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Transcript of Michael L. Metzker, Ph.D.

Date: July 31, 2025

Case: Qiagen Sciences, LLC -v- Tecan Genomics, Inc. (PTAB)

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WORLDWIDE COURT REPORTING & LITIGATION TECHNOLOGY

Conducted on July 31, 2025

<p>1 UNITED STATES PATENT AND TRADEMARK OFFICE</p> <p>2 -----</p> <p>3 BEFORE THE PATENT TRIAL AND APPEAL BOARD</p> <p>4 -----</p> <p>5 QIAGEN SCIENCES, LLC,</p> <p>6 Petitioner,</p> <p>7 v.</p> <p>8 TECAN GENOMICS, INC.,</p> <p>9 Patent Owner.</p> <p>10 -----</p> <p>11 IPR2025-00028</p> <p>12 U.S. Patent No. 10,036,012</p> <p>13 -----</p> <p>14 DEPOSITION OF</p> <p>15 MICHAEL L. METZKER, PH.D.</p> <p>16 NEW YORK, NEW YORK</p> <p>17 JULY 31, 2025</p> <p>18</p> <p>19</p> <p>20</p> <p>21 REPORTED BY: KIARA MILLER</p> <p>22 FILE NO.: 593985</p>	<p>3</p> <p>1 A P P E A R A N C E S:</p> <p>2</p> <p>3 ON BEHALF OF PETITIONER:</p> <p>4 QUINN EMANUEL URQUHART & SULLIVAN, LLP</p> <p>5 50 California Street</p> <p>6 22nd Floor</p> <p>7 San Francisco, California 94111</p> <p>8 BY: DAVID BILSKER, ESQ.</p> <p>9</p> <p>10 ON BEHALF OF PATENT OWNER:</p> <p>11 GOODWIN PROCTER LLP</p> <p>12 The New York Times Building</p> <p>13 620 Eighth Avenue</p> <p>14 New York, New York 10018</p> <p>15 BY: LINNEA P. CIPRIANO, ESQ.</p> <p>16</p> <p>17 ALSO PRESENT:</p> <p>18 RICHARD SAMUEL HERRON PHD, GOODWIN PROCTER ADVISOR</p> <p>19</p> <p>20</p> <p>21</p> <p>22</p>
<p>2</p> <p>1 Deposition of MICHAEL L. METZGER, PH.D., taken</p> <p>2 on behalf of PATENT OWNER, at 295 5th Avenue,</p> <p>3 New York, New York, commencing at 9:34 AM, JULY</p> <p>4 31, 2025, before Kiara Miller.</p> <p>5</p> <p>6</p> <p>7</p> <p>8</p> <p>9</p> <p>10</p> <p>11</p> <p>12</p> <p>13</p> <p>14</p> <p>15</p> <p>16</p> <p>17</p> <p>18</p> <p>19</p> <p>20</p> <p>21</p> <p>22</p>	<p>4</p> <p>1 MICHAEL METZKER, after having first</p> <p>2 been duly sworn by a Notary Public of the State of</p> <p>3 New York, was examined and testified as follows:</p> <p>4 EXAMINATION BY</p> <p>5 MS. CIPRIANO:</p> <p>6 Q Could you please state your name for the</p> <p>7 record.</p> <p>8 A Good morning. My name is Michael</p> <p>9 Metzker, M-e-t-z-k-e-r.</p> <p>10 Q I'm Linnea Cipriano, as I introduced</p> <p>11 myself a moment before. I'll be taking your</p> <p>12 deposition today.</p> <p>13 Have you been deposed before?</p> <p>14 A Yes.</p> <p>15 Q How many times?</p> <p>16 A Dozens.</p> <p>17 Q When was the most recent?</p> <p>18 A Let's see, I think it was, we're still</p> <p>19 in July, I think it was last month.</p> <p>20 Q More than likely these ground rules will</p> <p>21 be similar to those that you've sat through</p> <p>22 before, but just so we're on the same page,</p>

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<p>5</p> <p>1 I'd like to go through a few before we get 2 started.</p> <p>3 I will be asking you questions today and 4 your answers will be under oath as if you 5 were testifying in court.</p> <p>6 Do you understand that?</p> <p>7 A I do.</p> <p>8 Q During the course of the deposition your 9 counsel will likely make objections, but 10 unless you've been instructed not to answer, 11 I expect you to answer my question; is that 12 fair?</p> <p>13 A I do understand that.</p> <p>14 Q As you probably notice we have a court 15 reporter taking down everything that we say 16 today, that means you and I must speak 17 clearly. I will try not speak over you, I 18 would ask that you try not to speak over me.</p> <p>19 Does that make sense?</p> <p>20 A I understand that too.</p> <p>21 Q She can't record head nods or um-hmms, 22 so please answer verbally.</p>	<p>7</p> <p>1 able to answer my questions fully and 2 truthfully today?</p> <p>3 A No.</p> <p>4 Q Did you prepare for today's deposition?</p> <p>5 A Yes.</p> <p>6 Q Without revealing any privileged 7 communications with your counsel, at a high 8 level what did you do to prepare?</p> <p>9 A I met with counsel over the last day and 10 a half.</p> <p>11 Q With whom did you speak regarding this 12 deposition?</p> <p>13 A Both David Bilsker and Matt Troutman.</p> <p>14 Q Did you speak with anyone at Qiagen?</p> <p>15 A No.</p> <p>16 Q And about how long did you prepare?</p> <p>17 A I was here yesterday for about 18 eight hours and Monday was a half day, maybe 19 four and a half, five hours.</p> <p>20 Q Did you review any documents to prepare 21 for this deposition?</p> <p>22 A Yes.</p>
<p>6</p> <p>1 Does that make sense?</p> <p>2 A I understand that.</p> <p>3 Q We make take breaks from time to time, 4 let me know if you need a break. I'll just 5 ask that you answer any pending question 6 before we break.</p> <p>7 Does that make sense?</p> <p>8 A I do understand that.</p> <p>9 Q If you don't understand a question, 10 please ask me to rephrase it. I'm sure that 11 some of my questions will not be as clear as 12 I intend them to be, but if you answer a 13 question, I will assume you understood it.</p> <p>14 Does that make sense?</p> <p>15 A That's what I do.</p> <p>16 Q Okay. You understand that while I'm 17 asking you questions you're not permitted to 18 discuss the substance of your testimony with 19 your attorneys.</p> <p>20 Do you understand that?</p> <p>21 A I do.</p> <p>22 Q Is there any reason why you will not be</p>	<p>8</p> <p>1 Q Do you recall specifically which 2 documents you looked at?</p> <p>3 A Yes.</p> <p>4 Q What are they?</p> <p>5 A They would have been my IPR declaration 6 and references cited in the declaration.</p> <p>7 Q So you prepared the declaration as you 8 mentioned regarding Tecan patent ending in 9 '357, do you recall that?</p> <p>10 A I do.</p> <p>11 Q So I'm going to hand you -- it's my 12 understanding is it's Exhibit 1003 in the 13 IPR2025-00026 proceeding.</p> <p>14 (Whereupon, Declaration of 15 Michael L. Metzker, Ph.D., Case 16 IPR2025-00026 was previously 17 marked as Exhibit 1003.)</p> <p>18 Q Do you recognize this document?</p> <p>19 A I do.</p> <p>20 Q And what is it?</p> <p>21 A It is my declaration for IPR 2025-00026 22 regarding the '357 patent.</p>

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<p>9</p> <p>1 Q If we can turn to the last page in that</p> <p>2 the document, it's page marked 158.</p> <p>3 Do you see that?</p> <p>4 A I do.</p> <p>5 Q Is that your signature?</p> <p>6 A It is.</p> <p>7 Q At a high level, how did you prepare</p> <p>8 your declaration?</p> <p>9 A Typically I would first have a call with</p> <p>10 counsel describing what my -- what they're</p> <p>11 asking me to do, my activities. And then I</p> <p>12 would start reviewing documents and then</p> <p>13 there would be a -- I would do most of the</p> <p>14 technical writing, they would provide a</p> <p>15 skeleton for me to fill in where I need to</p> <p>16 fill in to finalize the declaration.</p> <p>17 Q And you said typically, is that how it</p> <p>18 happened in this case?</p> <p>19 A Yes.</p> <p>20 Q Okay. Did you consult with anyone other</p> <p>21 than the attorneys that you've mentioned</p> <p>22 here, in preparation of your declaration?</p>	<p>11</p> <p>1 Q Understood. If we could take a look</p> <p>2 at -- we'll come back to your declaration.</p> <p>3 If we could take a look at the Exhibit 1016.</p> <p>4 (Whereupon, Metzker CV was</p> <p>5 previously marked as Exhibit</p> <p>6 1016.)</p> <p>7 Q I've handed you what was previously</p> <p>8 marked as Exhibit 1016, do you recognize this</p> <p>9 document?</p> <p>10 A I do.</p> <p>11 Q What is it?</p> <p>12 A It's my CV dated February 22nd, 2024.</p> <p>13 Q And are there any updates between</p> <p>14 February 22nd, 2024 and now that are</p> <p>15 impactful to your experience?</p> <p>16 A There might be a couple of updates. I</p> <p>17 just submitted a new grant, may not have been</p> <p>18 listed here, just finished a study section</p> <p>19 with the NIH this month, probably wasn't</p> <p>20 listed here. I think we may have gotten</p> <p>21 another patent, so things like that.</p> <p>22 Q No changes to your employment?</p>
<p>10</p> <p>1 A No.</p> <p>2 Q Have you read patent owner's preliminary</p> <p>3 response that was filed after you submitted</p> <p>4 your declaration?</p> <p>5 A I believe so.</p> <p>6 Q Is there anything in that paper that</p> <p>7 would change or make you modify your opinions</p> <p>8 in your declaration?</p> <p>9 A Not that I'm aware of.</p> <p>10 Q Have you read the PTAB's institution</p> <p>11 decision?</p> <p>12 A I have.</p> <p>13 Q Same question. Anything in that that</p> <p>14 would make you change or modify your</p> <p>15 opinions?</p> <p>16 A No.</p> <p>17 Q Are there any the changes or corrections</p> <p>18 that you would like to make to your</p> <p>19 declaration as you sit here today?</p> <p>20 A No. Other than if we stumble over a</p> <p>21 typographical error maybe, but other than</p> <p>22 that, no.</p>	<p>12</p> <p>1 A No.</p> <p>2 Q Okay. Are you currently employed?</p> <p>3 A Yes.</p> <p>4 Q What is your current position?</p> <p>5 A So as stated here I'm the founder,</p> <p>6 president and CEO of a biotech company called</p> <p>7 RedVault Biosciences and I also own a wine</p> <p>8 company.</p> <p>9 Q That's very interesting. We can talk</p> <p>10 about that off the record.</p> <p>11 What do you consider your area of</p> <p>12 expertise?</p> <p>13 A I have a broad area of expertise. I've</p> <p>14 been in the field for more than 35 years.</p> <p>15 I've done a lot of technology development,</p> <p>16 particularly in biotechnology, molecular</p> <p>17 biology. Genomics. I've had a lot of</p> <p>18 development work in methods of preparing</p> <p>19 libraries for sequencing, generating</p> <p>20 technologies for sequencing itself.</p> <p>21 Developing methodologies for analyzing</p> <p>22 sequences, particularly in the area of HIV</p>

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<p>13</p> <p>1 transmission between individuals. I've been 2 involved with the characterizing variation in 3 the genome with respect to certain diseases, 4 such as diabetes. I've done some work in 5 cancer research. Done work in diagnostics. 6 So RedVault is involved in analyzing small 7 RNAs for urine to develop a point of care 8 task for identifying bacterias associated 9 with STDs. 10 So those are some examples. 11 I've done instrument work. We've 12 developed an actual fluorescent sequencer for 13 sequencing technologies and things like that. 14 Q Now, you talked a little bit about 15 RedVault, could you tell me a bit about your 16 work at LaserGen? 17 A I started LaserGen, it's my first 18 company in 2002. That really was spearheaded 19 from work I did in graduate school in 1994, 20 where I published one of the very first 21 papers on reversible terminator that is 22 currently used in many next generation</p>	<p>15</p> <p>1 sidelined, but that's what I did at LaserGen. 2 Q Was the focus of the LaserGen reversible 3 terminator all the way through 2012, which is 4 where I think you left, at least according to 5 your declaration? 6 A Right. Well, the reversible terminators 7 was a core chemistry that we then built a 8 platform around and really the goals was to 9 develop a next generation sequencing 10 instrument that utilized the novel reversible 11 terminators for lots of applications for DNA 12 sequencing. 13 Q Did that platform ever become 14 commercially available? 15 A It did not. As I mentioned, it was sold 16 to Agilent. Agilent had grand plans to 17 commercialize a clinical instrument for 18 cancer diagnostics. I wasn't involved in 19 that at that time, it's just secondhand news, 20 but at some point, new management came in and 21 they decided to shift gears. 22 Q Okay. You have been involved in patent</p>
<p>14</p> <p>1 sequencing methods, and I went ahead and 2 finished my Ph.D., then I went to Merck and 3 worked at Merck for a couple years and then 4 in 1999 I came back to Baylor College 5 Medicine where I was a tenured track 6 assistant professor. 7 During that time I decided I wanted to 8 start a company and build the company based 9 on sequencing technologies. So at that time 10 the National Human Genome Research Institute, 11 the NHGRI, was interested in funding new 12 programs for \$100,000 genome and the \$1,000 13 genome. So I was awarded a large grant 14 through Baylor. LaserGen was a 15 subcontractor, and we were chemically 16 synthesizing novel reversible terminators 17 that would be used in the next generation 18 sequencing. 19 That's what I did for about more than 20 10 years, we brought in investors, they took 21 the company to the next level, it sold to 22 Agilent and that technology now is kind of</p>	<p>16</p> <p>1 litigation before, right? 2 A Yes. 3 Q And on Page 2 of your CV, Exhibit 1016, 4 you have a list of prior expert experience. 5 Do you see that? 6 A I do. 7 Q Could you, to the extent you remember, 8 would you run down and let me know which of 9 these were patent cases? 10 A I don't think the 10X Bio-Rad was. It 11 was an ITC case. But the Plexxikon versus 12 Novartis was a patent case. Guardant versus 13 Foundation Medicine was a patent case. 14 Archer versus Qiagen was a patent case. 15 Illumina versus Natera was a patent case. 16 Illumina versus Complete Genomics was a 17 patent case. Complete Genomics versus 18 Illumina was a patent case. Ravgen versus 19 Ariosa Diagnostics was a patent case. The 20 Illumina Cambridge Limited versus MGI was a 21 patent case. 22 The Guardant versus Natera was not a</p>

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<p>17</p> <p>1 patent case; that was the false 2 advertisements case. DNA Genotek versus 3 Spectrum Solution was a patent case. 4 Twinstrand Bioscience versus Guardant was a 5 patent case. The Trustees of the University 6 of Pennsylvania versus Regenxbio -- and 7 Regenxbio versus Sarepta was a patent case. 8 Spectrum Solution versus DNA Genotek was 9 an IPR, but it did involve patent, obviously. 10 Invitae versus Natera is a patent case. 11 Natera versus NeoGenomics is a patent case. 12 Q So almost all of them? 13 A Almost all of them. I noted the 14 exceptions. 15 Q Yes, absolutely. Thank you. 16 And how many of these involved DNA 17 sequencing technologies? 18 A So going down the list, Guardant Health 19 versus Foundation Medicine is -- involves DNA 20 sequencing technologies. Archer versus 21 Qiagen. Illumina versus Natera. Illumina 22 versus Complete Genomics. Complete Genomics</p>	<p>19</p> <p>1 Q Okay. We can set your CV aside. 2 So you talk a little bit about some 3 technical background in your declaration. 4 I'd like to the talk through some of the 5 concepts. 6 PCR is polymerase chain reaction; is 7 that a fair -- we can -- we're on the same 8 page with that language? 9 A That's what it stands for. 10 Q Okay. And you explained that PCR can 11 amplify particular regions of genetic 12 material; is that fair? 13 A Well, PCR is a very broad term, and 14 there's lots of different ways to use PCR. 15 One of those ways, if you use two 16 target-specific primers, you can amplify 17 specific regions within the DNA sample that 18 you have. 19 Q And that's pretty typical, using two 20 primers to amplify a target section. That's, 21 generally, what we would call a typical PCR 22 reaction; is that fair?</p>
<p>18</p> <p>1 versus Illumina. Ravgen versus Ariosa was 2 more about blood collection, but was used for 3 isolating cell-free DNA for sequencing 4 purposes. 5 Illumina Cambridge versus MGI was 6 sequencing technology. The Guardant Health 7 versus Natera was false advertisement, but it 8 involved MRD tests that involved next 9 generation sequencing. DNA Genotek versus 10 Spectrum was saliva collection tubes that 11 were used for DNA sequencing application. 12 Twinstrand Biosciences versus Guardant 13 involved ways of quantitating variants by DNA 14 sequencing technologies. 15 Spectrum Solutions versus DNA Genotek 16 was, again, a saliva collection that would 17 be -- that sample would be used for next 18 generation sequencing. Invitae versus Natera 19 involves the analysis of sequencing data. 20 And Natera versus NeoGenomics involves MRD 21 testing, which uses next generation 22 sequencing technologies.</p>	<p>20</p> <p>1 MR. BILSKER: Objection. Vague and 2 ambiguous. Overbroad. 3 A As I said, PCR is a very broad term. 4 That's one way to do PCR. There's many ways 5 to do PCR. 6 Q Okay. If we look at the section of PCR 7 that's on Page 17 and 18 in your declaration, 8 starting at Paragraph 29, you have a figure 9 on Page 18 underneath Paragraph 30. 10 Do you see that? 11 A I do. 12 Q Okay. And this depicts a PCR reaction 13 where two primers are used, right? 14 A In this figure, two primers are being 15 shown. 16 Q Okay. And in order for the primers to 17 bind, the DNA must be what's call denatured; 18 is that fair? 19 MR. BILSKER: Objection. Vague and 20 ambiguous. Overbroad. Foundation. 21 A Could you repeat the question, please? 22 Q Sure. And maybe I can clarify a bit.</p>

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<p>21</p> <p>1 So in PCR -- in a PCR reaction, you</p> <p>2 start with, at least in the figure that is</p> <p>3 in -- on Page 18 of your declaration, you</p> <p>4 start with what's called a double stranded</p> <p>5 piece of DNA, right?</p> <p>6 A Right. So this is a figure that comes</p> <p>7 from a chapter that I wrote. And this is</p> <p>8 illustrating kind of the basics of how the</p> <p>9 PCR, as it was described by Kary Mullis in</p> <p>10 the 1980s, how this method would work.</p> <p>11 Q And so you're -- in this figure that you</p> <p>12 chose to include, you start with a double</p> <p>13 stranded piece of DNA, right?</p> <p>14 A In this illustration, just showing kind</p> <p>15 of the basic concepts of PCR, we do start</p> <p>16 with the double stranded -- we start with the</p> <p>17 double stranded DNA.</p> <p>18 Q And there is, in that first step of the</p> <p>19 figure, there's an arrow that's the labeled</p> <p>20 "primer extension."</p> <p>21 Do you see that?</p> <p>22 A Yes.</p>	<p>23</p> <p>1 Q And what is the purpose of separating a</p> <p>2 double strand into single strands?</p> <p>3 A So in the PCR reaction, there are also</p> <p>4 primers. And those primers are designed to</p> <p>5 hybridize to regions that, by design, will</p> <p>6 create a partially double stranded complex.</p> <p>7 Q Primers can't hybridize unless the</p> <p>8 double stranded DNA has been denatured; is</p> <p>9 that fair?</p> <p>10 MR. BILSKER: Objection. Form.</p> <p>11 A Not necessarily.</p> <p>12 Q What do you mean by that?</p> <p>13 A Well, there's things called triplex DNA,</p> <p>14 where double stranded DNA can be used, and</p> <p>15 you can hybridize a third strand of DNA.</p> <p>16 Q Is that what typically happens in a PCR</p> <p>17 reaction?</p> <p>18 MR. BILSKER: Objection. Form.</p> <p>19 A Again, there's many ways of doing PCR.</p> <p>20 In the example that I'm showing, which is,</p> <p>21 again, just kind of the basic PCR steps. In</p> <p>22 this example, you would denature the DNA, but</p>
<p>22</p> <p>1 Q And the shorter, I guess it's -- shorter</p> <p>2 lines with five blue ticks, those are the</p> <p>3 primers, right?</p> <p>4 A Well, again, this is kind of a</p> <p>5 high-level depiction of a primer and those</p> <p>6 picks are more like base pairs, where, by</p> <p>7 design, in this example, the primer has been</p> <p>8 designed to be complementary to a region</p> <p>9 within the DNA template.</p> <p>10 Q As you depict in the figure, one of the</p> <p>11 typical first steps of a PCR cycle is to</p> <p>12 separate the strands by denaturing them,</p> <p>13 right?</p> <p>14 MR. BILSKER: Objection. Form.</p> <p>15 A Well, again, this is kind of a general</p> <p>16 figure. There are actually other steps</p> <p>17 before you actually denature DNA. But I'm</p> <p>18 just -- I'm trying to describe the three</p> <p>19 basic steps in the basic PCR method described</p> <p>20 by Dr. Mullis. And one of those steps is to</p> <p>21 separate the double strand into single</p> <p>22 strands.</p>	<p>24</p> <p>1 that doesn't necessary mean it's true for all</p> <p>2 PCR methods.</p> <p>3 Q And once, in the figure that you have,</p> <p>4 once two primers hybridize to the single</p> <p>5 stranded pieces of DNA, each of the primers,</p> <p>6 in the reaction, extends.</p> <p>7 Can you explain what you mean by</p> <p>8 "extend" in the figure?</p> <p>9 A Sure. So in -- this is all happening in</p> <p>10 a tube. And in the tube is a solution. And</p> <p>11 in the solution, in addition to the DNA that</p> <p>12 we've been talking about and the primers that</p> <p>13 we've been talking about, there's an enzyme</p> <p>14 called a DNA polymerase. There are also</p> <p>15 nucleotides that are in the solution along</p> <p>16 with buffers that control the PH, magnesium</p> <p>17 ion, there can be some salts and all of that.</p> <p>18 But the idea is once the primer has</p> <p>19 hybridized to the template to create this</p> <p>20 partially double stranded complex, that's</p> <p>21 where the polymerase can then bind and start</p> <p>22 incorporating those nucleotides in solution</p>

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<p>25</p> <p>1 to create the complement strand of the</p> <p>2 template strand. And we call that extending.</p> <p>3 Q And the top of this figure creates a</p> <p>4 circle where you go from double stranded DNA</p> <p>5 down to single stranded DNA with the primers</p> <p>6 and extension, and then back to a double</p> <p>7 stranded DNA.</p> <p>8 Is that a fair representation of that</p> <p>9 first level of the figure at the top?</p> <p>10 A Well, we call it a cycle, not a circle.</p> <p>11 Q Sure.</p> <p>12 A And it is depicting the three steps.</p> <p>13 And in each cycle, if everything works</p> <p>14 perfectly, the number of DNA molecules that</p> <p>15 you make double. And over a number of</p> <p>16 cycles, it become what we call exponential</p> <p>17 and you can make millions of copies from your</p> <p>18 starting material.</p> <p>19 Q And what's being copied here is the</p> <p>20 sequence between the two primers, right?</p> <p>21 MR. BILSKER: Objection. Form.</p> <p>22 A In the example that I'm showing here,</p>	<p>27</p> <p>1 molecule, some of them end up amplifying</p> <p>2 those molecules, some of them continue to use</p> <p>3 single molecules.</p> <p>4 But typically are not cloned, where in</p> <p>5 sanger sequencing, all of our DNA templates</p> <p>6 were cloned into bacteria that we would have</p> <p>7 to culture and have large freezer farms of</p> <p>8 glycerol stocks of bacteria.</p> <p>9 Q I do remember that in my graduate school</p> <p>10 days, so understood.</p> <p>11 Next generation sequencing is often</p> <p>12 abbreviated NGS; is that fair?</p> <p>13 A That is fair.</p> <p>14 Q Okay. And just another vocabulary</p> <p>15 point, in order to use or perform a next</p> <p>16 generation sequencing, typically it's</p> <p>17 understood that you have to prepare what's</p> <p>18 called a sequencing library; is that</p> <p>19 accurate?</p> <p>20 A I think that's, generally, true for any</p> <p>21 sequencing technology.</p> <p>22 Q Okay. And what is a sequencing library,</p>
<p>26</p> <p>1 the primers define the area that gets copied.</p> <p>2 Q Okay. When we were talking about your</p> <p>3 background, you referred to next generations</p> <p>4 sequencing.</p> <p>5 Do you recall that?</p> <p>6 A I do.</p> <p>7 Q What is your understanding of what next</p> <p>8 generation sequencing is?</p> <p>9 A It is a broad field of different</p> <p>10 technologies that replaced what I called</p> <p>11 first generation sequencing or commonly known</p> <p>12 as sanger sequencing.</p> <p>13 Q And does that broad field of the</p> <p>14 technologies that replaced sanger sequencing,</p> <p>15 do they have commonalities, other than they</p> <p>16 just came next?</p> <p>17 MR. BILSKER: Objection. Form.</p> <p>18 A Very -- they have a couple general</p> <p>19 commonalities. For example, they don't</p> <p>20 require the use of electrophoresis, which was</p> <p>21 one of the main limitations of sanger</p> <p>22 sequencing. Most of them start with a single</p>	<p>28</p> <p>1 as you understand it?</p> <p>2 A So a library is not a building with</p> <p>3 books, but in molecular biology, we call a</p> <p>4 library the collection of DNA fragments, RNA</p> <p>5 fragments, nucleic acid fragments, for lack</p> <p>6 of a better word, that were derived from the</p> <p>7 sample that you want to analyze. Then you</p> <p>8 can manipulate that library in many different</p> <p>9 ways.</p> <p>10 Q So starting at Paragraph 39 of your</p> <p>11 declaration, you have a section that's</p> <p>12 entitled, "Applicable Standards."</p> <p>13 Do you see that?</p> <p>14 A I do.</p> <p>15 Q And the legal standards presented in</p> <p>16 this section over the next couple of pages,</p> <p>17 those were provided by counsel, right?</p> <p>18 A That's correct.</p> <p>19 Q And in your declaration you assumed a</p> <p>20 particular level of expertise for the person</p> <p>21 of ordinary skill in art.</p> <p>22 Do you recall that?</p>

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<p>29</p> <p>1 A I'm not sure I assumed anything.</p> <p>2 Q Okay.</p> <p>3 A I was provided some standards of how I</p> <p>4 should think about what a person of ordinary</p> <p>5 skill in the art would be relevant to the</p> <p>6 technology field.</p> <p>7 Q Okay. And how did you arrive at the</p> <p>8 skill set that you applied in this case?</p> <p>9 A Well, I considered some of things I was</p> <p>10 provided by counsel to consider, and then I</p> <p>11 also based it on my own experience in coming</p> <p>12 to what I thought the person of ordinary</p> <p>13 skill in the art -- the level of skill should</p> <p>14 be.</p> <p>15 Q Do you understand the patent owner</p> <p>16 provided a slightly different level of skill</p> <p>17 in their papers?</p> <p>18 A I believe I understand that.</p> <p>19 Q Would your opinions change if you</p> <p>20 applied patent owner's definition of the</p> <p>21 person of ordinary skill in the art?</p> <p>22 MR. BILSKER: Objection. Form.</p>	<p>31</p> <p>1 described here, where an adapter is an</p> <p>2 oligonucleotide, and it is something that you</p> <p>3 will attach typically to your target</p> <p>4 sequence.</p> <p>5 Q In your experience does an adapter serve</p> <p>6 a function?</p> <p>7 A It can.</p> <p>8 Q And what functions can an adapter serve?</p> <p>9 A It depends on what is included in the</p> <p>10 adapter.</p> <p>11 Q And what kinds of adapters have you used</p> <p>12 in your research?</p> <p>13 A I've used a variety of adapters.</p> <p>14 Q Can you give me some examples?</p> <p>15 A Sure.</p> <p>16 I've used adapters that were helpful for</p> <p>17 cloning. I've used adapters that were</p> <p>18 helpful for making sequencing libraries,</p> <p>19 particularly in next-generation sequencing.</p> <p>20 Adapters are absolutely required for</p> <p>21 next-generation sequencing because each</p> <p>22 platform has its own specific requirements</p>
<p>30</p> <p>1 A I don't think it would. I've come to</p> <p>2 the same conclusions.</p> <p>3 Q Okay. If we could look at the claim</p> <p>4 construction section of the declaration,</p> <p>5 which starts at, I think, Paragraph 53 -- or</p> <p>6 52, excuse me.</p> <p>7 Are you there?</p> <p>8 A Yes, I'm there.</p> <p>9 Q Now, the first sentence of this</p> <p>10 paragraph: "I understand the petitioner has</p> <p>11 taken the position that the term 'adapter'</p> <p>12 means any oligonucleotide that is attached to</p> <p>13 the target sequence."</p> <p>14 Do you see that?</p> <p>15 A I do see that sentence.</p> <p>16 Q What do you understand "adapter" to mean</p> <p>17 in your work?</p> <p>18 MR. BILSKER: Objection.</p> <p>19 Q Let me rephrase that.</p> <p>20 How do you use the term "adapter" in</p> <p>21 your work?</p> <p>22 A Well, I pretty much use it the way it's</p>	<p>32</p> <p>1 that the libraries need to be modified to run</p> <p>2 on the sequencers.</p> <p>3 Q Have you ever used an adapter that</p> <p>4 serves no purpose?</p> <p>5 MR. BILSKER: Objection. Form.</p> <p>6 A Not that I would recall. I usually</p> <p>7 don't use things that have no purpose.</p> <p>8 Q You note in your -- in that Paragraph 52</p> <p>9 that the patent, the '357 Patent, defines the</p> <p>10 term "adapter."</p> <p>11 Do you see that in the second sentence</p> <p>12 of Paragraph 52?</p> <p>13 A I do. And I actually quote where that</p> <p>14 comes from in the patent.</p> <p>15 Q Is there a reason why you didn't you use</p> <p>16 the definition provided in the patent in your</p> <p>17 analysis of the term "adapter"?</p> <p>18 MR. BILSKER: Objection. Form.</p> <p>19 A Yes.</p> <p>20 Q And why is that?</p> <p>21 A Well, it's stated in the next sentence</p> <p>22 that:</p>

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Conducted on July 31, 2025

<p>33</p> <p>1 "I'm also aware that during 2 prosecution the examiner reviewed the 3 above definition of 'adapter' and 4 concluded that since any 5 oligonucleotide can perform such 6 function, any oligonucleotide is 7 considered to anticipate this term." 8 And so based on that, that's my 9 understanding the basis of how the term 10 should be construed. 11 Q Do you have any understanding of the 12 legal standards that examiners use for claim 13 construction during prosecution? 14 MR. BILSKER: Objection. Form. 15 A I'm not an attorney, so I wouldn't know 16 what those standards are. 17 Q And so do you have any understanding of 18 whether the standard for claim construction 19 that an examiner uses is different than what 20 the PTAB will use? 21 MR. BILSKER: Objection. Form. 22 A I don't.</p>	<p>35</p> <p>1 Exhibit 1006.) 2 Q I've handed you what's been marked as 3 Exhibit 1006. Is this the publication that 4 you referred to as "Porreca" in your 5 declaration? 6 A Yes. 7 Q In general what does Porreca describe? 8 MR. BILSKER: Objection. Form. 9 A Well, I mean, I can read the abstract 10 for you. I mean, it really is trying to 11 develop methods and compositions to reduce 12 bias and increase the reproducibility of 13 multiplex analyses of genetic loci. And it 14 describes a number of preparative methods of 15 how that can be accomplished. 16 Q And do you have an understanding of what 17 is meant by "reduce bias" here? 18 A Yes. 19 Q And what does that mean? 20 A So for example, there are -- I like to 21 think of library preparation, next-generation 22 sequencing, analysis of next-generation</p>
<p>34</p> <p>1 Q And did you do any further analysis of 2 the specification or the prosecution history 3 other than this -- looking at this statement 4 by the examiner in your construction or 5 proposed construction of the term "adapter"? 6 A Well, I reviewed the specification, not 7 just this definition, but the entire 8 specification. I also looked at the entire 9 prosecution history, and this is where I came 10 down. 11 Q Okay. One of the articles that you 12 discuss in your declaration is referred to as 13 "Porreca." 14 Do you recall that? 15 A Yes. 16 Q And if we could -- I'll give it to you 17 to make sure that you have it handy. It's 18 Exhibit 1006. 19 (Whereupon, US Patent 20 Application Publication No. US 21 2012/0165202 A1 by Porreca et 22 al. was previously marked as</p>	<p>36</p> <p>1 sequencing as a workflow. And there are 2 various steps within the workflow that can 3 introduce errors, biases, for example, making 4 duplicates, creating errors that affect the 5 overall analyses. And so they're trying to 6 reduce all of those, which they call the 7 reduced bias and increasing the 8 reproducibility of the analysis. 9 Q And when you say that there are various 10 steps within the workflow that can introduce 11 errors, can you give me some examples of 12 those, please? 13 MR. BILSKER: Objection. Form. 14 A Sure. 15 For example, when starting with genomic 16 DNA, we typically break things up, and then 17 we try to put them back together. In 18 breaking DNA up, sometimes damages the DNA 19 bases themselves. So that when they're 20 analyzed, they may actually look like a 21 different base than what was in the sample. 22 That's one example.</p>

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<p>37</p> <p>1 Q Do you have any other examples?</p> <p>2 A Yes.</p> <p>3 Q And what are they?</p> <p>4 A So PCR can introduce errors. PCR can</p> <p>5 introduce duplications. Sequencing the</p> <p>6 actual process of decoding the four letters</p> <p>7 of the genetic code can incorrectly call a</p> <p>8 base. Once the reads are generated, the</p> <p>9 reads can be mapped to the genome and not be</p> <p>10 placed properly and provide errors. So</p> <p>11 there's lots of stages within the workflow</p> <p>12 where bias can occur.</p> <p>13 Q And when you say "PCR can introduce</p> <p>14 duplications," what do you mean by that?</p> <p>15 A So just give a brief example of a single</p> <p>16 molecule, and that's what I start with in my</p> <p>17 PCR. And when I go through the basic PCR</p> <p>18 protocol where I have two primers and I</p> <p>19 denature that single molecule of</p> <p>20 double-stranded DNA, and then I hybridize my</p> <p>21 primers, which I've designed to target a very</p> <p>22 specific region in the genome, and then</p>	<p>39</p> <p>1 Porreca reference, so you can have them</p> <p>2 handy.</p> <p>3 If we can take a look at Paragraph 73 of</p> <p>4 your declaration.</p> <p>5 A Yes. I'm there.</p> <p>6 Q And starting at Paragraph 73, you</p> <p>7 discuss what you've identified as</p> <p>8 Limitation 1A, and this limitation is the</p> <p>9 limitation that recites "ligating an adapter</p> <p>10 to each of a plurality of nucleic acid</p> <p>11 fragments."</p> <p>12 Do you see that?</p> <p>13 A I do.</p> <p>14 Q And in this section, you point to two</p> <p>15 disclosures in Porreca for support for your</p> <p>16 opinion that Porreca discloses this</p> <p>17 limitation, and I'd like to walk through</p> <p>18 those.</p> <p>19 First, you point to Paragraph 150 of</p> <p>20 Porreca, and that's quoted in the middle --</p> <p>21 or at the end of Paragraph 73.</p> <p>22 Do you see that?</p>
<p>38</p> <p>1 extend that, I make two copies.</p> <p>2 But in reality it only started from one</p> <p>3 copy. And so I mean, that's a simple example</p> <p>4 of what a duplication can be by PCR if, for</p> <p>5 example, there's just one molecule.</p> <p>6 Q Understood.</p> <p>7 And when you said, "PCR can introduce</p> <p>8 errors," what did you mean by that?</p> <p>9 A So in the extension step, we -- I</p> <p>10 previously described a DNA polymerase. DNA</p> <p>11 polymerases are enzymes that copy DNA. So</p> <p>12 they use one strand -- and the strands of DNA</p> <p>13 and RNA, they have information. The sequence</p> <p>14 of those letters provide a code. And so the</p> <p>15 job of the DNA polymerase is to copy one</p> <p>16 strand into another strand that faithfully</p> <p>17 reproduces that code. But there are</p> <p>18 instances where the DNA polymerase will</p> <p>19 insert or incorporate the wrong nucleotide,</p> <p>20 and that will be an error.</p> <p>21 Q Okay. I'm going to kind of go back and</p> <p>22 forth between your declaration and the</p>	<p>40</p> <p>1 A I do.</p> <p>2 Q Okay. And the second piece of</p> <p>3 information you point to in Porreca for this</p> <p>4 section of your declaration is Figure 4C, and</p> <p>5 you've depicted a copy of Figure 4C in the</p> <p>6 middle of Paragraph 74.</p> <p>7 Do you see that?</p> <p>8 A I do.</p> <p>9 Q And just so I understand these, the ends</p> <p>10 in Figure 4C, that's the adapter; is that</p> <p>11 accurate?</p> <p>12 A I'd say it's part of the adapter.</p> <p>13 Q Okay. And what do you mean by that?</p> <p>14 A Well, I think what Porreca is doing here</p> <p>15 is just illustrating what they call the</p> <p>16 differentiator tag. We can call it a UMI.</p> <p>17 But in reality the adapter would include</p> <p>18 other things in addition to these ends, but I</p> <p>19 think for simplicity they're just showing a</p> <p>20 very common method called ligation of an</p> <p>21 adapter to the end of a fragment.</p> <p>22 Q Okay. And Figure 4C is part of a larger</p>

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<p>41</p> <p>1 figure; is that fair? Figure 4 has two other 2 pieces. Do you recall that? 3 A I do. Figure 4 has A, B and C. And 4 their Figure 4 is just illustrating the 5 number of different well-known ways of 6 putting adapter on -- or appending an adapter 7 on the end of a DNA fragment. 8 Q And so if you could look at Figure 4 in 9 Porreca. It's on page marked 10 Exhibit 1006.0005. 11 A I'm there. 12 Q Okay. In Figure 4A could you explain 13 how the padlock probe incorporates the 14 adapter into a -- onto a target sequence? 15 A Sure. 16 So a padlock probe is a linear 17 oligonucleotide that has two targeting ends 18 or arms, as they depict them, which are 19 target-specific to the region of interest. 20 And those two targeting arms are connected by 21 an adapter that contains a number of 22 functions. As illustrated here it includes a</p>	<p>43</p> <p>1 appended to the targeting arms. And once the 2 targeting arms hybridize to the target, as 3 you can see in Figure 4A, there's a gap, the 4 gap can be just a break in the phosphodiester 5 bond, and a ligation reaction can take place 6 to ligate. Now, the probe containing the 7 adapter around the target sequence or a 8 polymerase if the gap is more than a 9 nucleotide long, a polymerase can extend the 10 targeting arm to the other targeting arm 11 creating a gap, by which again a ligase can 12 then ligate the padlock probe that has the 13 adapter based on hybridization to the target. 14 Q I'm not sure I understand. In 15 Figure 4A, where is the molecule of interest, 16 where would it be depicted if you were to 17 draw them? 18 A It would be right below the targeting 19 arms. 20 Q The targeting arms, is it fair to say 21 those are designed to hybridize with portions 22 of the molecule of interest?</p>
<p>42</p> <p>1 differentiator tag, PCR priming sites. It 2 may actually contain more functionality, but 3 I'm just describing what I see in Figure 4A. 4 Q And so in Figure 4A, the molecule of 5 interest is not depicted, right? 6 A That's correct. 7 Q So how does a padlock probe incorporate 8 an adapter to -- or let me use the word that 9 you -- 10 If I can rephrase my question. 11 In Figure 4A, how is the adapter 12 appended to the end of a DNA fragment? 13 MR. BILSKER: Objection. Form. 14 A So first, the way the padlock probe is 15 drawn there is a molecule of interest, it's 16 just not shown. Because as I mentioned the 17 padlock probe is a linear oligonucleotide and 18 they're actually showing it as a half circle 19 where the targeting arms appear as vertical 20 lines. 21 In this example, the adapter is already 22 attached to the targeting arms, it's already</p>	<p>44</p> <p>1 A They actually wrap around in forming the 2 double helix around the molecule of interest, 3 to which -- (indicating) I'm showing my 4 fingers coming together, which you can't 5 capture, to which there's a gap, whereby the 6 ligase will then ligate the probe arms 7 together around the target of interest. 8 Q So the targeting arm one is ligated to 9 targeting arm two; is that fair? 10 A Only when it's wrapped around the target 11 of interest. 12 Q Okay. 13 A They can't ligate to themselves in a 14 vacuum. 15 Q Understood. 16 A They have to be hybridized and wrapped 17 around the target of interest. 18 Q Okay. What's the next step, like, what 19 would be the purpose of attaching a padlock 20 probe to a double helix and the molecule of 21 interest? 22 MR. BILSKER: Objection. Form.</p>

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Conducted on July 31, 2025

<p>45</p> <p>1 A What is the purpose? Well, the purpose 2 is to enrich for a sequence of interest that 3 can then be characterized downstream. 4 Q How would you do that? 5 A Well, there's lot of ways to do that. 6 Q With the padlock probe, how would you -- 7 what's the next step after it binds to the 8 molecule of interest, what's the next step in 9 the process? 10 MR. BILSKER: Objection. Form. 11 A So the next step would be to introduce 12 an exonuclease into the reaction. 13 Q What does the exonuclease do? 14 A The exonuclease will degrade any of the 15 padlock probes that were not formed into a 16 circle. 17 Q After any of the padlock probes that are 18 not circular are degraded, what happens next? 19 A There's lots of things that can happen 20 next. I mean, lots of pathways -- different 21 pathways that can occur after the padlock 22 probe has been made into a circle.</p>	<p>47</p> <p>1 though, the way it's drawn here and I think 2 it's described several times, at least in 3 Porreca, that the targeting arms are 4 separated by some region of interest and they 5 will use a polymerase to copy the target, 6 which then gets ligated into a circle. So 7 the targeting arms and then the copied region 8 are all part of the template of interest that 9 gets characterized. 10 Q If we look at Figure 4B, can you explain 11 to me how the adapter is appended to the 12 molecule of interest in what's depicted in 13 4B? 14 A Yes, again 4A, or Figure 4, is just very 15 general ways of putting an adapter onto a 16 target. 17 In 4B this is using primers, which have 18 what we call five prime tails, and those 19 tails include adapter sequences. As depicted 20 here there would be a differentiator tag or a 21 UMI and then usually just shown as a line, 22 but it can be a priming side for PCR,</p>
<p>46</p> <p>1 Q If your goal is to enrich for the 2 sequence of interest, what would be the next 3 step? 4 A A next step could be to amplify. 5 Q How would you go about amplifying? 6 A Well, in the Figure 4A, there are two 7 PCR priming sites. So one example would be 8 to isolate the circle and then use PCR to 9 amplify that circle, the circle can be 10 degraded into a linear strand again, and it 11 can be characterized in a different way. 12 There's different ways to handle padlock 13 probe. 14 Q So at what stage, using a padlock probe, 15 at what stage do you have a molecule of 16 interest appended to the adapter? 17 A I think you have it here where you form 18 the circle, that's one example. 19 Q When you form the circle, the targeting 20 arms are attached or hybridized to a molecule 21 of interest, right? 22 A Well, what I was trying to explain</p>	<p>48</p> <p>1 sequencing side for sequencing. There can be 2 restriction enzyme sides for cloning, there's 3 lots of things that would go into any of 4 these adapters. 5 Q So I don't think that exactly answered 6 my question. 7 So if we look at Figure 4B, how is the 8 adapter appended to the molecule of interest. 9 We have a primer, you said it's a primer with 10 an adapter, but how do you end up with a 11 molecule of interest that's attached to the 12 adapter? 13 MR. BILSKER: Objection. Form. 14 A Oh, I guess I was describing the primer. 15 Once it's hybridized, it's just shown here as 16 the molecule of interest, it would be 17 extended and the copied template, which now 18 has copied the molecule of interest. Because 19 there's a five prime tail, now the adapter 20 has become a part of the molecule of 21 interest. 22 Q Understood, thank you.</p>

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<p>49</p> <p>1 So if we can look at limitation 1C, 2 which starts at Page 58, Paragraph 77 of your 3 declaration. 4 A I'm there. 5 Q So the limitation that you've label as 6 limitation 1C recites "An indexing site 7 unique to a subset of adapters." 8 Do you see that? 9 A I do. 10 Q For this limitation you rely on 11 Paragraph 149 of Porreca, I think that's 12 copied or quoted on the top of Page 59 of 13 your declaration. 14 Do you see that? 15 A I mean, it's one cite that I rely on. 16 Q And this, the portion that is quoted at 17 the top of Page 59, that refers to what's 18 called a bar code. 19 Do you see that? 20 A I do. 21 Q And I guess, do you understand how 22 Porreca uses the term bar code?</p>	<p>51</p> <p>1 systems affordable and fast, you can pool 2 samples. And that pooling of samples is 3 called multiplexing, but multiplexing has 4 been used for other descriptions as well, but 5 for this purpose, it's multiplexing is 6 pooling samples. 7 Q And what other ways has multiplexing 8 been used that you're referring to? 9 A Well, there's a common method called 10 multiplex PCR, and in that instance, you 11 would amplify in a single tube two or more 12 regions of interest and that can come from a 13 single sample, so that's why I think it's 14 context specific of how you use multiplex. 15 Q Understood. But you said Porreca uses 16 the -- or contemplates multiplexing in the 17 way that you first used it, the identifying 18 samples from different sources; is that 19 accurate? 20 MR. BILSKER: Objection. Form. 21 A Well, I mean it's just what Porreca says 22 and I quote "In addition bar codes may be</p>
<p>50</p> <p>1 A I do. 2 Q And how is that? 3 A So first bar codes are used for lots of 4 things, Porreca is using a bar code as a DNA 5 sequence that can be used for multiplexing 6 samples. 7 Q And this type of bar code is often 8 called a sample index; is that accurate? 9 MR. BILSKER: Objection. Form. 10 A There are lots of names. 11 I would not say it's a commonly used 12 term, but I wouldn't disagree that it hasn't 13 been referred to that before. 14 Q Okay. When you used the term multiplex, 15 what did you mean by that? 16 A Well, that's a good question. In 17 context of bar coding the way Porreca is 18 using it, multiplex would be to tag samples 19 from different individuals or different 20 sources, this is commonly done in next 21 generation sequencing because these machines 22 have so much throughput that to make the</p>	<p>52</p> <p>1 included, for example, to uniquely identify 2 individual samples (e.g. patient samples) 3 that may be mixed together" and that mixing I 4 call pooling, that would run on a single NGS 5 instrument. 6 Q So if we could look at Paragraph 149 7 of -- the full Paragraph 149 of Porreca that 8 you quoted from. 9 A My copy is marked up is that... 10 Q That is the way that it was filed. 11 A Okay. I just wanted to let you know. 12 Q I appreciate it. 13 MR. BILSKER: Really? 14 MS. CIPRIANO: Yeah. 15 MR. BILSKER: Because I have one 16 that's stamped as an exhibit that 17 doesn't have any marking on it. 18 MS. CIPRIANO: It has highlights? 19 MR. BILSKER: Mine doesn't have any 20 highlights on it. 21 THE WITNESS: So mine is marked -- 22 MR. BILSKER: I'm not saying that -- I</p>

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<p>53</p> <p>1 don't remember, but it seems like it 2 would have printed from the same 3 place, right, it's marked the same. 4 MS. CIPRIANO: Agreed. 5 MR. BILSKER: It's not a big deal, 6 we're just not going to attribute to 7 one person or another. 8 MS. CIPRIANO: No, agreed. 9 MR. BILSKER: We'll agree like if it 10 happens to be yours, it's not a waiver 11 of privilege or anything like that. 12 MS. CIPRIANO: I appreciate that. I 13 do believe it's what we pulled from 14 the -- at least what was served on us, 15 but we can agree both ways. 16 Q So if you look at Paragraph 149. 17 A Yes, I'm there. 18 Q And 149, refers, the first sentence 19 there it says, "the skilled artisan will 20 appreciate that as a part of a MIP library 21 preparation process, adaptors may be ligated 22 on to the ends of the molecules of interest."</p>	<p>55</p> <p>1 I'm going to look at Limitation 1E, and 2 that's around Paragraph 82, I have in my 3 notes. 4 A Yes. I'm there. 5 Q And the limitation that you've 6 identified as Limitation 1E, is the 7 limitation that requires amplifying the 8 adapter-ligated fragments into amplicons. 9 Do you see that? 10 A Yes. 11 Q And in Paragraph 82 you point to a 12 portion of Porreca that you quote from, that 13 is Paragraph 150. 14 Do you see that on Page 62? 15 A Yes. 16 Q And then there are two sections 17 highlighted in this, that you've bolded, I 18 guess I will say, in the quoted section. The 19 first is at the top, "For example, nucleic 20 acids comprising differentiator tag sequences 21 may be incorporated by ligation." 22 Do you see that?</p>
<p>54</p> <p>1 Do you see that sentence? 2 A I do. 3 Q Do you have an understanding of what a 4 MIP library preparation process is? 5 A Yes. 6 Q What is it? 7 A Well, it would be something similar to 8 what we saw in 4A. 9 Q What do you mean by that? 10 A So a MIP is, I think it's called a 11 molecule inversion probe and it is a single 12 oligonucleotide that has targeting arms as 13 one example depicted in 4A. 14 MS. CIPRIANO: So we've been going for 15 about an hour. Maybe it's time for a 16 break. 17 MR. BILSKER: Okay. That's fine. 18 (Whereupon, a recess was taken 19 from 10:39 AM until 10:54 AM.) 20 Q Welcome back. 21 A Thank you. 22 Q If we could go back to your declaration.</p>	<p>56</p> <p>1 A I do. 2 Q And then further down in that paragraph 3 you have another bolded sentence, "Another 4 method is to incorporate the differentiator 5 tag sequence into a PCR primer." 6 Do you see that? 7 A I do. 8 Q And I'd like to talk about the 9 mechanics, the differences of these two-ways 10 to incorporate the differentiator tag 11 sequence. 12 But is it accurate to say that with 13 ligation the fragments and adapters have 14 blunt ends and there's a ligation reaction 15 that sticks them together? 16 A I think that can be one way to do it. 17 Q How else does ligation work? 18 A Ligation works in a lot of ways, blunt 19 end ligation is one. There's a ligation 20 method called TA ligation, where a single A 21 base is added in a non-template way on the 22 ends of a fragment, and then that is</p>

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Conducted on July 31, 2025

<p>57</p> <p>1 hybridized with an adapter that has a</p> <p>2 T-overhang and a ligation reaction can occur</p> <p>3 in that way.</p> <p>4 Restriction enzymes have been frequently</p> <p>5 used for creating what we call sticky ends.</p> <p>6 And sticky ends can both occur on the DNA</p> <p>7 fragment of interest and on the adapters.</p> <p>8 And those come together, again, by</p> <p>9 hybridization, and then the gaps are closed</p> <p>10 by DNA ligase.</p> <p>11 So those are just some examples of how a</p> <p>12 ligation reaction can take place.</p> <p>13 Q You said where a single A base is added</p> <p>14 in a non-template way.</p> <p>15 What do you mean by "non-template way"?</p> <p>16 A So earlier I was describing for you the</p> <p>17 extension reaction where you have a primer</p> <p>18 that's hybridized to a single strand of</p> <p>19 template DNA that creates a partially double</p> <p>20 stranded. The polymerase will then use the</p> <p>21 single stranded portion of the template to</p> <p>22 copy the information from the template strand</p>	<p>59</p> <p>1 adapter-related sequences depending on what</p> <p>2 your application is for the target of</p> <p>3 interest.</p> <p>4 Q Okay. Thank you.</p> <p>5 And just so we're on the same page, the</p> <p>6 portion of Figure 4, Figure 4C that's</p> <p>7 reproduced at the bottom of Page 62, is</p> <p>8 that -- would you understand that to depict a</p> <p>9 blunt end ligation?</p> <p>10 A Yes. This example appears to be a blunt</p> <p>11 end ligation example.</p> <p>12 Q Okay. If we could look at Limitation</p> <p>13 1G, that starts at Paragraph 86 of your</p> <p>14 declaration.</p> <p>15 A I'm there.</p> <p>16 Q Okay. Limitation 1G is a limitation</p> <p>17 that recites identifying sequence reads with</p> <p>18 identical identifier and target sequences as</p> <p>19 duplicates.</p> <p>20 Do you see that?</p> <p>21 A I do see that sentence.</p> <p>22 Q So it's your opinion that Porreca</p>
<p>58</p> <p>1 into the new strand, and that's called</p> <p>2 template-dependent DNA polymerase.</p> <p>3 An independent addition is where there's</p> <p>4 no template available for the polymerase to</p> <p>5 copy from, this is the phenomenon, for</p> <p>6 example, taq polymerase has been used widely</p> <p>7 to add a single A base on the end of a</p> <p>8 fragment in a non-template way.</p> <p>9 Q Understood, thank you.</p> <p>10 And in this section, in the second</p> <p>11 bolded sentence in the paragraph that's</p> <p>12 quoted on Page 62 of your declaration, can</p> <p>13 you explain to me how you would incorporate</p> <p>14 the differentiator tag sequence into a PCR</p> <p>15 primer?</p> <p>16 A Yes. So both of these highlighted</p> <p>17 sections are widely-used methods in molecular</p> <p>18 biology, they've been used for decades.</p> <p>19 We've gone over this in Figure 4B, where we</p> <p>20 put a five prime tail on the end of a</p> <p>21 target-specific primer, and that tail can</p> <p>22 incorporate the differentiator tag and other</p>	<p>60</p> <p>1 teaches that differentiator tags can be used</p> <p>2 to identify whether particular nucleic acid</p> <p>3 molecules are copies of the same molecule,</p> <p>4 right?</p> <p>5 A Could you repeat that question.</p> <p>6 Q Sure. Let me rephrase it.</p> <p>7 If we look at the last sentence of</p> <p>8 Paragraph 86 of your declaration, just so</p> <p>9 we're on the same page, it reads, "Because</p> <p>10 each nucleic acid is tagged with a unique</p> <p>11 differentiator tag prior to amplification,</p> <p>12 molecules having identical nucleic acid</p> <p>13 sequences and bearing the same differentiator</p> <p>14 tags are duplicates of one another."</p> <p>15 Do you see that?</p> <p>16 A I do see that sentence.</p> <p>17 Q Does Porreca explain how the</p> <p>18 differentiator tag and the sequences are used</p> <p>19 to identify duplicates?</p> <p>20 A Yes.</p> <p>21 Q Where is that?</p> <p>22 A So, for example, if you go to</p>

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Conducted on July 31, 2025

<p>61</p> <p>1 Paragraph 85, I show an illustration of 2 Figure 5, where Porreca is showing pretty 3 much the overall workflow of characterizing a 4 library and identifying duplicate sequences. 5 And so on the left-hand side, Porreca is 6 starting with genomic DNA and they've made 7 their library where they've attached their 8 different DNA tags to the ends of DNA 9 fragments. And those DNA fragments are 10 illustrated by vertical lines. And then all 11 those vertical lines represent identical 12 sequences. And then they're highlighting a 13 particular locus that has either a T or an A. 14 That's their starting material, which 15 they then amplify and they're illustrating 16 some of the bias, such as creating duplicate 17 amplicons derived from the same DNA fragment 18 that's been tagged with a differentiator tag. 19 Identifying those duplicates and then 20 collapsing those -- the reads and the 21 differentiator tags into single consensus 22 sequences, based on both the identical target</p>	<p>63</p> <p>1 Q Does Porreca explain which element that 2 is sorted, which is done first in their 3 steps? 4 MR. BILSKER: Objection. Form. 5 A I guess I don't understand the question. 6 Q Right. 7 A Because I understand Porreca performs 8 sequencing on its library of fragments, and 9 generates sequence reads. And a single 10 sequence read would have both the target 11 sequence and the identifier sequence 12 together. 13 Q So there's a world where Porreca, the 14 way they analyze the data, Porreca first 15 sorts by the identifier sequence and then 16 sorts by the sequence; is that fair? 17 MR. BILSKER: Objection form. 18 A I'm not sure. Because when I look at 19 Figure 5, it is showing the alignment of 20 different reads that contain both the target 21 sequence and the identifier at the same time. 22 Q But it's possible that in this arrow of</p>
<p>62</p> <p>1 sequence and the differentiator tag. 2 MR. BILSKER: Can I just say one 3 thing. You said vertical lines, I 4 look at those lines -- 5 THE WITNESS: Sorry, horizontal lines. 6 MR. BILSKER: Okay. Just for the 7 record. 8 THE WITNESS: Understood. 9 MR. BILSKER: Or straight lines, I 10 think is probably what you're thinking 11 about, right. 12 Q The only lines, essentially, in Figure 5 13 are horizontal lines. 14 A Horizontal lines. 15 Q Understood. Let's break that down a 16 little bit. 17 It's your understanding that the longer, 18 if we look at Figure 5, the longer of the 19 horizontal lines, those all represent 20 identical sequences? 21 A Yes. That's commonly depicted in 22 sequence alignments.</p>	<p>64</p> <p>1 collapsing differentiator tag sequences, 2 Porreca first sorts by differentiator tag and 3 then looks at the nucleic acids that it's 4 depicted in the figure; is that accurate? 5 MR. BILSKER: Objection. Form. 6 A That's not my understanding of how 7 Porreca does this. 8 Q Where, in Porreca, does it explain how 9 that collapsing is done? 10 A Well, it's illustrated here in the 11 figure. I think the figure clearly 12 illustrates that it's looking at both the 13 target sequence and the differentiator tags 14 at the same time, to then collapse the reads 15 into consensus sequences. 16 So they're removing the duplicates and 17 ending up with the right proportion of target 18 sequences that they started with on the 19 left-hand side of the figure. 20 Q So based on the analysis that's done in 21 Porreca, how would a person of ordinary skill 22 know that the horizontal lines depicted in</p>

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Conducted on July 31, 2025

<p>65</p> <p>1 the figure are identical?</p> <p>2 A There are a number of ways to do it.</p> <p>3 One way is to do sequence alignment. Porreca</p> <p>4 is showing more than two, so we would call</p> <p>5 that a multiple sequence alignment. And</p> <p>6 there are many programs widely available to</p> <p>7 do such an analysis.</p> <p>8 Q Does Porreca explain how it compares</p> <p>9 sequences to determine if they're identical?</p> <p>10 A Well, I think the figure speaks for</p> <p>11 itself. And this is so common in the field</p> <p>12 that a person of ordinary skill in the art,</p> <p>13 just looking at the figure, would know how</p> <p>14 Porreca is doing this.</p> <p>15 Q So Porreca depicts single letters in --</p> <p>16 as a kind of break in the horizontal lines</p> <p>17 that we've been looking at.</p> <p>18 Do you have an understanding of why</p> <p>19 Porreca would have only represented a single</p> <p>20 nucleotide, here?</p> <p>21 A Sure.</p> <p>22 Q And why is that?</p>	<p>67</p> <p>1 Q I can point you to, I think it's</p> <p>2 Paragraph 127 I have in my notes of your</p> <p>3 declaration.</p> <p>4 A Yes. I'm there.</p> <p>5 Q So your discussion of Schmitt starts</p> <p>6 around Paragraph 100.</p> <p>7 But let's first start with -- okay.</p> <p>8 What is Exhibit 1007?</p> <p>9 A Well, it's a PCT patent application by</p> <p>10 Michael Schmitt, Jesse Salk and Larry Loeb</p> <p>11 that is titled Methods for Lowering the Error</p> <p>12 Rate of Massively Parallel DNA Sequencing</p> <p>13 Using Duplex Consensus Sequencing.</p> <p>14 Q And this is the Schmitt application that</p> <p>15 you rely on in your declaration?</p> <p>16 A Yes.</p> <p>17 Q So if we could look at your discussion</p> <p>18 of Limitation 1G, which starts at</p> <p>19 Paragraph 127 in your declaration. Just for</p> <p>20 the record, Limitation 1G, again, is</p> <p>21 identifying sequence reads with identical</p> <p>22 identifier and target sequences as</p>
<p>66</p> <p>1 A Well, so the region of interest is made</p> <p>2 up of many nucleotides, but within that</p> <p>3 region, there may be one variable site. And</p> <p>4 in this case, it would be a single nucleotide</p> <p>5 variant, we call it an SNV, and what Porreca</p> <p>6 is showing that two fragments have one copy,</p> <p>7 or one variant, T, these are letters that</p> <p>8 stand for the basis of DNA, and that the</p> <p>9 other variant is an A.</p> <p>10 And so we would call this a single</p> <p>11 nucleotide variant. And they're just</p> <p>12 illustrating the variant in these reads in</p> <p>13 Porreca Figure 5.</p> <p>14 Q Let's shift gears a little bit and talk</p> <p>15 about another reference that you testified</p> <p>16 about, and that's the Schmitt reference. And</p> <p>17 I'll hand you Exhibit 1007.</p> <p>18 (Whereupon, PCT Application No.</p> <p>19 WO 2013/142389 A1 was previously</p> <p>20 marked as Exhibit 1007.)</p> <p>21 Q Do you recognize Exhibit 1007?</p> <p>22 A I do.</p>	<p>68</p> <p>1 duplicates.</p> <p>2 Now, you rely -- I guess let's take a</p> <p>3 step back. In this section of your</p> <p>4 declaration, you talk about your opinion that</p> <p>5 the claims of the '357 Patent are obvious in</p> <p>6 light of a combination Schmitt and an article</p> <p>7 by McCloskey.</p> <p>8 Does that ring a bell?</p> <p>9 A Yes. But McCloskey is also a patent</p> <p>10 application.</p> <p>11 Q Apologies. Thank you for that</p> <p>12 clarification.</p> <p>13 And you rely on Schmitt for certain</p> <p>14 limitations and McCloskey for certain</p> <p>15 limitations; is that fair?</p> <p>16 MR. BILSKER: Objection. Form.</p> <p>17 A McCloskey for one limitation.</p> <p>18 Q Okay. So Limitation 1G, you only point</p> <p>19 to disclosures of Schmitt; is that accurate?</p> <p>20 A That's correct. Although that's not to</p> <p>21 say that those limitations cannot be found in</p> <p>22 McCloskey, but I only cite to Schmitt here in</p>

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Conducted on July 31, 2025

<p>69</p> <p>1 my declarations.</p> <p>2 Q Okay. And am I correct that you</p> <p>3 interpret the SMI tag in Schmitt to be the</p> <p>4 identifier sites of the claims?</p> <p>5 A I do, and I'm just looking at</p> <p>6 Paragraph 100 of my declaration, which has a</p> <p>7 table at the bottom of Page 78. And even</p> <p>8 though it says -- here's one of these typos,</p> <p>9 "the '241 Patent." It should be "the</p> <p>10 '357 Patent." And it's showing the</p> <p>11 identifier site being the equivalent of the</p> <p>12 single molecule identifier, or SMI, in</p> <p>13 Schmitt and the barcode in the McCloskey</p> <p>14 patent application.</p> <p>15 Q Okay. Now, on Paragraph 127 you cite to</p> <p>16 a section of Schmitt, and I'm looking at the</p> <p>17 middle of Page 98 where you're quoting</p> <p>18 from -- it looks like Paragraph 85 of</p> <p>19 Schmitt. This is a section right above</p> <p>20 Paragraph 128.</p> <p>21 Do you see that?</p> <p>22 A So are you in the second block quote?</p>	<p>71</p> <p>1 those five reads have the same SMI and have</p> <p>2 identical target sequences. So both of those</p> <p>3 are being evaluated in Schmitt in</p> <p>4 characterizing true PCR duplicates.</p> <p>5 Q So underneath of that figure, you've</p> <p>6 quoted, according to the footnote,</p> <p>7 Paragraph 14 of Schmitt. There's a quote,</p> <p>8 "A-C shows sequence reads brown sharing a</p> <p>9 unique set of SMI tags are grouped into</p> <p>10 paired families with members having strand</p> <p>11 identifiers in either the AB or BA</p> <p>12 orientation," or Alpha Beta or Beta Alpha</p> <p>13 orientation.</p> <p>14 Do you see that?</p> <p>15 A I do see that sentence.</p> <p>16 Q Okay. Now, can you point to anything in</p> <p>17 the description that explains that sequence</p> <p>18 reads are grouped into paired families by</p> <p>19 anything other than the SMI tags?</p> <p>20 MR. BILSKER: Objection. Asked and</p> <p>21 answered.</p> <p>22 A Again, I would point you to what's being</p>
<p>70</p> <p>1 Q I'm in the second block quote, yes.</p> <p>2 A Okay. Yes. Then I do see that.</p> <p>3 Q Okay. And at the bottom -- at the end</p> <p>4 of that section that's quoted at the end of</p> <p>5 Paragraph 127, you've bolded the statement</p> <p>6 "True PCR duplicates may be unambiguously</p> <p>7 identified by virtue of having a common</p> <p>8 (i.e., the same or identical) SMI sequence."</p> <p>9 Do you see that?</p> <p>10 A I do.</p> <p>11 Q Do you recall any mention in Schmitt</p> <p>12 where the PCR duplicates are sorted into</p> <p>13 families by anything other than the SMI tag?</p> <p>14 MR. BILSKER: Objection. Form.</p> <p>15 A Yes. So for example, if you go to</p> <p>16 Page 95, and I'm looking in Paragraph 125 at</p> <p>17 Figure 3, Schmitt is showing that the reads</p> <p>18 are being sorted and characterized both by</p> <p>19 the SMI and the target sequence.</p> <p>20 And so for example, looking at 3A, the</p> <p>21 top portion where the Alpha is on the left</p> <p>22 and the Beta is on the right, four out of</p>	<p>72</p> <p>1 depicted in Figure 3. I mean, it's clearly</p> <p>2 showing the grouping of the SMI tags and the</p> <p>3 sequences, which are -- in some cases are</p> <p>4 identical, in some cases they're not</p> <p>5 identical in the sequence alignments.</p> <p>6 Q But that's not what the text says,</p> <p>7 right?</p> <p>8 MR. BILSKER: Objection. Form.</p> <p>9 A Well, I think the text is just</p> <p>10 explaining part of what's in the figure.</p> <p>11 It's not a comprehensive explanation at all.</p> <p>12 For example, it refers to "brown," and the</p> <p>13 figures aren't brown.</p> <p>14 But again, looking at the figure, it's</p> <p>15 very obvious -- it would be obvious to a</p> <p>16 person of ordinary skill in the art that</p> <p>17 what's being displayed here are not just the</p> <p>18 SMI sequences but the actual target sequences</p> <p>19 themselves in showing where the sequences are</p> <p>20 identical and where the sequences differ.</p> <p>21 MS. CIPRIANO: Okay. If we can take a</p> <p>22 quick break. I might be almost done.</p>

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<p>1 THE WITNESS: Okay.</p> <p>2 (Whereupon, a recess was taken</p> <p>3 from 11:22 AM until 11:33 AM.)</p> <p>4 MS. CIPRIANO: I have no further</p> <p>5 questions for you on the UMI.</p> <p>6 THE WITNESS: Thank you.</p> <p>7 MR. BILSKER: I have no questions.</p> <p>8 (Whereupon, this examination was</p> <p>9 concluded at 11:33 AM.)</p> <p>10</p> <p>11</p> <p>12 _____</p> <p>13 MICHAEL L. METZKER PH.D.</p> <p>14</p> <p>15</p> <p>16 Subscribed and sworn to</p> <p>17 before me on this ____ day</p> <p>18 of _____, _____.</p> <p>19</p> <p>20 _____</p> <p>21 Notary Public</p> <p>22</p>	<p>1 C E R T I F I C A T E</p> <p>2</p> <p>3 I, KIARA MILLER,</p> <p>4 a Shorthand Reporter and Notary Public of the</p> <p>5 State of New York, do hereby certify:</p> <p>6</p> <p>7 That the witness whose examination is</p> <p>8 hereinbefore set forth, was duly sworn or</p> <p>9 affirmed by me, and the foregoing transcript is</p> <p>10 a true record of the testimony given by such</p> <p>11 witness.</p> <p>12</p> <p>13 I further certify that I am not related to any</p> <p>14 of the parties to this action by blood or</p> <p>15 marriage, and that I am in no way interested in</p> <p>16 the outcome of this matter.</p> <p>17</p> <p>18 <i>Kiara Miller</i></p> <p>19 _____</p> <p>20 KIARA MILLER</p> <p>21</p> <p>22</p>
<p>1 I N D E X</p> <p>2 WITNESS: MICHAEL METZKER</p> <p>3 EXAMINATION BY PAGE</p> <p>4 MS. CIPRIANO 4</p> <p>5</p> <p>6</p> <p>7 PREVIOUSLY DESCRIPTION PAGE</p> <p>8 MARKED</p> <p>9 Exhibit 1003 Declaration of 8</p> <p>10 Michael L. Metzker,</p> <p>11 Ph.D., Case</p> <p>12 IPR2025-00026</p> <p>13 Exhibit 1016 Metzker CV 11</p> <p>14 Exhibit 1006 US Patent 35</p> <p>15 Application</p> <p>16 Publication No. US</p> <p>17 2012/0165202 A1 by</p> <p>18 Porreca et al.</p> <p>19 Exhibit 1007 PCT Application No. 66</p> <p>20 WO 2013/142389 A1</p> <p>21</p> <p>22</p>	

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a-c	actual	advisor	already
71:8	13:12, 37:6,	3:18	42:21, 42:22
al	72:18	affect	also
34:21, 66:19,	actually	36:4	3:17, 12:7,
74:17, 74:20	22:16, 22:17,	affirmed	23:3, 24:14,
ab	32:13, 36:20,	75:9	29:11, 33:1,
71:11	42:2, 42:18,	affordable	34:8, 68:9
abbreviated	44:1	51:1	although
27:12	adapter	after	68:20
able	30:11, 30:16,	4:1, 10:3,	ambiguous
7:1	30:20, 31:1,	45:7, 45:17,	20:2, 20:20
about	31:5, 31:8,	45:21	amplicons
7:16, 7:17,	31:10, 32:3,	again	55:8, 61:17
12:10, 13:14,	32:10, 32:17,	18:16, 22:4,	amplification
13:15, 14:19,	33:3, 34:5,	22:15, 23:19,	60:11
18:2, 19:2,	39:9, 40:10,	23:21, 43:11,	amplify
24:12, 24:13,	40:12, 40:17,	46:10, 47:14,	19:11, 19:16,
26:2, 29:4,	40:21, 41:6,	57:8, 67:20,	19:20, 46:4,
46:5, 54:15,	41:14, 41:21,	71:22, 72:14	46:9, 51:11,
56:8, 62:11,	42:8, 42:11,	agilent	61:15
66:15, 66:16,	42:21, 43:7,	14:22, 15:16	amplifying
68:4	43:13, 46:16,	agree	27:1, 46:5,
above	47:11, 47:15,	53:9, 53:15	55:7
33:3, 69:19	47:19, 48:8,	agreed	analyses
absolutely	48:10, 48:12,	53:4, 53:8	35:13, 36:5
17:15, 31:20	48:19, 57:1	ahead	analysis
abstract	adapter-ligated	14:1	18:19, 32:17,
35:9	55:8	al	34:1, 35:22,
accomplished	adapter-related	34:22, 74:18	36:8, 64:20,
35:15	59:1	alignment	65:7
according	adapters	63:19, 65:3,	analyze
15:4, 71:6	31:11, 31:13,	65:5	28:7, 63:14
accurate	31:16, 31:17,	alignments	analyzed
27:19, 40:11,	31:20, 48:4,	62:22, 72:5	36:20
50:8, 51:19,	49:7, 56:13,	all	analyzing
56:12, 64:4,	57:7	15:3, 17:12,	12:21, 13:6
68:19	adaptors	17:13, 24:1,	another
acid	53:21	24:9, 24:17,	11:21, 27:14,
28:5, 39:10,	add	27:5, 36:6,	38:16, 53:7,
60:2, 60:10,	58:7	47:8, 61:10,	56:3, 60:14,
60:12	added	62:19, 72:11	66:15
acids	56:21, 57:13	almost	answer
55:20, 64:3	addition	17:12, 17:13,	5:10, 5:11,
action	24:11, 40:18,	72:22	5:22, 6:5, 6:12,
75:14	51:22, 58:3	along	7:1
activities	advertisement	24:15	answered
9:11	18:7	alpha	48:5, 71:21
	advertisements	70:21, 71:12	answers
	17:2		5:4

<p>anticipate 33:7</p> <p>any 6:5, 6:22, 7:6, 7:20, 10:17, 11:13, 27:20, 30:12, 33:4, 33:6, 33:11, 33:17, 34:1, 37:1, 45:14, 45:17, 48:3, 52:17, 52:19, 70:11, 75:13</p> <p>anyone 7:14, 9:20</p> <p>anything 10:6, 10:13, 29:1, 53:11, 70:13, 71:16, 71:19</p> <p>apologies 68:11</p> <p>appeal 1:3</p> <p>appear 42:19</p> <p>appears 59:10</p> <p>appended 42:12, 43:1, 46:16, 47:11, 48:8</p> <p>appending 41:6</p> <p>applicable 28:12</p> <p>application 18:11, 34:20, 59:2, 66:18, 67:9, 67:14, 68:10, 69:14, 74:15, 74:19</p> <p>applications 15:11</p> <p>applied 29:8, 29:20</p> <p>appreciate 52:12, 53:12,</p>	<p>53:20</p> <p>archer 16:14, 17:20</p> <p>area 12:11, 12:13, 12:22, 26:1</p> <p>aren't 72:13</p> <p>ariosa 16:19, 18:1</p> <p>arm 43:10, 44:8, 44:9</p> <p>arms 41:18, 41:20, 42:19, 42:22, 43:1, 43:2, 43:19, 43:20, 44:6, 46:20, 47:3, 47:7, 54:12</p> <p>around 15:8, 43:7, 44:1, 44:2, 44:7, 44:10, 44:17, 55:2, 67:6</p> <p>arrive 29:7</p> <p>arrow 21:19, 63:22</p> <p>art 28:21, 29:5, 29:13, 29:21, 65:12, 72:16</p> <p>article 68:6</p> <p>articles 34:11</p> <p>artisan 53:19</p> <p>aside 19:1</p> <p>asked 71:20</p> <p>asking 5:3, 6:17, 9:11</p> <p>assistant 14:6</p>	<p>associated 13:8</p> <p>assume 6:13</p> <p>assumed 28:19, 29:1</p> <p>attach 31:3</p> <p>attached 30:12, 42:22, 46:20, 48:11, 61:7</p> <p>attaching 44:19</p> <p>attorney 33:15</p> <p>attorneys 6:19, 9:21</p> <p>attribute 53:6</p> <p>available 15:14, 58:4, 65:6</p> <p>avenue 2:2, 3:13</p> <p>awarded 14:13</p> <p>aware 10:9, 33:1</p> <hr/> <p style="text-align: center;">B</p> <hr/> <p>ba 71:11</p> <p>back 11:2, 14:4, 25:6, 36:17, 38:21, 54:20, 54:22, 68:3</p> <p>background 19:3, 26:3</p> <p>bacteria 27:6, 27:8</p> <p>bacterias 13:8</p> <p>bar 49:18, 49:22, 50:3, 50:4, 50:7, 50:17,</p>	<p>51:22</p> <p>barcode 69:13</p> <p>base 22:6, 36:21, 37:8, 56:21, 57:13, 58:7</p> <p>based 14:8, 29:11, 33:8, 43:13, 61:22, 64:20</p> <p>bases 36:19</p> <p>basic 21:15, 22:19, 23:21, 37:17</p> <p>basics 21:8</p> <p>basis 33:9, 66:8</p> <p>baylor 14:4, 14:14</p> <p>bearing 60:13</p> <p>because 31:21, 42:16, 48:18, 50:21, 52:15, 60:9, 63:7, 63:18</p> <p>become 15:13, 25:16, 48:20</p> <p>been 4:2, 4:13, 5:10, 8:5, 11:17, 12:14, 13:1, 15:22, 22:7, 23:8, 24:12, 24:13, 35:2, 45:22, 50:13, 51:4, 51:8, 54:14, 57:4, 58:6, 58:18, 61:18, 65:17</p> <p>before 1:3, 2:4, 4:11, 4:13, 4:22, 5:1,</p>
---	---	---	--

6:6, 16:1, 22:17, 50:13, 73:17 behalf 2:2, 3:3, 3:10 being 20:14, 25:19, 69:11, 70:18, 71:3, 71:22, 72:17 believe 10:5, 29:18, 53:13 bell 68:8 below 43:18 beta 70:22, 71:12 better 28:6 between 11:13, 13:1, 25:20, 38:22 bias 35:12, 35:17, 36:7, 37:12, 61:16 biases 36:3 big 53:5 bilsker 3:8, 7:13, 20:1, 20:19, 22:14, 23:10, 23:18, 25:21, 26:17, 29:22, 30:18, 32:5, 32:18, 33:14, 33:21, 35:8, 36:13, 42:13, 44:22, 45:10, 48:13, 50:9, 51:20, 52:13, 52:15, 52:19, 52:22, 53:5, 53:9, 54:17,	62:2, 62:6, 62:9, 63:4, 63:17, 64:5, 68:16, 70:14, 71:20, 72:8, 73:7 bind 20:17, 24:21 binds 45:7 bio-rad 16:10 biology 12:17, 28:3, 58:18 bioscience 17:4 biosciences 12:7, 18:12 biotech 12:6 biotechnology 12:16 bit 13:14, 13:15, 19:2, 20:22, 62:16, 66:14 block 69:22, 70:1 blood 18:2, 75:14 blue 22:2 blunt 56:14, 56:18, 59:9, 59:10 board 1:3 bolded 55:17, 56:3, 58:11, 70:5 bond 43:5 books 28:3 both 7:13, 53:15, 57:6, 58:16,	61:22, 63:10, 63:20, 64:12, 70:18, 71:2 bottom 59:7, 69:7, 70:3 break 6:4, 6:6, 36:16, 43:4, 54:16, 62:15, 65:16, 72:22 breaking 36:18 breaks 6:3 brief 37:15 broad 12:13, 19:13, 20:3, 26:9, 26:13 brought 14:20 brown 71:8, 72:12, 72:13 buffers 24:16 build 14:8 building 3:12, 28:2 built 15:7 <hr/> C <hr/> california 3:5, 3:7 call 9:9, 19:21, 20:17, 25:2, 25:10, 25:16, 28:3, 36:6, 37:7, 40:15, 40:16, 47:18, 52:4, 57:5, 65:4, 66:5, 66:10	called 12:6, 21:4, 23:13, 24:14, 26:10, 27:18, 40:20, 49:18, 50:8, 51:3, 51:9, 54:10, 56:20, 58:1 cambridge 16:20, 18:5 came 14:4, 15:20, 26:16, 34:9 can't 5:21, 23:7, 44:4, 44:13 cancer 13:5, 15:18 cannot 68:21 capture 44:5 care 13:7 case 8:15, 9:18, 16:11, 16:12, 16:13, 16:14, 16:15, 16:17, 16:18, 16:19, 16:21, 17:1, 17:2, 17:3, 17:5, 17:7, 17:10, 17:11, 29:8, 66:4, 74:11 cases 16:9, 72:3, 72:4 cell-free 18:3 ceo 12:6 certain 13:3, 68:13, 68:14 certify 75:5, 75:13
---	--	--	---

chain 19:6 change 10:7, 10:14, 29:19 changes 10:17, 11:22 chapter 21:7 characterized 45:3, 46:11, 47:9, 70:18 characterizing 13:2, 61:3, 71:4 chemically 14:15 chemistry 15:7 chose 21:12 cipriano 3:15, 4:5, 4:10, 52:14, 52:18, 53:4, 53:8, 53:12, 54:14, 72:21, 73:4, 74:4 circle 25:4, 25:10, 42:18, 45:16, 45:22, 46:8, 46:9, 46:18, 46:19, 47:6 circular 45:18 cite 49:15, 68:22, 69:15 cited 8:6 claim 30:3, 33:12, 33:18 claims 68:5, 69:4 clarification 68:12	clarify 20:22 clear 6:11 clearly 5:17, 64:11, 72:1 clinical 15:17 cloned 27:4, 27:6 cloning 31:17, 48:2 closed 57:9 code 37:7, 38:14, 38:17, 49:18, 49:22, 50:4, 50:7 codes 50:3, 51:22 coding 50:17 collapse 64:14 collapsing 61:20, 64:1, 64:9 collection 18:2, 18:10, 18:16, 28:4 college 14:4 combination 68:6 come 11:2, 30:1, 51:12, 57:8 comes 21:6, 32:14 coming 29:11, 44:4 commencing 2:3 commercialize 15:17 commercially 15:14	common 40:20, 51:9, 65:11, 70:7 commonalities 26:15, 26:19 commonly 26:11, 50:11, 50:20, 62:21 communications 7:7 company 12:6, 12:8, 13:18, 14:8, 14:21 compares 65:8 complement 25:1 complementary 22:8 complete 16:16, 16:17, 17:22 complex 23:6, 24:20 compositions 35:11 comprehensive 72:11 comprising 55:20 concepts 19:5, 21:15 concluded 33:4, 73:9 conclusions 30:2 connected 41:20 consensus 61:21, 64:15, 67:13 consider 12:11, 29:10 considered 29:9, 33:7 construction 30:4, 33:13, 33:18, 34:4, 34:5 construed 33:10 consult 9:20 contain 42:2, 63:20 containing 43:6 contains 41:21 contemplates 51:16 context 50:17, 51:14 continue 27:2 control 24:16 copied 25:19, 26:1, 47:7, 48:17, 48:18, 49:12 copies 25:17, 38:1, 60:3 copy 38:3, 38:11, 38:15, 40:5, 47:5, 52:9, 57:22, 58:5, 66:6 core 15:7 correct 28:18, 42:6, 68:20, 69:2 corrections 10:17 could 4:6, 11:1, 11:3, 13:15, 16:7, 20:21, 30:3, 34:16, 41:8, 41:12, 46:4, 52:6, 54:22, 59:12,
--	---	---

60:5, 67:17 counsel 5:9, 7:7, 7:9, 9:10, 28:17, 29:10 couple 11:16, 14:3, 26:18, 28:16 course 5:8 court 5:5, 5:14 create 23:6, 24:19, 25:1 creates 25:3, 57:19 creating 36:4, 43:11, 57:5, 61:16 culture 27:7 current 12:4 currently 12:2, 13:22 cv 11:4, 11:12, 16:3, 19:1, 74:13 cycle 22:11, 25:10, 25:13 cycles 25:16	days 27:10 deal 53:5 decades 58:18 decided 14:7, 15:21 decision 10:11 declaration 8:5, 8:6, 8:7, 8:14, 8:21, 9:8, 9:16, 9:22, 10:4, 10:8, 10:19, 11:2, 15:5, 19:3, 20:7, 21:3, 28:11, 28:19, 30:4, 34:12, 35:5, 38:22, 39:4, 40:4, 49:3, 49:13, 54:22, 58:12, 59:14, 60:8, 67:3, 67:15, 67:19, 68:4, 69:6, 74:9 declarations 69:1 decoding 37:6 define 26:1 defines 32:9 definition 29:20, 32:16, 33:3, 34:7 degrade 45:14 degraded 45:18, 46:10 denature 22:17, 23:22, 37:19 denatured 20:17, 23:8	denaturing 22:12 depending 59:1 depends 31:9 depict 22:10, 41:18, 59:8 depicted 40:5, 42:5, 43:16, 47:12, 47:19, 54:13, 62:21, 64:4, 64:22, 72:1 depicting 25:12 depiction 22:5 depicts 20:12, 65:15 deposed 4:13 deposition 1:14, 2:1, 4:12, 5:8, 7:4, 7:12, 7:21 derived 28:6, 61:17 describe 22:18, 35:7 described 21:9, 22:19, 31:1, 38:10, 47:2 describes 35:14 describing 9:10, 42:3, 48:14, 57:16 description 71:17, 74:7 descriptions 51:4 design 22:7, 23:5 designed 22:8, 23:4,	37:21, 43:21 determine 65:9 develop 13:7, 15:9, 35:11 developed 13:12 developing 12:21 development 12:15, 12:18 diabetes 13:4 diagnostics 13:5, 15:18, 16:19 differ 72:20 differences 56:9 different 19:14, 26:9, 28:8, 29:16, 33:19, 36:21, 41:5, 45:20, 46:11, 46:12, 50:19, 51:18, 61:8, 63:20 differentiator 40:16, 42:1, 47:20, 55:20, 56:4, 56:10, 58:14, 58:22, 60:1, 60:11, 60:13, 60:18, 61:18, 61:21, 62:1, 64:1, 64:2, 64:13 disagree 50:12 discloses 39:16 disclosures 39:15, 68:19 discuss 6:18, 34:12, 39:7
D			
damages 36:18 data 18:19, 63:14 dated 11:12 david 3:8, 7:13 day 7:9, 7:18, 73:17			

discussion 67:5, 67:17 diseases 13:3 displayed 72:17 dna 15:11, 17:2, 17:8, 17:16, 17:19, 18:3, 18:9, 18:11, 18:13, 18:15, 19:17, 20:17, 21:5, 21:13, 21:17, 22:9, 22:17, 23:8, 23:13, 23:14, 23:15, 23:22, 24:5, 24:11, 24:14, 25:4, 25:5, 25:7, 25:14, 27:5, 28:4, 36:16, 36:18, 37:20, 38:10, 38:11, 38:12, 38:15, 38:18, 41:7, 42:12, 50:4, 57:6, 57:10, 57:19, 58:2, 61:6, 61:8, 61:9, 61:17, 66:8, 67:12 document 8:18, 9:2, 11:9 documents 7:20, 8:2, 9:12 doing 23:19, 40:14, 65:14 done 12:15, 13:4, 13:5, 13:11, 50:20, 63:2, 64:9, 64:20, 72:22 double 21:4, 21:12,	21:16, 21:17, 22:21, 23:2, 23:6, 23:8, 23:14, 24:20, 25:4, 25:6, 25:15, 44:2, 44:20, 57:19 double-stranded 37:20 down 5:15, 16:8, 17:18, 25:5, 34:10, 56:2, 62:15 downstream 45:3 dozens 4:16 dr 22:20 draw 43:17 drawn 42:15, 47:1 duly 4:2, 75:8 duplex 67:13 duplicate 61:4, 61:16 duplicates 36:4, 59:19, 60:14, 60:19, 61:19, 64:16, 68:1, 70:6, 70:12, 71:4 duplication 38:4 uplications 37:5, 37:14 during 5:8, 14:7, 33:1, 33:13 <hr/> E <hr/> e 52:2 each 24:5, 25:13,	31:21, 39:10, 60:10 earlier 57:16 eight 7:18 eighth 3:13 either 61:13, 71:11 electrophoresis 26:20 element 63:1 else 56:17 emanuel 3:4 employed 12:2 employment 11:22 end 27:1, 39:21, 40:21, 41:7, 42:12, 48:10, 56:19, 58:7, 58:20, 59:9, 59:11, 70:3, 70:4 ending 8:8, 64:17 ends 40:9, 40:18, 41:17, 53:22, 56:14, 56:22, 57:5, 57:6, 61:8 enrich 45:2, 46:1 entire 34:7, 34:8 entitled 28:12 enzyme 24:13, 48:2 enzymes 38:11, 57:4 equivalent 69:11	error 10:21, 38:20, 67:11 errors 36:3, 36:4, 36:11, 37:4, 37:10, 38:8 esq 3:8, 3:15 essentially 62:12 et 34:21, 74:18 evaluated 71:3 even 69:7 ever 15:13, 32:3 everything 5:15, 25:13 exactly 48:5 examination 4:4, 73:8, 74:3, 75:7 examined 4:3 examiner 33:2, 33:19, 34:4 examiners 33:12 example 22:7, 23:20, 23:22, 25:22, 26:19, 35:20, 36:3, 36:15, 36:22, 37:15, 38:3, 38:5, 42:21, 46:7, 46:18, 52:1, 54:13, 55:19, 58:6, 59:10, 59:11, 60:22, 70:15, 70:20, 72:12 examples 13:10, 31:14,
--	---	--	---

36:11, 37:1, 57:11 exceptions 17:14 excuse 30:6 exhibit 8:12, 8:17, 11:3, 11:5, 11:8, 16:3, 34:18, 35:1, 35:3, 41:10, 52:16, 66:17, 66:20, 66:21, 67:8, 74:9, 74:13, 74:14, 74:19 exonuclease 45:12, 45:13, 45:14 expect 5:11 experience 11:15, 16:4, 29:11, 31:5 expert 16:4 expertise 12:12, 12:13, 28:20 explain 24:7, 41:12, 46:22, 47:10, 58:13, 60:17, 63:1, 64:8, 65:8 explained 19:10 explaining 72:10 explains 71:17 explanation 72:11 exponential 25:16 extend 24:8, 38:1, 43:9	extended 48:17 extending 25:2 extends 24:6 extension 21:20, 25:6, 38:9, 57:17 extent 16:7	40:10, 40:22, 41:1, 41:3, 41:4, 41:8, 41:12, 42:3, 42:4, 42:11, 43:3, 43:15, 46:6, 47:10, 47:14, 48:7, 58:19, 59:6, 61:2, 62:12, 62:18, 63:19, 64:4, 64:11, 64:19, 65:1, 65:10, 65:13, 66:13, 70:17, 71:5, 72:1, 72:10, 72:14 figures 72:13 file 1:22 filed 10:3, 52:10 fill 9:15, 9:16 finalize 9:16 fine 54:17 fingers 44:4 finished 11:18, 14:2 first 4:1, 9:9, 13:17, 13:20, 21:18, 22:11, 25:9, 26:11, 30:9, 39:19, 42:14, 50:3, 51:17, 53:18, 55:19, 63:2, 63:14, 64:2, 67:7 five 7:19, 22:2, 47:18, 48:19, 58:20, 71:1	floor 3:6 fluorescent 13:12 focus 15:2 follows 4:3 footnote 71:6 foregoing 75:9 form 22:14, 23:10, 23:18, 25:21, 26:17, 29:22, 32:5, 32:18, 33:14, 33:21, 35:8, 36:13, 42:13, 44:22, 45:10, 46:17, 46:19, 48:13, 50:9, 51:20, 63:4, 63:17, 64:5, 68:16, 70:14, 72:8 formed 45:15 forming 44:1 forth 38:22, 75:8 found 68:21 foundation 16:13, 17:19, 20:20 founder 12:5 four 7:19, 37:6, 70:22 fragment 40:21, 41:7, 42:12, 56:22, 57:7, 58:8, 61:17 fragments 28:4, 28:5,
	F		
	fair 5:12, 19:7, 19:12, 19:22, 20:18, 23:9, 25:8, 27:12, 27:13, 41:1, 43:20, 44:9, 63:16, 68:15 faithfully 38:16 false 17:1, 18:7 families 70:13, 71:10, 71:18 farms 27:7 fast 51:1 february 11:12, 11:14 few 5:1 field 12:14, 26:9, 26:13, 29:6, 65:11 figure 20:8, 20:14, 21:2, 21:6, 21:11, 21:19, 22:10, 22:16, 24:3, 24:8, 25:3, 25:9, 40:4, 40:5,		

39:11, 55:8, 56:13, 61:9, 63:8, 66:6 francisco 3:7 freezer 27:7 frequently 57:4 full 52:7 fully 7:1 function 31:6, 33:6 functionality 42:2 functions 31:8, 41:22 funding 14:11 further 34:1, 56:2, 73:4, 75:13	18:18, 18:21, 26:8, 26:11, 27:11, 27:16, 50:21 generations 26:3 genetic 19:11, 35:13, 37:7 genome 13:3, 14:10, 14:12, 14:13, 37:9, 37:22 genomic 36:15, 61:6 genomics 1:8, 12:17, 16:16, 16:17, 17:22 genotek 17:2, 17:8, 18:9, 18:15 give 31:14, 34:16, 36:11, 37:15 given 75:10 glycerol 27:8 go 5:1, 25:4, 37:17, 38:21, 46:5, 48:3, 54:22, 60:22, 70:15 goal 46:1 goals 15:8 going 8:11, 17:18, 38:21, 53:6, 54:14, 55:1 gone 58:19 good 4:8, 50:16 goodwin 3:11, 3:18	gotten 11:20 graduate 13:19, 27:9 grand 15:16 grant 11:17, 14:13 ground 4:20 grouped 71:9, 71:18 grouping 72:2 guardant 16:12, 16:22, 17:4, 17:18, 18:6, 18:12 guess 22:1, 48:14, 49:21, 55:18, 63:5, 68:2	helix 44:2, 44:20 helpful 31:16, 31:18 here 7:17, 9:22, 10:19, 11:18, 11:20, 12:5, 25:19, 25:22, 31:1, 35:17, 40:14, 41:22, 46:17, 47:1, 47:20, 48:15, 64:10, 65:20, 68:22, 72:17 here's 69:8 hereby 75:5 hereinbefore 75:8 herron 3:18 high 7:7, 9:7 high-level 22:5 highlighted 55:17, 58:16 highlighting 61:12 highlights 52:18, 52:20 history 34:2, 34:9 hiv 12:22 horizontal 62:5, 62:13, 62:14, 62:19, 64:22, 65:16 hour 54:15 hours 7:18, 7:19 human 14:10 hybridization 43:13, 57:9
G		H	
gap 43:3, 43:4, 43:8, 43:11, 44:5 gaps 57:9 gears 15:21, 66:14 general 22:15, 26:18, 35:7, 47:15 generally 19:21, 27:20 generated 37:8 generates 63:9 generating 12:19 generation 13:22, 14:17, 15:9, 18:9,		half 7:10, 7:18, 7:19, 42:18 hand 8:11, 66:17 handed 11:7, 35:2 handle 46:12 handy 34:17, 39:2 happen 45:19 happened 9:18 happening 24:9 happens 23:16, 45:18, 53:10 head 5:21 health 17:18, 18:6	

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hybridize 23:5, 23:7, 23:15, 24:4, 37:20, 43:2, 43:21 hybridized 24:19, 44:16, 46:20, 48:15, 57:1, 57:18 <hr/> I <hr/> idea 24:18 identical 59:18, 60:12, 61:11, 61:22, 62:20, 65:1, 65:9, 67:21, 70:8, 71:2, 72:4, 72:5, 72:20 identified 39:7, 55:6, 70:7 identifier 59:18, 63:11, 63:15, 63:21, 67:22, 69:4, 69:11, 69:12 identifiers 71:11 identify 52:1, 60:2, 60:19 identifying 13:8, 51:17, 59:17, 61:4, 61:19, 67:21 illumina 16:15, 16:16, 16:18, 16:20, 17:21, 18:1, 18:5 illustrated 41:22, 61:10, 64:10 illustrates 64:12	illustrating 21:8, 40:15, 41:4, 61:15, 66:12 illustration 21:14, 61:1 impactful 11:15 inc 1:8 include 21:12, 40:17, 47:19 included 31:9, 52:1 includes 41:22 incorporate 38:19, 42:7, 56:4, 56:10, 58:13, 58:22 incorporated 55:21 incorporates 41:13 incorporating 24:22 incorrectly 37:7 increase 35:12 increasing 36:7 independent 58:3 index 50:8 indexing 49:6 indicating 44:3 individual 52:2 individuals 13:1, 50:19 information 38:13, 40:3, 57:22	insert 38:19 instance 51:10 instances 38:18 institute 14:10 institution 10:10 instructed 5:10 instrument 13:11, 15:10, 15:17, 52:5 intend 6:12 interest 41:19, 42:5, 42:15, 43:15, 43:22, 44:2, 44:7, 44:11, 44:17, 44:21, 45:2, 45:8, 46:2, 46:16, 46:21, 47:4, 47:8, 47:12, 48:8, 48:11, 48:16, 48:18, 48:21, 51:12, 53:22, 57:7, 59:3, 66:1 interested 14:11, 75:15 interesting 12:9 interpret 69:3 introduce 36:3, 36:10, 37:4, 37:5, 37:13, 38:7, 45:11 introduced 4:10 inversion 54:11 investors 14:20	invitae 17:10, 18:18 involve 17:9 involved 13:2, 13:6, 15:18, 15:22, 17:16, 18:8, 18:13 involves 17:19, 18:19, 18:20 ion 24:17 ipr 1:11, 8:5, 8:13, 8:16, 8:21, 17:9, 74:12 isolate 46:8 isolating 18:3 itc 16:11 itself 12:20, 65:11 <hr/> J <hr/> jesse 67:10 job 38:15 july 1:17, 2:3, 4:19 <hr/> K <hr/> kary 21:9 kiara 1:21, 2:4, 75:3, 75:20 kind 14:22, 21:8, 21:14, 22:4, 22:15, 23:21, 38:21, 65:16 kinds 31:11
--	--	--	--

know 6:4, 16:8, 33:15, 52:11, 64:22, 65:13 known 26:11 <hr/> L <hr/> label 49:5 labeled 21:19 lack 28:5 language 19:8 large 14:13, 27:7 larger 40:22 larry 67:10 lasergen 13:16, 13:17, 14:14, 15:1, 15:2 last 4:19, 7:9, 9:1, 60:7 least 15:4, 21:2, 47:2, 53:14 left 15:4, 70:21 left-hand 61:5, 64:19 legal 28:15, 33:12 let's 4:18, 62:15, 66:14, 67:7, 68:2 letters 37:6, 38:14, 65:15, 66:7 level 7:8, 9:7, 14:21, 25:9,	28:20, 29:13, 29:16 libraries 12:19, 31:18, 32:1 library 27:18, 27:22, 28:2, 28:4, 28:8, 35:21, 53:20, 54:4, 61:4, 61:7, 63:8 ligase 43:11, 44:6, 57:10 ligate 43:6, 43:12, 44:6, 44:13 ligated 44:8, 47:6, 53:21 ligating 39:9 ligation 40:20, 43:5, 55:21, 56:13, 56:14, 56:17, 56:18, 56:19, 56:20, 57:2, 57:12, 59:9, 59:11 light 68:6 likely 4:20, 5:9 limitation 39:8, 39:9, 39:17, 49:1, 49:5, 49:6, 49:10, 55:1, 55:5, 55:6, 55:7, 59:12, 59:16, 67:18, 67:20, 68:17, 68:18 limitations 26:21, 68:14, 68:15, 68:21 limited 16:20	line 47:21 linear 41:16, 42:17, 46:10 lines 22:2, 42:20, 61:10, 61:11, 62:3, 62:4, 62:5, 62:9, 62:12, 62:13, 62:14, 62:19, 64:22, 65:16 linnea 3:15, 4:10 list 16:4, 17:18 listed 11:18, 11:20 litigation 16:1 little 13:14, 19:2, 62:16, 66:14 llc 1:5 llp 3:4, 3:11 loci 35:13 locus 61:13 loeb 67:10 long 7:16, 43:9 longer 62:17, 62:18 look 11:1, 11:3, 20:6, 30:3, 36:20, 39:3, 41:8, 47:10, 48:7, 49:1, 52:6, 53:16, 55:1, 59:12, 60:7, 62:4, 62:18, 63:18,	67:17 looked 8:2, 34:8 looking 34:3, 64:12, 65:13, 65:17, 69:5, 69:16, 70:16, 70:20, 72:14 looks 64:3, 69:18 lot 12:15, 12:17, 45:5, 56:18 lots 15:11, 19:14, 37:11, 45:19, 45:20, 48:3, 50:3, 50:10 lowering 67:11 <hr/> M <hr/> m-e-t-z-k-e-r 4:9 machines 50:21 made 45:22, 61:6, 66:1 magnesium 24:16 main 26:21 make 5:9, 5:19, 6:1, 6:3, 6:7, 6:14, 10:7, 10:14, 10:18, 25:15, 25:17, 34:17, 38:1, 50:22 making 31:18, 36:3 management 15:20 manipulate 28:8 many 4:15, 13:22,
---	--	---	--

PLANET DEPOS

888.433.3767 | WWW.PLANETDEPOS.COM

17:16, 20:4, 23:19, 28:8, 65:6, 66:2 mapped 37:9 marked 8:17, 9:2, 11:5, 11:8, 34:22, 35:2, 41:9, 52:9, 52:21, 53:3, 66:20, 74:8 marking 52:17 marriage 75:15 massively 67:12 material 19:12, 25:18, 61:14 matt 7:13 matter 75:16 maybe 7:18, 10:21, 20:22, 54:15 mccloskey 68:7, 68:9, 68:14, 68:17, 68:22, 69:13 mean 23:12, 24:1, 24:7, 30:16, 35:9, 35:10, 35:19, 37:14, 38:3, 38:8, 40:13, 45:20, 49:15, 50:15, 51:21, 54:9, 57:15, 72:1 means 5:16, 30:12 meant 35:17 mechanics 56:9	medicine 14:5, 16:13, 17:19 members 71:10 mention 70:11 mentioned 8:8, 9:21, 15:15, 42:16 merck 14:2, 14:3 met 7:9 method 21:10, 22:19, 40:20, 51:9, 56:4, 56:20 methodologies 12:21 methods 12:18, 14:1, 24:2, 35:11, 35:14, 58:17, 67:11 metzger 2:1 metzker 1:15, 4:9, 8:15, 11:4, 73:13, 74:2, 74:10, 74:13 mg 16:20, 18:5 michael 1:15, 2:1, 4:8, 8:15, 67:10, 73:13, 74:2, 74:10 middle 39:20, 40:6, 69:17 might 11:16, 72:22 miller 1:21, 2:4, 75:3, 75:20 millions 25:17	mine 52:19, 52:21 mip 53:20, 54:4, 54:10 mixed 52:3 mixing 52:3 modified 32:1 modify 10:7, 10:14 molecular 12:16, 28:3, 58:17 molecule 27:1, 37:16, 37:19, 38:5, 42:4, 42:15, 43:15, 43:22, 44:2, 44:20, 45:8, 46:15, 46:20, 47:12, 48:8, 48:11, 48:16, 48:18, 48:20, 54:11, 60:3, 69:12 molecules 25:14, 27:2, 27:3, 53:22, 60:3, 60:12 moment 4:11 monday 7:18 month 4:19, 11:19 more 4:20, 12:14, 14:19, 18:2, 22:6, 42:2, 43:8, 51:11, 65:4 morning 4:8 most 4:17, 9:13,	26:22 mrd 18:8, 18:20 much 30:22, 50:22, 61:3 mullis 21:9, 22:20 multiple 65:5 multiplex 35:13, 50:14, 50:18, 51:10, 51:14 multiplexing 50:5, 51:3, 51:5, 51:7, 51:16 must 5:16, 20:17 myself 4:11 <hr/> N <hr/> name 4:6, 4:8 names 50:10 natera 16:15, 16:22, 17:10, 17:11, 17:21, 18:7, 18:18, 18:20 national 14:10 nd 3:6, 11:12, 11:14 necessarily 23:11 necessary 24:1 need 6:4, 9:15, 32:1 neogenomics 17:11, 18:20 new 1:16, 2:3,
---	---	--	---

PLANET DEPOS

888.433.3767 | WWW.PLANETDEPOS.COM

3:12, 3:14, 4:3, 11:17, 14:11, 15:20, 58:1, 75:5 news 15:19 next 13:22, 14:17, 14:21, 15:9, 18:8, 18:17, 18:21, 26:3, 26:7, 26:16, 27:11, 27:15, 28:16, 32:21, 44:18, 45:7, 45:8, 45:11, 45:18, 45:20, 46:2, 46:4, 50:20 next-generation 31:19, 31:21, 35:21, 35:22 ngs 27:12, 52:4 nhgri 14:11 nih 11:19 nods 5:21 non-template 56:21, 57:14, 57:15, 58:8 notary 4:2, 73:21, 75:4 note 32:8 noted 17:13 notes 55:3, 67:2 notice 5:14 novartis 16:12 novel 14:16, 15:10	nucleic 28:5, 39:10, 55:19, 60:2, 60:10, 60:12, 64:3 nucleotide 38:19, 43:9, 65:20, 66:4, 66:11 nucleotides 24:15, 24:22, 66:2 number 25:14, 25:15, 35:14, 41:5, 41:21, 65:2 <hr/> O <hr/> oath 5:4 objection 20:1, 20:19, 22:14, 23:10, 23:18, 25:21, 26:17, 29:22, 30:18, 32:5, 32:18, 33:14, 33:21, 35:8, 36:13, 42:13, 44:22, 45:10, 48:13, 50:9, 51:20, 63:4, 63:17, 64:5, 68:16, 70:14, 71:20, 72:8 objections 5:9 obvious 68:5, 72:15 obviously 17:9 occur 37:12, 45:21, 57:2, 57:6 office 1:1 often 27:11, 50:7	oh 48:14 okay 6:16, 9:20, 12:2, 15:22, 19:1, 19:10, 20:6, 20:12, 20:16, 26:2, 27:14, 27:22, 29:2, 29:7, 30:3, 34:11, 38:21, 40:2, 40:13, 40:22, 41:12, 44:12, 44:18, 50:14, 52:11, 54:17, 59:4, 59:12, 59:16, 62:6, 67:7, 68:18, 69:2, 69:15, 70:2, 70:3, 71:16, 72:21, 73:1 oligonucleotide 30:12, 31:2, 33:5, 33:6, 41:17, 42:17, 54:12 once 24:3, 24:4, 24:18, 37:8, 43:1, 48:15 one 13:20, 19:15, 20:4, 22:10, 22:20, 26:21, 34:11, 36:22, 38:2, 38:5, 38:12, 38:15, 44:8, 46:7, 46:18, 49:15, 52:15, 53:7, 54:13, 56:16, 56:19, 60:14, 62:2, 65:3, 66:3, 66:6, 66:7, 68:17, 69:8	only 38:2, 44:10, 62:12, 65:19, 68:18, 68:22 opinion 39:16, 59:22, 68:4 opinions 10:7, 10:15, 29:19 order 20:16, 27:15 ordinary 28:21, 29:4, 29:12, 29:21, 64:21, 65:12, 72:16 orientation 71:12, 71:13 other 9:20, 10:20, 10:21, 22:16, 26:15, 34:3, 37:1, 40:18, 41:1, 43:10, 51:4, 51:7, 58:22, 66:9, 70:13, 71:19 out 70:22 outcome 75:16 over 5:17, 5:18, 7:9, 10:20, 25:15, 28:16, 58:19 overall 36:5, 61:3 overbroad 20:2, 20:20 own 12:7, 29:11, 31:22 owner 1:9, 2:2, 3:10, 29:15 owner's 10:2, 29:20
---	--	---	---

P			
padlock	69:18, 69:20, 70:5, 70:16, 71:7	20:4, 20:5, 20:6, 20:12, 21:1, 21:9, 21:15, 22:11, 22:19, 23:3, 23:16, 23:19, 23:21, 24:2, 37:4, 37:13, 37:17, 38:4, 38:7, 42:1, 46:7, 46:8, 47:22, 51:10, 56:5, 58:14, 70:6, 70:12, 71:4	picks 22:6
41:13, 41:16, 42:7, 42:14, 42:17, 43:12, 44:19, 45:6, 45:15, 45:17, 45:21, 46:12, 46:14	parallel 67:12	part 40:12, 40:22, 47:8, 48:20, 53:20, 72:10	piece 21:5, 21:13, 40:2
page	partially 23:6, 24:20, 57:19	particular 19:11, 28:20, 60:2, 61:13	pieces 24:5, 41:2
4:22, 9:1, 9:2, 16:3, 19:8, 20:7, 20:9, 21:3, 41:9, 49:2, 49:12, 49:17, 55:14, 58:12, 59:5, 59:7, 60:9, 69:7, 69:17, 70:16, 74:3, 74:7	particulars 12:16, 12:22, 31:19	pct 66:18, 67:9, 74:19	place 43:5, 53:3, 57:12
pages	parties 75:14	pending 6:5	placed 37:10
28:16	patent 1:1, 1:3, 1:9, 1:12, 2:2, 3:10, 8:8, 8:22, 10:2, 11:21, 15:22, 16:9, 16:12, 16:13, 16:14, 16:15, 16:17, 16:18, 16:19, 16:21, 17:1, 17:3, 17:5, 17:7, 17:9, 17:10, 17:11, 29:15, 29:20, 32:9, 32:14, 32:16, 34:19, 67:9, 68:5, 68:9, 69:9, 69:10, 69:14, 74:14	pennsylvania 17:6	plans 15:16
paired		perfectly 25:14	platform 15:8, 15:13, 31:22
71:10, 71:18		perform 27:15, 33:5	please 4:6, 5:22, 6:10, 20:21, 36:12
pairs		performs 63:7	plexxikon 16:11
22:6		permitted 6:17	plurality 39:10
paper		person 28:20, 29:4, 29:12, 29:21, 53:7, 64:21, 65:12, 72:16	point 13:7, 15:20, 27:15, 39:14, 39:19, 40:3, 55:11, 67:1, 68:18, 71:16, 71:22
10:6		petitioner 1:6, 3:3, 30:10	polymerase 19:6, 24:14, 24:21, 38:10, 38:15, 38:18, 43:8, 43:9, 47:5, 57:20, 58:2, 58:4, 58:6
papers		ph 1:15, 2:1, 8:15, 14:2, 24:16, 73:13, 74:11	polymerases 38:11
13:21, 29:17		phd 3:18	pool 51:1
paragraph		phenomenon 58:5	pooling 51:2, 51:6, 52:4
20:8, 20:9, 28:10, 30:5, 30:10, 32:8, 32:12, 39:3, 39:6, 39:19, 39:21, 40:6, 49:2, 49:11, 52:6, 52:7, 53:16, 55:2, 55:11, 55:13, 56:2, 58:11, 59:13, 60:8, 61:1, 67:2, 67:6, 67:19, 69:6, 69:15,	pathways 45:20, 45:21	phosphodiester 43:4	porreca 34:13, 34:21,
	patient 52:2		
	pcr 19:6, 19:10, 19:13, 19:14, 19:21, 20:3,		

35:4, 35:7, 39:1, 39:15, 39:16, 39:20, 40:3, 40:14, 41:9, 47:3, 49:11, 49:22, 50:4, 50:17, 51:15, 51:21, 52:7, 55:12, 59:22, 60:17, 61:2, 61:5, 63:1, 63:7, 63:13, 63:14, 64:2, 64:7, 64:8, 64:21, 65:3, 65:8, 65:14, 65:15, 65:19, 66:5, 66:13, 74:18 portion 49:16, 55:12, 57:21, 59:6, 70:21 portions 43:21 position 12:4, 30:11 possible 63:22 preliminary 10:2 preparation 9:22, 35:21, 53:21, 54:4 preparative 35:14 prepare 7:4, 7:8, 7:16, 7:20, 9:7, 27:17 prepared 8:7 preparing 12:18 present 3:17 presented 28:15 president 12:6	pretty 19:19, 30:22, 61:2 previoiusly 74:7 previously 8:16, 11:5, 11:7, 34:22, 38:10, 66:19 prime 47:18, 48:19, 58:20 primer 21:20, 22:5, 22:7, 24:18, 48:9, 48:14, 56:5, 57:17, 58:15, 58:21 primers 19:16, 19:20, 20:13, 20:14, 20:16, 22:3, 23:4, 23:7, 24:4, 24:5, 24:12, 25:5, 25:20, 26:1, 37:18, 37:21, 47:17 priming 42:1, 46:7, 47:22 printed 53:2 prior 16:4, 60:11 privilege 53:11 privileged 7:6 probably 5:14, 11:19, 62:10 probe 41:13, 41:16, 42:7, 42:14, 42:17, 43:6, 43:12, 44:6, 44:20, 45:6,	45:22, 46:13, 46:14, 54:11 probes 45:15, 45:17 proceeding 8:13 process 37:6, 45:9, 53:21, 54:4 procter 3:11, 3:18 professor 14:6 programs 14:12, 65:6 properly 37:10 proportion 64:17 proposed 34:5 prosecution 33:2, 33:13, 34:2, 34:9 protocol 37:18 provide 9:14, 37:10, 38:14 provided 28:17, 29:3, 29:10, 29:16, 32:16 ptab 33:20 ptab's 10:10 public 4:2, 73:21, 75:4 publication 34:20, 35:3, 74:16 published 13:20 pulled 53:13 purpose 23:1, 32:4,	32:7, 44:19, 45:1, 51:5 purposes 18:4 put 36:17, 58:20 putting 41:6, 47:15 <hr/> Q <hr/> qiagen 1:5, 7:14, 16:14, 17:21 quantitating 18:13 question 5:11, 6:5, 6:9, 6:13, 10:13, 20:21, 42:10, 48:6, 50:16, 60:5, 63:5 questions 5:3, 6:11, 6:17, 7:1, 73:5, 73:7 quick 72:22 quinn 3:4 quote 32:13, 51:22, 55:12, 69:22, 70:1, 71:7 quoted 39:20, 49:12, 49:16, 52:8, 55:18, 58:12, 70:4, 71:6 quoting 69:17 <hr/> R <hr/> rate 67:12 ravgen 16:18, 18:1 reaction 19:6, 19:22,
---	--	---	---

20:12, 21:1, 23:3, 23:17, 24:6, 43:5, 45:12, 56:14, 57:2, 57:12, 57:17 read 10:2, 10:10, 35:9, 63:10 reads 37:8, 37:9, 59:17, 60:9, 61:20, 63:9, 63:20, 64:14, 66:12, 67:21, 70:17, 71:1, 71:8, 71:18 reality 38:2, 40:17 really 13:18, 15:8, 35:10, 52:13 reason 6:22, 32:15 recall 8:1, 8:9, 26:5, 28:22, 32:6, 34:14, 41:2, 70:11 recent 4:17 recess 54:18, 73:2 recites 39:9, 49:6, 59:17 recognize 8:18, 11:8, 66:21 record 4:7, 5:21, 12:10, 62:7, 67:20, 75:10 reduce 35:11, 35:17, 36:6 reduced 36:7	redvault 12:7, 13:6, 13:15 reference 39:1, 66:15, 66:16 references 8:6 referred 26:3, 34:12, 35:4, 50:13 referring 51:8 refers 49:17, 53:18, 72:12 regarding 7:11, 8:8, 8:22 regenxbio 17:6, 17:7 region 22:8, 37:22, 41:19, 47:4, 47:7, 66:1, 66:3 regions 19:11, 19:17, 23:5, 51:12 related 75:13 relevant 29:5 rely 49:10, 49:15, 67:15, 68:2, 68:13 remember 16:7, 27:9, 53:1 removing 64:16 repeat 20:21, 60:5 rephrase 6:10, 30:19, 42:10, 60:6 replaced 26:10, 26:14 reported 1:21	reporter 5:15, 75:4 represent 61:11, 62:19 representation 25:8 represented 65:19 reproduced 59:7 reproduces 38:17 reproducibility 35:12, 36:8 require 26:20 required 31:20 requirements 31:22 requires 55:7 research 13:5, 14:10, 31:12 respect 13:3 response 10:3 restriction 48:2, 57:4 revealing 7:6 reversible 13:21, 14:16, 15:2, 15:6, 15:10 review 7:20 reviewed 33:2, 34:6 reviewing 9:12 richard 3:18 right 15:6, 16:1, 20:13, 21:5,	21:6, 21:13, 22:3, 22:13, 25:20, 28:17, 42:5, 43:18, 46:21, 53:3, 60:4, 62:11, 63:6, 64:17, 69:19, 70:22, 72:7 ring 68:8 rna 28:4, 38:13 rnas 13:7 rules 4:20 run 16:8, 32:1, 52:4 <hr/> S <hr/> s 3:1 said 9:17, 20:3, 38:7, 48:9, 51:15, 57:13, 62:3 salk 67:10 salts 24:17 salvia 18:10, 18:16 same 4:22, 10:13, 19:7, 30:2, 53:2, 53:3, 59:5, 60:3, 60:9, 60:13, 61:17, 63:21, 64:14, 70:8, 71:1 sample 18:17, 19:17, 28:7, 36:21, 50:8, 51:13
--	--	--	---

samples 50:6, 50:18, 51:2, 51:6, 51:18, 52:2 samuel 3:18 san 3:7 sanger 26:12, 26:14, 26:21, 27:5 sarepta 17:7 sat 4:21 saw 54:8 say 5:15, 36:9, 37:13, 40:12, 43:20, 50:11, 55:18, 56:12, 62:2, 68:21 saying 52:22 says 51:21, 53:19, 69:8, 72:6 schmitt 66:16, 67:5, 67:10, 67:14, 68:6, 68:13, 68:19, 68:22, 69:3, 69:13, 69:16, 69:19, 70:11, 70:17, 71:3, 71:7 school 13:19, 27:9 sciences 1:5 second 32:11, 40:2, 58:10, 69:22, 70:1 secondhand 15:19 section 11:18, 19:20,	20:6, 28:11, 28:16, 30:4, 39:14, 40:4, 55:18, 58:10, 68:3, 69:16, 69:19, 70:4 sections 55:16, 58:17 see 4:18, 9:3, 16:5, 20:10, 21:21, 28:13, 30:14, 30:15, 32:11, 39:12, 39:22, 40:7, 42:3, 43:3, 49:8, 49:14, 49:19, 54:1, 55:9, 55:14, 55:22, 56:6, 59:20, 59:21, 60:15, 60:16, 69:21, 70:2, 70:9, 71:14, 71:15 seems 53:1 sense 5:19, 6:1, 6:7, 6:14 sentence 30:9, 30:15, 32:11, 32:21, 53:18, 54:1, 56:3, 58:11, 59:21, 60:7, 60:16, 71:15 separate 22:12, 22:21 separated 47:4 separating 23:1 sequence 25:20, 30:13, 31:4, 38:13, 41:14, 43:7, 45:2, 46:2,	50:5, 56:5, 56:11, 58:14, 59:17, 62:1, 62:22, 63:9, 63:10, 63:11, 63:15, 63:16, 63:21, 64:13, 65:3, 65:5, 67:21, 70:8, 70:19, 71:8, 71:17, 72:5 sequencer 13:12 sequencers 32:2 sequences 12:22, 47:19, 55:20, 59:1, 59:18, 60:13, 60:18, 61:4, 61:12, 61:22, 62:20, 64:1, 64:15, 64:18, 65:9, 67:22, 71:2, 72:3, 72:18, 72:19, 72:20 sequencing 12:19, 12:20, 13:13, 14:1, 14:9, 14:18, 15:9, 15:12, 17:17, 17:20, 18:3, 18:6, 18:9, 18:11, 18:14, 18:18, 18:19, 18:22, 26:4, 26:8, 26:11, 26:12, 26:14, 26:22, 27:5, 27:11, 27:16, 27:18, 27:21, 27:22, 31:18, 31:19, 31:21, 35:22, 36:1, 37:5, 48:1, 50:21, 63:8, 67:12,	67:13 serve 31:5, 31:8 served 53:14 serves 32:4 set 19:1, 29:8, 71:9, 75:8 several 47:2 sharing 71:8 shift 15:21, 66:14 shorter 22:1 shorthand 75:4 should 29:4, 29:13, 33:10, 69:9 show 61:1 showing 21:14, 23:20, 25:22, 40:19, 42:18, 44:3, 61:2, 63:19, 65:4, 66:6, 69:10, 70:17, 72:2, 72:19 shown 20:15, 42:16, 47:21, 48:15 shows 71:8 side 47:22, 48:1, 61:5, 64:19 sidelined 15:1 sides 48:2 signature 9:5 signature-plkal 75:18
---	--	--	--

similar 4:21, 54:7 simple 38:3 simplicity 40:19 since 33:4 single 22:21, 23:2, 24:4, 25:5, 26:22, 27:3, 37:15, 37:19, 51:11, 51:13, 52:4, 54:11, 56:20, 57:13, 57:18, 57:21, 58:7, 61:21, 63:9, 65:15, 65:19, 66:4, 66:10, 69:12 sit 10:19 site 49:6, 66:3, 69:11 sites 42:1, 46:7, 69:4 skeleton 9:15 skill 28:21, 29:5, 29:8, 29:13, 29:16, 29:21, 64:21, 65:12, 72:16 skilled 53:19 slightly 29:16 small 13:6 smi 69:3, 69:12, 70:8, 70:13, 70:19, 71:1, 71:9, 71:19,	72:2, 72:18 snv 66:5 sold 14:21, 15:15 solution 17:3, 17:8, 24:10, 24:11, 24:15, 24:22 solutions 18:15 some 6:11, 13:4, 13:10, 15:20, 19:2, 19:4, 24:17, 27:1, 27:2, 29:3, 29:9, 31:14, 36:11, 47:4, 57:11, 61:16, 72:3, 72:4 something 31:2, 54:7 sometimes 36:18 sorry 62:5 sorted 63:2, 70:12, 70:18 sorts 63:15, 63:16, 64:2 sources 50:20, 51:18 speak 5:16, 5:17, 5:18, 7:11, 7:14 speaks 65:10 spearheaded 13:18 specific 19:17, 31:22, 37:22, 51:14 specifically 8:1 specification 34:2, 34:6,	34:8 spectrum 17:3, 17:8, 18:10, 18:15 stage 46:14, 46:15 stages 37:11 stamped 52:16 stand 66:8 standard 33:18 standards 28:12, 28:15, 29:3, 33:12, 33:16 stands 19:9 start 9:12, 14:8, 21:2, 21:4, 21:12, 21:15, 21:16, 24:21, 26:22, 37:16, 67:7 started 5:2, 13:17, 38:2, 64:18 starting 20:8, 25:18, 28:10, 36:15, 39:6, 61:6, 61:14 starts 30:5, 49:2, 59:13, 67:5, 67:18 state 4:2, 4:6, 75:5 stated 12:5, 32:21 statement 34:3, 70:5 states 1:1 stds 13:9	step 21:18, 38:9, 44:18, 45:7, 45:8, 45:11, 46:3, 46:4, 68:3 steps 22:11, 22:16, 22:19, 22:20, 23:21, 25:12, 36:2, 36:10, 63:3 sticks 56:15 sticky 57:5, 57:6 still 4:18 stocks 27:8 straight 62:9 strand 22:21, 23:2, 23:15, 25:1, 25:2, 38:12, 38:16, 46:10, 57:18, 57:22, 58:1, 71:10 stranded 21:4, 21:13, 21:16, 21:17, 23:6, 23:8, 23:14, 24:5, 24:20, 25:4, 25:5, 25:7, 57:20, 57:21 strands 22:12, 22:22, 23:2, 38:12 street 3:5 study 11:18 stumble 10:20 subcontractor 14:15 submitted 10:3, 11:17
---	---	---	---

subscribed 73:16	64:13, 71:9, 71:19, 72:2	43:10, 43:18, 43:20, 44:8, 44:9, 46:19, 47:3, 47:7, 54:12	15:11
subset 49:7	tail 48:19, 58:20, 58:21	task 13:8	testified 4:3, 66:15
substance 6:18	tails 47:18, 47:19	teaches 60:1	testifying 5:5
sullivan 3:4	take 6:3, 11:1, 11:3, 39:3, 43:5, 57:12, 68:2, 72:21	tecn 1:8, 8:8	testimony 6:18, 75:10
support 39:15	taken 2:1, 30:11, 54:18, 73:2	technical 9:14, 19:3	testing 18:21
sure 6:10, 20:22, 24:9, 25:11, 29:1, 31:15, 34:17, 36:14, 41:15, 43:14, 60:6, 63:18, 65:21	taking 4:11, 5:15	technologies 12:20, 13:13, 14:9, 17:17, 17:20, 18:14, 18:22, 26:10, 26:14	tests 18:8
sworn 4:2, 73:16, 75:8	talk 12:9, 19:2, 19:4, 56:8, 66:14, 68:4	technology 12:15, 14:22, 18:6, 27:21, 29:6	text 72:6, 72:9
synthesizing 14:16	talked 13:14	tell 13:15	thank 17:15, 48:22, 54:21, 58:9, 59:4, 68:11, 73:6
systems 51:1	talking 24:12, 24:13, 26:2	template 22:9, 24:19, 25:2, 47:8, 48:17, 57:19, 57:21, 57:22, 58:4	themselves 36:19, 44:13, 72:19
T	taq 58:6	template-depende- nt 58:2	thing 62:3
t-overhang 57:2	target 19:20, 30:13, 31:3, 37:21, 41:14, 43:2, 43:7, 43:13, 44:7, 44:10, 44:17, 47:5, 47:16, 59:2, 59:18, 61:22, 63:10, 63:20, 64:13, 64:17, 67:22, 70:19, 71:2, 72:18	templates 27:5	things 11:21, 13:13, 23:13, 29:9, 32:7, 36:16, 40:18, 45:19, 48:3, 50:4
ta 56:20	target-specific 19:16, 41:19, 58:21	tenured 14:5	think 4:18, 4:19, 11:20, 15:4, 16:10, 27:20, 29:4, 30:1, 30:5, 35:21, 40:14, 40:19, 46:17, 47:1, 48:5, 49:11, 51:13, 54:10, 56:16, 62:10, 64:11, 65:10, 67:1, 72:9
table 69:7	targeting 41:17, 41:20, 42:19, 42:22, 43:1, 43:2,	term 19:13, 20:3, 30:11, 30:20, 32:10, 32:17, 33:7, 33:9, 34:5, 49:22, 50:12, 50:14	thinking 62:10
tag 40:16, 42:1, 47:20, 50:18, 55:20, 56:5, 56:10, 58:14, 58:22, 60:11, 60:18, 61:18, 62:1, 64:1, 64:2, 69:3, 70:13		terminator 13:21, 15:3	third 23:15
tagged 60:10, 61:18		terminators 14:16, 15:6,	thought 29:12
tags 60:1, 60:14, 61:8, 61:21,			

three 22:18, 25:12 through 4:21, 5:1, 14:14, 15:3, 19:4, 37:17, 39:17 throughput 50:22 ticks 22:2 time 6:3, 14:7, 14:9, 15:19, 54:15, 63:21, 64:14 times 3:12, 4:15, 47:2 titled 67:11 today 4:12, 5:3, 5:16, 7:2, 10:19 today's 7:4 together 36:17, 44:4, 44:7, 52:3, 56:15, 57:8, 63:12 took 14:20 top 25:3, 25:9, 49:12, 49:17, 55:19, 70:21 track 14:5 trademark 1:1 transcript 75:9 transmission 13:1 trial 1:3 triplex 23:13	troutman 7:13 true 24:1, 27:20, 70:6, 71:4, 75:10 trustees 17:5 truthfully 7:2 try 5:17, 5:18, 36:17 trying 22:18, 35:10, 36:5, 46:22 tube 24:10, 51:11 tubes 18:10 turn 9:1 twinstrand 17:4, 18:12 two 19:15, 19:19, 20:13, 20:14, 24:4, 25:20, 37:18, 38:1, 39:14, 41:1, 41:17, 41:20, 44:9, 46:6, 51:11, 55:16, 65:4, 66:6 two-ways 56:9 type 50:7 typical 19:19, 19:21, 22:11 typically 9:9, 9:17, 23:16, 27:4, 27:16, 31:3, 36:16 typographical 10:21	typos 69:8 <hr/> U <hr/> um-hmms 5:21 umi 40:16, 47:21, 73:5 unambiguously 70:6 under 5:4 underneath 20:9, 71:5 understand 5:6, 5:13, 5:20, 6:2, 6:8, 6:9, 6:16, 6:20, 28:1, 29:15, 29:18, 30:10, 30:16, 40:9, 43:14, 49:21, 59:8, 63:5, 63:7 understanding 8:12, 26:7, 33:9, 33:11, 33:17, 35:16, 54:3, 62:17, 64:6, 65:18 understood 6:13, 11:1, 27:10, 27:17, 38:6, 44:15, 48:22, 51:15, 58:9, 62:8, 62:15 unique 49:7, 60:10, 71:9 uniquely 52:1 united 1:1 university 17:5 unless 5:10, 23:7	until 54:19, 73:3 updates 11:13, 11:16 urine 13:7 urquhart 3:4 use 19:14, 19:15, 26:20, 27:2, 27:15, 30:20, 30:22, 32:7, 32:15, 33:12, 33:20, 38:12, 42:8, 46:8, 47:5, 51:14, 57:20 uses 18:21, 33:19, 49:22, 51:15 using 19:19, 46:14, 47:17, 50:4, 50:18, 67:13 usually 32:6, 47:21 utilized 15:10 <hr/> V <hr/> vacuum 44:14 vague 20:1, 20:19 variable 66:3 variant 66:5, 66:7, 66:9, 66:11, 66:12 variants 18:13 variation 13:2 variety 31:13 various 36:2, 36:9
--	---	--	--

verbally 5:22	41:5, 45:5, 46:12, 47:15, 51:7, 53:15, 56:18, 65:2	word 28:6, 42:8	0
versus 16:11, 16:12, 16:14, 16:15, 16:16, 16:17, 16:18, 16:20, 16:22, 17:2, 17:4, 17:6, 17:7, 17:8, 17:10, 17:11, 17:19, 17:20, 17:21, 17:22, 18:1, 18:5, 18:7, 18:9, 18:12, 18:15, 18:18, 18:20	we'll 11:2, 53:9 we're 4:18, 4:22, 19:7, 53:6, 59:5, 60:9 we've 13:11, 24:12, 24:13, 54:14, 58:19, 65:17 welcome 54:20 well-known 41:5 went 14:1, 14:2 whereby 44:5 whereupon 8:14, 11:4, 34:19, 54:18, 66:18, 73:2, 73:8 whether 33:18, 60:2 widely 58:6, 65:6 widely-used 58:17 wine 12:7 within 19:17, 22:9, 36:2, 36:10, 37:11, 66:2 without 7:6 witness 52:21, 62:5, 62:8, 73:1, 73:6, 74:2, 75:7, 75:11 wo 66:19, 74:20	work 12:18, 13:4, 13:5, 13:11, 13:16, 13:19, 21:10, 30:17, 30:21, 56:17 worked 14:3 workflow 36:1, 36:2, 36:10, 37:11, 61:3 works 25:13, 56:18 world 63:13 wouldn't 33:15, 50:12 wrap 44:1 wrapped 44:10, 44:16 writing 9:14 wrong 38:19 wrote 21:7	00026 8:13, 8:16, 8:21, 74:12 00028 1:11 0165202 34:21, 74:17
vertical 42:19, 61:10, 61:11, 62:3			1
virtue 70:7			10 14:20, 16:10 10,036,012 1:12 100 67:6, 69:6 10018 3:14 1003 8:12, 8:17, 74:9 1006 34:18, 35:1, 35:3, 74:14 1006.0005 41:10 1007 66:17, 66:20, 66:21, 67:8, 74:19 1016 11:3, 11:6, 11:8, 16:3, 74:13 10:39 am 54:19 10:54 am 54:19 11 74:13 11:22 am 73:3 11:33 am 73:3, 73:9 125 70:16 127 67:2, 67:19,
vocabulary 27:14			
W		Y	
waiver 53:10 walk 39:17 want 28:7 wanted 14:7, 52:11 way 15:3, 20:4, 30:22, 42:14, 46:11, 47:1, 50:17, 51:17, 52:10, 56:16, 56:21, 57:3, 57:14, 57:15, 58:8, 63:14, 65:3, 75:15 ways 18:13, 19:14, 19:15, 20:4, 23:19, 28:9,		yeah 52:14 years 12:14, 14:3, 14:20 yesterday 7:17 york 1:16, 2:3, 3:12, 3:14, 4:3, 75:5	
		\$	
		\$1,000 14:12 \$100,000 14:12	

69:15, 70:5 128 69:20 14 71:7 142389 66:19, 74:20 149 49:11, 52:6, 52:7, 53:16, 53:18 150 39:19, 55:13 158 9:2 17 20:7 18 20:7, 20:9, 21:3 1980 21:10 1994 13:19 1999 14:4 1a 39:8 1c 49:1, 49:6 1e 55:1, 55:6 1g 59:13, 59:16, 67:18, 67:20, 68:18 <hr/> 2 <hr/> 2002 13:18 2012 15:3, 34:21, 74:17 2013 66:19, 74:20 2024 11:12, 11:14 2025 1:11, 1:17,	2:4, 8:13, 8:16, 8:21, 74:12 22 3:6, 11:12, 11:14 241 69:9 29 20:8 295 2:2 <hr/> 3 <hr/> 30 20:9 31 1:17, 2:4 35 12:14, 74:14 357 8:9, 8:22, 32:9, 68:5, 69:10 39 28:10 3a 70:20 <hr/> 4 <hr/> 4a 41:12, 42:3, 42:4, 42:11, 43:3, 43:15, 46:6, 47:14, 54:8, 54:13 4b 47:10, 47:13, 47:17, 48:7, 58:19 4c 40:4, 40:5, 40:10, 40:22, 59:6 <hr/> 5 <hr/> 50 3:5 52 30:6, 32:8,	32:12 53 30:5 58 49:2 59 49:12, 49:17 593985 1:22 5th 2:2 <hr/> 6 <hr/> 62 55:14, 58:12, 59:7 620 3:13 66 74:19 <hr/> 7 <hr/> 73 39:3, 39:6, 39:21 74 40:6 77 49:2 78 69:7 <hr/> 8 <hr/> 82 55:2, 55:11 85 61:1, 69:18 86 59:13, 60:8 <hr/> 9 <hr/> 94111 3:7 95 70:16 98 69:17 9:34 am 2:3	
--	---	--	--