So we need to know how do we reliably

NOTE Confidence: 0.941371867777778

00:09:01.489 --> 00:09:03.870 identify the specific subsets.

NOTE Confidence: 0.941371867777778

00:09:03.870 --> 00:09:06.030 So with immune cell substate sets

NOTE Confidence: 0.941371867777778

00:09:06.030 --> 00:09:08.470 you might use CD3 for a T cell.

NOTE Confidence: 0.943608066666667

00:09:10.500 --> 00:09:13.818 CD14 for a type of monocyte,

NOTE Confidence: 0.943608066666667

00:09:13.820 --> 00:09:16.332 which are the right markers to use for

NOTE Confidence: 0.943608066666667

00:09:16.332 --> 00:09:18.210 different types of vesicles and then

NOTE Confidence: 0.943608066666667

00:09:18.210 --> 00:09:20.135 how to reliably assay them Once you  
NOTE Confidence: 0.943608066666667

00:09:20.135 --> 00:09:22.096 know which ones you want to assay,  
NOTE Confidence: 0.943608066666667

00:09:22.100 --> 00:09:25.220 how do you do the assay in a reliable way.  
NOTE Confidence: 0.943608066666667

00:09:25.220 --> 00:09:27.775 So we're in an in between space.  
NOTE Confidence: 0.943608066666667

00:09:27.780 --> 00:09:29.558 There's a lot of cellular biology and  
NOTE Confidence: 0.943608066666667

00:09:29.558 --> 00:09:31.616 this is pretty mature at this point and  
NOTE Confidence: 0.943608066666667

00:09:31.616 --> 00:09:33.500 there's a lot of molecular diagnostics.  
NOTE Confidence: 0.943608066666667

00:09:33.500 --> 00:09:37.178 It's individual molecule assessed in mass,  
NOTE Confidence: 0.943608066666667

00:09:37.180 --> 00:09:39.760 these are packets and so.  
NOTE Confidence: 0.943608066666667

00:09:39.760 --> 00:09:41.248 We're looking at packets of informations  
NOTE Confidence: 0.943608066666667

00:09:41.248 --> 00:09:43.200 and sets of packets of those informations.  
NOTE Confidence: 0.943608066666667

00:09:43.200 --> 00:09:45.900 So it's a fundamentally different  
NOTE Confidence: 0.943608066666667

00:09:45.900 --> 00:09:47.520 type of bioinformatics.  
NOTE Confidence: 0.943608066666667

00:09:47.520 --> 00:09:49.260 And Joshua Welch,  
NOTE Confidence: 0.943608066666667

00:09:49.260 --> 00:09:52.160 in my group Staff Scientist,  
NOTE Confidence: 0.943608066666667

00:09:52.160 --> 00:09:55.704 has led the development of three software



NOTE Confidence: 0.943608066666667

00:09:55.704 --> 00:09:58.912 tools which help improve the rigor and

NOTE Confidence: 0.943608066666667

00:09:58.912 --> 00:10:00.464 reproducibility of three important

NOTE Confidence: 0.943608066666667

00:10:00.464 --> 00:10:02.807 tools that we use for characterizing.

NOTE Confidence: 0.943608066666667

00:10:02.810 --> 00:10:05.820 Individual E v's and repertoires of E

NOTE Confidence: 0.943608066666667

00:10:05.820 --> 00:10:08.084 V's and I'm going to walk through these.

NOTE Confidence: 0.943608066666667

00:10:08.090 --> 00:10:10.700 One that's targeted at EV flow

NOTE Confidence: 0.943608066666667

00:10:10.700 --> 00:10:12.850 cytometry for single E V's,

NOTE Confidence: 0.943608066666667

00:10:12.850 --> 00:10:14.490 resisted pulse sensing that

NOTE Confidence: 0.943608066666667

00:10:14.490 --> 00:10:16.130 measures size and concentration,

NOTE Confidence: 0.943608066666667

00:10:16.130 --> 00:10:20.050 and MPA pass and Multiplex analysis system,

NOTE Confidence: 0.943608066666667

00:10:20.050 --> 00:10:22.275 which is useful for assessing

NOTE Confidence: 0.943608066666667

00:10:22.275 --> 00:10:23.165 repertoires broadly.

NOTE Confidence: 0.9201268

00:10:26.490 --> 00:10:30.286 So these are the specific

NOTE Confidence: 0.9201268

00:10:30.286 --> 00:10:33.640 tonologies I'm going to talk about.

NOTE Confidence: 0.9201268

00:10:33.640 --> 00:10:39.650 So for single EV studies we've produced

NOTE Confidence: 0.9201268

00:10:39.650 --> 00:10:43.475 these advanced protocols for labeling,  
NOTE Confidence: 0.9201268

00:10:43.480 --> 00:10:45.568 sorting, a framework which I'll show  
NOTE Confidence: 0.9201268

00:10:45.568 --> 00:10:48.881 you or how to do and organize and  
NOTE Confidence: 0.9201268

00:10:48.881 --> 00:10:51.784 report the the studies and then also  
NOTE Confidence: 0.9201268

00:10:51.784 --> 00:10:53.800 do your assays in a calibrated way.  
NOTE Confidence: 0.9201268

00:10:53.800 --> 00:10:55.240 So rather than an arbitrary  
NOTE Confidence: 0.9201268

00:10:55.240 --> 00:10:56.392 unit sharing calibrated units,  
NOTE Confidence: 0.9201268

00:10:56.400 --> 00:10:58.655 which if in everybody probably  
NOTE Confidence: 0.9201268

00:10:58.655 --> 00:10:59.557 does philositometry,  
NOTE Confidence: 0.9201268

00:10:59.560 --> 00:11:01.456 you know your skills are actually.  
NOTE Confidence: 0.9201268

00:11:01.460 --> 00:11:02.591 Arbitrary, they're not  
NOTE Confidence: 0.9201268

00:11:02.591 --> 00:11:04.099 calibrated for essence units.  
NOTE Confidence: 0.946765108235294

00:11:06.940 --> 00:11:08.708 And then I'm going to show you how  
NOTE Confidence: 0.946765108235294

00:11:08.708 --> 00:11:09.865 we're stepping towards integrating  
NOTE Confidence: 0.946765108235294

00:11:09.865 --> 00:11:11.540 this into a comprehensive Atlas.  
NOTE Confidence: 0.946765108235294

00:11:11.540 --> 00:11:15.930 So our labeling protocols and

NOTE Confidence: 0.946765108235294  
00:11:15.930 --> 00:11:19.860 sorting we're done on the Astrios,  
NOTE Confidence: 0.946765108235294  
00:11:19.860 --> 00:11:23.100 basically a next generation Moflow XDP  
NOTE Confidence: 0.946765108235294  
00:11:23.100 --> 00:11:28.760 den air system with this we showed here.  
NOTE Confidence: 0.946765108235294  
00:11:28.760 --> 00:11:29.444 Remember this picture?  
NOTE Confidence: 0.946765108235294  
00:11:29.444 --> 00:11:31.040 I'm going to come back to it.  
NOTE Confidence: 0.946765108235294  
00:11:31.040 --> 00:11:32.615 These are vesicles that we  
NOTE Confidence: 0.946765108235294  
00:11:32.615 --> 00:11:34.590 can see from DC 2.4 cells.  
NOTE Confidence: 0.946765108235294  
00:11:34.590 --> 00:11:36.720 And this bottom thing is not  
NOTE Confidence: 0.946765108235294  
00:11:36.720 --> 00:11:38.320 a population of vesicles,  
NOTE Confidence: 0.946765108235294  
00:11:38.320 --> 00:11:40.678 it's the noise of the instrument.  
NOTE Confidence: 0.946765108235294  
00:11:40.680 --> 00:11:42.759 So you can see how we're really  
NOTE Confidence: 0.946765108235294  
00:11:42.760 --> 00:11:46.078 hugging the bottom limits of detection.  
NOTE Confidence: 0.946765108235294  
00:11:46.080 --> 00:11:48.120 We can't separate the vesicles  
NOTE Confidence: 0.946765108235294  
00:11:48.120 --> 00:11:50.480 from the noise any better because  
NOTE Confidence: 0.946765108235294  
00:11:50.480 --> 00:11:52.462 of the limits of detection and.  
NOTE Confidence: 0.946765108235294

00:11:52.462 --> 00:11:54.754 Polystyrene beads can't be used as  
NOTE Confidence: 0.946765108235294

00:11:54.754 --> 00:11:56.440 calibrators because they refract.  
NOTE Confidence: 0.946765108235294

00:11:56.440 --> 00:11:58.120 They have a different refractive index,  
NOTE Confidence: 0.946765108235294

00:11:58.120 --> 00:12:01.445 so they they fundamentally can't  
NOTE Confidence: 0.946765108235294

00:12:01.445 --> 00:12:03.440 be actual calibrators.  
NOTE Confidence: 0.946765108235294

00:12:03.440 --> 00:12:05.855 This is what happens when your particles  
NOTE Confidence: 0.946765108235294

00:12:05.855 --> 00:12:08.560 are lower than the limited fraction.  
NOTE Confidence: 0.946765108235294

00:12:08.560 --> 00:12:08.987 Fortunately,  
NOTE Confidence: 0.946765108235294

00:12:08.987 --> 00:12:11.549 HIV and most of our extracellular  
NOTE Confidence: 0.946765108235294

00:12:11.549 --> 00:12:13.840 vesicles is about the same size,  
NOTE Confidence: 0.946765108235294

00:12:13.840 --> 00:12:15.884 so we took advantage of that and  
NOTE Confidence: 0.946765108235294

00:12:15.884 --> 00:12:20.220 took two different HIV variants,  
NOTE Confidence: 0.946765108235294

00:12:20.220 --> 00:12:23.280 one that was.  
NOTE Confidence: 0.946765108235294

00:12:23.280 --> 00:12:25.622 CCR5 trophic and one that's CX  
NOTE Confidence: 0.946765108235294

00:12:25.622 --> 00:12:28.394 CR4 trophic and labeled 1 red,  
NOTE Confidence: 0.946765108235294

00:12:28.400 --> 00:12:29.306 labeled 1 green,

NOTE Confidence: 0.946765108235294  
00:12:29.306 --> 00:12:31.118 which showed that we could sort  
NOTE Confidence: 0.946765108235294  
00:12:31.118 --> 00:12:33.193 them and that they remain retain  
NOTE Confidence: 0.946765108235294  
00:12:33.193 --> 00:12:35.080 their trophism for their specific  
NOTE Confidence: 0.948080688888889  
00:12:37.680 --> 00:12:39.880 for their specific type and  
NOTE Confidence: 0.948080688888889  
00:12:39.880 --> 00:12:41.640 the recipient cell line.  
NOTE Confidence: 0.948080688888889  
00:12:41.640 --> 00:12:43.720 So this shows fidelity.  
NOTE Confidence: 0.948080688888889  
00:12:43.720 --> 00:12:45.632 It's not something you're ever  
NOTE Confidence: 0.948080688888889  
00:12:45.632 --> 00:12:47.796 going to do if you're producing.  
NOTE Confidence: 0.948080688888889  
00:12:47.796 --> 00:12:48.912 Therapeutic vessels and  
NOTE Confidence: 0.948080688888889  
00:12:48.912 --> 00:12:50.809 you want to produce a lot.  
NOTE Confidence: 0.948080688888889  
00:12:50.810 --> 00:12:52.250 It's feasible to do this  
NOTE Confidence: 0.948080688888889  
00:12:52.250 --> 00:12:53.402 if you're studying viruses,  
NOTE Confidence: 0.948080688888889  
00:12:53.410 --> 00:12:55.170 but I think our group,  
NOTE Confidence: 0.948080688888889  
00:12:55.170 --> 00:12:57.060 Vanderbilt's group and a group and  
NOTE Confidence: 0.948080688888889  
00:12:57.060 --> 00:12:58.985 the Netherlands may be the only  
NOTE Confidence: 0.948080688888889

00:12:58.985 --> 00:13:00.809 groups who've ever really done this  
NOTE Confidence: 0.948080688888889

00:13:00.810 --> 00:13:03.806 and it takes like 48 hours nonstop.  
NOTE Confidence: 0.948080688888889

00:13:03.810 --> 00:13:08.070 We set up COTS in the lab and all of that.  
NOTE Confidence: 0.948080688888889

00:13:08.070 --> 00:13:09.650 So it's it's not a  
NOTE Confidence: 0.9805073

00:13:11.850 --> 00:13:13.650 scalable approach.  
NOTE Confidence: 0.931374129411765

00:13:16.600 --> 00:13:19.032 So when we did this we we had  
NOTE Confidence: 0.931374129411765

00:13:19.032 --> 00:13:21.548 so many people look at our data  
NOTE Confidence: 0.931374129411765

00:13:21.548 --> 00:13:23.040 and say how did you do that?  
NOTE Confidence: 0.931374129411765

00:13:23.040 --> 00:13:26.920 I just, I just don't believe those results.  
NOTE Confidence: 0.931374129411765

00:13:26.920 --> 00:13:29.202 And so we we took that challenge  
NOTE Confidence: 0.931374129411765

00:13:29.202 --> 00:13:31.039 on and we said okay,  
NOTE Confidence: 0.931374129411765

00:13:31.040 --> 00:13:33.908 we're going to prove it and we  
NOTE Confidence: 0.931374129411765

00:13:33.908 --> 00:13:36.494 need to help each other be able  
NOTE Confidence: 0.931374129411765

00:13:36.494 --> 00:13:37.736 to look at each other's data  
NOTE Confidence: 0.931374129411765

00:13:37.736 --> 00:13:39.119 and know what we can believe,  
NOTE Confidence: 0.931374129411765

00:13:39.120 --> 00:13:40.912 what we what we can trust in terms

NOTE Confidence: 0.931374129411765

00:13:40.912 --> 00:13:42.627 of the integrity of the data.

NOTE Confidence: 0.931374129411765

00:13:42.630 --> 00:13:45.066 So this led to this formation of

NOTE Confidence: 0.931374129411765

00:13:45.066 --> 00:13:47.874 a Tri society ISAF ISAC ISTH flow

NOTE Confidence: 0.931374129411765

00:13:47.874 --> 00:13:50.520 cytometry group where we worked on

NOTE Confidence: 0.931374129411765

00:13:50.602 --> 00:13:53.206 how can we improve the rigor in

NOTE Confidence: 0.931374129411765

00:13:53.206 --> 00:13:55.408 the field so that we can speak the

NOTE Confidence: 0.931374129411765

00:13:55.408 --> 00:13:57.190 same language for EV flow cytometry

NOTE Confidence: 0.931374129411765

00:13:57.190 --> 00:14:00.910 and improve our data approach.

NOTE Confidence: 0.931374129411765

00:14:00.910 --> 00:14:05.534 So this was the product of a

NOTE Confidence: 0.931374129411765

00:14:05.534 --> 00:14:08.378 surprisingly long time working with

NOTE Confidence: 0.931374129411765

00:14:08.378 --> 00:14:11.348 groups who do things differently.

NOTE Confidence: 0.931374129411765

00:14:11.350 --> 00:14:14.890 And we basically set out these

NOTE Confidence: 0.931374129411765

00:14:14.890 --> 00:14:20.108 guidelines to help basically tell you

NOTE Confidence: 0.931374129411765

00:14:20.110 --> 00:14:21.510 when you're designing your experiment,

NOTE Confidence: 0.931374129411765

00:14:21.510 --> 00:14:22.354 you're setting things up.

NOTE Confidence: 0.931374129411765

00:14:22.354 --> 00:14:23.921 What do you need to do to  
NOTE Confidence: 0.931374129411765

00:14:23.921 --> 00:14:24.909 help people reproduce it?  
NOTE Confidence: 0.931374129411765

00:14:24.910 --> 00:14:26.506 How do you prove that what  
NOTE Confidence: 0.931374129411765

00:14:26.506 --> 00:14:27.910 you're looking at is Ev's?  
NOTE Confidence: 0.931374129411765

00:14:27.910 --> 00:14:29.482 How do you validate it across  
NOTE Confidence: 0.931374129411765

00:14:29.482 --> 00:14:30.268 instruments and settings?  
NOTE Confidence: 0.931374129411765

00:14:30.270 --> 00:14:34.142 And how do you make your data shareable,  
NOTE Confidence: 0.931374129411765

00:14:34.142 --> 00:14:36.830 transparent, and ideally interoperable?  
NOTE Confidence: 0.93824092

00:14:39.510 --> 00:14:42.390 So this is where Josh's coding  
NOTE Confidence: 0.93824092

00:14:42.390 --> 00:14:43.670 and technology development skills  
NOTE Confidence: 0.93824092

00:14:43.670 --> 00:14:45.270 have really come into play.  
NOTE Confidence: 0.93824092

00:14:45.270 --> 00:14:47.712 He tackled both the single EV  
NOTE Confidence: 0.93824092

00:14:47.712 --> 00:14:49.917 analysis problem and the EV  
NOTE Confidence: 0.93824092

00:14:49.917 --> 00:14:52.427 repertoire problem in flow cytometry.  
NOTE Confidence: 0.93824092

00:14:52.430 --> 00:14:55.412 Single EV analysis Low is highly  
NOTE Confidence: 0.93824092

00:14:55.412 --> 00:14:58.470 quantitative, but it has terrible



NOTE Confidence: 0.93824092  
00:14:58.470 --> 00:14:59.815 sensitivity for single EV's.  
NOTE Confidence: 0.93824092  
00:14:59.815 --> 00:15:01.832 If you do it on a bead based  
NOTE Confidence: 0.93824092  
00:15:01.832 --> 00:15:03.387 way like a Multiplex way,  
NOTE Confidence: 0.93824092  
00:15:03.390 --> 00:15:04.350 it's high throughput.  
NOTE Confidence: 0.93824092  
00:15:04.350 --> 00:15:06.235 It's multi, multi, parametric but.  
NOTE Confidence: 0.93824092  
00:15:06.235 --> 00:15:08.310 It's only semi quantitative and  
NOTE Confidence: 0.93824092  
00:15:08.310 --> 00:15:10.801 you can't really assess the full  
NOTE Confidence: 0.93824092  
00:15:10.801 --> 00:15:11.995 range of complexity.  
NOTE Confidence: 0.93824092  
00:15:12.000 --> 00:15:15.650 So for single EV flow cytometry  
NOTE Confidence: 0.93824092  
00:15:15.650 --> 00:15:17.066 he developed FCM passive  
NOTE Confidence: 0.93824092  
00:15:17.066 --> 00:15:18.280 software that basically  
NOTE Confidence: 0.928587020555555  
00:15:20.800 --> 00:15:22.684 derives the collection angle of the  
NOTE Confidence: 0.928587020555555  
00:15:22.684 --> 00:15:24.626 actual optics of the actual machine  
NOTE Confidence: 0.928587020555555  
00:15:24.626 --> 00:15:26.516 at the time that you're doing.  
NOTE Confidence: 0.928587020555555  
00:15:26.520 --> 00:15:28.557 So if the engineer came in and  
NOTE Confidence: 0.928587020555555

00:15:28.560 --> 00:15:29.928 fiddled with the alignment,  
NOTE Confidence: 0.928587020555555

00:15:29.928 --> 00:15:31.638 it would you'd have to,  
NOTE Confidence: 0.928587020555555

00:15:31.640 --> 00:15:34.440 it wouldn't be the same collection angle.  
NOTE Confidence: 0.928587020555555

00:15:34.440 --> 00:15:35.875 But once you've derived the collection angle,  
NOTE Confidence: 0.928587020555555

00:15:35.880 --> 00:15:38.160 if you collect the proper calibrators,  
NOTE Confidence: 0.928587020555555

00:15:38.160 --> 00:15:41.392 then you can convert your data from those  
NOTE Confidence: 0.928587020555555

00:15:41.392 --> 00:15:44.736 flow cytometry arbitrary units using ME  
NOTE Confidence: 0.928587020555555

00:15:44.736 --> 00:15:48.696 theory to calibrated SI standard units  
NOTE Confidence: 0.928587020555555

00:15:48.696 --> 00:15:51.320 of nanometers and for your fluorescence.  
NOTE Confidence: 0.928587020555555

00:15:51.320 --> 00:15:53.360 You can also calibrate with molecular  
NOTE Confidence: 0.928587020555555

00:15:53.418 --> 00:15:55.873 equivalence of soluble fluorescents and  
NOTE Confidence: 0.928587020555555

00:15:55.873 --> 00:15:58.838 generate calibrated fluorescence as well.  
NOTE Confidence: 0.933065435714286

00:16:01.520 --> 00:16:04.355 This led to. A bunch of papers,  
NOTE Confidence: 0.933065435714286

00:16:04.360 --> 00:16:06.280 a bunch of our reports.  
NOTE Confidence: 0.933065435714286

00:16:06.280 --> 00:16:08.909 This is still something that we're trying  
NOTE Confidence: 0.933065435714286

00:16:08.909 --> 00:16:11.381 to get out and use more commonly so

NOTE Confidence: 0.933065435714286  
00:16:11.381 --> 00:16:13.410 that we can more actively engage with  
NOTE Confidence: 0.933065435714286  
00:16:13.410 --> 00:16:16.000 and sort of share data with each other.  
NOTE Confidence: 0.933065435714286  
00:16:16.000 --> 00:16:20.552 For the EV repertoire analysis this involves.  
NOTE Confidence: 0.933065435714286  
00:16:20.552 --> 00:16:22.904 We prototyped a lot of this using  
NOTE Confidence: 0.933065435714286  
00:16:22.904 --> 00:16:25.320 the Miltonie Multiplex Exosome Kit,  
NOTE Confidence: 0.933065435714286  
00:16:25.320 --> 00:16:28.038 which is a bead set of almost 40 beads.  
NOTE Confidence: 0.938934430769231  
00:16:30.060 --> 00:16:32.560 Several micronic piece which capture  
NOTE Confidence: 0.938934430769231  
00:16:32.560 --> 00:16:36.260 based on one antibody type 1 epitope type  
NOTE Confidence: 0.938934430769231  
00:16:36.260 --> 00:16:38.972 captures the vesicles and then you go in  
NOTE Confidence: 0.938934430769231  
00:16:38.972 --> 00:16:42.056 and you detect with a different antibody.  
NOTE Confidence: 0.938934430769231  
00:16:42.060 --> 00:16:45.312 And so with this I'm going to walk you  
NOTE Confidence: 0.938934430769231  
00:16:45.312 --> 00:16:46.939 through some of the results that we see.  
NOTE Confidence: 0.938934430769231  
00:16:46.940 --> 00:16:49.600 But he's written the software to really  
NOTE Confidence: 0.938934430769231  
00:16:49.600 --> 00:16:51.503 facilitate the complexity of this and  
NOTE Confidence: 0.938934430769231  
00:16:51.503 --> 00:16:53.820 all of that data that has to get analyzed  
NOTE Confidence: 0.938934430769231

00:16:53.820 --> 00:16:56.300 all at once in a calibrated way compared

NOTE Confidence: 0.938934430769231

00:16:56.300 --> 00:16:59.740 between experiments etcetera, so.

NOTE Confidence: 0.938934430769231

00:16:59.740 --> 00:17:03.390 This is a heat map showing several

NOTE Confidence: 0.938934430769231

00:17:03.390 --> 00:17:05.564 experiments we did with different

NOTE Confidence: 0.938934430769231

00:17:05.564 --> 00:17:08.060 antibody capture B combinations,

NOTE Confidence: 0.938934430769231

00:17:08.060 --> 00:17:12.458 different biofluid types and it's all

NOTE Confidence: 0.938934430769231

00:17:12.460 --> 00:17:16.564 calibrated again and with fluorescence

NOTE Confidence: 0.938934430769231

00:17:16.564 --> 00:17:19.007 and and the appropriate controls.

NOTE Confidence: 0.938934430769231

00:17:19.007 --> 00:17:22.739 So what you can see is CSF is unique,

NOTE Confidence: 0.938934430769231

00:17:22.740 --> 00:17:24.576 it's it's sort of standing off on its own,

NOTE Confidence: 0.938934430769231

00:17:24.580 --> 00:17:27.807 it's very different from plasm and serum.

NOTE Confidence: 0.938934430769231

00:17:27.810 --> 00:17:29.676 Plasma and Serum are relatively similar

NOTE Confidence: 0.938934430769231

00:17:29.676 --> 00:17:32.070 to each other and in the way that

NOTE Confidence: 0.938934430769231

00:17:32.070 --> 00:17:34.529 PCA and RT Sneeze are parsing them.

NOTE Confidence: 0.938934430769231

00:17:34.530 --> 00:17:37.050 So that's what we've done for that

NOTE Confidence: 0.938934430769231

00:17:37.050 --> 00:17:39.170 and now we have worked,

NOTE Confidence: 0.938934430769231

00:17:39.170 --> 00:17:41.666 we're working on stitching those together

NOTE Confidence: 0.938934430769231

00:17:41.666 --> 00:17:44.052 into a more comprehensive Atlas type

NOTE Confidence: 0.938934430769231

00:17:44.052 --> 00:17:46.579 approach where we can integrate single EV

NOTE Confidence: 0.938934430769231

00:17:46.579 --> 00:17:49.167 data with Multiplex EV repertoire data.

NOTE Confidence: 0.964405694

00:17:51.610 --> 00:17:54.330 There's also. Resistive pulse sensing.

NOTE Confidence: 0.964405694

00:17:54.330 --> 00:17:56.786 So if any of you are doing small

NOTE Confidence: 0.964405694

00:17:56.786 --> 00:17:58.862 particle work you may use a Nano

NOTE Confidence: 0.964405694

00:17:58.862 --> 00:18:00.166 site nanoparticle tracking analyzer.

NOTE Confidence: 0.964405694

00:18:00.170 --> 00:18:01.386 Resistive pulse sensing like

NOTE Confidence: 0.964405694

00:18:01.386 --> 00:18:03.210 a Spectradine or an eyes on.

NOTE Confidence: 0.964405694

00:18:03.210 --> 00:18:05.880 SO this is specifically resistive pulse

NOTE Confidence: 0.964405694

00:18:05.880 --> 00:18:08.390 sensing that works with the output and

NOTE Confidence: 0.964405694

00:18:08.390 --> 00:18:10.010 interface of the spectradine instruments.

NOTE Confidence: 0.964405694

00:18:10.010 --> 00:18:12.404 Those use little chips and what we

NOTE Confidence: 0.964405694

00:18:12.404 --> 00:18:14.980 found was if we took the same sample

NOTE Confidence: 0.964405694

00:18:14.980 --> 00:18:17.770 and reran it on a set of chips  
NOTE Confidence: 0.964405694

00:18:17.770 --> 00:18:19.996 we get a different result every  
NOTE Confidence: 0.964405694

00:18:19.996 --> 00:18:22.230 time just with the standard beads.  
NOTE Confidence: 0.964405694

00:18:22.230 --> 00:18:24.310 And that's not good.  
NOTE Confidence: 0.964405694

00:18:24.310 --> 00:18:27.007 So we developed a way to use this  
NOTE Confidence: 0.964405694

00:18:27.007 --> 00:18:30.784 bike in and then reanalyze the  
NOTE Confidence: 0.964405694

00:18:30.784 --> 00:18:32.669 data to normalize the data.  
NOTE Confidence: 0.964405694

00:18:32.670 --> 00:18:36.510 So essentially it appropriately scales  
NOTE Confidence: 0.937931276923077

00:18:38.550 --> 00:18:40.517 so that it is calibrated and it  
NOTE Confidence: 0.937931276923077

00:18:40.517 --> 00:18:42.148 makes a difference in your data.  
NOTE Confidence: 0.937931276923077

00:18:42.150 --> 00:18:47.560 So the plot on your left is of data  
NOTE Confidence: 0.937931276923077

00:18:47.560 --> 00:18:49.870 that was not processed with RPS pass,  
NOTE Confidence: 0.937931276923077

00:18:49.870 --> 00:18:51.788 and you can see there's a huge.  
NOTE Confidence: 0.937931276923077

00:18:51.790 --> 00:18:55.549 Variation in those when we look at  
NOTE Confidence: 0.937931276923077

00:18:55.549 --> 00:18:58.760 that with where the the spike has been  
NOTE Confidence: 0.937931276923077

00:18:58.760 --> 00:19:00.806 used to appropriately scale the data,

NOTE Confidence: 0.937931276923077  
00:19:00.806 --> 00:19:03.336 you can see that we can more  
NOTE Confidence: 0.937931276923077  
00:19:03.336 --> 00:19:06.830 clearly discriminate the the, the,  
NOTE Confidence: 0.937931276923077  
00:19:06.830 --> 00:19:10.071 the qualities of the size of and  
NOTE Confidence: 0.937931276923077  
00:19:10.071 --> 00:19:12.509 the concentration of those Ev's.  
NOTE Confidence: 0.937931276923077  
00:19:12.510 --> 00:19:16.720 So all three of these we're working with.  
NOTE Confidence: 0.937931276923077  
00:19:16.720 --> 00:19:18.380 Baylor and other collaborators  
NOTE Confidence: 0.937931276923077  
00:19:18.380 --> 00:19:20.870 to to work on integrating these  
NOTE Confidence: 0.937931276923077  
00:19:20.946 --> 00:19:23.160 into tools that people can access  
NOTE Confidence: 0.937931276923077  
00:19:23.160 --> 00:19:27.440 comprehensively and shared data.  
NOTE Confidence: 0.9272475  
00:19:29.920 --> 00:19:33.076 So this has been relatively quick.  
NOTE Confidence: 0.9272475  
00:19:33.080 --> 00:19:34.475 You know, this is something  
NOTE Confidence: 0.9272475  
00:19:34.475 --> 00:19:35.870 we've really been working hard  
NOTE Confidence: 0.9272475  
00:19:35.918 --> 00:19:39.120 on for the last 5-6 years,  
NOTE Confidence: 0.9272475  
00:19:39.120 --> 00:19:40.728 but that has made the difference  
NOTE Confidence: 0.9272475  
00:19:40.728 --> 00:19:42.587 as the field has gone from being  
NOTE Confidence: 0.9272475

00:19:42.587 --> 00:19:44.230 able to go from Western lots.

NOTE Confidence: 0.9272475

00:19:44.230 --> 00:19:46.240 To flow cytometry where we don't

NOTE Confidence: 0.9272475

00:19:46.240 --> 00:19:48.200 know our limits of detection,

NOTE Confidence: 0.9272475

00:19:48.200 --> 00:19:50.160 now we know our limits of detection,

NOTE Confidence: 0.9272475

00:19:50.160 --> 00:19:52.212 we can articulate them and really

NOTE Confidence: 0.9272475

00:19:52.212 --> 00:19:54.319 reproducibly state what the results are.

NOTE Confidence: 0.911931973333333

00:19:57.040 --> 00:19:59.476 So then okay, we've done it.

NOTE Confidence: 0.911931973333333

00:19:59.480 --> 00:20:02.318 Can the whole field do it.

NOTE Confidence: 0.911931973333333

00:20:02.320 --> 00:20:06.552 In November we had a an EV

NOTE Confidence: 0.911931973333333

00:20:06.552 --> 00:20:08.400 reference material study.

NOTE Confidence: 0.911931973333333

00:20:08.400 --> 00:20:10.325 This was really spearheaded by

NOTE Confidence: 0.911931973333333

00:20:10.325 --> 00:20:12.170 Joshua Walsh who who recognized

NOTE Confidence: 0.911931973333333

00:20:12.170 --> 00:20:14.585 that as much as we try to.

NOTE Confidence: 0.911931973333333

00:20:14.590 --> 00:20:18.664 Teach everybody what they need to know.

NOTE Confidence: 0.911931973333333

00:20:18.670 --> 00:20:21.024 And we want the data to

NOTE Confidence: 0.911931973333333

00:20:21.024 --> 00:20:22.446 actually be consistent,



NOTE Confidence: 0.9119319733333333  
00:20:22.446 --> 00:20:24.710 to be calibrated, to be well,  
NOTE Confidence: 0.9119319733333333  
00:20:24.710 --> 00:20:25.830 to 1st be capable,  
NOTE Confidence: 0.9119319733333333  
00:20:25.830 --> 00:20:27.790 but then to be calibrated,  
NOTE Confidence: 0.9119319733333333  
00:20:27.790 --> 00:20:30.868 to have the whole data set  
NOTE Confidence: 0.9119319733333333  
00:20:30.870 --> 00:20:32.030 reproducible, etcetera.  
NOTE Confidence: 0.9315017  
00:20:35.510 --> 00:20:37.750 It's too complex when it's a really  
NOTE Confidence: 0.9315017  
00:20:37.750 --> 00:20:41.138 complex sample, so we use this.  
NOTE Confidence: 0.9315017  
00:20:41.138 --> 00:20:43.536 Fluorescent recombinant EV reference  
NOTE Confidence: 0.9315017  
00:20:43.536 --> 00:20:44.880 material from Anne Hendricks,  
NOTE Confidence: 0.9315017  
00:20:44.880 --> 00:20:49.960 which is now available from Sigma Millipore.  
NOTE Confidence: 0.9315017  
00:20:49.960 --> 00:20:52.120 It's called recombinant exosomes.  
NOTE Confidence: 0.9315017  
00:20:52.120 --> 00:20:54.975 From them, they rebranded it just because  
NOTE Confidence: 0.9315017  
00:20:54.975 --> 00:20:57.240 they thought that would sell better.  
NOTE Confidence: 0.9315017  
00:20:57.240 --> 00:21:00.140 Sigma did not, not.  
NOTE Confidence: 0.9315017  
00:21:00.140 --> 00:21:03.048 And and to really get at the heart of  
NOTE Confidence: 0.9315017

00:21:03.048 --> 00:21:05.794 where we need to make inroads in the field,  
NOTE Confidence: 0.9315017

00:21:05.800 --> 00:21:08.330 we went to the manufacturers.  
NOTE Confidence: 0.9315017

00:21:08.330 --> 00:21:10.706 So Josh basically offered all of  
NOTE Confidence: 0.9315017

00:21:10.706 --> 00:21:12.290 the instrument manufacturers the  
NOTE Confidence: 0.9315017

00:21:12.359 --> 00:21:14.129 opportunity to take a sample.  
NOTE Confidence: 0.9315017

00:21:14.130 --> 00:21:18.082 We shipped it off to them and send us back  
NOTE Confidence: 0.9315017

00:21:18.082 --> 00:21:21.288 the results with a a set of sort of criteria.  
NOTE Confidence: 0.9315017

00:21:21.290 --> 00:21:22.648 We want you to report back this,  
NOTE Confidence: 0.9315017

00:21:22.650 --> 00:21:24.610 this, this and this.  
NOTE Confidence: 0.9315017

00:21:24.610 --> 00:21:25.602 So everything was fair.  
NOTE Confidence: 0.9315017

00:21:25.602 --> 00:21:26.842 It was transparent up front.  
NOTE Confidence: 0.9315017

00:21:26.850 --> 00:21:29.818 This is what we wanted because  
NOTE Confidence: 0.9315017

00:21:29.818 --> 00:21:30.970 when you buy instruments,  
NOTE Confidence: 0.9315017

00:21:30.970 --> 00:21:33.245 you need the manufacturer to be able  
NOTE Confidence: 0.9315017

00:21:33.245 --> 00:21:35.846 to tell you how to properly use it.  
NOTE Confidence: 0.9315017

00:21:35.850 --> 00:21:37.320 In this study,

NOTE Confidence: 0.9315017  
00:21:37.320 --> 00:21:39.770 this is only the beginnings,  
NOTE Confidence: 0.9315017  
00:21:39.770 --> 00:21:41.443 but this shows I won't go into  
NOTE Confidence: 0.9315017  
00:21:41.443 --> 00:21:42.690 all of the results.  
NOTE Confidence: 0.9315017  
00:21:42.690 --> 00:21:45.266 But basically in that wheel of all  
NOTE Confidence: 0.9315017  
00:21:45.266 --> 00:21:48.970 the criteria we'd like to have met,  
NOTE Confidence: 0.9315017  
00:21:48.970 --> 00:21:51.118 some are very good and others  
NOTE Confidence: 0.9315017  
00:21:51.118 --> 00:21:53.529 are in the process of learning.  
NOTE Confidence: 0.9315017  
00:21:53.530 --> 00:21:56.848 And so if we redid this today,  
NOTE Confidence: 0.9315017  
00:21:56.850 --> 00:21:59.398 some of those on the bottom row.  
NOTE Confidence: 0.9315017  
00:21:59.400 --> 00:22:01.560 Would be either nearly completely  
NOTE Confidence: 0.9315017  
00:22:01.560 --> 00:22:03.720 filled in or filled in.  
NOTE Confidence: 0.9315017  
00:22:03.720 --> 00:22:05.298 So this was really a good  
NOTE Confidence: 0.9315017  
00:22:05.298 --> 00:22:06.711 opportunity to work with industry  
NOTE Confidence: 0.9315017  
00:22:06.711 --> 00:22:08.559 to start trying to pioneer this.  
NOTE Confidence: 0.9315017  
00:22:08.560 --> 00:22:13.355 So OK, so if we can do it,  
NOTE Confidence: 0.9315017

00:22:13.360 --> 00:22:14.880 how do we share this with the field?  
NOTE Confidence: 0.9315017

00:22:14.880 --> 00:22:17.393 So this needs to be centralized,  
NOTE Confidence: 0.9315017

00:22:17.393 --> 00:22:17.906 accessible.  
NOTE Confidence: 0.9315017

00:22:17.906 --> 00:22:21.497 So we've been working with Baylor as  
NOTE Confidence: 0.9315017

00:22:21.497 --> 00:22:25.296 part of the ERCC common fund effort  
NOTE Confidence: 0.9315017

00:22:25.296 --> 00:22:28.716 to develop the Nanoflow repository.  
NOTE Confidence: 0.9315017

00:22:28.720 --> 00:22:34.466 And so that's the beginnings of a  
NOTE Confidence: 0.9315017

00:22:34.466 --> 00:22:38.040 shared way to deposit the data.  
NOTE Confidence: 0.9315017

00:22:38.040 --> 00:22:41.713 This Baylor Group is also as  
NOTE Confidence: 0.9315017

00:22:41.713 --> 00:22:44.078 in parallel the XRNA Alice.  
NOTE Confidence: 0.9315017

00:22:44.080 --> 00:22:45.672 So you can see what I was talking  
NOTE Confidence: 0.9315017

00:22:45.672 --> 00:22:47.075 about before where we want to  
NOTE Confidence: 0.9315017

00:22:47.075 --> 00:22:48.275 tie the surface phenotyping data,  
NOTE Confidence: 0.9315017

00:22:48.280 --> 00:22:50.480 the individual EV phenotyping data,  
NOTE Confidence: 0.9315017

00:22:50.480 --> 00:22:52.336 the repertoire phenotyping data  
NOTE Confidence: 0.9315017

00:22:52.336 --> 00:22:55.120 then along with the RNA cargo.

NOTE Confidence: 0.9315017

00:22:55.120 --> 00:22:56.600 Data.

NOTE Confidence: 0.9315017

00:22:56.600 --> 00:23:01.410 They do have the the skeleton and

NOTE Confidence: 0.9315017

00:23:01.410 --> 00:23:03.360 the background and the infrastructure

NOTE Confidence: 0.9315017

00:23:03.360 --> 00:23:08.199 of the xrna Atlas there.

NOTE Confidence: 0.9315017

00:23:08.200 --> 00:23:10.817 So this is moving us closer to

NOTE Confidence: 0.9315017

00:23:10.817 --> 00:23:11.879 doing what we want to do,

NOTE Confidence: 0.9315017

00:23:11.880 --> 00:23:13.716 which is to be able to look at subsets,

NOTE Confidence: 0.9315017

00:23:13.720 --> 00:23:15.760 identify markers to pull out

NOTE Confidence: 0.9315017

00:23:15.760 --> 00:23:18.200 subsets to look at the RNA.

NOTE Confidence: 0.9315017

00:23:18.200 --> 00:23:20.600 Then we hit a roadblock which

NOTE Confidence: 0.9315017

00:23:20.600 --> 00:23:23.908 was one I expected, but.

NOTE Confidence: 0.9315017

00:23:23.910 --> 00:23:25.642 You know,

NOTE Confidence: 0.9315017

00:23:25.642 --> 00:23:28.818 most RNA seq methods require

NOTE Confidence: 0.9315017

00:23:28.818 --> 00:23:32.310 nanogram levels of RNA.

NOTE Confidence: 0.9315017

00:23:32.310 --> 00:23:34.266 When you get to the subsets,

NOTE Confidence: 0.9315017

00:23:34.270 --> 00:23:36.364 you're probably in less than 100  
NOTE Confidence: 0.9315017

00:23:36.364 --> 00:23:38.514 picogram kind of range of RNA.  
NOTE Confidence: 0.9315017

00:23:38.514 --> 00:23:40.488 So we tried to we decided to  
NOTE Confidence: 0.9315017

00:23:40.488 --> 00:23:42.707 test whether or not we could use  
NOTE Confidence: 0.9315017

00:23:42.710 --> 00:23:44.498 single cell sequencing methods,  
NOTE Confidence: 0.9315017

00:23:44.498 --> 00:23:47.789 not in the single cell mode but in.  
NOTE Confidence: 0.9315017

00:23:47.790 --> 00:23:51.115 Bulk using that as the library preparation  
NOTE Confidence: 0.9315017

00:23:51.115 --> 00:23:54.110 method for looking at EVRN A's.  
NOTE Confidence: 0.940253531111111

00:23:57.230 --> 00:23:58.812 And remember I showed you the picture  
NOTE Confidence: 0.940253531111111

00:23:58.812 --> 00:24:00.831 of the DC 2.4 E V's on the flow  
NOTE Confidence: 0.940253531111111

00:24:00.831 --> 00:24:02.668 cytometer and I said remember these?  
NOTE Confidence: 0.940253531111111

00:24:02.670 --> 00:24:06.030 That's the cell line we chose.  
NOTE Confidence: 0.940253531111111

00:24:06.030 --> 00:24:07.314 It grows like weeds.  
NOTE Confidence: 0.940253531111111

00:24:07.314 --> 00:24:09.240 It's a little mouse dendritic cell  
NOTE Confidence: 0.940253531111111

00:24:09.304 --> 00:24:11.408 line that Ken Rock made back in the  
NOTE Confidence: 0.940253531111111

00:24:11.408 --> 00:24:13.141 1990s and it feeds itself GMCSF.

NOTE Confidence: 0.9402535311111111  
00:24:13.141 --> 00:24:14.496 So these are the happiest  
NOTE Confidence: 0.9402535311111111  
00:24:14.496 --> 00:24:15.580 cells you could ever.  
NOTE Confidence: 0.9402535311111111  
00:24:15.580 --> 00:24:19.820 Want they grow like weeds,  
NOTE Confidence: 0.9402535311111111  
00:24:19.820 --> 00:24:21.857 and they've also had a lot of  
NOTE Confidence: 0.9402535311111111  
00:24:21.857 --> 00:24:23.420 manipulation in their background.  
NOTE Confidence: 0.9402535311111111  
00:24:23.420 --> 00:24:26.820 So I outlined here all of the background  
NOTE Confidence: 0.9402535311111111  
00:24:26.820 --> 00:24:30.002 that I kind of ignored until one of  
NOTE Confidence: 0.9402535311111111  
00:24:30.002 --> 00:24:34.199 our reviewers pushed us to instead of  
NOTE Confidence: 0.9402535311111111  
00:24:34.199 --> 00:24:38.168 TEM get cryoem to really hammer out  
NOTE Confidence: 0.9402535311111111  
00:24:38.168 --> 00:24:40.883 the exact size of these and what we  
NOTE Confidence: 0.9402535311111111  
00:24:40.883 --> 00:24:43.760 couldn't see in the TEM on the left.  
NOTE Confidence: 0.9402535311111111  
00:24:43.760 --> 00:24:45.615 You can see really clearly on the  
NOTE Confidence: 0.9402535311111111  
00:24:45.615 --> 00:24:47.524 right we have retroviral capsules or  
NOTE Confidence: 0.9402535311111111  
00:24:47.524 --> 00:24:50.320 something that looks awfully a lot like them.  
NOTE Confidence: 0.9402535311111111  
00:24:50.320 --> 00:24:52.608 And I got a call from the lab  
NOTE Confidence: 0.9402535311111111

00:24:52.608 --> 00:24:54.877 who was helping us with this.  
NOTE Confidence: 0.9402535311111111

00:24:54.880 --> 00:24:56.065 Not a call,  
NOTE Confidence: 0.9402535311111111

00:24:56.065 --> 00:24:58.040 it was worse than that,  
NOTE Confidence: 0.9402535311111111

00:24:58.040 --> 00:25:01.155 an e-mail that was carbon copied to  
NOTE Confidence: 0.9402535311111111

00:25:01.155 --> 00:25:04.036 the then scientific director of all of  
NOTE Confidence: 0.9402535311111111

00:25:04.036 --> 00:25:06.691 NIH saying what do you not understand  
NOTE Confidence: 0.9402535311111111

00:25:06.691 --> 00:25:10.120 about B SL1 samples for a B SL1 lab.  
NOTE Confidence: 0.93622824

00:25:12.930 --> 00:25:17.018 This cell line is sold by Sigma and  
NOTE Confidence: 0.93622824

00:25:17.018 --> 00:25:20.768 mercury pour has a B SL1 cell line.  
NOTE Confidence: 0.93622824

00:25:20.770 --> 00:25:23.927 And I say look, I'm really sorry,  
NOTE Confidence: 0.93622824

00:25:23.930 --> 00:25:25.088 I don't know what that is.  
NOTE Confidence: 0.93622824

00:25:25.090 --> 00:25:26.890 It could be a mishmash of  
NOTE Confidence: 0.93622824

00:25:26.890 --> 00:25:28.930 rearrangements of any of those things.  
NOTE Confidence: 0.93622824

00:25:28.930 --> 00:25:30.784 In this background it's also a  
NOTE Confidence: 0.93622824

00:25:30.784 --> 00:25:32.865 mouse cell line and they have  
NOTE Confidence: 0.93622824

00:25:32.865 --> 00:25:34.810 lots of endogenous rector viruses,



NOTE Confidence: 0.93622824

00:25:34.810 --> 00:25:38.351 so I have no idea what that is, but.

NOTE Confidence: 0.93622824

00:25:38.351 --> 00:25:40.759 I'll I'll get to the bottom of it.

NOTE Confidence: 0.93622824

00:25:40.760 --> 00:25:42.256 And this is now.

NOTE Confidence: 0.93622824

00:25:42.256 --> 00:25:45.519 I've lost count of how many years later

NOTE Confidence: 0.93622824

00:25:45.520 --> 00:25:47.186 we decided we wanted to apply this

NOTE Confidence: 0.93622824

00:25:47.186 --> 00:25:48.925 RN A/C approach to those because I

NOTE Confidence: 0.93622824

00:25:48.925 --> 00:25:50.720 wanted to figure out what's what is it.

NOTE Confidence: 0.93622824

00:25:50.720 --> 00:25:53.030 I don't want to just do a PCR for this,

NOTE Confidence: 0.93622824

00:25:53.030 --> 00:25:53.880 that and the other thing,

NOTE Confidence: 0.93622824

00:25:53.880 --> 00:25:56.680 I want to know what's in it.

NOTE Confidence: 0.93622824

00:25:56.680 --> 00:25:58.810 So we've done Proteomics and we've

NOTE Confidence: 0.93622824

00:25:58.810 --> 00:26:01.272 done RNAC and it turns out we find

NOTE Confidence: 0.93622824

00:26:01.272 --> 00:26:03.151 a dominant species and it turns

NOTE Confidence: 0.93622824

00:26:03.151 --> 00:26:05.089 out it's Mouse Maloney virus which

NOTE Confidence: 0.93622824

00:26:05.089 --> 00:26:06.920 was part of its background.

NOTE Confidence: 0.90623035

00:26:10.150 --> 00:26:13.510 So that is a xenotropic virus,  
NOTE Confidence: 0.90623035

00:26:13.510 --> 00:26:16.030 meaning it doesn't go from mouse cells to us,  
NOTE Confidence: 0.90623035

00:26:16.030 --> 00:26:18.022 it just stays between mouse cells and it  
NOTE Confidence: 0.90623035

00:26:18.022 --> 00:26:20.038 doesn't go from mouse cell to mouse cell  
NOTE Confidence: 0.90623035

00:26:20.038 --> 00:26:21.949 unless the cells are actively dividing,  
NOTE Confidence: 0.90623035

00:26:21.950 --> 00:26:26.550 which, well, those do. And so  
NOTE Confidence: 0.94427896

00:26:29.230 --> 00:26:30.766 another reason I wanted  
NOTE Confidence: 0.94427896

00:26:30.766 --> 00:26:32.910 to go down this crazy Rd.  
NOTE Confidence: 0.94427896

00:26:32.910 --> 00:26:34.902 is because of the hers where  
NOTE Confidence: 0.94427896

00:26:34.902 --> 00:26:36.940 we know that those modulate.  
NOTE Confidence: 0.94427896

00:26:36.940 --> 00:26:38.540 Responses to immunotherapy or  
NOTE Confidence: 0.94427896

00:26:38.540 --> 00:26:40.540 their indications that they do.  
NOTE Confidence: 0.94427896

00:26:40.540 --> 00:26:43.020 And so we wanted to have a pipeline,  
NOTE Confidence: 0.94427896

00:26:43.020 --> 00:26:46.276 a method that would allow us to elucidate  
NOTE Confidence: 0.94427896

00:26:46.276 --> 00:26:48.887 the presence or absence or the types  
NOTE Confidence: 0.94427896

00:26:48.887 --> 00:26:51.850 of herbs in our human EV samples.

NOTE Confidence: 0.94427896

00:26:51.850 --> 00:26:53.666 So we're collaborating with

NOTE Confidence: 0.94427896

00:26:53.666 --> 00:26:55.936 Kendall Jensen at Tijan and

NOTE Confidence: 0.94427896

00:26:55.936 --> 00:26:58.192 Yasmine Belkade's group at NIAID.

NOTE Confidence: 0.94427896

00:26:58.192 --> 00:27:00.302 She's just accepted the position

NOTE Confidence: 0.94427896

00:27:00.302 --> 00:27:01.942 to run the Pasteur Institute.

NOTE Confidence: 0.94427896

00:27:01.942 --> 00:27:03.154 So unfortunately we're going

NOTE Confidence: 0.94427896

00:27:03.154 --> 00:27:04.250 to lose her soon.

NOTE Confidence: 0.94427896

00:27:04.250 --> 00:27:06.112 But we're working very hard to get

NOTE Confidence: 0.94427896

00:27:06.112 --> 00:27:09.434 this all tied together before she

NOTE Confidence: 0.94427896

00:27:09.434 --> 00:27:12.130 goes to have a comprehensive pipeline

NOTE Confidence: 0.94427896

00:27:12.130 --> 00:27:14.370 that would include conventional RN,

NOTE Confidence: 0.94427896

00:27:14.370 --> 00:27:15.450 A's and the Hearse.

NOTE Confidence: 0.94427896

00:27:15.450 --> 00:27:17.846 So I want to show you some results

NOTE Confidence: 0.94427896

00:27:17.846 --> 00:27:20.310 of all these tools that we've been.

NOTE Confidence: 0.94427896

00:27:20.310 --> 00:27:22.947 Working on and here a couple of the examples.

NOTE Confidence: 0.94427896

00:27:22.950 --> 00:27:27.030 I'll show you a little bit of kidney cancer,  
NOTE Confidence: 0.94427896

00:27:27.030 --> 00:27:31.990 prostate cancer, colon cancer, CNS diseases.  
NOTE Confidence: 0.94427896

00:27:31.990 --> 00:27:33.990 But first,  
NOTE Confidence: 0.94427896

00:27:33.990 --> 00:27:36.906 if you could live a day in my shoes,  
NOTE Confidence: 0.94427896

00:27:36.910 --> 00:27:41.710 you get a question just about every day.  
NOTE Confidence: 0.94427896

00:27:41.710 --> 00:27:43.047 I want to start a study and  
NOTE Confidence: 0.94427896

00:27:43.047 --> 00:27:44.548 I want to look at exosomes.  
NOTE Confidence: 0.94427896

00:27:44.550 --> 00:27:47.741 That's what people say to me and I want  
NOTE Confidence: 0.94427896

00:27:47.741 --> 00:27:50.210 to know what kind of blood tube I need.  
NOTE Confidence: 0.94427896

00:27:50.210 --> 00:27:52.355 And that's a really hard what  
NOTE Confidence: 0.94427896

00:27:52.355 --> 00:27:53.370 do you want to do with it?  
NOTE Confidence: 0.94427896

00:27:53.370 --> 00:27:56.230 What do you, what do you want to look at.  
NOTE Confidence: 0.94427896

00:27:56.230 --> 00:27:58.913 So to help us figure out what is  
NOTE Confidence: 0.94427896

00:27:58.913 --> 00:28:00.610 our right blood collection tube,  
NOTE Confidence: 0.94427896

00:28:00.610 --> 00:28:05.450 we decided to compare for the SST tubes,  
NOTE Confidence: 0.94427896

00:28:05.450 --> 00:28:08.170 EDTA tubes and the street

NOTE Confidence: 0.94427896

00:28:08.170 --> 00:28:09.265 DNA&amp;RNA complete tubes.

NOTE Confidence: 0.94427896

00:28:09.265 --> 00:28:11.455 This is the comparisons that we

NOTE Confidence: 0.94427896

00:28:11.455 --> 00:28:14.345 did to suss out the impacts of

NOTE Confidence: 0.94427896

00:28:14.345 --> 00:28:16.680 platelets and not platelets and.

NOTE Confidence: 0.94427896

00:28:16.680 --> 00:28:18.198 Ways that you do the spins,

NOTE Confidence: 0.94427896

00:28:18.200 --> 00:28:20.629 we counted the particles that were remaining

NOTE Confidence: 0.94427896

00:28:20.629 --> 00:28:23.210 after we did the depletions etcetera.

NOTE Confidence: 0.94427896

00:28:23.210 --> 00:28:28.004 And what you see is that we had

NOTE Confidence: 0.94427896

00:28:28.004 --> 00:28:33.240 a surprise which is that CD62 P,

NOTE Confidence: 0.94427896

00:28:33.240 --> 00:28:33.765 CD242A,

NOTE Confidence: 0.94427896

00:28:33.765 --> 00:28:36.390 some platelet markers were not

NOTE Confidence: 0.94427896

00:28:36.390 --> 00:28:38.052 only elevated in samples where

NOTE Confidence: 0.94427896

00:28:38.052 --> 00:28:39.750 you froze the sample and then

NOTE Confidence: 0.94427896

00:28:39.808 --> 00:28:41.198 you spin out the platelets,

NOTE Confidence: 0.94427896

00:28:41.200 --> 00:28:42.160 which is a terrible idea,

NOTE Confidence: 0.94427896

00:28:42.160 --> 00:28:44.939 but a lot of people do it.  
NOTE Confidence: 0.94427896

00:28:44.940 --> 00:28:47.775 It was also elevated in the struck DNA tubes.  
NOTE Confidence: 0.94427896

00:28:47.780 --> 00:28:49.095 So maybe something with the  
NOTE Confidence: 0.94427896

00:28:49.095 --> 00:28:50.758 fixation of the struck DNA tube  
NOTE Confidence: 0.94427896

00:28:50.758 --> 00:28:52.258 that's causing shedding of these  
NOTE Confidence: 0.94729798

00:28:54.740 --> 00:28:55.388 these vesicles.  
NOTE Confidence: 0.94729798

00:28:55.388 --> 00:28:57.980 And so we've we've looked at this further.  
NOTE Confidence: 0.94729798

00:28:57.980 --> 00:29:00.402 But you know it's this kind of  
NOTE Confidence: 0.94729798

00:29:00.402 --> 00:29:02.081 quantitative analysis that helps us  
NOTE Confidence: 0.94729798

00:29:02.081 --> 00:29:04.016 assess the not only the integrity  
NOTE Confidence: 0.94729798

00:29:04.016 --> 00:29:06.050 but also the repertoire and the  
NOTE Confidence: 0.94729798

00:29:06.121 --> 00:29:08.292 relative abundance of these different  
NOTE Confidence: 0.94729798

00:29:08.292 --> 00:29:11.124 types of vesicles in the solution.  
NOTE Confidence: 0.94729798

00:29:11.130 --> 00:29:12.168 So for us, for our lab,  
NOTE Confidence: 0.94729798

00:29:12.170 --> 00:29:13.490 for our protocols,  
NOTE Confidence: 0.94729798

00:29:13.490 --> 00:29:17.650 we're doing SST tubes and complete RNA tubes.

NOTE Confidence: 0.94729798

00:29:17.650 --> 00:29:19.870 I I actually think that plasma

NOTE Confidence: 0.94729798

00:29:19.870 --> 00:29:21.489 DTA tubes are also great.

NOTE Confidence: 0.88469275375

00:29:23.730 --> 00:29:25.530 I spoke to somebody earlier

NOTE Confidence: 0.88469275375

00:29:25.530 --> 00:29:26.610 today about oncosomes.

NOTE Confidence: 0.88469275375

00:29:26.610 --> 00:29:28.615 These are large vesicles shed

NOTE Confidence: 0.88469275375

00:29:28.615 --> 00:29:31.096 by tumor cells which are like

NOTE Confidence: 0.88469275375

00:29:31.096 --> 00:29:32.850 larger than 800 nanometers,

NOTE Confidence: 0.88469275375

00:29:32.850 --> 00:29:35.250 sometimes larger than a Micron.

NOTE Confidence: 0.88469275375

00:29:35.250 --> 00:29:36.555 So every platelet depleting protocol

NOTE Confidence: 0.88469275375

00:29:36.555 --> 00:29:38.702 that you do to spin out the platelets

NOTE Confidence: 0.88469275375

00:29:38.702 --> 00:29:40.286 is going to remove the oncosomes.

NOTE Confidence: 0.9100634

00:29:42.990 --> 00:29:46.426 I I don't have a good solution.

NOTE Confidence: 0.9100634

00:29:46.430 --> 00:29:47.828 If you want to study those,

NOTE Confidence: 0.9100634

00:29:47.830 --> 00:29:50.918 I think you have to go directly to

NOTE Confidence: 0.9100634

00:29:50.918 --> 00:29:53.430 processing the onpisomes separately.

NOTE Confidence: 0.9100634

00:29:53.430 --> 00:29:58.150 So our approach is showing us good  
NOTE Confidence: 0.9100634

00:29:58.150 --> 00:30:00.509 fidelity and differences in tumor types.  
NOTE Confidence: 0.9100634

00:30:00.510 --> 00:30:04.030 So Long story short we compared a bunch  
NOTE Confidence: 0.9100634

00:30:04.030 --> 00:30:06.430 of different EV's from different tumors.  
NOTE Confidence: 0.9100634

00:30:06.430 --> 00:30:07.950 This is something that  
NOTE Confidence: 0.9100634

00:30:07.950 --> 00:30:09.609 we've already published and.  
NOTE Confidence: 0.9100634

00:30:09.610 --> 00:30:12.042 You can see Epcam is more commonly spread  
NOTE Confidence: 0.9100634

00:30:12.042 --> 00:30:14.856 or sort of more highly expressed in these  
NOTE Confidence: 0.9100634

00:30:14.856 --> 00:30:17.210 samples from the epithelial tumors and  
NOTE Confidence: 0.9100634

00:30:17.210 --> 00:30:20.322 from the Seglio bus, I mean, it's good.  
NOTE Confidence: 0.9100634

00:30:20.322 --> 00:30:22.965 You wouldn't expect Epicam so much in those.  
NOTE Confidence: 0.9100634

00:30:22.970 --> 00:30:26.170 The tetraspanins and CD44 are  
NOTE Confidence: 0.9100634

00:30:26.170 --> 00:30:30.210 up in the in in both.  
NOTE Confidence: 0.9100634

00:30:30.210 --> 00:30:33.450 So for kidney cancers,  
NOTE Confidence: 0.9100634

00:30:33.450 --> 00:30:37.356 there's not a great molecular handle.  
NOTE Confidence: 0.9100634

00:30:37.360 --> 00:30:39.200 For pulling out kidney cancers.



NOTE Confidence: 0.9100634

00:30:39.200 --> 00:30:43.400 So Marsha Lenahan and Maria Marino

NOTE Confidence: 0.9100634

00:30:43.400 --> 00:30:45.608 have this amazing set of cohorted

NOTE Confidence: 0.9100634

00:30:45.608 --> 00:30:48.152 patients and data and studies and

NOTE Confidence: 0.9100634

00:30:48.152 --> 00:30:50.160 information they've learned about

NOTE Confidence: 0.9100634

00:30:50.160 --> 00:30:53.436 one hippo window and and other

NOTE Confidence: 0.9100634

00:30:53.440 --> 00:30:56.800 forms of hereditary kidney cancers.

NOTE Confidence: 0.9100634

00:30:56.800 --> 00:30:59.880 So we worked with them to look at some

NOTE Confidence: 0.9100634

00:30:59.880 --> 00:31:02.600 of the different tumor types that we

NOTE Confidence: 0.9100634

00:31:02.600 --> 00:31:04.932 could prototype with and then begin to

NOTE Confidence: 0.9100634

00:31:04.932 --> 00:31:06.630 look at those samples and patients.

NOTE Confidence: 0.9100634

00:31:06.630 --> 00:31:11.190 And so we tried a battery of different

NOTE Confidence: 0.9100634

00:31:11.190 --> 00:31:14.574 markers and we found some that

NOTE Confidence: 0.9100634

00:31:14.574 --> 00:31:17.310 really hadn't been expected and they

NOTE Confidence: 0.9100634

00:31:17.310 --> 00:31:18.750 have some of the same features.

NOTE Confidence: 0.9100634

00:31:18.750 --> 00:31:20.976 Some of them are also Tetra spanners

NOTE Confidence: 0.9100634

00:31:20.976 --> 00:31:22.950 and they're also Stemmus markers.  
NOTE Confidence: 0.9100634

00:31:22.950 --> 00:31:24.822 So this is consistent with what we saw  
NOTE Confidence: 0.9100634

00:31:24.822 --> 00:31:26.870 was really elevated in the other two types.  
NOTE Confidence: 0.9100634

00:31:26.870 --> 00:31:29.229 So maybe we're finding that there's kind  
NOTE Confidence: 0.9100634

00:31:29.229 --> 00:31:33.240 of a a malignant signature as opposed  
NOTE Confidence: 0.9100634

00:31:33.240 --> 00:31:36.910 to a specific type of tumor signature.  
NOTE Confidence: 0.9100634

00:31:36.910 --> 00:31:40.550 In the types of markers they express.  
NOTE Confidence: 0.9100634

00:31:40.550 --> 00:31:43.088 Then we worked with collaborators to  
NOTE Confidence: 0.9100634

00:31:43.088 --> 00:31:46.894 look at the EV profiles in malignant CSF  
NOTE Confidence: 0.9100634

00:31:46.894 --> 00:31:51.910 samples and other CSF samples including  
NOTE Confidence: 0.9100634

00:31:51.910 --> 00:31:54.990 autoimmune diseases and viral diseases.  
NOTE Confidence: 0.9100634

00:31:54.990 --> 00:31:56.756 And you can see again here  
NOTE Confidence: 0.9100634

00:31:56.756 --> 00:31:57.986 we see that same pattern.  
NOTE Confidence: 0.925158445

00:32:00.870 --> 00:32:01.590 And so  
NOTE Confidence: 0.940253542

00:32:03.790 --> 00:32:04.990 once we have the markers,  
NOTE Confidence: 0.940253542

00:32:04.990 --> 00:32:06.470 we do the pull down.

NOTE Confidence: 0.940253542

00:32:06.470 --> 00:32:08.990 Do we actually see differences in the RNA?

NOTE Confidence: 0.940253542

00:32:08.990 --> 00:32:13.198 So this was one of my early proof of

NOTE Confidence: 0.940253542

00:32:13.198 --> 00:32:15.028 principles examples where we just took

NOTE Confidence: 0.940253531

00:32:17.430 --> 00:32:18.694 a thoracentesis sample from

NOTE Confidence: 0.940253531

00:32:18.694 --> 00:32:20.590 a patient of mine who had

NOTE Confidence: 0.957025865

00:32:22.790 --> 00:32:24.842 very metastatic prostate cancer,

NOTE Confidence: 0.957025865

00:32:24.842 --> 00:32:28.718 which is PSMA positive the therapeutic

NOTE Confidence: 0.957025865

00:32:28.718 --> 00:32:30.986 tap and in the therapeutic tap in

NOTE Confidence: 0.957025865

00:32:30.986 --> 00:32:32.842 the biospecimen protocol we're able

NOTE Confidence: 0.957025865

00:32:32.842 --> 00:32:36.140 to pull down the PSMA positive EV's.

NOTE Confidence: 0.957025865

00:32:36.140 --> 00:32:38.790 Compared to the PSMA negative

NOTE Confidence: 0.957025865

00:32:38.790 --> 00:32:41.250 EV's compared to the bulk sample.

NOTE Confidence: 0.957025865

00:32:41.250 --> 00:32:44.449 And you can see there are several

NOTE Confidence: 0.957025865

00:32:44.450 --> 00:32:45.990 RNA's which are highly associated

NOTE Confidence: 0.957025865

00:32:45.990 --> 00:32:47.735 with the PSMA positive ones,

NOTE Confidence: 0.957025865

00:32:47.735 --> 00:32:49.920 which you would have missed if  
NOTE Confidence: 0.957025865

00:32:49.920 --> 00:32:52.090 you were looking at the soup of  
NOTE Confidence: 0.957025865

00:32:52.090 --> 00:32:53.744 everything because there's so many  
NOTE Confidence: 0.957025865

00:32:53.744 --> 00:32:56.032 other kinds of vesicles that compete  
NOTE Confidence: 0.957025865

00:32:56.032 --> 00:32:58.487 in that type of identification.  
NOTE Confidence: 0.957025865

00:32:58.490 --> 00:32:59.972 So I thought PSMA was going  
NOTE Confidence: 0.957025865

00:32:59.972 --> 00:33:01.450 to be a great marker,  
NOTE Confidence: 0.957025865

00:33:01.450 --> 00:33:03.490 but really what marker should we be using?  
NOTE Confidence: 0.939559504827587

00:33:06.500 --> 00:33:08.866 I insinuated and I really feel like  
NOTE Confidence: 0.939559504827587

00:33:08.866 --> 00:33:11.141 the markers that we choose are not  
NOTE Confidence: 0.939559504827587

00:33:11.141 --> 00:33:13.634 going to be the same markers that we  
NOTE Confidence: 0.939559504827587

00:33:13.634 --> 00:33:15.853 use in the context of intact tissue.  
NOTE Confidence: 0.939559504827587

00:33:15.860 --> 00:33:18.100 It may relate more to their phenotype.  
NOTE Confidence: 0.939559504827587

00:33:18.100 --> 00:33:21.760 So we did a large screen of 170  
NOTE Confidence: 0.939559504827587

00:33:21.760 --> 00:33:24.732 different EV surface markers across  
NOTE Confidence: 0.939559504827587

00:33:24.732 --> 00:33:27.720 some of those kidney cancer patients.

NOTE Confidence: 0.939559504827587  
00:33:27.720 --> 00:33:30.680 Other CSF sample was just a massive cohort.  
NOTE Confidence: 0.939559504827587  
00:33:30.680 --> 00:33:31.946 So if you're squinting at this  
NOTE Confidence: 0.939559504827587  
00:33:31.946 --> 00:33:33.240 from the back of the room,  
NOTE Confidence: 0.939559504827587  
00:33:33.240 --> 00:33:34.640 you can see there's sort of a  
NOTE Confidence: 0.939559504827587  
00:33:34.640 --> 00:33:37.600 tartan Plaid kind of pattern.  
NOTE Confidence: 0.939559504827587  
00:33:37.600 --> 00:33:39.970 There's a sample down here  
NOTE Confidence: 0.939559504827587  
00:33:39.970 --> 00:33:42.020 where it's all blown out.  
NOTE Confidence: 0.939559504827587  
00:33:42.020 --> 00:33:43.280 It turned out,  
NOTE Confidence: 0.939559504827587  
00:33:43.280 --> 00:33:45.752 turned out that person had a radioisotopic  
NOTE Confidence: 0.939559504827587  
00:33:45.752 --> 00:33:48.120 treatment for metastatic prostate  
NOTE Confidence: 0.939559504827587  
00:33:48.120 --> 00:33:51.540 cancer couple weeks before and was  
NOTE Confidence: 0.939559504827587  
00:33:51.540 --> 00:33:53.228 having ramped up marrow production.  
NOTE Confidence: 0.939559504827587  
00:33:53.228 --> 00:33:55.800 I don't have any other samples like that,  
NOTE Confidence: 0.939559504827587  
00:33:55.800 --> 00:33:57.840 but clearly this is. Not,  
NOTE Confidence: 0.939559504827587  
00:33:57.840 --> 00:34:00.824 we're not going to understand much from that.  
NOTE Confidence: 0.939559504827587

00:34:00.824 --> 00:34:03.032 But then there are sections where  
NOTE Confidence: 0.939559504827587

00:34:03.032 --> 00:34:05.140 you see more of some workers,  
NOTE Confidence: 0.939559504827587

00:34:05.140 --> 00:34:07.464 less of other markers and in sets.  
NOTE Confidence: 0.939559504827587

00:34:07.464 --> 00:34:09.847 And if you break down those sets and  
NOTE Confidence: 0.939559504827587

00:34:09.847 --> 00:34:12.239 you look and you say CSF versus serum,  
NOTE Confidence: 0.939559504827587

00:34:12.240 --> 00:34:14.799 they're really different.  
NOTE Confidence: 0.939559504827587

00:34:14.800 --> 00:34:16.260 Looking at PC A's,  
NOTE Confidence: 0.939559504827587

00:34:16.260 --> 00:34:18.872 if you look at tumors versus immune,  
NOTE Confidence: 0.939559504827587

00:34:18.872 --> 00:34:20.732 this tumor, that tumor,  
NOTE Confidence: 0.939559504827587

00:34:20.732 --> 00:34:22.916 they're also very separable.  
NOTE Confidence: 0.939559504827587

00:34:22.920 --> 00:34:23.402 Well,  
NOTE Confidence: 0.939559504827587

00:34:23.402 --> 00:34:27.258 some of them are separable more than others.  
NOTE Confidence: 0.939559504827587

00:34:27.260 --> 00:34:31.122 And then looking at the CSF samples from  
NOTE Confidence: 0.939559504827587

00:34:31.122 --> 00:34:33.414 patients with or without brain tumors,  
NOTE Confidence: 0.939559504827587

00:34:33.420 --> 00:34:35.748 you can also see that there  
NOTE Confidence: 0.939559504827587

00:34:35.748 --> 00:34:37.180 are differences that we see.

NOTE Confidence: 0.941511525

00:34:40.620 --> 00:34:45.065 So we also worked with Steve Jacobson

NOTE Confidence: 0.941511525

00:34:45.065 --> 00:34:50.100 in NININDS and he studies both Ms.

NOTE Confidence: 0.941511525

00:34:50.100 --> 00:34:53.940 and he MTSP, the HTLV associated

NOTE Confidence: 0.941511525

00:34:53.940 --> 00:34:56.139 tropical Myelotis myelo.

NOTE Confidence: 0.790203765

00:34:59.920 --> 00:35:02.840 \*\*\*\*\* peripheresis.

NOTE Confidence: 0.790203765

00:35:02.840 --> 00:35:06.984 So these are CSF samples from those

NOTE Confidence: 0.790203765

00:35:06.984 --> 00:35:10.450 patients and patients also with who carry

NOTE Confidence: 0.790203765

00:35:10.450 --> 00:35:12.400 the HTLV virus but are asymptomatic.

NOTE Confidence: 0.790203765

00:35:12.400 --> 00:35:15.262 That's what the AC's are or other

NOTE Confidence: 0.790203765

00:35:15.262 --> 00:35:17.848 viral diseases and you can see

NOTE Confidence: 0.790203765

00:35:17.848 --> 00:35:21.160 that turns out that hand patients,

NOTE Confidence: 0.790203765

00:35:21.160 --> 00:35:23.866 the ones with active disease associated

NOTE Confidence: 0.790203765

00:35:23.866 --> 00:35:26.450 with HTLV in the nervous system.

NOTE Confidence: 0.790203765

00:35:26.450 --> 00:35:31.009 Have higher CDA than CD2E V counts.

NOTE Confidence: 0.790203765

00:35:31.010 --> 00:35:33.630 We've followed that up with

NOTE Confidence: 0.790203765

00:35:33.630 --> 00:35:35.726 another set of samples,  
NOTE Confidence: 0.790203765

00:35:35.730 --> 00:35:37.788 again that size and as as well  
NOTE Confidence: 0.790203765

00:35:37.788 --> 00:35:39.815 as other markers and we still  
NOTE Confidence: 0.790203765

00:35:39.815 --> 00:35:41.247 see that robust difference.  
NOTE Confidence: 0.790203765

00:35:41.250 --> 00:35:42.814 It's it's it's very,  
NOTE Confidence: 0.790203765

00:35:42.814 --> 00:35:44.769 it's not a massive magnitude,  
NOTE Confidence: 0.790203765

00:35:44.770 --> 00:35:47.210 but it's very consistent.  
NOTE Confidence: 0.790203765

00:35:47.210 --> 00:35:48.986 So which of these EV markers  
NOTE Confidence: 0.790203765

00:35:48.986 --> 00:35:50.570 relate to the biological state,  
NOTE Confidence: 0.790203765

00:35:50.570 --> 00:35:52.362 meaning the biological state  
NOTE Confidence: 0.790203765

00:35:52.362 --> 00:35:55.050 of the cell that made them?  
NOTE Confidence: 0.790203765

00:35:55.050 --> 00:35:59.306 And this is work that was done with a  
NOTE Confidence: 0.790203765

00:35:59.306 --> 00:36:02.170 colleague who had a really interesting  
NOTE Confidence: 0.790203765

00:36:02.170 --> 00:36:04.730 biological phenotype they were studying.  
NOTE Confidence: 0.790203765

00:36:04.730 --> 00:36:06.926 They made some knockout cell lines.  
NOTE Confidence: 0.790203765

00:36:06.930 --> 00:36:08.855 And what you see here is our



NOTE Confidence: 0.790203765

00:36:08.855 --> 00:36:10.250 B plus antibody control,

NOTE Confidence: 0.790203765

00:36:10.250 --> 00:36:11.846 the knockout line, the control line,

NOTE Confidence: 0.790203765

00:36:11.850 --> 00:36:13.728 the knockout line, the control line.

NOTE Confidence: 0.790203765

00:36:13.730 --> 00:36:16.810 And what you can see is that

NOTE Confidence: 0.790203765

00:36:16.810 --> 00:36:19.750 there are some genes that are just

NOTE Confidence: 0.790203765

00:36:19.750 --> 00:36:22.556 missing from the knockouts and there

NOTE Confidence: 0.790203765

00:36:22.556 --> 00:36:24.914 are some genes that are missing.

NOTE Confidence: 0.790203765

00:36:24.920 --> 00:36:26.000 From the controls.

NOTE Confidence: 0.790203765

00:36:26.000 --> 00:36:28.520 So we're really getting a sense of

NOTE Confidence: 0.790203765

00:36:28.592 --> 00:36:31.879 changes in these related to that.

NOTE Confidence: 0.790203765

00:36:31.880 --> 00:36:34.710 So we're really just starting

NOTE Confidence: 0.790203765

00:36:34.710 --> 00:36:36.800 to apply these and learn more.

NOTE Confidence: 0.790203765

00:36:36.800 --> 00:36:40.400 We have also found a pattern

NOTE Confidence: 0.790203765

00:36:40.400 --> 00:36:41.902 in metastatic potential,

NOTE Confidence: 0.790203765

00:36:41.902 --> 00:36:44.318 so match sets of cell lines that have

NOTE Confidence: 0.790203765

00:36:44.318 --> 00:36:45.828 different metastatic potential on  
NOTE Confidence: 0.790203765

00:36:45.828 --> 00:36:51.040 their markers and so now we want to  
NOTE Confidence: 0.790203765

00:36:51.040 --> 00:36:53.789 move forward further with that so.  
NOTE Confidence: 0.790203765

00:36:53.789 --> 00:36:54.487 You know,  
NOTE Confidence: 0.790203765

00:36:54.487 --> 00:36:56.232 I started talking about the  
NOTE Confidence: 0.790203765

00:36:56.232 --> 00:36:57.650 commotion in the blood,  
NOTE Confidence: 0.790203765

00:36:57.650 --> 00:37:01.781 or as one of the earlier professor said,  
NOTE Confidence: 0.790203765

00:37:01.781 --> 00:37:03.803 the the mess that is the  
NOTE Confidence: 0.790203765

00:37:03.803 --> 00:37:05.128 extracellular space these days.  
NOTE Confidence: 0.938576255

00:37:08.130 --> 00:37:11.104 I think the reason why we've wrangled and  
NOTE Confidence: 0.938576255

00:37:11.104 --> 00:37:13.880 learned so much from the immune system is  
NOTE Confidence: 0.938576255

00:37:13.953 --> 00:37:16.529 being able to be so systematic about it.  
NOTE Confidence: 0.938576255

00:37:16.530 --> 00:37:20.498 And so I've tried to begin to wrangle the  
NOTE Confidence: 0.938576255

00:37:20.498 --> 00:37:22.610 extracellular space into the same way,  
NOTE Confidence: 0.938576255

00:37:22.610 --> 00:37:24.082 to establish some foundations  
NOTE Confidence: 0.938576255

00:37:24.082 --> 00:37:25.922 to make a consistent Atlas,

NOTE Confidence: 0.938576255

00:37:25.930 --> 00:37:28.856 to then begin to study the specific

NOTE Confidence: 0.938576255

00:37:28.856 --> 00:37:30.730 markers related to tumors,

NOTE Confidence: 0.938576255

00:37:30.730 --> 00:37:32.230 relating them to phenotypes and

NOTE Confidence: 0.938576255

00:37:32.230 --> 00:37:33.730 all of these other things.

NOTE Confidence: 0.938576255

00:37:33.730 --> 00:37:37.446 So the survey that I sent you guys

NOTE Confidence: 0.938576255

00:37:37.450 --> 00:37:40.948 and thank you for those who who went

NOTE Confidence: 0.938576255

00:37:40.948 --> 00:37:44.060 slog through it to to to humor me.

NOTE Confidence: 0.938576255

00:37:44.060 --> 00:37:45.020 The the bottom line is,

NOTE Confidence: 0.938576255

00:37:45.020 --> 00:37:47.660 do we need an extracellular ontology?

NOTE Confidence: 0.938576255

00:37:47.660 --> 00:37:50.340 We have a cellular ontology.

NOTE Confidence: 0.938576255

00:37:50.340 --> 00:37:52.538 But when you take a liquid biopsy,

NOTE Confidence: 0.938576255

00:37:52.540 --> 00:37:54.004 you have no idea.

NOTE Confidence: 0.938576255

00:37:54.004 --> 00:37:55.834 There's nothing that's a single

NOTE Confidence: 0.938576255

00:37:55.834 --> 00:37:58.350 marker that can tell you that a

NOTE Confidence: 0.938576255

00:37:58.350 --> 00:38:01.260 vesicle came from an exosomal pathway.

NOTE Confidence: 0.938576255

00:38:01.260 --> 00:38:01.858 In fact,  
NOTE Confidence: 0.938576255

00:38:01.858 --> 00:38:03.353 the biologists are really kind  
NOTE Confidence: 0.938576255

00:38:03.353 --> 00:38:05.323 of working out all the specifics  
NOTE Confidence: 0.938576255

00:38:05.323 --> 00:38:07.018 of the exosomal pathway anyway.  
NOTE Confidence: 0.938576255

00:38:07.020 --> 00:38:09.204 So then you try to frame the  
NOTE Confidence: 0.938576255

00:38:09.204 --> 00:38:10.759 ontology of the extracellular  
NOTE Confidence: 0.938576255

00:38:10.759 --> 00:38:13.479 space in the related ontologies.  
NOTE Confidence: 0.938576255

00:38:13.480 --> 00:38:14.712 So I just mentioned,  
NOTE Confidence: 0.938576255

00:38:14.712 --> 00:38:16.880 do we need an extra cellular one?  
NOTE Confidence: 0.938576255

00:38:16.880 --> 00:38:18.040 There's a cellular one,  
NOTE Confidence: 0.93270605

00:38:20.560 --> 00:38:21.960 I don't know, you guys can tell me,  
NOTE Confidence: 0.93270605

00:38:21.960 --> 00:38:25.080 but I have asked my liquid biopsy colleagues,  
NOTE Confidence: 0.93270605

00:38:25.080 --> 00:38:28.470 is it probably pretty true that  
NOTE Confidence: 0.93270605

00:38:28.470 --> 00:38:31.340 you categorize the things that  
NOTE Confidence: 0.93270605

00:38:31.340 --> 00:38:33.720 you're using for biomarkers,  
NOTE Confidence: 0.93270605

00:38:33.720 --> 00:38:36.912 classify them really by what it is

NOTE Confidence: 0.93270605

00:38:36.912 --> 00:38:38.760 that you isolated or how you isolated?

NOTE Confidence: 0.93270605

00:38:38.760 --> 00:38:40.360 I say yeah, okay so.

NOTE Confidence: 0.818847855714286

00:38:42.710 --> 00:38:44.926 And the nano material

NOTE Confidence: 0.818847855714286

00:38:44.926 --> 00:38:47.123 field is super detailed.

NOTE Confidence: 0.818847855714286

00:38:47.123 --> 00:38:50.740 They have a Nano Nano Particle Ontology,

NOTE Confidence: 0.818847855714286

00:38:50.740 --> 00:38:53.590 the NPO, that's all about formulation,

NOTE Confidence: 0.818847855714286

00:38:53.590 --> 00:38:55.630 this is the shell, this is the surface,

NOTE Confidence: 0.818847855714286

00:38:55.630 --> 00:38:59.310 this is the, it's extensive.

NOTE Confidence: 0.818847855714286

00:38:59.310 --> 00:39:01.266 So how do we just approach

NOTE Confidence: 0.818847855714286

00:39:01.266 --> 00:39:03.230 the mess that's in between?

NOTE Confidence: 0.818847855714286

00:39:03.230 --> 00:39:08.014 So hence the survey and I didn't ask

NOTE Confidence: 0.818847855714286

00:39:08.014 --> 00:39:10.447 the question I wanted to ask because

NOTE Confidence: 0.818847855714286

00:39:10.447 --> 00:39:12.865 it was so strongly objected to.

NOTE Confidence: 0.818847855714286

00:39:12.870 --> 00:39:14.788 My first question was going to be

NOTE Confidence: 0.818847855714286

00:39:14.790 --> 00:39:17.184 what do you think an exozone is?

NOTE Confidence: 0.818847855714286

00:39:17.190 --> 00:39:19.875 A BCD? But people decided  
NOTE Confidence: 0.818847855714286

00:39:19.875 --> 00:39:22.023 that was too controversial,  
NOTE Confidence: 0.818847855714286

00:39:22.030 --> 00:39:27.706 so instead we asked more obliquely,  
NOTE Confidence: 0.818847855714286

00:39:27.710 --> 00:39:30.630 maybe obtusely.  
NOTE Confidence: 0.818847855714286

00:39:30.630 --> 00:39:33.374 This is a selection of ways to  
NOTE Confidence: 0.818847855714286

00:39:33.374 --> 00:39:34.550 classify extracellular vesicles.  
NOTE Confidence: 0.818847855714286

00:39:34.550 --> 00:39:37.678 Which one do you think is most central  
NOTE Confidence: 0.818847855714286

00:39:37.678 --> 00:39:41.310 to harmonizing with later system?  
NOTE Confidence: 0.818847855714286

00:39:41.310 --> 00:39:42.372 4 vesicles,  
NOTE Confidence: 0.818847855714286

00:39:42.372 --> 00:39:45.027 the largest proportion that the  
NOTE Confidence: 0.818847855714286

00:39:45.027 --> 00:39:48.929 highest answer is based on biological  
NOTE Confidence: 0.818847855714286

00:39:48.929 --> 00:39:50.586 considerations like Biogenesis.  
NOTE Confidence: 0.818847855714286

00:39:50.586 --> 00:39:54.250 And so I think that message of the  
NOTE Confidence: 0.818847855714286

00:39:54.345 --> 00:39:57.095 EV community of what distinguishes  
NOTE Confidence: 0.818847855714286

00:39:57.095 --> 00:40:00.578 A vesicle from a non vesicle and  
NOTE Confidence: 0.818847855714286

00:40:00.578 --> 00:40:02.612 an exosome and microparticles

NOTE Confidence: 0.818847855714286  
00:40:02.612 --> 00:40:05.922 or other things it's getting  
NOTE Confidence: 0.818847855714286  
00:40:05.922 --> 00:40:08.690 through in response to the non  
NOTE Confidence: 0.818847855714286  
00:40:08.690 --> 00:40:09.950 vesicular extracellular particles.  
NOTE Confidence: 0.863050828571428  
00:40:11.990 --> 00:40:16.344 Even the EV people, the ISAF people,  
NOTE Confidence: 0.863050828571428  
00:40:16.350 --> 00:40:18.670 say we don't know what the Biogenesis is.  
NOTE Confidence: 0.863050828571428  
00:40:18.670 --> 00:40:21.415 For the most part, the top answer is based  
NOTE Confidence: 0.863050828571428  
00:40:21.415 --> 00:40:24.023 on biochemical considerations, composition.  
NOTE Confidence: 0.863050828571428  
00:40:24.023 --> 00:40:26.428 Is it a lipid bilayer? What's in it?  
NOTE Confidence: 0.938911752666667  
00:40:29.230 --> 00:40:30.700 Informally? And I don't know if this  
NOTE Confidence: 0.938911752666667  
00:40:30.700 --> 00:40:32.389 is ever going to get published or not,  
NOTE Confidence: 0.938911752666667  
00:40:32.390 --> 00:40:34.982 but we did a we did a beta test.  
NOTE Confidence: 0.938911752666667  
00:40:34.990 --> 00:40:37.192 I used my friends and colleagues  
NOTE Confidence: 0.938911752666667  
00:40:37.192 --> 00:40:39.110 at NIH as Guinea pigs.  
NOTE Confidence: 0.938911752666667  
00:40:39.110 --> 00:40:41.546 We have a couple of listservs for  
NOTE Confidence: 0.938911752666667  
00:40:41.546 --> 00:40:43.861 the liquid biopsy group and the EV  
NOTE Confidence: 0.938911752666667

00:40:43.861 --> 00:40:46.349 interest group and we sent it to them  
NOTE Confidence: 0.938911752666667

00:40:46.350 --> 00:40:49.590 and it was even more extreme when we  
NOTE Confidence: 0.938911752666667

00:40:49.590 --> 00:40:52.590 focused on the liquid biopsy groups.  
NOTE Confidence: 0.938911752666667

00:40:52.590 --> 00:40:54.228 It's about composition, what is it,  
NOTE Confidence: 0.938911752666667

00:40:54.230 --> 00:40:57.170 what we're looking at and the  
NOTE Confidence: 0.938911752666667

00:40:57.170 --> 00:40:59.446 EV folks about everything,  
NOTE Confidence: 0.938911752666667

00:40:59.446 --> 00:41:03.106 not just vesicles and when asked about  
NOTE Confidence: 0.938911752666667

00:41:03.106 --> 00:41:05.251 everything without dividing into vesicles  
NOTE Confidence: 0.938911752666667

00:41:05.251 --> 00:41:08.258 or non vehicular extracellular particles.  
NOTE Confidence: 0.938911752666667

00:41:08.260 --> 00:41:11.697 The EV group still focused on Biogenesis,  
NOTE Confidence: 0.938911752666667

00:41:11.700 --> 00:41:15.060 so I'm working on the analysis of who  
NOTE Confidence: 0.938911752666667

00:41:15.060 --> 00:41:18.378 answered what and it should be interesting.  
NOTE Confidence: 0.938911752666667

00:41:18.380 --> 00:41:21.690 But I've been at meetings where  
NOTE Confidence: 0.938911752666667

00:41:21.690 --> 00:41:23.459 people stand up and they ask me  
NOTE Confidence: 0.938911752666667

00:41:23.460 --> 00:41:25.700 why do you care what it's called,  
NOTE Confidence: 0.938911752666667

00:41:25.700 --> 00:41:27.420 if it's a good biomarker?



NOTE Confidence: 0.938911752666667  
00:41:27.420 --> 00:41:27.944 And honestly,  
NOTE Confidence: 0.938911752666667  
00:41:27.944 --> 00:41:29.778 if the biomarkers is a good biomarker,  
NOTE Confidence: 0.938911752666667  
00:41:29.780 --> 00:41:30.530 that's great.  
NOTE Confidence: 0.938911752666667  
00:41:30.530 --> 00:41:33.155 It's just if you want to stitch  
NOTE Confidence: 0.938911752666667  
00:41:33.155 --> 00:41:35.569 the data together and understand  
NOTE Confidence: 0.938911752666667  
00:41:35.569 --> 00:41:39.246 how our data relates to each other.  
NOTE Confidence: 0.938911752666667  
00:41:39.250 --> 00:41:41.740 Everybody who does omics and assays  
NOTE Confidence: 0.938911752666667  
00:41:41.740 --> 00:41:44.491 and atlases knows that there has to  
NOTE Confidence: 0.938911752666667  
00:41:44.491 --> 00:41:46.850 be a common framework it's set on.  
NOTE Confidence: 0.938911752666667  
00:41:46.850 --> 00:41:48.730 So I just want to.  
NOTE Confidence: 0.938911752666667  
00:41:48.730 --> 00:41:49.484 In addition,  
NOTE Confidence: 0.938911752666667  
00:41:49.484 --> 00:41:51.820 I really have to thank you all  
NOTE Confidence: 0.938911752666667  
00:41:51.820 --> 00:41:53.170 for inviting me to come speak.  
NOTE Confidence: 0.938911752666667  
00:41:53.170 --> 00:41:55.290 It's really an honor for me as a  
NOTE Confidence: 0.938911752666667  
00:41:55.290 --> 00:41:57.810 young scientist to speak to you guys  
NOTE Confidence: 0.938911752666667

00:41:57.810 --> 00:42:01.329 learn from you, get your feedback.  
NOTE Confidence: 0.938911752666667

00:42:01.330 --> 00:42:03.976 I also need to thank the laboratory  
NOTE Confidence: 0.938911752666667

00:42:03.976 --> 00:42:07.910 pathology kind of that be my mentors.  
NOTE Confidence: 0.938911752666667

00:42:07.910 --> 00:42:10.278 Past, present and current.  
NOTE Confidence: 0.938911752666667

00:42:10.278 --> 00:42:14.265 As you know I was thinking last night  
NOTE Confidence: 0.938911752666667

00:42:14.270 --> 00:42:15.908 I couldn't say this takes a village.  
NOTE Confidence: 0.938911752666667

00:42:15.910 --> 00:42:19.389 This actually takes like lots of villages.  
NOTE Confidence: 0.938911752666667

00:42:19.390 --> 00:42:23.910 So these are some of the villages  
NOTE Confidence: 0.938911752666667

00:42:23.910 --> 00:42:26.168 who have and they're continuing to  
NOTE Confidence: 0.938911752666667

00:42:26.168 --> 00:42:28.454 help me and I'll take questions.  
NOTE Confidence: 0.938911752666667

00:42:28.454 --> 00:42:31.713 But as a sneak peek I had bought  
NOTE Confidence: 0.938911752666667

00:42:31.713 --> 00:42:34.359 on behalf of our residency program  
NOTE Confidence: 0.938911752666667

00:42:34.359 --> 00:42:36.617 director some slides about the.  
NOTE Confidence: 0.938911752666667

00:42:36.620 --> 00:42:41.068 Residency program at at NIH If there  
NOTE Confidence: 0.938911752666667

00:42:41.068 --> 00:42:43.252 are folks who are interested in it  
NOTE Confidence: 0.938911752666667

00:42:43.252 --> 00:42:46.722 at lunch and I'll just e-mail it to

NOTE Confidence: 0.938911752666667  
00:42:46.722 --> 00:42:48.620 anybody who's interested, thank you.  
NOTE Confidence: 0.937378342857143  
00:42:59.370 --> 00:43:01.058 Should I open the chat and see if  
NOTE Confidence: 0.937378342857143  
00:43:01.058 --> 00:43:02.810 there are questions in the chat? Okay  
NOTE Confidence: 0.873622808  
00:43:10.850 --> 00:43:13.540 act stating for CME credit.  
NOTE Confidence: 0.873622808  
00:43:13.540 --> 00:43:16.564 Texting for CME credit, so I don't  
NOTE Confidence: 0.873622808  
00:43:16.564 --> 00:43:19.060 think those are questions. Yeah,  
NOTE Confidence: 0.8570157625  
00:43:21.340 --> 00:43:24.940 refer to analyze the EV in  
NOTE Confidence: 0.8570157625  
00:43:24.940 --> 00:43:28.739 the context of that area.  
NOTE Confidence: 0.8570157625  
00:43:28.740 --> 00:43:33.380 Yeah, there's a whole group of ISA which  
NOTE Confidence: 0.8570157625  
00:43:33.380 --> 00:43:37.860 is interested in not only the Ev's,  
NOTE Confidence: 0.8570157625  
00:43:37.860 --> 00:43:41.374 the host Ev's, but also the Ev's.  
NOTE Confidence: 0.8570157625  
00:43:41.380 --> 00:43:44.530 Of you know, across the microbiome  
NOTE Confidence: 0.8570157625  
00:43:44.530 --> 00:43:47.544 or infections, that's become a very  
NOTE Confidence: 0.8570157625  
00:43:47.544 --> 00:43:49.812 interesting part of COVID work.  
NOTE Confidence: 0.8570157625  
00:43:49.812 --> 00:43:52.980 Kendall's done some work on that at Tgen.  
NOTE Confidence: 0.8570157625

00:43:52.980 --> 00:43:54.855 Several people have have done  
NOTE Confidence: 0.8570157625

00:43:54.855 --> 00:43:57.900 a lot of work on that. Yeah.  
NOTE Confidence: 0.8570157625

00:43:57.900 --> 00:44:00.805 In that context how do you  
NOTE Confidence: 0.8570157625

00:44:00.805 --> 00:44:02.220 differentiate the post PR?  
NOTE Confidence: 0.937378342857143

00:44:05.220 --> 00:44:07.875 Yeah, so it depends on your assay, right.  
NOTE Confidence: 0.937378342857143

00:44:07.875 --> 00:44:11.565 So if you. Have species specific  
NOTE Confidence: 0.937378342857143

00:44:11.570 --> 00:44:13.616 antibody clones that can begin to  
NOTE Confidence: 0.937378342857143

00:44:13.616 --> 00:44:15.634 differentiate some of it and that's  
NOTE Confidence: 0.937378342857143

00:44:15.634 --> 00:44:17.810 been applied in some model systems.  
NOTE Confidence: 0.9452853

00:44:20.410 --> 00:44:22.618 I don't know if it's been  
NOTE Confidence: 0.9452853

00:44:22.618 --> 00:44:24.090 applied in clinical settings.  
NOTE Confidence: 0.938995675

00:44:27.250 --> 00:44:30.750 And then in terms of the informatics  
NOTE Confidence: 0.938995675

00:44:30.750 --> 00:44:32.390 for you know like RNA analysis it  
NOTE Confidence: 0.938995675

00:44:32.390 --> 00:44:34.050 would it would be based on the genomes.  
NOTE Confidence: 0.95836772

00:44:36.980 --> 00:44:39.860 There's certainly overlap where you can't  
NOTE Confidence: 0.95836772

00:44:39.860 --> 00:44:43.192 discriminate I would imagine my final

NOTE Confidence: 0.95836772

00:44:43.192 --> 00:44:48.820 question on the basis of the buy markers,

NOTE Confidence: 0.95836772

00:44:48.820 --> 00:44:52.220 the efforts pull down subset,

NOTE Confidence: 0.95836772

00:44:52.220 --> 00:44:55.740 yeah that's what we're doing and

NOTE Confidence: 0.95836772

00:44:55.740 --> 00:44:58.440 that's why we had we've had such an

NOTE Confidence: 0.95836772

00:44:58.440 --> 00:45:00.630 extensive focus on which markers to use.

NOTE Confidence: 0.95836772

00:45:00.630 --> 00:45:02.830 And then once we do the pull downs,

NOTE Confidence: 0.95836772

00:45:02.830 --> 00:45:05.465 how do you make that work robustly for

NOTE Confidence: 0.95836772

00:45:05.465 --> 00:45:08.305 the very small amount that you pull down?

NOTE Confidence: 0.95836772

00:45:08.310 --> 00:45:11.019 So one thing that struck me and

NOTE Confidence: 0.95836772

00:45:11.019 --> 00:45:13.590 I think anybody who's interested

NOTE Confidence: 0.95836772

00:45:13.590 --> 00:45:16.590 in doing liquid biopsies of EV's

NOTE Confidence: 0.95836772

00:45:16.590 --> 00:45:18.920 should probably understand this in

NOTE Confidence: 0.95836772

00:45:18.920 --> 00:45:21.446 a milliliter of blood, you know,

NOTE Confidence: 0.95836772

00:45:21.446 --> 00:45:24.230 you might have 3 circulating tumor cells,

NOTE Confidence: 0.95836772

00:45:24.230 --> 00:45:26.030 10 circulating tumor cells.

NOTE Confidence: 0.95836772

00:45:26.030 --> 00:45:27.830 There's something on the order  
NOTE Confidence: 0.95836772

00:45:27.830 --> 00:45:29.635 of about a billion EV's.  
NOTE Confidence: 0.95836772

00:45:29.635 --> 00:45:32.190 And there's something on the order of  
NOTE Confidence: 0.938995675

00:45:35.030 --> 00:45:38.201 10 to the 16th versus 10 to the 18th,  
NOTE Confidence: 0.938995675

00:45:38.201 --> 00:45:42.107 like a billion billion lipoprotein particles.  
NOTE Confidence: 0.938995675

00:45:42.110 --> 00:45:44.590 So, So those since they're so close and  
NOTE Confidence: 0.938995675

00:45:44.590 --> 00:45:46.589 overlapping in size with the vesicles,  
NOTE Confidence: 0.938995675

00:45:46.590 --> 00:45:49.830 those become the main complicator.  
NOTE Confidence: 0.919367286666667

00:45:51.870 --> 00:45:54.982 And it's what I like about the affinity  
NOTE Confidence: 0.919367286666667

00:45:54.982 --> 00:45:57.507 pull down part is that you can.  
NOTE Confidence: 0.968431054

00:45:59.810 --> 00:46:02.090 Directly interrogate A membrane receptor,  
NOTE Confidence: 0.968431054

00:46:02.090 --> 00:46:03.143 another membrane receptor,  
NOTE Confidence: 0.968431054

00:46:03.143 --> 00:46:05.249 and know that you're dealing with  
NOTE Confidence: 0.968431054

00:46:05.249 --> 00:46:06.841 something that is likely something that  
NOTE Confidence: 0.968431054

00:46:06.841 --> 00:46:09.038 has a little bit by later because it  
NOTE Confidence: 0.968431054

00:46:09.038 --> 00:46:10.970 has a Tetra span and then thanks, yeah,

NOTE Confidence: 0.95367142

00:46:16.610 --> 00:46:17.408 the best questions.

NOTE Confidence: 0.884472171428571

00:46:46.570 --> 00:46:49.930 Yeah. So, So, yes, yes and yes.

NOTE Confidence: 0.884472171428571

00:46:49.930 --> 00:46:51.574 So the the question for folks

NOTE Confidence: 0.884472171428571

00:46:51.574 --> 00:46:53.288 online who maybe didn't hear it was

NOTE Confidence: 0.9469625333333333

00:46:55.850 --> 00:46:59.426 are there. Impacts of the cellular,

NOTE Confidence: 0.9469625333333333

00:46:59.430 --> 00:47:02.346 the state of the cell in terms of its

NOTE Confidence: 0.9469625333333333

00:47:02.346 --> 00:47:04.359 metabolism or other stressors that

NOTE Confidence: 0.9469625333333333

00:47:04.359 --> 00:47:06.789 affect the type of vesicles produced

NOTE Confidence: 0.9469625333333333

00:47:06.790 --> 00:47:10.022 And are there impacts also on the ways

NOTE Confidence: 0.9469625333333333

00:47:10.022 --> 00:47:15.149 that cells receive vesicles. So you know

NOTE Confidence: 0.950317

00:47:17.350 --> 00:47:20.584 2004 Arnie Levine showed that P53 was

NOTE Confidence: 0.950317

00:47:20.584 --> 00:47:22.869 central regulator of producing exosome.

NOTE Confidence: 0.950317

00:47:22.870 --> 00:47:26.405 So there's. From way back there's there's

NOTE Confidence: 0.950317

00:47:26.405 --> 00:47:28.600 been an understanding that genotoxic stress

NOTE Confidence: 0.950317

00:47:28.600 --> 00:47:30.798 hence my interest as a radiation oncologist.

NOTE Confidence: 0.950317

00:47:30.800 --> 00:47:38.160 Radiation kicks off a surge of these and  
NOTE Confidence: 0.950317

00:47:38.160 --> 00:47:43.448 no so you can give a sublethal dose and  
NOTE Confidence: 0.950317

00:47:43.448 --> 00:47:46.532 it it stimulates the exozone pathway.  
NOTE Confidence: 0.950317

00:47:46.532 --> 00:47:50.615 So so this is there's clearly a.  
NOTE Confidence: 0.950317

00:47:50.615 --> 00:47:53.035 Very wide heterogeneous range  
NOTE Confidence: 0.950317

00:47:53.035 --> 00:47:55.860 of types of vesicles.  
NOTE Confidence: 0.950317

00:47:55.860 --> 00:47:56.664 There are these exosomes,  
NOTE Confidence: 0.950317

00:47:56.664 --> 00:47:58.302 they're small ones made in the vesicles or  
NOTE Confidence: 0.950317

00:47:58.302 --> 00:47:59.854 other types that are shut off the surface.  
NOTE Confidence: 0.950317

00:47:59.860 --> 00:48:00.724 There are the,  
NOTE Confidence: 0.950317

00:48:00.724 --> 00:48:02.740 I guess you could call them apoptosomes,  
NOTE Confidence: 0.950317

00:48:02.740 --> 00:48:04.708 the ones that are shut in  
NOTE Confidence: 0.950317

00:48:04.708 --> 00:48:06.020 the context of apoptosis.  
NOTE Confidence: 0.94276945

00:48:08.100 --> 00:48:10.795 I think we are only scratching the  
NOTE Confidence: 0.94276945

00:48:10.795 --> 00:48:13.420 surface of those different types the  
NOTE Confidence: 0.956606625

00:48:15.460 --> 00:48:18.580 in the 80s or 90s they originally described.



NOTE Confidence: 0.956606625

00:48:18.580 --> 00:48:20.905 These little vesicles and microscopy

NOTE Confidence: 0.956606625

00:48:20.905 --> 00:48:23.113 is platelet dust where they just

NOTE Confidence: 0.956606625

00:48:23.113 --> 00:48:24.739 kind of kick out the garbage.

NOTE Confidence: 0.956606625

00:48:24.740 --> 00:48:27.340 So there was first an idea that these

NOTE Confidence: 0.956606625

00:48:27.340 --> 00:48:30.099 are garbage bags and there was this idea

NOTE Confidence: 0.956606625

00:48:30.099 --> 00:48:31.774 that they're sophisticated endocrine

NOTE Confidence: 0.956606625

00:48:31.774 --> 00:48:34.259 systems of communicating between cells.

NOTE Confidence: 0.956606625

00:48:34.260 --> 00:48:36.409 I think it's both and and a

NOTE Confidence: 0.956606625

00:48:36.409 --> 00:48:38.260 lot of stuff in between.

NOTE Confidence: 0.956606625

00:48:38.260 --> 00:48:40.836 So for me, I'm going to be

NOTE Confidence: 0.956606625

00:48:40.836 --> 00:48:43.194 looking for different types of

NOTE Confidence: 0.956606625

00:48:43.194 --> 00:48:45.884 vesicles with different types of.

NOTE Confidence: 0.956606625

00:48:45.890 --> 00:48:47.570 Aberrant DNA damage,

NOTE Confidence: 0.942266198

00:48:50.490 --> 00:48:53.370 Addux and other things.

NOTE Confidence: 0.942266198

00:48:53.370 --> 00:48:57.850 So yes genotoxic stress increases

NOTE Confidence: 0.942266198

00:48:57.850 --> 00:49:00.970 exosome production per se.  
NOTE Confidence: 0.942266198

00:49:00.970 --> 00:49:03.650 Also probably stress and loving  
NOTE Confidence: 0.942266198

00:49:03.650 --> 00:49:09.702 There also is starving cells to this  
NOTE Confidence: 0.942266198

00:49:09.702 --> 00:49:12.478 is really kind of related to some  
NOTE Confidence: 0.942266198

00:49:12.478 --> 00:49:14.134 work that Raghu Glory has talked  
NOTE Confidence: 0.942266198

00:49:14.134 --> 00:49:16.019 a lot about which is that the.  
NOTE Confidence: 0.942266198

00:49:16.020 --> 00:49:17.272 Pancreatic cells,  
NOTE Confidence: 0.942266198

00:49:17.272 --> 00:49:19.776 which are essentially just  
NOTE Confidence: 0.942266198

00:49:19.776 --> 00:49:21.654 ravenous for resources,  
NOTE Confidence: 0.942266198

00:49:21.660 --> 00:49:23.720 take up these therapeutic  
NOTE Confidence: 0.942266198

00:49:23.720 --> 00:49:25.780 vesicles that he produces.  
NOTE Confidence: 0.942266198

00:49:25.780 --> 00:49:27.772 And he thinks that that's because  
NOTE Confidence: 0.942266198

00:49:27.772 --> 00:49:29.580 of their metabolic state and  
NOTE Confidence: 0.942266198

00:49:29.580 --> 00:49:31.260 receptor affinity for vesicles  
NOTE Confidence: 0.942266198

00:49:31.260 --> 00:49:32.940 compared to surrounding tissue,  
NOTE Confidence: 0.942266198

00:49:32.940 --> 00:49:34.944 which doesn't seem to pick up

NOTE Confidence: 0.942266198

00:49:34.944 --> 00:49:36.620 those therapeutic vesicles as well.

NOTE Confidence: 0.942266198

00:49:36.620 --> 00:49:41.500 But any type of vesicle that you look at,

NOTE Confidence: 0.942266198

00:49:41.500 --> 00:49:44.518 you can find.

NOTE Confidence: 0.942266198

00:49:44.520 --> 00:49:46.680 Yin and Yang and a lot of these things.

NOTE Confidence: 0.942266198

00:49:46.680 --> 00:49:52.550 So there are the E V's or exosomes that

NOTE Confidence: 0.942266198

00:49:52.550 --> 00:49:55.192 cause essentially vaccinating effects,

NOTE Confidence: 0.942266198

00:49:55.192 --> 00:49:56.520 tumor stimulation.

NOTE Confidence: 0.942266198

00:49:56.520 --> 00:49:59.430 There are other vesicles which are

NOTE Confidence: 0.942266198

00:49:59.430 --> 00:50:02.675 clearly inhibitory that promote a more

NOTE Confidence: 0.942266198

00:50:02.675 --> 00:50:04.879 mildly suppressor type phenotype,

NOTE Confidence: 0.942266198

00:50:04.880 --> 00:50:07.040 which are which which do what?

NOTE Confidence: 0.942266198

00:50:07.040 --> 00:50:09.180 Until we systematically start

NOTE Confidence: 0.942266198

00:50:09.180 --> 00:50:11.320 breaking the groups apart,

NOTE Confidence: 0.942266198

00:50:11.320 --> 00:50:12.340 there are a lot of mysteries

NOTE Confidence: 0.942266198

00:50:12.340 --> 00:50:13.360 that are hard to unravel.

NOTE Confidence: 0.80877167

00:50:22.650 --> 00:50:22.890 Yeah.  
NOTE Confidence: 0.9452853

00:50:25.050 --> 00:50:28.050 So that's a really good question.  
NOTE Confidence: 0.9452853

00:50:28.050 --> 00:50:29.478 As with all of it, part of  
NOTE Confidence: 0.9452853

00:50:29.478 --> 00:50:30.448 the answer is it depends.  
NOTE Confidence: 0.9201268

00:50:33.850 --> 00:50:37.306 So if you make synthetic  
NOTE Confidence: 0.9201268

00:50:37.306 --> 00:50:38.726 ones and you inject them,  
NOTE Confidence: 0.9201268

00:50:38.730 --> 00:50:42.048 it depends on how you made them.  
NOTE Confidence: 0.9201268

00:50:42.050 --> 00:50:43.205 They could just.  
NOTE Confidence: 0.9201268

00:50:43.205 --> 00:50:45.515 Go first pass and get largely  
NOTE Confidence: 0.9201268

00:50:45.515 --> 00:50:47.822 taken up in the spleen or the  
NOTE Confidence: 0.9201268

00:50:47.822 --> 00:50:49.230 liver and they may make only  
NOTE Confidence: 0.9201268

00:50:49.230 --> 00:50:50.330 kind of one round through,  
NOTE Confidence: 0.9201268

00:50:50.330 --> 00:50:51.710 so it might really matter  
NOTE Confidence: 0.9201268

00:50:51.710 --> 00:50:53.090 which way you inject them.  
NOTE Confidence: 0.950316921

00:50:55.930 --> 00:50:58.138 In other cases where you've made  
NOTE Confidence: 0.950316921

00:50:58.138 --> 00:50:59.610 them under other conditions,

NOTE Confidence: 0.950316921

00:50:59.610 --> 00:51:01.330 they circle around quite

NOTE Confidence: 0.950316921

00:51:01.330 --> 00:51:03.050 a bit longer beforehand.

NOTE Confidence: 0.950316921

00:51:03.050 --> 00:51:06.560 I I think the common understanding

NOTE Confidence: 0.950316921

00:51:06.560 --> 00:51:09.101 is that probably the turnover

NOTE Confidence: 0.950316921

00:51:09.101 --> 00:51:11.843 overall is something like 6 hours.

NOTE Confidence: 0.950316921

00:51:11.850 --> 00:51:16.010 But it's relatively rapid.

NOTE Confidence: 0.950316921

00:51:16.010 --> 00:51:19.210 But for me as a radiation oncologist,

NOTE Confidence: 0.950316921

00:51:19.210 --> 00:51:21.782 I don't feel like I need to run in

NOTE Confidence: 0.950316921

00:51:21.782 --> 00:51:24.834 and get a sample in the first hour.

NOTE Confidence: 0.950316921

00:51:24.834 --> 00:51:27.208 There are a lot of things that happen

NOTE Confidence: 0.950316921

00:51:27.210 --> 00:51:29.688 2448 hours later that take that long

NOTE Confidence: 0.950316921

00:51:29.690 --> 00:51:33.866 to start to manifest and be able to be

NOTE Confidence: 0.950316921

00:51:33.866 --> 00:51:36.280 discernible even if you did seamless

NOTE Confidence: 0.950316921

00:51:36.280 --> 00:51:38.410 offstaining in the affected tissue.

NOTE Confidence: 0.93522182

00:51:42.790 --> 00:51:44.308 It seems to be quite rapid,

NOTE Confidence: 0.9335445833333333

00:52:00.910 --> 00:52:04.130 so I think there's a myth that  
NOTE Confidence: 0.9335445833333333

00:52:04.130 --> 00:52:07.230 every vesicle a cell relieves,  
NOTE Confidence: 0.9335445833333333

00:52:07.230 --> 00:52:09.490 shoots out and heads straight  
NOTE Confidence: 0.9335445833333333

00:52:09.490 --> 00:52:11.298 for the bloodstream and.  
NOTE Confidence: 0.9335445833333333

00:52:11.300 --> 00:52:14.580 Circulates and then whatever  
NOTE Confidence: 0.9335445833333333

00:52:14.580 --> 00:52:16.380 the kidneys, clear some.  
NOTE Confidence: 0.9335445833333333

00:52:16.380 --> 00:52:20.252 There lots of urine studies which look at  
NOTE Confidence: 0.9335445833333333

00:52:20.252 --> 00:52:22.438 vascular populations deliver clear some.  
NOTE Confidence: 0.9335445833333333

00:52:22.438 --> 00:52:25.000 I suspect that the clearance is dependent  
NOTE Confidence: 0.9335445833333333

00:52:25.069 --> 00:52:27.631 on the surface markers like selectins  
NOTE Confidence: 0.9335445833333333

00:52:27.631 --> 00:52:29.339 and organ specific distributions,  
NOTE Confidence: 0.9301903

00:52:31.580 --> 00:52:31.980 but  
NOTE Confidence: 0.93270617

00:52:34.180 --> 00:52:36.180 I wish I knew who first said this,  
NOTE Confidence: 0.93270617

00:52:36.180 --> 00:52:37.860 but I've heard it said that.  
NOTE Confidence: 0.93773775

00:52:40.600 --> 00:52:45.040 The blood is sort of our ocean within.  
NOTE Confidence: 0.93773775

00:52:45.040 --> 00:52:49.840 So in the context of organisms evolving

NOTE Confidence: 0.93773775

00:52:49.840 --> 00:52:54.320 through mammals and vertebrates to have a

NOTE Confidence: 0.93773775

00:52:54.320 --> 00:52:58.020 circulating system that those circulating

NOTE Confidence: 0.93773775

00:52:58.020 --> 00:53:01.860 systems strikingly reflect the oceans and

NOTE Confidence: 0.93773775

00:53:01.860 --> 00:53:03.920 those salinity conditions. Other things

NOTE Confidence: 0.92111244

00:53:06.680 --> 00:53:10.510 there's a researcher at MIT.

NOTE Confidence: 0.92111244

00:53:10.510 --> 00:53:13.642 Who? Sally Chisholm,

NOTE Confidence: 0.92111244

00:53:13.642 --> 00:53:17.038 who discovered when she was a

NOTE Confidence: 0.92111244

00:53:17.038 --> 00:53:18.990 postdoc Prochlorococcus bacteria,

NOTE Confidence: 0.92111244

00:53:18.990 --> 00:53:21.610 which are responsible for some

NOTE Confidence: 0.92111244

00:53:21.610 --> 00:53:24.230 ridiculous amount of the world's

NOTE Confidence: 0.92111244

00:53:24.230 --> 00:53:25.870 CO2 metabolism in the oceans.

NOTE Confidence: 0.92111244

00:53:25.870 --> 00:53:28.020 Like when you fly over in some areas are kind

NOTE Confidence: 0.92111244

00:53:28.070 --> 00:53:30.023 of green and some areas are kind of blue.

NOTE Confidence: 0.92111244

00:53:30.030 --> 00:53:32.430 It's different. Prochloroccus.

NOTE Confidence: 0.92111244

00:53:32.430 --> 00:53:37.250 They shed vesicles and there's a

NOTE Confidence: 0.92111244

00:53:37.250 --> 00:53:39.590 thought that's part of how they.  
NOTE Confidence: 0.92111244

00:53:39.590 --> 00:53:41.650 Communicate and cross regulate.  
NOTE Confidence: 0.92111244

00:53:41.650 --> 00:53:45.550 But I think in our compact systems,  
NOTE Confidence: 0.92111244

00:53:45.550 --> 00:53:48.304 there's probably a great deal of  
NOTE Confidence: 0.92111244

00:53:48.304 --> 00:53:50.822 vesicle release that impacts the  
NOTE Confidence: 0.92111244

00:53:50.822 --> 00:53:53.052 local tumor microenvironment and  
NOTE Confidence: 0.92111244

00:53:53.052 --> 00:53:55.584 is not necessarily part of what  
NOTE Confidence: 0.92111244

00:53:55.590 --> 00:53:58.841 processes out and which which  
NOTE Confidence: 0.92111244

00:53:58.841 --> 00:54:00.863 stay and which get processed out.  
NOTE Confidence: 0.92111244

00:54:00.870 --> 00:54:04.263 We talked a little bit about it at dinner.  
NOTE Confidence: 0.92111244

00:54:04.270 --> 00:54:06.394 We just we have to find better ways of  
NOTE Confidence: 0.92111244

00:54:06.394 --> 00:54:08.348 studying the extracellular spaces I think.  
NOTE Confidence: 0.927310497272727

00:54:23.740 --> 00:54:25.588 So I was hoping I could find  
NOTE Confidence: 0.927310497272727

00:54:25.588 --> 00:54:26.860 unique sorts of things,  
NOTE Confidence: 0.927310497272727

00:54:26.860 --> 00:54:29.218 but that's not what I'm finding.  
NOTE Confidence: 0.927310497272727

00:54:29.220 --> 00:54:33.077 I'm finding patterns among classes of cells



NOTE Confidence: 0.927310497272727

00:54:33.077 --> 00:54:37.149 as opposed to unique this versus that.

NOTE Confidence: 0.927310497272727

00:54:37.150 --> 00:54:38.913 And I I think you know, if you think

NOTE Confidence: 0.927310497272727

00:54:38.913 --> 00:54:40.599 about anatomic pathology and how you

NOTE Confidence: 0.927310497272727

00:54:40.599 --> 00:54:42.866 take a chunk of tissue and you look at it,

NOTE Confidence: 0.927310497272727

00:54:42.870 --> 00:54:46.874 so PSMA, that's pretty indicative of a

NOTE Confidence: 0.927310497272727

00:54:46.874 --> 00:54:49.614 prostate cancer cell in a certain state.

NOTE Confidence: 0.927310497272727

00:54:49.614 --> 00:54:52.429 If you took a chunk of prostate tissue out,

NOTE Confidence: 0.927310497272727

00:54:52.430 --> 00:54:54.910 if you took a piece of my perotid,

NOTE Confidence: 0.927310497272727

00:54:54.910 --> 00:54:57.616 you'd also see high levels of PSMA.

NOTE Confidence: 0.927310497272727

00:54:57.616 --> 00:55:02.962 So PSMA is not really a good

NOTE Confidence: 0.927310497272727

00:55:02.962 --> 00:55:07.148 prostate cancer necessarily marker.

NOTE Confidence: 0.927310497272727

00:55:07.150 --> 00:55:12.601 So I I actually think the the

NOTE Confidence: 0.927310497272727

00:55:12.601 --> 00:55:14.856 best classifying markers will will

NOTE Confidence: 0.927310497272727

00:55:14.856 --> 00:55:17.431 probably not be exactly the same

NOTE Confidence: 0.927310497272727

00:55:17.431 --> 00:55:19.525 as those which have been defined

NOTE Confidence: 0.927310497272727

00:55:19.525 --> 00:55:22.190 so far in intact tissue contexts.

NOTE Confidence: 0.62401146

00:55:24.310 --> 00:55:24.400 All

NOTE Confidence: 0.887546576666667

00:55:29.080 --> 00:55:31.960 right, I put everybody to sleep.

NOTE Confidence: 0.887546576666667

00:55:31.960 --> 00:55:32.998 Thank you, everybody.