

UNITED STATES PATENT AND TRADEMARK OFFICE
BEFORE THE PATENT TRIAL AND APPEAL BOARD

INTEGRATED DNA TECHNOLOGIES, INC.,

Petitioner,

VS.

TECAN GENOMICS, INC.

Patent Owner.

Case IPR2024-01504

U.S. Patent No. 11,098,357

Case IPR2025-00015

Case IPR2025-00016

U.S. Patent Nos. 10,036,012 and 10,876,108

DEPOSITION OF

DR. CARLOS BUSTAMANTE, VOL I

New York, New York

Wednesday, October 22, 2025

Reported By:

CATHI IRISH, RPR, CRR, CLVS

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October 22, 2025

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9:09 a.m.

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Deposition of DR. CARLOS

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BUSTAMANTE, Vol I, held at the offices of

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Goodwin Procter, 620 Eighth Avenue,

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New York, New York, before Cathi

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Irish, a Registered Professional

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Reporter, Certified Realtime Reporter,

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and Notary Public of the State of

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New York.

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1 A P P E A R A N C E S:

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7 BY: THOMAS S. FLETCHER, ESQ.

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10

11

12 GOODWIN PROCTER LLP

13 Attorneys for Patent Owner

14 and the Witness

15 The New York Times Building

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17 New York, New York 10018

18 BY: TIMOTHY J. BEAVERS, ESQ.

19 LINNEA P. CIPRIANO, ESQ.

20 (via Zoom)

21

22

23 ALSO PRESENT:

24 MICHAEL LEVY, Danaher Corporation

25

1 ----- I N D E X -----

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3	CARLOS BUSTAMANTE	MR. FLETCHER	6
4		MR. HOAGLAND-HANSON	176

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1 ----- EXHIBITS CON'T -----

2 EXHIBIT NUMBER DESCRIPTION PAGE

3 IDT Exhibit 1120, Claim Construction 62

4 - Scenario 4.3

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1 D R. C A R L O S B U S T A M A N T E,
2 called as a witness, having been duly
3 sworn by a Notary Public, was examined
4 and testified as follows:

5 MR. FLETCHER: Let me just
6 announce who's here for the record.

7 This is Thomas Fletcher from
8 Williams & Connolly for the
9 petitioner, Integrated DNA
10 Technologies. With me are my
11 colleagues, Patrick Hynds and Kevin
12 Hoagland-Hanson. Also present is
13 Michael Levy from Danaher, the parent
14 of Integrated DNA Technologies.

15 MR. BEAVERS: And this is Timothy
16 Beavers from Goodwin Procter. I'm
17 joined by my colleague remotely,
18 Linnea Cipriano, and we are
19 representing the patent owner, Tecan
20 Genomics, Incorporated.

21 EXAMINATION

22 BY MR. FLETCHER:

23 Q. Good morning, Dr. Bustamante.

24 A. Good morning.

25 Q. I've given you two documents,

1 Exhibits 2035 and Exhibit 1004. Do you
2 have those in front of you?

3 A. I do.

4 Q. Do you recognize those?

5 A. I do.

6 Q. Exhibit 2035 is your declaration
7 in this matter; correct?

8 A. That is correct.

9 Q. And your declaration analyzes
10 Exhibit 1004 which is U.S. patent number
11 11,098,357; correct?

12 A. Correct.

13 Q. Can we agree to refer to Exhibit
14 1004 as the '357 patent?

15 A. Happy to do so.

16 Q. Dr. Bustamante, when were you
17 retained?

18 A. I was retained late last year, as
19 I recall.

20 Q. Late 2024; correct?

21 A. I believe that's right.

22 Q. Had you seen Exhibit 1004 before
23 you were retained?

24 A. I'm not sure. I don't recall
25 having seen it before.

1 Q. At the time that you were being
2 retained, did you know you were being
3 retained by the patent owner Tecan
4 Genomics?

5 A. That I believe was in the
6 documents I received.

7 Q. So at the beginning of your work
8 on this matter, you knew that you had been
9 retained by the patent owner Tecan
10 Genomics; correct?

11 A. I think that's right.

12 Q. If we could go to paragraph 4 of
13 your declaration, please. Paragraph 4
14 states that you have considered the
15 contents of the '357 patent. Do you see
16 that?

17 A. I do.

18 Q. What did you mean by that?

19 A. The document as we see before us,
20 as well as the priority date and the
21 information provided to me by the
22 attorneys on the legal standards for
23 considering the document.

24 Q. Focusing on this phrase, "the
25 contents of the '357 patent," did you, for

1 example, consider the figures in the
2 patent?

3 A. I did.

4 Q. And you considered the claims at
5 the end of the patent?

6 A. I did.

7 Q. The next part of the sentence
8 reads that you considered the file history
9 of the patent and related patents. Do you
10 see that?

11 A. I do.

12 Q. And what did you consider for
13 purposes of that?

14 A. This is the information provided
15 on the prosecution of the patent, the
16 prosecution of the parent from which they
17 were derived as well as related materials
18 provided.

19 Q. Next up you say that you
20 considered prior art publications. Do you
21 see that?

22 A. I do.

23 Q. Which prior art publications did
24 you consider?

25 A. I considered the publications

1 that are in my -- in my report.

2 Q. There were many other
3 publications that were discussed by
4 Dr. Sims and cited in IDT's petition that
5 are not discussed in your declaration.
6 Fair?

7 A. That's fair.

8 Q. Does that mean you did not
9 consider those publications?

10 A. Not necessarily. I referred to
11 the prior art that was part of the
12 petition, and then in reviewing Dr. Sims'
13 material I also reviewed exhibits that
14 were provided.

15 Q. So you reviewed all the
16 publications that were cited by Dr. Sims
17 and IDT; is that fair?

18 MR. BEAVERS: Objection, form.

19 THE WITNESS: I reviewed
20 Dr. Sims' declaration and as necessary
21 referred back to exhibits that were
22 provided.

23 BY MR. FLETCHER:

24 Q. And you were provided with all of
25 the exhibits to Dr. Sims' declaration;

1 correct?

2 A. I was provided with many
3 exhibits. I can't tell you that I checked
4 every single one that was in Sims but
5 certainly the main prior art that was
6 referenced and additional materials I felt
7 necessary to form my opinion.

8 Q. The next portion of your sentence
9 says that you've considered the two
10 petitions for inter partes review. Do you
11 see that, Dr. Bustamante?

12 A. I do.

13 Q. And what were you referring to
14 here?

15 A. The petitions related to this
16 patent.

17 Q. What do you mean by the petitions
18 related to this patent?

19 A. So there were multiple petitions
20 that I was provided, including the
21 petition related to the '357.

22 Q. And Dr. Bustamante, I'll try to
23 be more clear. When you said the two
24 petitions here, were you referring to
25 other petitions filed by Integrated DNA

1 Technologies or are you referring to a
2 petition filed by QIAGEN?

3 A. The QIAGEN petitions are in a
4 separate report and there are separate
5 reports that have been filed.

6 Q. Here you were referring to the
7 IDT petitions?

8 A. That's correct.

9 Q. And you reviewed the declaration
10 of Dr. Sims; correct?

11 A. I did.

12 Q. Do you know Dr. Sims?

13 A. I don't.

14 Q. Was there any sequence in which
15 you reviewed these materials?

16 A. Yes.

17 Q. What was that sequence?

18 A. I reviewed the materials as they
19 were provided to me. I can't recollect
20 the exact order but I believe the first
21 documents I was provided were the patent
22 and the petitions. Obviously, I didn't
23 review Dr. Sims' declaration until he gave
24 the declaration. It happened afterwards.

25 Q. You mean deposition?

1 A. Sorry, the declaration and his
2 deposition.

3 Q. Dr. Bustamante, at the time that
4 you started your analysis for this matter,
5 you knew that Tecan wanted to maintain the
6 validity of its patents; correct?

7 MR. BEAVERS: Objection, form.

8 THE WITNESS: I understood that
9 Tecan was in an inter partes review
10 and required scientific expertise to
11 evaluate the petition and the
12 response.

13 BY MR. FLETCHER:

14 Q. And you understood that Tecan's
15 desired outcome of the proceeding would be
16 maintaining the validity of its patents;
17 correct?

18 MR. FLETCHER: Objection, form.

19 THE WITNESS: I'm not a lawyer
20 and I'm not part of the legal
21 decision-making but I understood that
22 they wanted my review of their patent
23 and the responses.

24 BY MR. FLETCHER:

25 Q. You understood that they were the

1 patent owner; correct?

2 A. I did understand that, yes.

3 Q. And you've been involved in
4 patent disputes before this one; correct?

5 A. I have.

6 Q. Separate; correct?

7 A. That's correct.

8 Q. You understand the patent owner
9 wants to maintain the validity of their
10 patent; correct?

11 A. Again, I can't disclose anything
12 that's been shared under privilege but in
13 general, patent owners want to retain the
14 patent they have been issued. I think
15 that's a fair statement.

16 Q. If we could go to paragraph 19.

17 A. Give me a minute to get there.

18 Q. You list in paragraph 19 prior
19 instances of testimony as an expert
20 witness in the past five years; correct?

21 A. That's correct.

22 Q. Have you provided any testimony
23 as a fact witness in the past five years?

24 A. Not to my recollection. I
25 believe that is the extent of my sworn

1 testimony in the last five years.

2 Q. Have you provided sworn testimony
3 prior to these instances that you list in
4 paragraph 19?

5 A. I have.

6 Q. And can you recall the names of
7 those proceedings?

8 A. They were cases before the state
9 court in Texas involving two companies in
10 a trade secret matter.

11 Q. Do you remember the names of
12 either of the companies?

13 A. I don't want to be inaccurate.
14 It was around 2016.

15 Q. Was one of them NuGEN or Tecan?

16 A. No.

17 Q. You have also previously worked
18 as an expert witness without providing
19 testimony; correct?

20 A. I'm trying to recall. I
21 wasn't -- I'm not sure I can speak to that
22 right here right now.

23 Q. You provided expert reports that
24 were not for purposes of testifying;
25 correct?

1 A. Other kind of expert reports,
2 sure.

3 MR. FLETCHER: Dr. Bustamante,
4 I'm handing you and your counsel what
5 I have marked as Exhibit 1122.
6 (IDT Exhibit 1122, report, marked
7 for identification.)

8 MR. BEAVERS: Just to preserve
9 our objections here and for the
10 record, we're going to object to
11 relevance, scope, completion and
12 foundation at this point. Thank you.

13 MR. FLETCHER: Any other
14 objection?

15 MR. BEAVERS: We're good for now.

16 BY MR. FLETCHER:

17 Q. Dr. Bustamante, do you recognize
18 Exhibit 1122?

19 A. I do recognize this.

20 Q. This is a report that you've
21 previously provided that was not for
22 purposes of testifying; correct?

23 A. That is correct, I have not
24 reviewed this report and I am not prepared
25 to discuss it today.

1 Q. Okay. You prepared this Exhibit
2 1122; correct?

3 A. I did.

4 Q. And in this report, you concluded
5 that there was, quote, strong evidence
6 that a DNA sample of primarily European
7 descent also contains Native American
8 ancestry from an ancestor in the sample's
9 pedigree six to 10 generations ago;
10 correct?

11 A. That's what it states. As I
12 said, I haven't reviewed this report and
13 am not prepared to discuss it today.

14 Q. That was a conclusion you formed
15 at the time; correct?

16 A. That's what I wrote at the time.

17 Q. I would like to turn your
18 attention to the first paragraph under
19 Request. The final sentence reads, "The
20 identity of the sample donor, Elizabeth
21 Warren, was not known to the analyst
22 during the time the work was performed."

23 Do you see that?

24 A. I do.

25 Q. Why was that important?

1 A. Again, I'm not prepared to
2 discuss this report today.

3 Q. When you prepared this report,
4 you chose to shield the analyst from the
5 knowledge of the source of the sample
6 donor; correct?

7 A. Again, I'm not prepared to
8 discuss that report today.

9 Q. I'm trying to understand the
10 difference in your methodology when you're
11 providing unsworn report versus sworn
12 testimony. I'm not going to ask you about
13 the details of this. I don't care about
14 the outcome of this.

15 A. I understand.

16 Q. The question, Dr. Bustamante, is
17 why when you performed this research did
18 you shield the analyst from the identity
19 of the sample donor?

20 MR. BEAVERS: Objection, form,
21 scope.

22 THE WITNESS: Again I'm not
23 prepared to discuss this report or
24 anything about the report at this
25 time.

1 BY MR. FLETCHER:

2 Q. What do you mean you're not
3 prepared?

4 A. I haven't reviewed this, there
5 are reasons I can't discuss this report
6 today.

7 Q. What are the reasons you can't
8 discuss this report?

9 MR. BEAVERS: Objection, form,
10 scope.

11 THE WITNESS: As I said, I
12 can't -- I am not prepared to discuss
13 this report today.

14 BY MR. FLETCHER:

15 Q. I'm asking you about your
16 memory --

17 A. I understand.

18 Q. -- and your process. So as a
19 matter of fact, Dr. Bustamante, can you
20 recall why the identity of the sample
21 donor was kept from the analyst when the
22 work was performed?

23 MR. BEAVERS: Objection, form,
24 scope, asked and answered.

25 THE WITNESS: As I said,

1 unfortunately I'm not prepared to
2 discuss this report or anything
3 related to this example.

4 BY MR. FLETCHER:

5 Q. Is the problem you have no
6 factual memory of the events seven years
7 ago?

8 MR. BEAVERS: Objection,
9 argumentative.

10 THE WITNESS: Unfortunately, I'm
11 not prepared to discuss this report
12 today.

13 BY MR. FLETCHER:

14 Q. So I've heard that. I'd like to
15 understand why, Dr. Bustamante, why are
16 you unable to answer my question about the
17 process that was used in this research.

18 MR. BEAVERS: Objection, form,
19 scope, if there be a question there.

20 THE WITNESS: I am not at liberty
21 to discuss this report today.

22 BY MR. FLETCHER:

23 Q. You mean you're contractually
24 prohibited from discussing this report?

25 MR. BEAVERS: Objection. Tom,

1 would it be okay to discuss whether or
2 not there's a confidentiality issue
3 here? Because that may be an issue.

4 MR. FLETCHER: If you need to go
5 off the record, by all means take a
6 break.

7 (Witness conferring with
8 Counsel.)

9 BY MR. FLETCHER:

10 Q. Dr. Bustamante, why was the
11 identity of the sample donor kept from the
12 analyst for purposes of this report,
13 Exhibit 1122?

14 A. In general I am not at liberty to
15 discuss much about this project due to
16 third-party contractual obligations.

17 Q. Let me make sure I understand,
18 Dr. Bustamante. You can't discuss the
19 thinking behind the methodology in this
20 report because you are bound by a
21 confidentiality agreement?

22 A. I can't speak to the contract. I
23 can speak in general to why one would
24 blind a sample, if that would be helpful.

25 Q. Why would one blind a sample,

1 Dr. Bustamante?

2 A. In general it is good scientific
3 practice to limit information provided to
4 an analyst because it isn't relevant to
5 the analysis.

6 Q. And in this report, there were
7 two analysts, correct, you and your
8 colleague?

9 A. That is correct.

10 Q. And the report states that the
11 identity was not known to the analyst. Do
12 you see that?

13 A. I do.

14 Q. Was the identity not known to
15 both of you?

16 A. Again there are many aspects of
17 this I cannot discuss due to third-party
18 contractual obligations.

19 Q. I'd hate to get you in trouble
20 with Senator Warren so we'll put this one
21 aside.

22 A. Okay. Appreciate that.

23 Q. No one wants to be in her
24 crosshairs. So we're turning to the
25 declaration, Exhibit 2035. Are you there?

1 A. Yes.

2 Q. For purposes of this work, you
3 did know the identity of the party that
4 you were performing the work for; correct?

5 A. I did.

6 Q. Could you please go to paragraph
7 83 of your report?

8 The end of paragraph 83 you state
9 that you have analyzed this term based on
10 the claims and specification of the '357
11 patent and the prosecution of the '357
12 patent and related patents; correct?

13 A. Give me one second just to reread
14 this section.

15 (Witness perusing document.)

16 I'd like to quickly note that
17 there's a typographical error in 81 and 82
18 where in quotes indexing site should be
19 identifier site. Happy to answer your
20 question if you could restate it.

21 Q. Thank you, Dr. Bustamante.

22 Okay, so why don't I ask a new
23 question.

24 This portion of your declaration
25 starting at paragraph 79 is about how the

1 person of ordinary skill in the art would
2 understand the claims of the patent,
3 Exhibit 1004; correct?

4 A. That is correct.

5 Q. And to do that analysis, you
6 reviewed the claims in the specification
7 and the prosecution history; correct?

8 A. I did.

9 Q. And you considered the whole
10 specification of the patent; correct?

11 A. Correct.

12 Q. You reviewed all the figures of
13 the patent; correct?

14 A. I have reviewed the figures in
15 the patent.

16 Q. You reviewed all of the examples
17 in the patent; correct?

18 A. I reviewed the examples in the
19 patent.

20 Q. So if you could also go to the
21 final page of Exhibit 1004 to the claim
22 language.

23 A. Okay.

24 Q. The final limitation of claim 1
25 reads, "identifying sequence reads with

1 identical identifier and target sequences
2 as duplicates."

3 Do you see that?

4 A. I do.

5 Q. And you interpreted that language
6 from the perspective of the person of
7 ordinary skill in the art; correct?

8 A. That is correct.

9 Q. I'm going to refer to this
10 limitation as the identifying limitation.
11 Is that okay with you?

12 A. That's fine.

13 Q. In paragraph 84, you offer your
14 conclusion about what this language means;
15 correct?

16 A. Let me briefly review.

17 (Witness perusing document.)

18 Could you repeat the question?

19 Q. In paragraph 84, you offer your
20 conclusion about what this language means;
21 correct?

22 A. I offer a conclusion as to what
23 this language means.

24 Q. Your conclusion was that it means
25 identifying sequence reads as duplicates

1 through a step that uses both the
2 identifier and at least a portion of the
3 target sequence; correct?

4 A. That's correct.

5 Q. That's still your opinion today;
6 right?

7 A. That is my opinion.

8 Q. As you understand this
9 identifying limitation, it is a
10 information processing limitation;
11 correct?

12 A. Exactly what do you mean by an
13 information processing?

14 Q. Let me do it in the context of
15 the other steps or limitations of the
16 claim.

17 Do you have the claim language in
18 front of you?

19 A. I do.

20 Q. The method begins, ligating an
21 adaptor. Do you see that?

22 A. I do.

23 Q. That is a physical step; correct?

24 A. Physical, chemical, yes.

25 Q. The next limitation begins,

1 amplifying the adaptor-ligated fragments.

2 Do you see that?

3 A. I do.

4 Q. And that is a physical, chemical
5 step; correct?

6 A. Correct.

7 Q. The third limitation begins
8 sequencing the amplicons to produce
9 sequence reads. Do you see that?

10 A. I do.

11 Q. And that limitation combines a
12 physical step of performing sequencing and
13 the generation of information; correct?

14 A. I would agree with that.

15 Q. The final limitation, the
16 identifying limitation involves the
17 manipulation or analysis of the
18 information; correct?

19 A. I would agree that that is an
20 analysis step.

21 Q. There is no physical chemistry
22 happening in the identifying limitation;
23 correct?

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: Sitting here right

1 now today, I would say this would
2 largely be performed through some form
3 of sequence analysis. It doesn't
4 limit necessarily what could be done
5 through additional confirmation but as
6 written, I would consider this largely
7 an analysis step.

8 BY MR. FLETCHER:

9 Q. This analysis could happen within
10 the scientist's head; correct?

11 A. No. Well, depending on who the
12 scientist is, but in general you would use
13 a computational tool to carry out this
14 work.

15 Q. It is most typically done on a
16 computer; correct?

17 A. I would agree with that.
18 Although you would spot-check it by
19 looking at it by eye and so on. Computer
20 aided, let's call it that.

21 Q. Computer-aided limitation. In
22 paragraph 94 of your declaration you offer
23 some further analysis on the meaning of
24 the word duplicate which is included in
25 your prior conclusion; correct?

1 A. I see that's what I wrote in
2 paragraph 94.

3 Q. In paragraph 94 you formed the
4 conclusion that a duplicate means
5 sequences -- the sequences possess an
6 identical sequence. Do you see that?

7 A. I do.

8 MR. BEAVERS: Objection,
9 misstates the report.

10 MR. FLETCHER: Mr. Beavers, did I
11 misread it?

12 MR. BEAVERS: Yes.

13 MR. FLETCHER: I'll reread it.

14 BY MR. FLETCHER:

15 Q. Dr. Bustamante, your conclusion
16 was that the term duplicate means the
17 sequences possess an identical sequence?

18 MR. BEAVERS: Same objection.

19 THE WITNESS: If I could read the
20 entirety of paragraph 94, I understand
21 the Board invited the parties to
22 explain whether the term duplicate
23 means the sequences possess an
24 identical sequence.

25 ///

1 BY MR. FLETCHER:

2 Q. Thank you for clarifying. So
3 what is your opinion about the meaning of
4 the word duplicate?

5 A. As I state in 95, it is my
6 opinion that based on the claim language
7 specification and prosecution history, a
8 POSA would understand that term duplicate
9 sequencing reads to mean reads containing
10 identical sequences as the original
11 nucleic acid fragment.

12 MR. FLETCHER: Thank you for
13 making that clear.

14 Dr. Bustamante, I have prepared
15 some demonstratives to help us
16 understand how you analyze the claims.
17 So I'm handing you a demonstrative
18 that I've labeled Claim Construction -
19 Scenario 1 and I premarked it as IDT
20 Exhibit 1113.

21 MR. BEAVERS: Just so the record
22 is clear and we preserve our
23 objection, I'll be objecting to scope,
24 relevance, foundation, hearsay, and
25 completion.

1 (IDT Exhibit 1113, Claim
2 Construction - Scenario 1, marked for
3 identification.)

4 BY MR. FLETCHER:

5 Q. Dr. Bustamante, in my
6 demonstrative Scenario 1 I presented you
7 with two sequences. Do you see that?

8 A. This is my first time reviewing
9 this so give me a moment.

10 (Witness perusing document.)

11 Q. Dr. Bustamante, I have identified
12 in red where I made the sequences have
13 different bases. Do you see that?

14 A. I do see in red that the sequence
15 on the top has a C and the sequence at the
16 bottom has a G.

17 Q. So Dr. Bustamante, to make sure I
18 understand your interpretation of the
19 phrase "duplicate sequencing reads," are
20 the two sequences presented on my slide,
21 Exhibit 1113, duplicate sequencing reads
22 as you understand the term?

23 A. I would say sitting here today
24 right now seeing this demonstrative for
25 the first time and having no additional

1 context around how the experiment was
2 carried out, I would be hesitant to
3 provide a strong opinion on this. Again,
4 this is a demonstrative, I haven't seen
5 this, I don't know how this was derived.
6 With that caveat, by the definition of
7 identical, the top sequence and the bottom
8 sequence are not identical. So by that
9 definition, without any other context, you
10 would not necessarily conclude that these
11 are duplicates.

12 Q. And they are not duplicate
13 sequence reads as you understand that term
14 because the final base pair in these
15 sequences is a mismatch; correct?

16 A. Yes, again this is -- these are
17 two sequences on paper. I have no idea
18 how they were derived and it's a
19 hypothetical. But again, it stands to
20 reason they are not identical so they
21 would not be labeled duplicates.

22 Q. And I can provide you more
23 context. I invented these. I made them
24 up to try to understand your opinions so
25 they are not real sequencing reads. As

1 you understand the term identical, because
2 there is a mismatch in the final base pair
3 in these hypothetical reads, these would
4 not be duplicate sequencing reads;
5 correct?

6 A. In this example, this
7 hypothetical example, with no additional
8 information, using the definition of
9 identical, they are not identical so they
10 would not be considered duplicate
11 sequencing reads, by that definition.

12 (IDT Exhibit 1114, Claim
13 Construction - Scenario 1.1, marked
14 for identification.)

15 MR. FLETCHER: I'll now hand you
16 what I've labeled Claim Construction -
17 Scenario 1.1 which I have premarked as
18 Exhibit 1114.

19 MR. BEAVERS: For the record, to
20 preserve our objections I'll be
21 objecting to relevancy, hearsay,
22 foundation, scope and then completion.

23 BY MR. FLETCHER:

24 Q. Dr. Bustamante, in this scenario,
25 there is a single base mismatch but it is

1 towards the middle of the target sequence
2 region. Do you see that?

3 A. Let me review this.

4 (Witness perusing document.)

5 Again, to reiterate my caveat,
6 given no additional information, given
7 that this is a demonstrative, given I have
8 no additional context with which to
9 evaluate the question, I would say I do
10 see that there's a difference in the top
11 sequence and the bottom sequence where
12 there's a T in the top sequence and an A
13 in the bottom sequence.

14 Q. So the location of a mismatch
15 between two sequencing reads does not bear
16 on whether two sequencing reads are
17 duplicates as you understand that term;
18 correct?

19 A. So as I lay out in my report, in
20 the context of DNA sequencing you often go
21 through a processing step where you would
22 assign quality scores and you may fail to
23 make a call or not make a call at a given
24 position. Assuming this has gone through
25 quality control and processing, again this

1 is an artificial example, with all the
2 limited information I have in front of me,
3 I would say that short of processes that
4 would include trimming or where the target
5 sequence is defined as a portion of the
6 read as opposed to the entire read, in
7 this example you have shown it as the
8 entire target sequence, that would be
9 considered a difference between the top
10 strand and the bottom strand. Sorry, the
11 top sequence and the bottom sequence.
12 They are not strands.

13 Q. So that we're clear, the two
14 hypothetical sequence reads presented in
15 Claim Construction - Scenario 1.1 are not
16 duplicate sequencing reads as you
17 understand that term in the context of the
18 patent?

19 A. As I understand the term, given
20 this example and no additional context,
21 these would not be labeled as duplicate
22 sequencing reads.

23 (IDT Exhibit 1115, Claim
24 Construction - Scenario 2, marked for
25 identification.)

1 MR. FLETCHER: I'll hand you
2 Claim Construction - Scenario 2. I'll
3 give you the title and the exhibit
4 number and then you can object,
5 Mr. Beavers. Another set of
6 hypothetical sequence reads that I've
7 prepared that I've labeled Claim
8 Construction - Scenario 2 and I've
9 premarked as Exhibit 1115.

10 MR. BEAVERS: And for the record
11 we'll be objecting to relevancy,
12 hearsay, foundation, completion and
13 scope.

14 BY MR. FLETCHER:

15 Q. Dr. Bustamante, in this
16 demonstrative Exhibit 1115, there is a
17 single base mismatch in the portion of the
18 sequences that I have labeled unique
19 identifier. Do you see that?

20 A. I do.

21 Q. And if I understand your opinion
22 about the meaning of duplicate sequence
23 reads, I believe you would agree that
24 these two are not duplicate sequencing
25 reads as you understand that term in the

1 context of this patent; correct?

2 A. Again, caveating my answer to say
3 I have no additional context with which to
4 evaluate this except the document as
5 presented that I have never seen before,
6 the sequence on the top is different than
7 the sequence at the bottom which means
8 they are not identical which means they
9 would not be considered duplicate
10 sequencing reads because the unique
11 identifiers are not identical.

12 Q. Dr. Bustamante, what is a Hamming
13 distance?

14 A. A Hamming distance is a measure
15 of similarity or difference between
16 strings.

17 Q. Would you agree that a Hamming
18 distance is the number of positions at
19 which the corresponding symbols are
20 different between two strings?

21 A. Depending on the context but yes,
22 that's usually I would say a standard
23 definition, the added distance.

24 Q. As you understand claim 1 of the
25 '357 patent, the duplicate sequences must

1 have a Hamming distance of zero; correct?

2 A. They need to be identical which
3 would imply a Hamming distance of zero.

4 Q. You can set these scenarios
5 aside. There are more coming but I would
6 like to go back to your declaration.

7 If you could go back to paragraph
8 84, please.

9 A. Yes.

10 Q. You state that identifying --
11 sorry, start over.

12 In your claim construction
13 opinion, you refer to a step that uses
14 both the identifier and at least a portion
15 of the target sequence; is that correct?

16 A. That is correct.

17 Q. What do you mean by "at least a
18 portion"?

19 A. Well, one could imagine a
20 scenario where the read that is produced
21 by sequencing is longer than the intended
22 target sequence of an experiment. So in
23 some contexts, the analyst may limit or
24 may choose to limit analysis to the
25 identifier site and the portion of the

1 read that contains the target sequence.

2 Q. Can a single base be a portion?

3 MR. BEAVERS: Objection, form.

4 THE WITNESS: In general, a
5 target sequence would be more than a
6 single base.

7 BY MR. FLETCHER:

8 Q. I agree. So can a single base be
9 a portion of a target sequence?

10 A. A single base by definition would
11 be a portion of a target sequence but in
12 general, the target sequence would be
13 longer than a single base.

14 (IDT Exhibit 1116, Claim
15 Construction - Scenario 3, marked for
16 identification.)

17 MR. FLETCHER: I'll hand you
18 Exhibit 1116, another demonstrative
19 exhibit that I prepared of
20 hypothetical sequencing reads entitled
21 Claim Construction - Scenario 3.

22 MR. BEAVERS: And for the record,
23 we'll be objecting to relevancy,
24 hearsay, foundation, scope and
25 completion.

1 MR. FLETCHER: I'd love to cure
2 the completion objections but the
3 slide is the slide.

4 BY MR. FLETCHER:

5 Q. Dr. Bustamante, in Claim
6 Construction - Scenario 3 I've given you
7 two sequencing reads with identical unique
8 identifiers in the first three bases of
9 the target sequence match; correct?

10 A. Let me evaluate this for a
11 minute.

12 Again, I'm seeing this for the
13 first time and have no additional context
14 with which to evaluate. But I understand
15 the question and see that the unique
16 identifiers match in the first three bases
17 of the target sequence match.

18 Q. After the first three bases of
19 the target sequence, there are various
20 mismatches; correct?

21 A. That is correct.

22 Q. So focusing on the first three
23 base pairs of the first three bases of the
24 target sequence ATC, do you see that?

25 A. I do.

1 Q. As you understand this patent,
2 can we call the ATC at the beginning of
3 the target sequence a, quote, portion of
4 the target sequence?

5 A. Again, I think this is a
6 difficult to evaluate constructed
7 scenario. In general, we would want to
8 identify duplicate sequencing reads which
9 have unique identifier sites that are
10 identical and target sequences that are
11 identical.

12 Q. Dr. Bustamante, I'm trying to
13 understand how your understanding of this
14 claim works. So for Claim Construction -
15 Scenario 3, are the first three base pairs
16 of the target -- I'm sorry, I keep
17 misspeaking -- are the first three bases
18 of the target sequence a, quote, portion
19 of the target sequence?

20 A. They are a very small portion of
21 the target sequence.

22 Q. Can you please retrieve Claim
23 Construction - Scenario 1 in your pile of
24 exhibits? That's Exhibit 1113; correct?

25 A. That's right.

1 Q. And in this scenario, the only
2 mismatch between the two sequence reads
3 was the final base; correct?

4 A. That is correct.

5 Q. Would you consider the entirety
6 of the target sequence minus the last base
7 a portion of the target sequence?

8 A. Again, I think that would need to
9 be predefined in order to trim that base.
10 I think in general you would be limiting
11 calls to where you have unique identifier
12 and unique target sequence. But if you
13 predefined it where in reality I think you
14 need to make a distinction between what is
15 the sequencing read and what is the target
16 sequence where you would predefine what
17 counts as the portion that you'd want to
18 make identical or required to be identical
19 as part of the duplicate process.

20 Q. Dr. Bustamante, sometimes I think
21 in math which can be hard to translate to
22 words but I will try.

23 A. Okay.

24 Q. I'm trying to understand whether
25 there's a difference between these

1 concepts of target sequence and portioning
2 target sequence. And so I'll now share
3 what I understood from your opinion and
4 I'd like you to correct me if I'm wrong.

5 A. Okay.

6 Q. I had understood that for a
7 target sequence of N bases, a portion of
8 that target sequence would be any number
9 of bases from 1 to N minus 1. Do you
10 understand my scenario?

11 A. I think I'm tracking.

12 Q. And was my understanding the same
13 as your understanding of the claims?

14 MR. BEAVERS: Objection, form.

15 THE WITNESS: So as I wrote here,
16 at least a portion of the target
17 sequence and as I've written
18 throughout my opinion, you're really
19 looking for a complete match between
20 the unique identifier on both of the
21 sequencing reads and the target
22 sequence, so N match N as it were, if
23 that makes sense to you, sir, in my
24 answer.

25 ///

1 BY MR. FLETCHER:

2 Q. It makes sense to me. Hopefully,
3 it will make sense to the Board.

4 That's where I think maybe some
5 of these concepts then come together. So
6 if you could refer back to Scenario 3,
7 Exhibit 1116, as you understand the claims
8 of the '357 patent, you could compare the
9 sequence reads and you would conclude that
10 they are not duplicates, even though they
11 have the same unique identifier and a
12 portion of the target sequence matches?

13 A. Yeah, I think it stands to reason
14 that given the sequence dissimilarity
15 here, you would not consider these
16 duplicate sequencing reads.

17 Q. Even though the identifiers match
18 and a portion of the sequences match?

19 A. That is correct but the vast
20 majority of the target sequence does not
21 match and when I was considering this
22 concept of a portion, it should have been
23 predefined and not post. In this example
24 it's a particularly contrived example but
25 I think that it stands to reason that you

1 would not consider these duplicate
2 sequencing reads.

3 Q. And in Scenario 1, Exhibit 1113,
4 the identifiers match and a portion of the
5 target sequence matches. And here again,
6 you would say these are not duplicate
7 sequencing reads; correct?

8 A. By the strict definition you
9 would not consider these duplicate
10 sequencing reads.

11 Q. And as you understand this
12 patent, it has a strict definition for
13 duplicates; correct?

14 A. The patent requires the unique
15 identifiers to be identical and the target
16 sequence to be identical. What I would
17 say a caveat that if there were a
18 particular reason to predefine a region
19 for matching, that could be part of the
20 workflow but it would be predefined. I
21 think in general by the plain and clear
22 understanding, you would require unique
23 identifiers to be identical and the target
24 sequence to be identical to call them
25 duplicates.

1 Q. You can put Scenario 3 aside for
2 a while. If we could go back to paragraph
3 84. In paragraph 84, you interpret the
4 claims as requiring, quote, a step that
5 uses both the identifier and at least a
6 portion of the target sequence. Do you
7 see that?

8 A. I do.

9 Q. What do you mean by a step?

10 A. As we were discussing a often
11 computer-aided analysis that would compare
12 the sequencing reads that are produced by
13 the instrument to each other and
14 comparing, identifying those that have
15 identical identifier site and identical
16 target sequence.

17 Q. And as you understand the claims,
18 this step requires that these strings of
19 information be compared at the same time,
20 and by these strings of information, I
21 mean the identifier sequence and the
22 target sequence; is that correct?

23 A. They would be -- you would need
24 matching of the identifier site and
25 matching of the target sequence and

1 because match equals match, right, you
2 would require both to match. In terms of
3 order of processing, you would often do it
4 at the same time, but I guess one could in
5 theory match identifier sites, match
6 identical sites -- sorry, match identifier
7 sequences, match target sequence and
8 retain those that are perfect matches, but
9 in general you would often carry this out
10 at the same time.

11 Q. Probably should have used one of
12 my demonstratives.

13 A. That's okay.

14 MR. FLETCHER: Let me try to make
15 sure, I think I understand your
16 opinion.

17 (IDT Exhibit 1121, Claim
18 Construction - Scenario 5, marked for
19 identification.)

20 MR. FLETCHER: Dr. Bustamante,
21 I'll hand you what I have entitled
22 Claim Construction - Scenario 5 which
23 has more contrived sequence reads,
24 more contrived processing steps. It's
25 all contrived by me to understand your

1 opinions.

2 THE WITNESS: Okay.

3 MR. FLETCHER: I have pre-labeled
4 it IDT Exhibit 1121.

5 THE WITNESS: Can we take a break
6 before; is that okay?

7 MR. FLETCHER: That's fine.

8 (Recess taken from 10:14 a.m. to
9 10:30 a.m.)

10 MR. FLETCHER: I handed you,
11 Dr. Bustamante, what I've labeled as
12 Claim Construction - Scenario 5 which
13 I premarked as Exhibit 1121.

14 MR. BEAVERS: And I'm going to
15 for the record object for scope,
16 foundation, relevance, hearsay and
17 completion, and I don't think we yet
18 have a copy of them. Thank you.

19 MR. FLETCHER: You now have a
20 copy; correct?

21 MR. BEAVERS: Yes.

22 BY MR. FLETCHER:

23 Q. Dr. Bustamante, I was attempting
24 in scenario 5 to illustrate a workflow to
25 understand your interpretation of the

1 claims and your use of the phrase "step."

2 So on the left-hand side of my slide I
3 have eight sequence reads. Do you see
4 that?

5 A. Give me a moment to review this.

6 (Witness perusing document.)

7 Q. Nine sequence reads.

8 A. I believe there are nine, yeah.

9 (Witness perusing document.)

10 Q. What I have depicted in the
11 center of the slide is a grouping of those
12 sequence reads based on the eight bases of
13 the unique identifiers and thus created
14 three bins of sequence reads. Do you see
15 that?

16 A. I believe I'm tracking.

17 Q. And then on the right-hand side
18 of the slide I have compared the target
19 sequence portions of these sequences and
20 removed duplicates of the target sequences
21 such that Bin 1 now contains two reads,
22 Bin 2 now contains one read, and Bin 3
23 contains one read. Do you see that?

24 A. I see how in step 1 the reads are
25 clustered, if we could use that, by unique

1 identifier or what you labeled as unique
2 identifier in that you've reduced the
3 number of sequences in each of the bins
4 by, I believe, identifying duplicates.

5 Q. And Dr. Bustamante, if this
6 workflow was carried out by computer
7 program in two different comparisons, a
8 first comparison looking only at the
9 unique identifiers and a second comparison
10 looking only at the target sequences, is
11 that one step as you use that term in your
12 claim construction opinion or is that two
13 steps?

14 MR. BEAVERS: Objection, form.

15 THE WITNESS: Could you simplify
16 that question for me?

17 BY MR. FLETCHER:

18 Q. I can try. I'm trying to
19 understand what you mean by a step and how
20 one delimits things that happen on a
21 computer which might be invisible to a
22 user.

23 I'm representing to you that in
24 my hypothetical, in this clustering that
25 has occurred in the middle of the slide,

1 only the unique identifier sequences were
2 compared. In the right-hand side of the
3 slide, only the target sequence portions
4 of the reads were compared. And those two
5 comparisons happened in sequence. Is that
6 one step or two steps?

7 MR. BEAVERS: Objection, form,
8 incomplete hypothetical.

9 THE WITNESS: Still a bit of a
10 long question. Could we break that
11 down?

12 BY MR. FLETCHER:

13 Q. I don't think so. I'll ask a
14 different question.

15 Dr. Bustamante, how would you
16 understand the meaning of step in your
17 claim construction opinion?

18 A. Well, I would go back to the
19 claims and say the step being performed
20 here is identifying sequencing reads with
21 identical identifier and target sequences
22 as duplicates, and that requires you to
23 have unique identifier and unique target
24 sequence in order to tag sequencing reads
25 as duplicates.

1 Q. And so this analysis requires the
2 comparison of some sequence reads;
3 correct?

4 A. That is correct.

5 Q. And if the comparison is split
6 into two components such that first
7 identifiers are compared and then target
8 sequences are compared, is that still a
9 single step?

10 A. I would say that because the
11 condition is an if and only if, you could
12 start with a string comparison that would
13 begin with a character comparison, right,
14 so it depends on how granular you want to
15 be in defining the recipe by which it's
16 carried out; right? Just like any recipe,
17 you could further break down a step into
18 multiple steps that it comprises. So at
19 the end you need to have unique identifier
20 and -- you need to have identical unique
21 identifiers or identical identifiers and
22 identical target sequence in order to
23 identify the sequences as duplicates.

24 Q. And if my workflow compares the
25 identifiers first, then clusters them

1 based on that, then compares target
2 sequences, that is still a single step;
3 correct?

4 A. I would say so, right, and by
5 analogy I could start by comparing the
6 first character and the second character,
7 then the third character and if they are
8 identical you keep going. If they are not
9 identical, you would flag them as not
10 identical and it could be done from left
11 to right or right to left.

12 Q. I'd like to just make sure that's
13 very clear. In your opinion, if the
14 sequence comparison preceded one character
15 at a time, that would still be a single
16 step even though there are end comparisons
17 being made in sequence?

18 A. It's an if and only --

19 MR. BEAVERS: Objection, form.

20 THE WITNESS: I want to give you
21 time.

22 MR. BEAVERS: I'm good.

23 THE WITNESS: Okay. Because
24 string comparison encompasses
25 character comparison, you could go

1 down the representation, right, coded
2 in ASCII and so on; right? So I think
3 it is all part of the single
4 identifier step. You could break it
5 down as far as you would like, it is
6 still that single process by which
7 you're comparing the unique
8 identifier, ensuring that's identical
9 and the target sequencing and ensuring
10 that is identical.

11 BY MR. FLETCHER:

12 Q. You can set aside Scenario 5. If
13 you could please go back to your
14 declaration to paragraph 100. You
15 understand that the Board invited the
16 parties to address some aspects of this
17 word "comprising"; correct?

18 A. I understand.

19 Q. And you framed the question as
20 "Whether the prior art meets the claim if
21 the prior art discloses a method of
22 identifying as duplicates those sequence
23 reads with identical labels and target
24 fragments and in addition, also identifies
25 other sequence reads as duplicates,

1 including sequence reads with some
2 mutations resulting from PCR or SBS
3 error."

4 Do you see that?

5 A. I do.

6 Q. And what is your answer to that
7 question?

8 A. What's the question?

9 Q. The one the Board asked you.

10 A. (Witness perusing document.)

11 MR. FLETCHER: I'll hand you
12 Claim Construction - Scenario 4. You
13 knew it was coming, which I have
14 marked as IDT Exhibit 1117.

15 (IDT Exhibit 1117, Claim
16 Construction - Scenario 4, marked for
17 identification.)

18 MR. BEAVERS: For the record,
19 I'll object on foundation, hearsay,
20 completion, relevance and scope.

21 BY MR. FLETCHER:

22 Q. In Scenario 4, Dr. Bustamante, I
23 have provided four hypothetical sequence
24 reads on the left, two of which are
25 identical with zero mismatches. The third

1 sequence which has a single base mismatch
2 to the first two reads and a fourth
3 sequence that has a different single base
4 mismatch with the first two reads. Do you
5 see that on the left?

6 A. I do.

7 Q. And in my hypothetical, the
8 information processing method results in
9 calling all of these sequence duplicates
10 and produces a deduplicated sequence that
11 matches the top two sequences. Do you see
12 that?

13 A. I do.

14 Q. As you understand the claims of
15 the '357 patent, has this method
16 identified sequence reads with identical
17 identifier and target sequences as
18 duplicates?

19 A. It has.

20 Q. Even though it also identified as
21 duplicates two sequences that do not have
22 an identical match?

23 A. That is correct. It practices
24 the first two sequences because they are
25 identical, have been identified as

1 duplicates by the method discussed here,
2 unique identifier and unique target
3 sequence.

4 MR. FLETCHER: So now I'll hand
5 you Claim Construction - Scenario 4.1
6 which is Exhibit 1118.

7 (IDT Exhibit 1118, Claim
8 Construction - Scenario 4.1, marked
9 for identification.)

10 MR. BEAVERS: For the record,
11 I'll object based on relevancy, scope,
12 hearsay, foundation and completeness.

13 MR. FLETCHER: I wish I knew how
14 to cure that objection.

15 BY MR. FLETCHER:

16 Q. Dr. Bustamante, in Scenario 4.1 I
17 have removed the second read sequence as
18 compared to Scenario 4. Do you see that?

19 A. I do.

20 Q. And in this hypothetical
21 processing step the method has called all
22 three of these reads as duplicates and
23 produced a sequence matching the first
24 sequence. Do you see that?

25 A. I do.

1 Q. As you understand the claims of
2 the '357 patent, this method has not
3 identified sequence reads with identical
4 identifier and target sequences as
5 duplicates; correct?

6 A. Well, first, again caveat, it's
7 my first time seeing this. This was not
8 provided to me earlier and I have no
9 additional context of what you've
10 represented in this hypothetical. So in
11 this context, the unique identifiers are
12 all identical for the three sequences and
13 the two bottom sequences, so let's label
14 them 1, 2 and 3 from the top. So sequence
15 2 and sequence 3 differ by one nucleotide
16 from the top sequence. If the top
17 sequence had been derived using the
18 Scenario 4 or the first two of Scenario 4
19 had been collapsed and had already labeled
20 duplicates, then it would have practiced
21 the identifying step. So if you'd have
22 done that duplication identification first
23 and that's how you arrived at that top
24 sequence, then it is essentially a
25 comprising of what is in Scenario 4.

1 Q. Okay. But let me clarify that
2 they are similar but they are not
3 interconnected that way. Scenario 4.1 in
4 this hypothetical, my sequencer has only
5 given me three sequencing reads and then
6 my methodology has deduplicated those
7 reads to a single sequence. As I
8 understand your opinion, I think you would
9 say the method of identifying sequence
10 reads in the '357 patent is not met by
11 this information processing on Scenario
12 4.1?

13 A. If there are no additional reads
14 in the dataset that are identical in
15 identifier and target sequence to the
16 first sequence, as is the case in Scenario
17 4 which is flagged for duplicate
18 identification, in the contrived example
19 where these are the only three sequences
20 produced and in which there wasn't a
21 decision made to consider the portions of
22 the sequencing reads that are identical as
23 identical, if that had not been
24 predefined, then you would not have
25 defined duplicates in that way. In the

1 event that the first sequence was derived
2 because there were other identical
3 sequencing reads and already marked as
4 duplicates or in the event the analyst
5 decision had been to look through and
6 define the regions here as not part of the
7 matching criteria, then that would be the
8 case.

9 (IDT Exhibit 1119, Claim
10 Construction - Scenario 4.2, marked
11 for identification.)

12 MR. FLETCHER: I'll now hand you
13 Scenario 4.2 which I have labeled as
14 Exhibit 1119.

15 MR. BEAVERS: So for the record
16 I'll object based on relevancy,
17 hearsay, scope, foundation and
18 completeness.

19 BY MR. FLETCHER:

20 Q. Dr. Bustamante, in this
21 hypothetical scenario that I've created,
22 there are three sequencing reads on the
23 left and my algorithm compares the
24 identifier and target sequence strings and
25 removes duplicate sequences to create what

1 is shown on the right, the deduplicated
2 sequence. Do you follow that
3 hypothetical?

4 A. I believe so. Again, I'm seeing
5 this for the first time and I don't see
6 any red so I presume there are no
7 mismatches and that they are -- you're
8 representing that they are identical
9 sequences in both target and unique
10 identifier, if I understand the question
11 correctly.

12 Q. Correct.

13 A. Okay.

14 Q. And in this scenario, with the
15 representations, you would agree that this
16 algorithm has identified sequencing reads
17 with identical identifier and target
18 sequences as duplicates as you understand
19 that limitation of the patent?

20 A. Again, given the caveat that
21 having seen this for the first time having
22 no additional context, the fact that you
23 have unique identifier and unique -- you
24 have unique identifiers that are identical
25 and target sequences that are identical,

1 this would meet the definition of
2 duplicate sequencing reads.

3 (IDT Exhibit 1120, Claim
4 Construction - Scenario 4.3, marked
5 for identification.)

6 MR. FLETCHER: Now I'll hand you
7 Claim Construction - Scenario 4.3
8 which I've labeled as Exhibit 1120.

9 MR. BEAVERS: For the record,
10 we'll be objecting for relevance, for
11 hearsay, scope, foundation and
12 completeness.

13 BY MR. FLETCHER:

14 Q. Dr. Bustamante, in Scenario 4.3 I
15 have presented you on the left with the
16 same three sequence reads as Scenario 4.2
17 and the same end result on the right-hand
18 side, but I have changed the algorithm I
19 used to generate that sequence and I've
20 labeled it as consensus sequence. And I
21 generated the sequence on the right by
22 comparing the three strings on the left
23 character by character by character one
24 character at a time and selecting the most
25 common character at each position. Do you

1 follow that scenario?

2 A. I follow the scenario.

3 Q. As you understand the claims and
4 have interpreted them, does my Scenario
5 4.3 identify sequence reads with identical
6 identifier and target sequences as
7 duplicates?

8 MR. BEAVERS: Objection, scope.

9 THE WITNESS: So the fact that
10 you have arrived at a sequence called
11 the consensus sequence does not impact
12 the fact that identifying sequences
13 that have unique identifier and unique
14 target sequence by the method here
15 would identify them as duplicates.

16 BY MR. FLETCHER:

17 Q. Dr. Bustamante, I'll re-ask or
18 attempt to clarify my question.

19 The difference between Scenario
20 4.2 and Scenario 4.3 is a difference in
21 the algorithm, the hypothetical algorithm
22 that goes from the left to the right. And
23 in Scenario 4.3 the algorithm compares the
24 reads character by character and selects
25 the most frequent character at the

1 position, but the algorithm does not at
2 any point compare the entirety of the
3 unique identifier or the target sequence.

4 Do you understand that hypothetical?

5 A. I do understand that
6 hypothetical.

7 Q. So then with that in mind, do you
8 believe that that hypothetical algorithm
9 constitutes identifying sequence reads
10 with identical identifier and target
11 sequences as duplicates?

12 MR. BEAVERS: Objection, scope.

13 THE WITNESS: So as I understand
14 the question, if you have arrived at
15 the sequence on the right-hand side as
16 the consensus sequence of the
17 identical reads on the left-hand side,
18 does that not also mean because they
19 are identical across all of the
20 characters, pair wise identical, that
21 you have also identified duplicates
22 through comparison of the unique
23 identifier string and the target
24 sequence strings? The end result is
25 the identical sequence and one can

1 imagine scenarios where the consensus
2 sequence arrives at a sequence that is
3 identical to the sequence that would
4 be the matching sequence when pair
5 wise unique identifier and target
6 sequences are identical. So the fact
7 that you have derived a consensus
8 sequence in my opinion first is not
9 the same thing as duplicate
10 identification necessarily. It's a
11 consensus sequence. And secondly, it
12 doesn't impact the fact that you still
13 have duplicate sequencing reads here
14 by the definition of reads that
15 contain unique identifier and unique
16 target sequence in a pair wise fashion
17 across the three.

18 BY MR. FLETCHER:

19 Q. So Dr. Bustamante, does it or
20 does it not constitute identifying
21 sequence reads with identical identifier
22 and target sequences as duplicates as you
23 understand that limitation?

24 MR. BEAVERS: Objection, scope,
25 asked and answered.

1 THE WITNESS: As I've tried to
2 answer, the fact that the consensus
3 sequence happens to be identical to
4 the duplication signature, if we want
5 to call it that, of identical unique
6 identifier and identical target
7 sequence in a pair wise fashion across
8 the three sequences does not mean that
9 you do not also have duplicate
10 sequencing reads here by the
11 definitions of the stated patent.

12 BY MR. FLETCHER:

13 Q. I contrived this example so that
14 the consensus sequence and the
15 deduplication produced the same sequence.
16 What I'm trying to understand from you,
17 Dr. Bustamante, is whether it matters that
18 the algorithms are different for purposes
19 of the claims of the '357 patent.

20 A. Whenever the reads have unique
21 identifier and unique target sequence, and
22 you have used matching to identify the
23 pair of unique identifier and unique
24 target sequence, and used that to identify
25 duplicates, you identify duplicates by the

1 definition of the patent.

2 Q. So in this Scenario 4.3 where a
3 consensus sequence has been generated by
4 character by character pair wise
5 comparison, have sequence reads with
6 identical identifier and target sequence
7 been identified as duplicates?

8 MR. BEAVERS: Objection, scope.

9 THE WITNESS: Again, you have as
10 I understand it, again I'm seeing this
11 for the first time, I have no
12 additional information on how this was
13 derived except that from what I can
14 glean the document Scenario 4.2 and
15 document Scenario 4.3 differ in what
16 you've labeled deduplication in 4.2
17 and what you've labeled consensus
18 sequencing in 4.3.

19 BY MR. FLETCHER:

20 Q. That's correct, that's the only
21 difference.

22 A. Okay. So the fact of the
23 consensus sequence happens to be identical
24 to all of the sequencing reads does not
25 obviate the fact that comparison of the

1 unique identifier and the target sequence
2 pair wise would identify them as
3 duplicates. It is an extra piece of
4 information that is provided here. It is
5 the consensus sequence, I would agree that
6 is the consensus sequence. I see nothing
7 in this document that labels these as
8 duplicates.

9 Q. So as you understand the patent
10 and the claims, if the algorithm that is
11 being used is a consensus sequence
12 generation algorithm, is merely doing
13 character by character analysis and
14 selecting the most common character
15 definition, it is not practicing the
16 identifying limitation of claim 1?

17 MR. BEAVERS: Objection, scope,
18 asked and answered.

19 THE WITNESS: In my opinion, the
20 calculation of a consensus sequence,
21 whatever it happens to be, does not
22 take away from the fact that duplicate
23 reads would be identified using the
24 unique identifier that are identical
25 in target sequences that are

1 identical, unique identifiers that are
2 identical and target sequences that
3 are identical in at least a pair wise
4 fashion. So there are multiple ways
5 that can be accomplished. You start
6 with a pair, define those as
7 identical, add another sequence that
8 is identical to that, that would
9 arrive at the same conclusion
10 regarding duplication that would also
11 have the same consensus sequence. So
12 simply by calculating a consensus
13 sequence you, in my opinion, do not
14 get around the fact that these have
15 unique identifier and unique target
16 sequence in a pair wise fashion and in
17 a multi way fashion.

18 BY MR. FLETCHER:

19 Q. You just said you do not get
20 around to that fact. Did I hear you
21 correctly?

22 A. We could calculate additional
23 statistics, we could calculate the Wordle
24 or the word distribution, the base
25 distribution, as it were. At each site,

1 the logo is often done in motif and it
2 would be all identical for each of the
3 columns.

4 Q. So perhaps I misunderstood your
5 opinions. Let me try and reexamine them.

6 Dr. Bustamante, is it your
7 opinion that a algorithm that generates a
8 consensus sequence from two or more reads
9 can practice the limitation of identifying
10 sequence reads with identical identifier
11 and target sequences as duplicates?

12 MR. BEAVERS: Objection, scope,
13 asked and answered.

14 THE WITNESS: I think I addressed
15 that in my previous answer. The fact
16 that you calculate a consensus
17 sequence does not take away from the
18 fact that comparing unique identifier
19 and target sequence and finding they
20 are identical for pair wise or multi
21 way reads identifies them as
22 duplicates.

23 BY MR. FLETCHER:

24 Q. I agree it does not take away
25 from the fact, Dr. Bustamante, but I'm

1 positing to you a hypothetical algorithm
2 that does not compare identifier strings
3 or target sequence strings, it instead
4 proceeds character by character to
5 generate a consensus sequence. Can that
6 meet the requirement of identifying
7 sequence reads with identical identifier
8 and target sequences as duplicates?

9 MR. BEAVERS: Objection, scope,
10 asked and answered.

11 THE WITNESS: I can try again.
12 The consensus sequence from the set of
13 reads may arrive at the identical
14 sequence as one would use to match
15 unique identifier and target sequence
16 in identifying duplicate reads. It
17 may or may not. Depends on the
18 scenario. In this scenario that
19 you've provided for me where all the
20 sequences are identical, it stands to
21 reason that it will give you the same
22 answer and oftentimes different
23 methods may give you the same answer.
24 That doesn't take away from the fact
25 that using unique identifiers and

1 target sequence to identify sequences
2 that are identical in the unique
3 identifier and the target sequence
4 identifies duplicates by the
5 definition of the patent. As long as
6 the unique identifier are identical
7 and the target sequence are identical,
8 then the patent would identify them or
9 embodiment of the patent would
10 identify them as identical sequencing
11 reads.

12 BY MR. FLETCHER:

13 Q. And that method can be performed
14 character by character or identifier and
15 then target sequence or entire string in
16 any order?

17 A. As we've said, it's an if and
18 only if statement; right? So as you break
19 down the comparison, you would be -- you
20 could match -- you would start matching
21 and keep matching until the end of a
22 string and then define them as identical.
23 If as you're matching you find they are
24 not identical, you would stop matching.
25 So I think the comprising language here

1 means you can break that down into
2 multiple ways of doing the comparison of
3 unique identifier and target sequence and
4 as long as the unique identifier and the
5 target sequence are identical across at
6 least pairs of reads, you would flag them
7 by this method as duplicates.

8 Q. Okay, let's put the scenarios to
9 the side for now.

10 A. Okay.

11 Q. I'm fond of them. We may come
12 back but why don't we turn back to the
13 patent, Exhibit 1004.

14 If you could please go to column
15 16 of the patent.

16 Column 16, line 47 or so begins
17 with example 1 entitled Identification of
18 Duplicate Sequencing Reads with NuGEN
19 Ovation Target Enrichment Library System.
20 Do you see that?

21 A. I do.

22 Q. Have you used this NuGEN system
23 in your research before?

24 A. I'm not sure if I have. We've
25 done a lot of experiments and we have

1 collaborated with a lot of people so we
2 may have. I honestly can't recall one way
3 or another.

4 Q. In your research, Dr. Bustamante,
5 is it fair to say that your colleagues do
6 the sequencing and you analyze the
7 sequencing data?

8 A. No, we sometimes do the
9 sequencing in the laboratory and have done
10 so, so it's under my direction.

11 Q. When you have done the
12 sequencing, what sequencers have you used?

13 A. We've used a lot of sequencers.

14 Q. Which one is your preferred
15 sequencer?

16 A. That's a difficult question given
17 that we work with a lot of sequencing
18 vendors. I'm going to say we have a lot
19 of sequencers we use. We use the Illumina
20 system, we use the Oxford Nanopore system,
21 we've in the past also worked with other
22 sequencing vendors.

23 Q. Can you recall ever using any
24 NuGEN product?

25 A. Again, I'm sitting here right

1 now, I can't point to a particular paper
2 that I recall using NuGEN. That doesn't
3 mean that we haven't in the past or
4 collaborators haven't in the past. I
5 can't tell you with certainty.

6 Q. So I would like to discuss this
7 example 1 --

8 A. Okay.

9 Q. -- and compare it to the claim so
10 you may need to do a little flipping back
11 and forth.

12 A. Okay.

13 Q. At line 59, they describe the
14 library generation for example 1; correct?

15 A. That's what is stated here,
16 library generation.

17 Q. And they describe using an
18 identifier site having a random six base
19 sequence; correct?

20 A. Give me a minute to consult here
21 the document.

22 (Witness perusing document.)

23 I see that it says step 3 and
24 identifier site having a random six base
25 sequence, if that's what you're referring

1 to.

2 Q. I am.

3 A. Okay.

4 Q. So the adaptor being used in
5 example 1 contains a unique identifier;
6 right?

7 A. It includes an identifier site.

8 Q. And six nucleotides is between
9 about one and about eight nucleotides;
10 correct?

11 A. I would agree with that
12 statement.

13 Q. This adaptor being described at
14 the bottom of column 16 also contains an
15 indexing site; correct?

16 A. It says to an indexing site so I
17 believe it does.

18 Q. This adaptor also contains a
19 primer binding site; correct?

20 A. It says it contains a sequence
21 compatible with an Illumina forward
22 sequencing priming site.

23 Q. If we carry over to column 17,
24 the example states that the adaptors were
25 ligated onto the end-repaired DNA;

1 correct?

2 A. Give me a minute just to read
3 down column 17.

4 (Witness perusing document.)

5 That's what it states in that
6 first complete sentence, I believe.

7 Q. So now to try to do this
8 efficiently, if you could please refer to
9 claim 1.

10 A. Okay.

11 Q. You would agree that the
12 methodology being described in example 1
13 of the '357 patent meets this first
14 ligating an adaptor limitation; correct?

15 A. I would agree with that.

16 Q. Okay. So back to column 17.
17 Line 21.

18 A. Okay.

19 Q. "These libraries were enriched by
20 30 cycles of PCR using NuGEN library
21 enrichment primers"; correct?

22 A. Give me one second to reread
23 this.

24 (Witness perusing document.)

25 I'm sorry, your question was

1 about line?

2 Q. My question was first to direct
3 you to line 21 indicating that these
4 libraries were enriched by 30 cycles of
5 PCR. Do you see that?

6 A. I do. I had heard 31. That's
7 why I wanted to double check. Okay, yes.
8 Line 21.

9 Q. So Dr. Bustamante, you would
10 agree the method being described in
11 example 1 meets the second limitation of
12 claim 1, the amplifying the
13 adaptor-ligated fragments into amplicons;
14 correct?

15 A. I believe that is correct.

16 Q. Now if we could go to line 32 of
17 column 17, I guess also line 29 to 32,
18 there's a reference to sequencing these
19 libraries on an Illumina MiSeq DNA
20 sequencer. Do you see that?

21 A. I do.

22 Q. And you would agree that this
23 method of example 1 meets the third
24 limitation of claim 1, the sequencing the
25 amplicons to produce sequence reads that

1 include identifier and target sequences;
2 correct?

3 A. That would be met by sequencing
4 on a MiSeq instrument.

5 Q. Dr. Bustamante, I would like to
6 discuss the data analysis pipeline of
7 example 1 of the '357 patent. At line 32
8 the first step of, quote, our data
9 analysis pipeline is sequence alignment
10 with bowtie aligner; correct?

11 A. I'm sorry, where are you pointing
12 to?

13 Q. I may have given you the wrong
14 line. Line 39.

15 A. So I would say the data analysis
16 starts on line 32 where they do processing
17 of the reads. That's where the data
18 analysis would start.

19 Q. That's fair. And in that first
20 step the reads are now split into two
21 different files effectively, a file
22 containing the identifier reads and a file
23 containing the target sequence reads; is
24 that fair?

25 A. So there are three kinds of reads

1 that are generated by the instrument, what
2 is normally called the forward, the index
3 and the reverse. So in this case, the
4 index read which is 14 bases was split
5 into eight bases for the indexing read,
6 which would be call them the sample
7 indexes, and then six bases which would be
8 the random bases which would be the
9 identifier sites.

10 Q. After this initial processing,
11 the next step is sequence alignment with
12 bowtie aligner; correct?

13 A. There is a step where they do
14 data analysis by doing a standard
15 alignment by the bowtie aligner.

16 Q. What do you mean by standard
17 alignment?

18 A. So bowtie was one of the short
19 read aligners that was available at the
20 time to do read alignment to the genome.

21 Q. To be clear, the bowtie program
22 will align the target sequence to a
23 reference genome; is that correct?

24 A. Yes, bowtie is a short read
25 aligner that is used to align reads to

1 reference genomes or other sequence files
2 of interest but usually it's referencing
3 genome.

4 Q. And the end result of doing the
5 bowtie alignment is that each read gets
6 tagged with a genomic start position;
7 correct?

8 A. That is one of the outputs of the
9 bowtie aligner.

10 Q. The next step in the patent's
11 data analysis pipeline is to group the
12 sequencing reads that start at the same
13 genomic position and check them against
14 the identifier sequences; correct?

15 A. So by my read, sequencing reads
16 that start at the same genomic position
17 were checked against the random bases;
18 i.e., the file with identifier sites, to
19 see if they have the same or different set
20 of random bases ligated to them.

21 Q. And the inventors describe in
22 example 1 that where two sequences have
23 the same starting genomic coordinate and
24 the same identifier sequence, they were
25 considered to have come from the same

1 event; correct?

2 A. That's what they write.

3 Q. And they were considered as a
4 single sequencing read for purposes of
5 their analysis; correct?

6 A. Give me a second. What they
7 write is that the two sequences with the
8 same starting genomic coordinates have the
9 same set of random base and -- excuse me,
10 "Where two sequences with the same
11 starting genomic coordinate had the same
12 set of random bases, they were considered
13 to have come from the same initial DNA
14 ligation event, regardless of which
15 Ovation Target Enrichment targeting probe
16 was used to generate the sequence fragment
17 in question."

18 Q. The inventors then continued,
19 stated that "These sequences therefore,
20 did not provide unique information about
21 the starting genomic DNA and are
22 considered as one sequencing read for the
23 purpose of variant analysis"; correct?

24 A. That is what they write.

25 Q. They then note that two sequence

1 reads with the same starting genomic
2 coordinate but different identifier
3 sequences were derived from unique events
4 and each were considered to be discrete
5 sequencing reads; correct?

6 MR. BEAVERS: Objection,
7 misstates the record.

8 THE WITNESS: I think what they
9 write is considered to be both -- were
10 considered to both be valid sequencing
11 reads for the purpose of variant
12 identification.

13 BY MR. FLETCHER:

14 Q. I was trying to paraphrase.

15 A. That's fine.

16 Q. Do you agree with my
17 paraphrasing?

18 A. I would say they write valid.

19 You said --

20 Q. Discrete.

21 A. Discrete. Okay.

22 Q. Whereas in the prior discussion,
23 two sequences with the same identifiers
24 and the same genomic start coordinates
25 were deemed a single read; correct?

1 A. That's what they wrote.

2 Q. They then refer the reader of the
3 patent to Figure 5 for the analysis
4 results demonstrating the identification
5 of the duplicate reads; correct?

6 A. They reference Figure 5.

7 Q. In example 1, duplicate reads are
8 identified using the genomic start
9 coordinate and the identifier sequence;
10 correct?

11 A. So they use mapping as part of
12 their workflow because they are using this
13 capture product. And when they use the
14 mapping sequencing reads that start at the
15 same genomic position and have
16 identical -- have the same set of random
17 bases that were considered -- excuse me,
18 where the two sequences -- I'm just
19 reading from here, "Where two sequences
20 with the same starting genomic coordinate
21 had the same set of random bases, they
22 were considered to have come from the same
23 initial DNA ligation."

24 Q. So in example 1 the inventors
25 identified duplicates using a genomic

1 start coordinate and the identifier
2 sequence; correct?

3 A. The genomic start coordinate was
4 used as part of the identification
5 process, given the experimental set up
6 where they have probes that could have
7 overlapped and pulled the native fragment
8 for sequencing.

9 Q. The answer though is yes, the
10 information that was used to identify
11 duplicates in example 1 was the genomic
12 start coordinate and the identifier
13 sequence; correct?

14 A. Included the genomic coordinate
15 as part of the workflow.

16 Q. The workflow is not described as
17 including a comparison of the target
18 sequence; correct?

19 A. Let's turn to Figure 5. So my
20 read here of that section and of Figure 5
21 is that they were interested in
22 identifying, quote, apparent duplicates
23 using conventional measures of duplicate
24 reads; i.e., reads mapped using bowtie and
25 all reads with the same start and end

1 genomic coordinates were counted as
2 duplicates as part of the demonstration of
3 the technology they were using, standard
4 workflow.

5 Q. Thank you for referring us to
6 Figure 5. Figure 5 and example 1
7 distinguish between, quote, apparent
8 duplicates and, quote, true duplicates;
9 correct?

10 A. That's what it states here in
11 quotation marks, duplicates and identifier
12 sequence introduced through ligation.

13 Q. And the apparent duplicates are
14 identified using only the genomic
15 coordinates; correct?

16 A. That is my understanding.

17 Q. And the true duplicates are
18 identified by the inventors using the
19 genomic coordinates and the identifier
20 sequences; correct?

21 A. In quotation mark true
22 duplicates, in quotation mark apparent
23 duplicates, that's what it states here.

24 Q. As you understand and have
25 interpreted claim 1, the methods and

1 research described in example 1 are
2 outside the scope of claim 1 because there
3 has been no comparison of the target
4 sequences?

5 A. I can't point to a section here
6 where they specifically discuss comparing
7 the target sequence and ensuring that they
8 are identical. I believe this example is
9 meant to illustrate the value of using the
10 identifier sites in a standard workflow,
11 in a standard analysis workflow that uses
12 the mapping coordinates to identify
13 apparent duplicates.

14 Q. So Dr. Bustamante, the answer to
15 my question is as you understand claim 1,
16 claim 1 was not practiced or described in
17 example 1 because the inventors in example
18 1 used only the genomic coordinates rather
19 than the target sequence?

20 MR. BEAVERS: Objection, form.

21 THE WITNESS: They used at least
22 the genomic -- they used the genomic
23 coordinates in describing Figure 5 and
24 the value afforded by using identifier
25 sites to move from, quote, apparent

1 duplicates to true duplicates. It is
2 unclear, it is not apparent here
3 whether or not additional steps were
4 used in this example that ended up
5 comparing the target sequence and
6 ensuring those were identical in their
7 definition of true duplicates. It is
8 not stated here in an obvious way for
9 me to determine.

10 BY MR. FLETCHER:

11 Q. In the absence of that statement,
12 you would say if all they did was use the
13 genomic coordinates, they were not
14 practicing claim 1 as you understand it?

15 MR. BEAVERS: Objection, form.

16 THE WITNESS: In order to
17 practice claim 1, you need to be
18 comparing the identifier sites and the
19 target sequence sites and ensuring
20 they are identical.

21 BY MR. FLETCHER:

22 Q. And so when the inventors were
23 identifying, quote, true duplicates in
24 example 1 and Figure 5, it is your opinion
25 that they were not practicing the claimed

1 method; correct?

2 MR. BEAVERS: Objection, form.

3 THE WITNESS: I don't have

4 additional information one way or

5 another that tells me whether or not

6 they were using the target sequence as

7 a step -- target sequence comparison

8 as part of the comparison of the tags.

9 They might have been. It's not

10 apparent here. I'm not sure that was

11 the purpose of this experiment. If

12 they did do that, then they would have

13 been practicing the invention.

14 BY MR. FLETCHER:

15 Q. Returning to Figure 5, you'd

16 agree on the bottom of the figure that a,

17 quote, true duplicate is one that has the

18 same identifier sequence and the same

19 start and end mapping coordinates as that

20 concept is used in Figure 5; correct?

21 A. Again, true in quotation mark

22 duplicates for purpose of experiment 1 is

23 defined as an identifier sequence --

24 through the identifier sequence that's

25 introduced to differentiate fragments that

1 have the same start and end mapping
2 coordinates, many of which are presumably
3 identical.

4 Q. Dr. Bustamante, you did not
5 discuss example 1 in your declaration;
6 correct?

7 A. I don't believe I go through
8 example 1 in the declaration but we could
9 double check. I think that's correct.

10 Q. Did you consider example 1 in
11 forming your claim construction opinions?

12 A. I did.

13 Q. Would you agree that the Board
14 should account for example 1 in forming
15 its construction of the claims of this
16 patent?

17 A. I'm not a lawyer or a judge and
18 I'm in no position to weigh in on the
19 Board's procedures. I'm sure they
20 consider the totality of the evidence
21 presented to them.

22 Q. You do in your declaration
23 discuss example 5; correct?

24 A. I do.

25 Q. So you would agree that it is

1 relevant and important for the Board to
2 consider the examples; correct?

3 A. Absolutely. And I'd like to make
4 a kind of clarification which is given the
5 very short nature of the reads that are
6 produced by the MiSeq, it is very likely
7 the case that when you have identical
8 start and end positions, those reads that
9 are aligning to those positions are nearly
10 if not always identical.

11 Q. And by very short you're
12 referring to the 36 base --

13 A. Yeah, the 36 base pair forward
14 read.

15 Q. Can you please go further down
16 column 17 to line 64?

17 A. In example 3?

18 Q. In example 1. It's column 17.

19 A. Okay.

20 Q. This paragraph that goes from
21 column 17, line 64 to column 18, line 5 is
22 part of example 1; correct?

23 A. It is.

24 Q. And in this passage, the
25 inventors are explaining the logic behind

1 example 1; correct?

2 A. They are.

3 Q. And their logic is that the
4 identifier sequence can be used to help
5 distinguish the true duplicates from the
6 apparent duplicates; correct?

7 A. In the context of when sequences
8 of two libraries are identical. So if two
9 libraries have identical target sequence,
10 their duplication status is unknown since
11 they could occur by chance in any library.
12 If an identifier sequence is used in
13 combination with the library reads, the
14 status can be determined. Duplicate and
15 identical is distinctly different.

16 Q. The phrase target sequence didn't
17 actually appear in what we're reading;
18 correct?

19 A. It says if the sequences of the
20 two libraries are identical. I would
21 presume that would be target sequence.

22 Q. I was just making sure you're
23 distinguishing what's in quotes.

24 A. And that's separate from the
25 discussion of Figure 5.

1 Q. It follows the discussion of
2 Figure 5; correct?

3 A. It does but it's not in the same
4 paragraph.

5 Q. Now, turning to --

6 A. In some sense that is a place
7 where they are discussing identical target
8 sequence in example 1.

9 Q. The final paragraph of example 1
10 then discusses the data that appear in
11 Figure 5; correct?

12 A. I believe that data comes from
13 Figure 5.

14 Q. So turning to example 2.

15 A. I see example 2.

16 Q. Column 18, line 38 it reads,
17 "Standard PCR duplicate removal software
18 such as PICARD, Markduplicates, and/or
19 SAMtools rmdup is used to identify and
20 remove the PCR duplicates."

21 Do you see that?

22 A. I do.

23 Q. And Dr. Bustamante, you are
24 familiar with each of PICARD,
25 Markduplicates and SAMtools rmdup;

1 correct?

2 A. I'm certainly familiar with
3 PICARD and SAMtools. I think I've used
4 Markduplicates but in general I'm familiar
5 with these sort of workflows.

6 Q. These were software workflows
7 that were available to a person of
8 ordinary skill in the art; correct?

9 A. They were available at the time.

10 Q. And they were standard software
11 at the time?

12 A. They were part of many workflows.

13 Q. You would agree that NuGEN/Tecan
14 did not invent the idea of using software
15 to identify duplicate sequences; correct?

16 MR. BEAVERS: Objection, form.

17 THE WITNESS: I don't believe
18 they've claimed that.

19 BY MR. FLETCHER:

20 Q. They haven't claimed it and they
21 didn't invent that idea; correct?

22 MR. BEAVERS: Objection, form.

23 THE WITNESS: I'm happy to agree
24 that people have been using software
25 to analyze DNA sequences for a long

1 time.

2 BY MR. FLETCHER:

3 Q. Okay. And this patent, the '357
4 patent, does not disclose any new software
5 for identifying duplicates; correct?

6 A. As I understand it, the
7 patentability of software is something we
8 could spend a lot of time on. So my
9 understanding is that the patent is really
10 focused on the method for detecting
11 duplicate sequencing reads that makes use
12 of the combined, as you've stated,
13 physical, chemical steps and the
14 computational steps, or analytical steps.

15 Q. And my point, Dr. Bustamante, is
16 that the patent is premised on the idea
17 that people already have software that can
18 carry out the steps that are being
19 described in the patent; correct?

20 MR. BEAVERS: Objection, form.

21 THE WITNESS: Well, I'm not sure
22 I would agree with that. They --
23 these steps aren't necessarily, and
24 certainly aren't, what we would
25 describe as the identifying step.

1 They are using different approaches.

2 BY MR. FLETCHER:

3 Q. You would agree that the existing
4 standard software is being used to carry
5 out the methods described in the patent;
6 correct?

7 MR. BEAVERS: Objection, form.

8 THE WITNESS: No, what it states
9 here is they are performing standard
10 PCR duplicate removal leaving behind
11 for analysis any instance of multiple
12 cDNA fragments that happen to have the
13 same sequence. That's what they
14 wrote.

15 BY MR. FLETCHER:

16 Q. So in example 2, the standard
17 software is being used to compare sequence
18 reads that include an eight base
19 identifier site and a target sequence;
20 correct?

21 A. It states here that the pool of
22 identifier sites is used for the detection
23 of PCR duplicates. Those PCR duplicates
24 will contain identical identifier sites;
25 whereas, two different cDNA molecules will

1 ligate to two different adapters
2 containing two different identifier sites,
3 so using the identifier sites, they are
4 looking for PCR duplicates where the
5 identifier sites are identical and used in
6 conjunction with the rest of the
7 sequencing reads.

8 Q. And to achieve that goal of
9 analyzing the eight base identifier and
10 target sequence, the inventors described
11 using standard duplicate removal software
12 to identify and remove the PCR duplicates;
13 correct?

14 A. That's what they wrote.

15 Q. So in view of these three
16 standard options, you would agree that the
17 claim does not require any particular
18 algorithm for identifying duplicates;
19 correct?

20 A. It -- the claim certainly
21 requires that the identifier sequences be
22 identical and that the target sequences be
23 identical.

24 Q. The claim does not require any
25 specific software package that does that;

1 correct?

2 A. I don't see any software package
3 in the limitation.

4 Q. So let's go to example 5 then.

5 A. Okay.

6 Q. Example 5 is entitled
7 Identification of Duplicate Sequencing
8 Reads in Determining mRNA Expression
9 Levels; correct?

10 A. That is correct.

11 Q. And the library generation step
12 is described at line 64; correct?

13 A. Starts on line 64 of column 19 as
14 I read it.

15 Q. And the example discloses some of
16 the sequences that were used for the
17 library generation; correct?

18 A. That is correct.

19 Q. These sequences in column 20 are
20 the sequences for the adaptors that were
21 used in example 5; correct?

22 A. That's what I understand.

23 Q. The adaptors contain an eight
24 base pair identifier site which is
25 indicated by the capital N; correct?

1 A. That is correct.

2 Q. And the reverse adaptors also
3 contained a six base pair index sequence
4 that is identified by underlining;
5 correct?

6 A. That is correct in my
7 declaration, I believe.

8 Q. All of these adaptors were
9 obtained by NuGEN from Integrated DNA
10 Technologies; correct?

11 A. They were ordered from IDT as it
12 states here, as I understand it.

13 Q. And at line 52, we get a
14 description of the sequencing runs that
15 were performed on the Illumina GAIIX MiSeq
16 or HiSeq; correct?

17 A. That's what they wrote.

18 Q. And they did two reads rather
19 than three as in example 1; correct?

20 A. That's what I understand.

21 Q. The first read was a 50 base
22 first read; correct?

23 A. That's what they wrote.

24 Q. And the six base second; correct?

25 A. That's my understanding.

1 Q. This second read was for
2 sequencing the six base pair index;
3 correct?

4 A. That's my understanding.

5 Q. So the substantive read was the
6 50 base pair first read; correct?

7 MR. BEAVERS: Objection, form.

8 THE WITNESS: If by substantive
9 we mean the read that contains the
10 target sequence and the identifier
11 site.

12 BY MR. FLETCHER:

13 Q. That is what I meant.

14 A. Okay.

15 Q. And that is 14 base pairs longer
16 than the first read that we saw in example
17 1; correct?

18 A. That's -- yes, 36 is 14 bases
19 shorter than 50.

20 Q. I believe you described the
21 sequencing run in the first example in
22 your words as very short; correct?

23 A. Short, probably said very short.

24 Q. Which word would you use or none?

25 A. 36 is certainly kind of the -- on

1 the shorter end of the distribution that
2 you would want to use.

3 Q. In this example they did 50
4 base --

5 A. That's correct. They are doing
6 cDNA sequencing here so that was I would
7 say relatively standard to do single N 50
8 base pair reads for cDNA.

9 Q. We then get to the data analysis
10 in line 55 of column 20; correct?

11 A. Column 20, line 55 starts data
12 analysis.

13 Q. And first they demultiplexed the
14 sequencing information using the index
15 reads; correct?

16 A. That's what I understand.

17 Q. They then did sequence
18 comparisons using the 50 bases; correct?

19 A. Correct.

20 Q. They did not use a portion of the
21 sequencing read, they used the entire
22 sequencing read; correct?

23 A. They write here, "The first 50
24 bases of the target sequence read are
25 compared to each other."

1 Q. And that was the entire --

2 A. That's the entire and that
3 includes the eight base pairs of
4 identical -- of identifier site, so 42
5 bases of sequence and eight bases of
6 identifier site.

7 Q. And on that basis they identified
8 and removed duplicates; correct?

9 A. That is correct.

10 Q. And in this example, after having
11 removed duplicates, they aligned the reads
12 to the reference genome; correct?

13 A. That is correct.

14 Q. And I believe you mentioned in
15 your declaration that these various data
16 processing steps and the pipeline can be
17 rearranged; correct?

18 A. They can. I would say example 5
19 is a different experiment all together
20 than example 1. Example 1 is a target of
21 I believe 300 or so genes. This is an
22 mRNA expression through cDNA sequencing
23 experiment, so the goal here is to
24 estimate expression levels based on the
25 number of reads that map to the reference

1 transcriptome or the reference genome.

2 Q. So now moving to example 6.

3 MR. BEAVERS: I know we've been
4 going for a while. I don't know how
5 much more you have on this.

6 MR. FLETCHER: There's more but
7 if it's a convenient time to break for
8 lunch.

9 MR. BEAVERS: Yeah, let's break
10 for lunch.

11 (Lunch recess taken at
12 11:56 a.m.)

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1 A F T E R N O O N S E S S I O N

2 (Time noted: 12:46 p.m.)

3 D R. C A R L O S B U S T A M A N T E,

4 resumed and testified as follows:

5 CONTINUED EXAMINATION

6 BY MR. FLETCHER:

7 Q. Dr. Bustamante, welcome back from
8 lunch.

9 A. Thank you.

10 Q. If you could please turn to
11 example 6 of the patent at column 21.

12 A. (Witness perusing document.)

13 Q. This example involves the use of
14 the Ovation Library System for low
15 complexity samples; correct?

16 A. That's what I read.

17 Q. And the inventors have generated
18 four libraries; correct?

19 A. That's what I read from table 1.

20 Q. Each of the libraries was
21 generated from a single amplicon; correct?

22 A. That's my understanding.

23 Q. And the person of ordinary skill
24 in the art would understand from that that
25 in theory, every nucleic acid in that

1 library is supposed to have the same
2 target sequence; correct?

3 A. That would be my understanding.

4 Q. Nevertheless, it's possible for
5 nucleic acids in a single amplicon to have
6 different sequences due to, for example,
7 PCR errors; correct?

8 A. That is correct.

9 Q. And in these libraries, the
10 inventors used a 0 to 8 nucleotide random
11 sequence in the adaptor; correct?

12 A. That's what they wrote.

13 Q. These identifiers were used to
14 identify duplicates; correct?

15 A. That is my understanding.

16 Q. The other piece of information
17 that was used to identify duplicates was
18 the length of the identifier; correct?

19 A. As I read here, any paired end
20 reads that shared the same length and
21 sequence of these random bases with any
22 other paired end read was called a
23 duplicate.

24 Q. So in this example 6, the
25 inventors were not practicing claim 1 of

1 the patent as you understand it; correct?

2 MR. BEAVERS: Objection, form.

3 THE WITNESS: Well, so they write
4 that "the 0 to 8 nucleotides of random
5 sequence contained in the adaptors
6 ligated to the amplicon were treated
7 as an identifier sequence and used to
8 mark duplicates."

9 So they are comparing the random
10 sequences which are used as identifier
11 sites and ensuring that those are
12 identical. Any paired end reads that
13 share the same length and sequence is
14 being used, so that is, I believe,
15 marking identical and because they are
16 targeting one single molecule that
17 targets sequences essentially known.

18 BY MR. FLETCHER:

19 Q. So in your opinion,
20 Dr. Bustamante, were the inventors in
21 example 6 practicing the method of claim
22 1?

23 MR. BEAVERS: Objection, form.

24 THE WITNESS: In my opinion, if
25 the sequence of the target molecule

1 was identical and the random sequences
2 were identical, then they would be
3 practicing the claimed invention.

4 BY MR. FLETCHER:

5 Q. If that were the case, but is
6 that what is described in example 6?

7 A. It says, "Any paired end reads
8 that shared the same length and sequence
9 of these random bases with any other
10 paired end read was called a duplicate."

11 And presumably they are
12 identifying paired end reads based on 125
13 nucleotide forward and 25 nucleotide
14 reverse reads to identify those reads so
15 they are comparing those.

16 Q. And where do you see it described
17 that they are comparing the target
18 sequences?

19 A. With the -- well, the paired end
20 reads would be the forward and reverse
21 reads; right? Those are what are paired.

22 Q. So to be clear, Dr. Bustamante,
23 is it your opinion that the example
24 described in example 6 is a method that
25 practices claim 1 or not?

1 A. If my understanding is correct
2 that they are looking at paired end reads
3 that share the same length and sequence,
4 so the paired end reads would be denoted
5 by the forward and reverse and they would
6 read into the index. Those paired end
7 reads have the same start and end by the
8 design and then they would read into the
9 index which is of random size, and as long
10 as those sequences were identical, they
11 would mark them as duplicates. So if they
12 are using those paired end reads and the
13 read information and you're using -- you
14 have a mapping of which pair of reads go
15 together, and those indexes are identical
16 in size and sequence of the indexes with
17 the sequence essentially being known from
18 the forward and reverse, then you would
19 mark those as duplicates.

20 Q. Let's maybe go line by line to
21 make sure we all understand what's
22 happening in example 6 and we're on the
23 same page. At line 10 it reads, "Purified
24 libraries were mixed and sequenced as a
25 multiplex on the Illumina MiSeq to produce

1 125 nucleotide forward, 8 nucleotide index
2 1, 8 nucleotide index 2 and 25 nucleotide
3 reverse reads."

4 Do you agree?

5 A. That's what it says. The line
6 before that says that the Ovation Library
7 System was used to generate four
8 libraries, each from a single amplicon.
9 There was one amplicon that was generated.

10 Q. And that amplicon is intended to
11 have the same nucleic acid in it, but as
12 we all know, due to PCR errors may contain
13 various sequences; correct?

14 A. That is correct. There may well
15 be errors introduced as part of the PCR
16 process.

17 Q. The example then states that
18 "Because all amplicon reads start and end
19 at the same sequence coordinates, the
20 traditional method of marking library PCR
21 duplicates (marking reads that start and
22 end at the same genomic coordinates as
23 duplicates) cannot be used."

24 Do you see that?

25 A. I do.

1 Q. It then stated that "Instead, the
2 0 to 8 nucleotides of random sequence
3 contained in the adaptors ligated to the
4 amplicon or treated as an identifier
5 sequence and used to mark duplicates";
6 correct?

7 A. That is correct.

8 Q. So in this example the inventors
9 used only the identifier sequence to
10 identify duplicates; correct?

11 MR. BEAVERS: Objection, form.

12 THE WITNESS: It doesn't say that
13 that's the only thing they used. It
14 says, "the 0 to 8 nucleotide of random
15 sequence contained in the adaptor
16 ligated to the amplicon were treated
17 as an identifier sequence and used to
18 mark duplicates."

19 Part of what gets used.

20 BY MR. FLETCHER:

21 Q. The next sentence reads, "Any
22 paired end reads that shared these same
23 length and sequence of these random bases
24 with any other paired end read was called
25 a duplicate"; correct?

1 A. That's what it states.

2 Q. The only pieces of information
3 used to call duplicates in example 6 were
4 the length and sequence of the random
5 bases; correct?

6 A. Which has any paired end reads
7 that share the same length and sequence,
8 share the same length and sequence of
9 these bases with any other paired end read
10 was marked as a duplicate. So it would
11 stand to reason using the length and
12 sequence, at least the length and the
13 sequence of the adaptors ligated to the
14 amplicon to mark duplicates and because
15 it's a single amplicon, they essentially
16 know the target sequence and that is part
17 of what can be checked in the forward and
18 reverse reads, say in the event you might
19 not have an insert, for example.

20 Q. So Dr. Bustamante, is it your
21 testimony that if you have some knowledge
22 about the target sequence, as they did
23 here because it's a single amplicon,
24 that's sufficient for satisfying the
25 identifying limitation in claim 1?

1 A. No, I think you need to compare
2 the sequence and the identifier sites.
3 What I think has potentially been left out
4 of this is the fact that they are using
5 the forward and reverse reads in defining
6 the paired end reads. They have to use
7 that to define the paired end reads. That
8 comes from the 125 and the 25 and then the
9 indexes are part of that. So I think
10 inherent in any analysis of paired end
11 reads would be reading the sequence that's
12 the 125 and the 25 to define the paired
13 ends.

14 Q. If you can focus on the sentence
15 in line 20. "Any paired end reads that
16 shared the same length and sequence," do
17 you see that?

18 A. Yes.

19 Q. Same length, is that they are
20 referring to the random bases or to the
21 paired end read?

22 A. It's a simple amplicon, they are
23 all -- so I would say any paired end reads
24 that shared the same length and sequence
25 of these random bases. So length and

1 sequences of the random bases is what is
2 discussed as shared. But in defining
3 paired end reads, you're going to define
4 that based on forward and reverse reads
5 that are marked in the sequencing files.

6 Q. There's no statement here that
7 the target sequences were aligned and
8 compared; correct?

9 A. There presumably would be no need
10 for alignment because it's a single
11 amplicon. They wouldn't do alignment.
12 They would read the first 125 base pairs
13 and the other 25 reverse reads to ensure
14 you had a proper read, paired end reads.
15 So to get the paired end reads you need to
16 analyze the forward and the reverse reads.
17 You can't do that in vacuo.

18 Q. Based on the information
19 presented here in example 6, you believe
20 this method falls outside the scope of
21 claim 1 as you understand it?

22 MR. BEAVERS: Objection, form.

23 THE WITNESS: I'm not sure that's
24 what I stated. I think if in the
25 defining of the paired end reads and

1 the analysis of the forward and
2 reverse reads they are comparing and
3 comparing the sequences, then it would
4 obviously fall in if those were
5 identical. I think it really comes
6 down to how are they defining their
7 paired end reads, and in my
8 experience, you define that based on
9 the sequence of the read 1 file and
10 the read 2 file.

11 BY MR. FLETCHER:

12 Q. So let's clarify. The paired end
13 read, they are comparing the forward read
14 and the reverse read to see if there's
15 complementary; correct?

16 A. No.

17 Q. What are they doing?

18 A. They are reading from the two
19 ends of the molecule, right, so this would
20 be on the essentially 5' end of the target
21 sequence and this would be reading from
22 the complement on the 3' end. They don't
23 necessarily overlap. We don't know the
24 length of the amplicon.

25 Q. Why do you believe to pair a

1 forward and a reverse read that they need
2 to have compared a read 1 with another
3 read 1?

4 A. So in sequencing you often get
5 all kinds of products including adaptor
6 primer that you're sequencing, so you
7 would check. Sometimes you don't always
8 get the second read. So they are clearly
9 saying paired ends read so they have done
10 the work of ensuring that you have for
11 every forward, you have a reverse. And
12 presumably I think a POA would understand
13 that you would also look to make sure they
14 have the insert in there by checking the
15 sequence of the 125 and the sequence of
16 the 25 to ensure it is what you expect it
17 to be.

18 Q. In order to practice the
19 identifying limitation of claim 1 and use
20 the target sequence to identify a
21 duplicate, you need to compare a forward
22 read with another forward read; correct?

23 A. You need to compare the
24 sequencing reads, at least a portion of
25 the sequencing reads to ensure that you

1 have the target sequence and identical
2 target sequence for the portions that
3 you're comparing. So my read of this
4 would be the 125 and the 25 are checked to
5 be what you expect the sequence to be,
6 which would come from both the known
7 sequence you're targeting and the other
8 reads that they get. So if for whatever
9 reason this amplicon had one difference
10 from the reference genome but you read
11 read 1 and read 2 and they all contained
12 that, this would be identical, marked
13 identical as meeting the criteria for
14 paired end read. And then you would only
15 mark as duplicates those that had
16 identical length and sequence of the
17 random sequence.

18 BY MR. FLETCHER:

19 Q. To your understanding, you
20 believe example 6 is an example of a
21 method that practices claim 1 of the
22 patent?

23 A. If my --

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: -- is correct, the

1 way I read this experiment is you have
2 a single amplicon, you've sequenced
3 the first 125 base pairs of that
4 amplicon, let's call it from the
5 Watson strand, and 25 nucleotides from
6 the Crick strand, you ensure that they
7 are a true good paired end read based
8 on the fact they should be tagged and
9 in sequence files and that they are
10 containing the interim you expect them
11 to contain, and you would also
12 presumably compare against the rest of
13 read 1 and read 2, which you expect to
14 all be identical except for the
15 component of the identifier site which
16 would vary across the unique molecules
17 that have been ligated with the
18 adaptor sequence, which you would
19 infer from index reads, from the
20 random sequence.

21 BY MR. FLETCHER:

22 Q. And if your understanding was
23 incorrect and they, in fact, did not
24 compare a forward read to another forward
25 read and instead called duplicates using

1 only the 0 to 8 nucleotide random sequence
2 contained in the adaptor, you would agree
3 that the method of example 6 does not
4 practice the claim; correct?

5 MR. BEAVERS: Objection, form.

6 THE WITNESS: I think in reading
7 the sequence, you're going to go
8 through and check to see that you have
9 the amplicon and look to make sure
10 that you're properly getting the
11 amplicon you expect by comparing both
12 again the reference that you built
13 this on and the reads themselves.
14 It's a rather straightforward
15 experiment in that sense. You have
16 one amplicon, you've read 125
17 nucleotides from one end, 25
18 nucleotides from the other, they say
19 they are limiting themselves to paired
20 end reads, those have to be defined.
21 You sometimes only get read 1 and
22 sometimes you only get read 2 so in
23 the process of defining these paired
24 end reads, you would logically expect
25 that you have what you expect to have.

1 BY MR. FLETCHER:

2 Q. To practice the claimed method
3 you also have to compare one read to
4 another read; correct?

5 A. You would have to compare one
6 read to another read and that's being done
7 both with the target sequence and with the
8 identifier sites here based on the 0 to 8
9 nucleotides of random sequence. Both of
10 those criteria would be met before they
11 would call them duplicates. For example,
12 if there was no insert that didn't contain
13 the amplicon, those would not be part of
14 the paired end reads and they have to
15 compare the amplicon to ensure they have
16 proper insert.

17 Q. So hypothetically if the Board
18 reads this example differently and
19 concludes that the only string of data
20 that was being compared were the 0 to 8
21 nucleotides of random sequence, if you
22 accept that hypothetical, which you may
23 disagree with, you would agree it's not
24 practicing claim 1; correct?

25 MR. BEAVERS: Objection, form.

1 THE WITNESS: Again, my read of
2 this is based as a person of ordinary
3 skill in the arts at the time, that
4 you would look at sequence data in
5 read 1 and read 2 to define where you
6 have paired end reads. So that is
7 being done. And that's being done
8 along read 1, along read 2 and they
9 are properly tagged as coming from the
10 same set. And then you look in the 0
11 to 8 nucleotide random sequence to
12 define whether they are duplicates or
13 not based on those being identical.
14 You would only mark them as duplicates
15 if you had a true pair end read which
16 include 125 nucleotides of sequence,
17 25 nucleotides of sequence and then
18 identical identifier sites based on 0
19 to 8 nucleotides of random sequence.

20 BY MR. FLETCHER:

21 Q. Dr. Bustamante, I worry we might
22 be getting confused because of the paired
23 end. At least as I thought I understood
24 your opinions to be practicing the method,
25 you would need to compare one paired end

1 set to a second paired end set and call
2 one of those a duplicate of the other. Or
3 have I misunderstood and you can call a
4 duplicate as between a forward read and a
5 reverse read?

6 A. No, the forward and reverse reads
7 mark a paired end read, okay, so you
8 define N paired end reads, some number of
9 paired end reads from that set of paired
10 end reads, then compare the 0 to 8
11 nucleotides of random sequence contained
12 in the sequencing that accompanies those
13 paired end reads. And when those 0 to 8
14 nucleotides are identical in length and
15 sequence, you mark those as duplicates.
16 And you would mark the set of paired end
17 reads that contain identical 0 to 8
18 nucleotides of random sequence as
19 duplicates, having defined the paired end
20 reads as containing the 125 nucleotides
21 and 25 nucleotides on the other strand as
22 you expect from the amplicon and as you
23 see from read 1 and read 2 as usually
24 defined from Illumina standard sequencing.

25 Q. If there had been a single based

1 PCR error in one of those paired end
2 reads, it would still have the same
3 length; correct?

4 A. Depending on the PCR error but
5 the length would be -- so if you had a PCR
6 error on the barcode, then the barcodes
7 would not be identical, the identifier
8 sites, and so you would not tag those as
9 identical and duplicates.

10 Q. If there was a single base
11 mismatch PCR error in the forward read,
12 you would still have 125 nucleotides worth
13 of forward reads sequence; correct?

14 A. You would. And depending on
15 where that error was, let's imagine the
16 amplicon's 400 base pairs long, the error
17 could be in the internal segment and you
18 wouldn't know it because you only have the
19 125 nucleotides on one end and 25
20 nucleotides on the other end.

21 Q. If it was in the 125 in the
22 forward primer, the method of claim 6
23 would still mark that sequence as a
24 duplicate of the others if it had the same
25 0 to 8 nucleotides of random sequencing;

1 correct?

2 MR. BEAVERS: Objection, form.

3 THE WITNESS: I think this is one
4 of those examples where at least a
5 portion of the target sequences is
6 being used to define. Given this
7 experiment that would make sense that
8 you know what you're targeting, you
9 have the readout from read 1 and read
10 2 and you're using the experimental
11 design to inform the components that
12 you're relying on, which include the
13 sequence identity of the target and
14 the sequence identity of the
15 identifier sites.

16 BY MR. FLETCHER:

17 Q. So, for example, in this
18 amplicon, the inventors know that the
19 reads all start and end in the same
20 sequence coordinates; correct?

21 A. They presume that, yeah.

22 Q. So having that knowledge of the
23 target sequence, the start and end
24 sequence coordinates is enough to be using
25 a portion of the target sequence to call a

1 duplicate; correct?

2 A. But you also rely on the
3 integrity of the paired end reads which
4 you can only define based on the
5 sequencing data itself.

6 Q. So it's your testimony that you
7 believe the POA would understand from
8 this example 6 that the various paired end
9 reads were compared to each other in their
10 125 plus 25 nucleotides of sequence were
11 compared to each other in their entirety;
12 is that correct?

13 MR. BEAVERS: Objection, form.

14 THE WITNESS: It is my
15 understanding that in order to define
16 properly paired end reads, you would
17 make use of the 125 nucleotide forward
18 and the 25 nucleotide reverse reads
19 because you can get all kinds of
20 chimeric molecules. So if you read
21 that chimeric molecule and it has
22 something unexpected in the 125 or in
23 the 25, you would not label them as
24 properly paired end reads from the
25 target region that you're interested

1 in.

2 BY MR. FLETCHER:

3 Q. Where in this example do you then
4 believe the inventors compared one paired
5 end read to another paired end read?

6 A. Well, it says here, paired end
7 reads that shared the same length and
8 sequence of these random bases with other
9 paired end reads were called duplicates,
10 so presumably they have gone through in
11 defining the paired end reads to include
12 those that have forward and reverse reads
13 that are fully consistent with the target
14 sequence of interest, excluding chimeric
15 molecules and other artifacts that you
16 would not want to use in defining a proper
17 paired end read from the target region of
18 interest.

19 Q. It's your belief that as part of
20 the data process in example 6, for
21 example, if there was a single base PCR
22 error detected in the first 125
23 nucleotides of the forward read, that
24 sequence was eliminated?

25 A. You may have if that sequence is

1 contained in the molecules that have
2 identical 0 to 8 nucleotide random
3 sequence of identical length in sequence,
4 you would retain that information. So if
5 the identifier sites are identical, you
6 would -- and all of them contained and are
7 tagging a PCR duplicate, then presumably
8 that could be identified although because
9 those themselves if they are duplicated
10 you would be able to tag. Depends on when
11 in the cycle the PCR error occurred.

12 Q. So, for example, if the PCR error
13 occurred several cycles in, it is possible
14 for nucleic acids to have the same random
15 identifier including their adaptor but
16 different target sequences; correct?

17 A. But they would be identical to
18 each other.

19 Q. If we posit that the mismatch is
20 created eight cycles into a 20 cycle PCR,
21 wouldn't there be two populations of
22 nucleic acids with this same adaptor but
23 different target sequences?

24 A. Depending on when it occurred in
25 the cycle.

1 Q. And I postulated it happened in
2 cycle 8.

3 A. You would mark all those that
4 have the same sequence and the same
5 adaptor as duplicates so you could detect
6 duplicates of the PCR duplicates and
7 duplicates of the original molecule.

8 Q. In my example you would agree
9 there are two populations of nucleic
10 acids. They have the same identifiers and
11 the same target sequence except that the
12 second population is a single case
13 mismatch.

14 A. You would not call the PCR
15 duplicate with the error a duplicate of
16 the original molecule, but you would be
17 able to detect duplicates of the
18 introduced error if it was early enough in
19 the cycle where you had multiple examples
20 that showed up in the sequencing reads.
21 It's completely consistent with the
22 specification.

23 Q. And so then where in the example
24 6 data analysis pipeline was the 125
25 nucleotide target sequence compared to the

1 other 25 nucleotide target sequences?

2 A. I would say in the definition of
3 proper paired end reads that would then go
4 forward into the analysis starting online
5 20. So to define paired end reads, you
6 would logically check to make sure you
7 have an insert. Because you know what
8 you're targeting you would use the 125
9 from the forward and the 25 from the
10 reverse to ensure you had the right
11 insert.

12 Q. And I think this is common ground
13 but let's assume hypothetically that that
14 is not how someone would understand this
15 example. And if only the identifier
16 sequences were compared, you would agree
17 you are not practicing claim 1 of the
18 method; correct?

19 A. If they made use of no
20 information from the target sequence, as
21 we clearly stated you need to use at least
22 a portion of the target sequence, which in
23 my opinion would be from the forward and
24 reverse reads. So they are only comparing
25 identifier sites, but that's not what it

1 says here. It says they are comparing
2 paired end reads with identical identifier
3 sites using length and sequence to
4 determine identifier sites.

5 Q. But in your opinion if you are
6 comparing the identifier and some
7 information from the target sequence, you
8 are practicing claim 1?

9 A. Depending on the experiment, yes.
10 In this case, I would say logically
11 comparing the forward and the reverse
12 reads to define paired end reads and then
13 among the read 1 and read 2.

14 Q. Let's move on to example 7.
15 Example 7, the inventors were preparing a
16 restriction of the entire human genome;
17 correct?

18 A. They were preparing Reduced
19 Representation Bisulfite Libraries.

20 Q. So this is different than the
21 single amplicon; correct?

22 A. That is correct.

23 Q. This is a high-complexity sample;
24 correct?

25 A. Complexity is a relative measure

1 and in some sense depends on the depth of
2 sequencing. But I would certainly agree
3 that it is a more complex library than
4 single amplicon.

5 Q. They then purified library so
6 that they gather the short fragments;
7 correct?

8 A. Give me a minute.

9 (Witness perusing document.)

10 So they selected for short
11 fragments, as I understand, and then these
12 short fragments are ligated with adaptor
13 sequence comprising an index site and
14 identifier site for 6 or 8 random
15 nucleotides.

16 Q. And here the identifier sites are
17 then used to identify duplicates; correct?

18 A. Correct, in conjunction with the
19 target sequence.

20 Q. Where does it say that the target
21 sequence was used?

22 A. It must be used because the
23 number of 6 and 8 random nucleotides is
24 smaller than the number of fragments that
25 you are expecting to get. So just 6 or 8

1 random nucleotides matching without the
2 target sequence matching wouldn't identify
3 duplicates. If you're doing reduced
4 representation sequencing, you're going to
5 target thousands of sites, so just
6 ligating 6 or 8 random nucleotides, you
7 wouldn't identify duplicates.

8 Q. Example 7 doesn't actually tell
9 us anything about the data processing
10 pipeline other than the fact that the
11 identifier site was used; correct?

12 MR. BEAVERS: Objection, form.

13 THE WITNESS: It says that the
14 identifier site was used in revealing
15 the true number of duplicates in the
16 pool, and without the identifier
17 sites, the number of apparent or
18 perceived duplicates would be greater
19 than the number of true duplicates.
20 But it stands to reason that they must
21 be comparing the target sequence
22 flanking the 6 or 8 random
23 nucleotides, otherwise they couldn't
24 be duplicates.

25 ///

1 BY MR. FLETCHER:

2 Q. In Figure 5 in example 1, the
3 apparent and true duplicates were
4 identified using start and end genomic
5 coordinates; correct?

6 A. That is what they laid out in
7 Figure 5 and in the accompanying text.

8 Q. So when they are discussing true
9 and apparent duplicates in example 7, they
10 may also have been using genomic start and
11 end coordinates; correct?

12 MR. BEAVERS: Objection, form.

13 THE WITNESS: They don't make
14 that clear.

15 BY MR. FLETCHER:

16 Q. If they had been using genomic
17 start and end coordinates for the sequence
18 comparison, they still nevertheless would
19 have been practicing claim 1; correct?

20 MR. BEAVERS: Objection, form.

21 THE WITNESS: As long as they are
22 comparing the target sequence and the
23 identifier site and ensuring that the
24 target sequence across pairs and the
25 identifier site across the same pairs

1 is identical, that's what's necessary
2 to identify duplicates.

3 BY MR. FLETCHER:

4 Q. But if they only compared the
5 genomic start and end coordinates, they
6 wouldn't be practicing claim 1?

7 A. Claim 1 requires you to have
8 compared the target sequence as part of
9 the comparison of the sequencing reads
10 themselves.

11 Q. Perhaps you could turn to Figure
12 2A, please.

13 A. Of the patent?

14 Q. Yes.

15 A. Okay.

16 Q. Figure 2A represents part of the
17 library generation being used in the
18 examples in the patent; correct?

19 A. That is my understanding.

20 Q. And Figure 2A is prior art to the
21 '357 patent; right?

22 A. Could you point to the reference
23 that you are indicating as prior art.

24 Q. I was asking you if you consider
25 this to be background well known at the

1 time of the '357 patent. If you don't
2 know that's fine. I just thought you did.

3 A. Again, this is a figure. I'm not
4 sure if this figure has appeared somewhere
5 else and is a cartoon. It's hard to know
6 what prior art you're referring to.

7 Q. The diagram is called Single
8 Primer Extension Reaction; correct?

9 A. That's what it states.

10 Q. What is single primer extension?

11 A. Single primer extension in this
12 context means a genomic region of interest
13 with a single primer and extending through
14 and then often using a universal primer
15 and subsequent amplification reactions.
16 That's where you would see a primer
17 binding to the genomic DNA.

18 Q. So this third step in Figure 2A
19 is the single primer extension; correct?

20 A. That would be single primer
21 extension as indicated in the figure.

22 Q. And single primer extension in
23 this context is distinct from
24 amplification by PCR primers; correct?

25 A. That is correct.

1 Q. They generate different outcomes?

2 MR. BEAVERS: Objection, form.

3 THE WITNESS: Single primer

4 extension generates an extension

5 product.

6 BY MR. FLETCHER:

7 Q. PCR amplification starts by

8 generating an extension product; right?

9 A. Extension is part of PCR.

10 Q. So the product of a single primer

11 extension and the product of a PCR are the

12 same; right?

13 A. No.

14 Q. How are you distinguishing them

15 if they both start with a primer

16 extension?

17 A. PCR polymer is chain reaction.

18 The operative word here is chain reaction.

19 It's a process by which you are undergoing

20 multiple rounds of extension, annealing,

21 separation, fill-in using the template

22 generated by the extension and the

23 original template to produce nearly

24 exponential amplification of the target.

25 Extension is just extension is one step

1 and may or may not be followed by PCR.

2 Q. But you would agree that the very
3 first event in the PCR is
4 indistinguishable from the single primer
5 extension; right?

6 MR. BEAVERS: Objection, form.

7 THE WITNESS: I would say
8 extension is part of PCR.

9 BY MR. FLETCHER:

10 Q. Extension is used in PCR so they
11 are, in fact, the same?

12 A. They are not the same.

13 Q. They are the same at the
14 beginning of the PCR?

15 A. The steps in PCR include an
16 extension step. Each round has an
17 extension step.

18 Q. And so single primer extension
19 and the beginning of a PCR step produce
20 the same products; right?

21 MR. BEAVERS: Objection, form.

22 THE WITNESS: I would
23 respectfully disagree because the
24 products of PCR are the end result of
25 the multiple cycles, not just of one

1 single extension step.

2 BY MR. FLETCHER:

3 Q. Why don't we go to paragraph 53
4 of your declaration. Paragraph 53 you are
5 providing the Board with background
6 information about processing of sequencing
7 data; is that fair?

8 A. That's correct.

9 Q. These are concepts that were well
10 known to the POA as of the priority date
11 of the '357 patent; correct?

12 A. There were standard workflows for
13 next-generation sequencing analysis at the
14 time.

15 Q. So, for example, the person of
16 ordinary skill in the art would have known
17 that an Illumina sequencer outputs a FASTQ
18 file?

19 A. That's correct.

20 Q. The person of ordinary skill in
21 the art would have known that the FASTQ
22 file purports the sequence of the nucleic
23 acid and the quality of the base calls in
24 that sequence; correct?

25 A. Yes, that's what the Q stands

1 for.

2 Q. A person of ordinary skill in the
3 art would have been familiar with this
4 seven-module data processing pipeline you
5 lay out in paragraph 53; correct?

6 A. Or something akin to it. I wrote
7 it as a seven-step process. You can break
8 it up into more steps or fewer steps but
9 these are I would say are relatively
10 standard set of steps one would take in
11 analyzing NGS sequencing data.

12 Q. As of the priority date of the
13 '357 patent; correct?

14 A. That's correct as I read by the
15 early 2010s.

16 Q. The modules could be swapped out
17 of the particular analysis pipeline as the
18 researcher's needs required; correct?

19 A. That's right, and you wouldn't
20 always take all steps. So, for example,
21 if you didn't have a reference genome, you
22 wouldn't do reference genome alignment.
23 For example, if you only had one sample
24 you wouldn't do sample demultiplexing. So
25 this is an illustrative list of the types

1 of steps one would normally take in doing
2 an analysis.

3 Q. I would now like to talk about
4 the reference genome alignment module that
5 I believe you discuss starting at
6 paragraph 54. The person of ordinary
7 skill in the art as of the priority date
8 would have been familiar with reference
9 genome alignment; correct?

10 A. I would say that's accurate.

11 Q. The reference genome alignment is
12 performed using software typically;
13 correct?

14 A. Yes, that is correct.

15 Q. The reference genome alignment
16 tool compares the sequencing read to the
17 reference genome in order to do the
18 aligning; correct?

19 A. That is correct.

20 Q. So the reference genome alignment
21 tool uses at least a portion of the target
22 sequence to align it to the genome;
23 correct?

24 A. That would be accurate.

25 Q. And the output of that alignment

1 is a tagging of the sequencing read;
2 correct?

3 A. Depends on what you mean by
4 tagging but usually the algorithm would
5 return the locations where the read maps
6 and that may be one or more locations
7 depending on the read and the quality of
8 the reference genome. And of course,
9 reference genomes aren't perfect so
10 sometimes they don't contain all the
11 sequence that is truly in the genome of
12 the organism being sequenced and it also
13 has collapsing of areas. But in general,
14 it returns the locations where the read
15 would map and that may be more than one
16 location.

17 Q. So in paragraph 59 you talk a
18 little bit about this tag so I'd like to
19 make sure we're on the same page about
20 that.

21 A. 59.

22 Q. Paragraph 59 you wrote, "The file
23 output from reference genome alignment
24 programs tag each read with its respective
25 characteristics"; correct?

1 A. That's right. Usually a
2 location. That's usually what is
3 returned.

4 Q. And that location conveys
5 information about the sequence read;
6 correct?

7 A. Yes, and because the genome is
8 annotated, you would know, for example, is
9 this read mapping to a known gene, to an
10 axon, to an axon boundary, et cetera.

11 Q. So the genomic coordinates that
12 you receive from reference genome
13 alignment tell you something about the
14 sequence of the sequencing read?

15 A. The reference genome alignment
16 tells you where it aligns, where the
17 locations are and depending on the
18 annotations available or the organism
19 under study it would tell you something
20 about the source of the material from
21 which it was derived, say the coding
22 sequence for a given gene or if it's a
23 cDNA library, the location from which it
24 could be derived as original mRNA and so
25 on and so forth.

1 Q. So to be clear about the way this
2 data analysis pipeline works, the
3 alignment tools uses the sequencing read
4 as its input; correct?

5 A. Uh-huh.

6 Q. And the output you still have the
7 sequencing read; correct?

8 A. You've not consumed the
9 sequencing read in the alignment process.

10 Q. What you've added is you have
11 added some information like a genomic
12 start coordinate; correct?

13 A. That's correct, genomic
14 coordinates would be accurate,
15 corresponding genomic coordinates to best
16 mapping elevation.

17 Q. So I'm going ahead to paragraph
18 61 of your declaration. You talk about
19 duplicate identification in the prior art;
20 correct?

21 A. In the prior art, yes.

22 Q. You wrote, "For example, by 2012
23 it was standard practice to assess the
24 genomic alignment coordinates (start,
25 strand, and end) of each read against

1 other sequencing reads to determine
2 duplicates."

3 Did I read that correctly?

4 A. That's what I wrote.

5 Q. And that's true, in your opinion
6 that was standard practice as of 2012?

7 A. Yes, and I believe I provided
8 references there.

9 Q. So you would agree that
10 NuGEN/Tecan did not invent the idea of
11 deduplicating sequence reads?

12 MR. BEAVERS: Objection, form.

13 THE WITNESS: Identifying
14 duplicates was a long-standing problem
15 in the field.

16 BY MR. FLETCHER:

17 Q. If we go to paragraph 64, you
18 state that "In order to address issues
19 with duplicate identification, researchers
20 developed 'molecular barcodes,' leveraging
21 the principle of sample barcoding at the
22 molecular level"; correct?

23 A. That's what I wrote.

24 Q. So you would agree that by 2012,
25 the idea of using molecular barcodes also

1 was already known to the POSA; correct?

2 A. As I've written, this was a
3 problem people were working on, the use of
4 molecular barcodes.

5 Q. Paragraph 65, you note that
6 molecular barcode sequences took up
7 valuable space on the sequencing read; is
8 that correct?

9 A. That's correct.

10 Q. So you agree that the person of
11 ordinary skill in the art would have
12 wanted to use the shortest possible
13 molecular barcode that they could in view
14 of the complexity of their sample, given
15 this real estate problem?

16 MR. BEAVERS: Objection, form.

17 THE WITNESS: I would say there's
18 a trade-off between the length of the
19 barcode and its ability to uniquely
20 tag the population of molecules and
21 the length of a sequencing read that
22 is allocated to target sequence
23 identification.

24 BY MR. FLETCHER:

25 Q. And that was a recognized

1 trade-off as of 2012; correct?

2 A. You have a fixed amount of real
3 estate and so you have to decide how much
4 is barcode and how much is target
5 sequence.

6 Q. So that's a yes?

7 A. Yes.

8 Q. I'm going to now hand you what
9 has previously been marked as Exhibit
10 1011. Exhibit 1011 is the Iafrate patent
11 application; correct?

12 A. That is correct.

13 Q. This is a reference that you
14 reviewed as part of your work on this
15 case; correct?

16 A. I did.

17 Q. And 1011 has 52 pages; correct?

18 A. It appears to have 52 pages.

19 Q. And in your discussion in your
20 declaration, your discussion spans from
21 paragraph 104 to 105 for Iafrate; correct?

22 A. That's correct.

23 Q. In paragraph 104 you noted that a
24 portion of the adaptor-ligated nucleic
25 acid fragment is then amplified, quote,

1 with a first adaptor primer and a first
2 target specific primer, close quote, and
3 the resulting amplicons are then amplified
4 by a second adaptor primer in a second
5 target specific primer.

6 Do you see that?

7 A. That's correct.

8 Q. Why did you share that
9 observation with the Board?

10 A. The Iafrate patent?

11 Q. You're turning to page 51, the
12 beginning of the claims?

13 A. I am. That's what I cited in my
14 declaration. So I am pointing to the
15 claims in Iafrate.

16 Q. For example, claim 1 of Iafrate
17 includes steps A through D and eight
18 different wherein clauses; correct?

19 A. That's correct, and whereins
20 within whereins apparently.

21 Q. Several wherein clauses?

22 A. Yes.

23 Q. In paragraph 104 you call out
24 aspects of this claim; correct?

25 A. That is correct.

1 Q. Why?

2 A. To discuss what I feel is the
3 relevant components of Iafrate's which do
4 not discuss identifier sites, but they
5 describe in detail the adaptors, but these
6 are in my reading not assessing identifier
7 sites.

8 Q. You understand this to be
9 describing the anchored multiplex PCR;
10 correct?

11 A. It is describing a method for
12 targeting nucleotide sequences that
13 include multiple steps as part of the
14 targeting, including different rounds of
15 target-specific parameters.

16 Q. Have you used the multiple rounds
17 of target-specific primers in your
18 research?

19 A. I have used various forms of
20 amplicon sequencing, including targeting
21 schemes where you begin with a broad
22 genomic neighborhood and narrow down to a
23 target of interests defining that initial
24 PCR such that you are not amplifying what
25 could be a known duplicate, a nested PCR

1 in some sense.

2 Q. When you were doing nested PCR
3 were you using the Archer AMP products?

4 A. I'm not sure I was. We usually
5 developed our own and it really depends on
6 the collaborator or post doc doing the
7 project, but I don't believe I published
8 in Archer, although maybe some of the work
9 we did on cancer, some of the
10 collaborators may have done part of the
11 experiment with Archer. I can't tell you
12 that.

13 MR. BEAVERS: We've been going
14 for a little over an hour. Do you
15 want to take a break?

16 (Recess taken from 1:51 p.m. to
17 2:07 p.m.)

18 BY MR. FLETCHER:

19 Q. Dr. Bustamante, I'll hand you now
20 an exhibit previously marked 1012 and
21 1013. And we'll be using these in the
22 next module.

23 So first of all, Dr. Bustamante,
24 do you recognize Exhibit 1012?

25 A. I do recognize Exhibit 1012.

1 Q. What is Exhibit 1012?

2 A. It is the Kivioja paper.

3 Q. Do you know Dr. Kivioja?

4 A. I don't.

5 Q. Dr. Bustamante, this Exhibit 1012
6 is a reference that you analyze in your
7 declaration; correct?

8 A. That is correct.

9 Q. If we go to paragraph 106 of your
10 declaration, please.

11 A. I'm at paragraph 106.

12 Q. Here you state that the Kivioja
13 reference teaches the use of the unique
14 molecular identifier; correct?

15 A. I'm not sure I'd put it exactly
16 like that but let me -- can you repeat?

17 Q. I'll ask it a different way. For
18 example, in the Kivioja reference, the
19 authors teach using a molecular barcode
20 sequence; correct?

21 A. They make use of molecular
22 barcode sequence in the Kivioja paper.

23 Q. You, for example, depict that
24 molecular barcode in the excerpt that you
25 reproduce in your paragraph 107; correct?

1 A. That is correct, I reproduce
2 parts of the paper in the declaration.

3 Q. And in paragraph 110 you
4 reproduce a portion of the paper that
5 depicts the process used by the authors in
6 Kivioja; correct?

7 A. That is correct, I believe sub
8 figure A of Figure 1.

9 Q. And this methodology allows the
10 authors in Kivioja to identify duplicate
11 reads; correct?

12 A. Well, my read of Kivioja is that
13 they are interested in counting unique
14 molecules as part of their process.

15 Q. And counting unique molecules
16 involves figuring out which molecules are
17 duplicates and counting them; correct?

18 A. It involves dealing with
19 duplicate reads one way or another.

20 Q. And what you wrote in paragraph
21 110 is that the identifying sequences can
22 then be used in conjunction with other
23 information generated during sequencing
24 data processing to call duplicate reads
25 that are predicted to arise from the same

1 molecule in the original sample; correct?

2 A. That is what I wrote.

3 Q. So you would agree that the

4 Kivioja methodology involves the

5 identification of duplicates; correct?

6 A. I've written that as part of the

7 sequencing data processing they call

8 duplicates.

9 Q. And one piece of information that

10 the Kivioja authors use to identify

11 duplicates is the sequence of the

12 molecular barcode; correct?

13 A. That is correct.

14 Q. And the other piece of

15 information that the authors in Kivioja

16 use to identify duplicates is the genomic

17 alignment; correct?

18 A. That is correct.

19 Q. And as shown in the figure that

20 you excerpt in paragraph 110, the authors

21 in Kivioja, for example, in the bottom

22 left identify multiple copies of

23 sequencing reads shown to have the green

24 sequence with the purple UMI. Do you see

25 that?

1 A. I do.

2 Q. And they group those sequences as
3 a single UMI; correct?

4 A. All the ones that have the purple
5 are one UMI and then all the ones that
6 have the red are a second UMI.

7 Q. In the Kivioja method a variety
8 of sequencing reads are generated and the
9 ones that have the same molecular barcode
10 and the same genomic alignment are treated
11 as duplicates; correct?

12 A. Yes, they also make use of what
13 they call gene and position.

14 Q. What is the significance to you
15 that they are using the gene and position?

16 A. That they are often interested in
17 using the molecular coordinates and
18 annotation as part of the process.

19 Q. If we could go to paragraph 128,
20 please. You note in paragraph 128 that
21 any reads that do not map to the
22 referenced genome are discarded.

23 A. That's what I wrote.

24 Q. What's the significance of that
25 observation?

1 A. That they need mapping
2 information for their method in order to
3 call duplicates.

4 Q. If a method discards sequence
5 information, does it fall outside the
6 scope of claim 1?

7 A. I wouldn't necessarily say that.

8 Q. I agree. I'm just trying to
9 understand why you made this observation.

10 A. As I read Kivioja and pretty
11 clear from the title and the applications,
12 they are mostly interested in molecular
13 counting as the application. And so in
14 order to do that, you're often interested
15 in counting in the context of a genomic
16 position either for gene expression or for
17 identifying extra copies of chromosomes
18 and so on and so forth.

19 Q. In paragraph 129 you state that
20 once Kivioja has determined the gene and
21 position, it teaches that this information
22 is used in conjunction with the label to
23 group sequences into unique groups which
24 Kivioja calls UMIs; correct?

25 A. That's what I wrote.

1 Q. Kivioja considers each sequencing
2 read contained within a UMI group as a
3 duplicate; correct?

4 A. Of other reads in the same
5 grouping, yes, that's what I wrote.

6 Q. So we're clear, Kivioja teaches a
7 method of identifying duplicate sequences
8 using the molecular barcode and the gene
9 and position information; correct?

10 A. That is part of the Kivioja
11 method.

12 Q. And the gene and position
13 information is generated by aligning the
14 target sequence to the reference genome;
15 correct?

16 A. By aligning sequencing reads to
17 the reference genome.

18 Q. The sequencing read contains the
19 target sequence; correct?

20 A. It depends.

21 Q. The sequencing read contains the
22 target sequence; correct?

23 A. The sequencing reads are what
24 they align. Whether there's actual target
25 sequence in there is dependent on the

1 context but the application and the
2 experiment being done.

3 Q. You have Exhibit 1012 in front of
4 you; right?

5 A. 1012, yes.

6 Q. You see the text in bold in the
7 left column, the abstract effectively?

8 A. I do.

9 Q. The final sentence of the
10 abstract teaches "Use of this method can
11 improve accuracy of almost any
12 next-generation sequencing method."

13 Do you see that?

14 A. I do.

15 Q. Do you agree with that?

16 A. Depends what you mean by almost
17 but it could improve the accuracy of
18 next-generation sequencing methods.

19 Q. And the person of ordinary skill
20 reading this would have understood that
21 Kivioja was teaching a method that can
22 improve sequencing accuracy; correct?

23 A. That's what they wrote.

24 Q. If we could go to paragraph 125
25 of your declaration.

1 You offer in paragraph 125 the
2 opinion "that both Kivioja and Iafrate
3 fail to disclose all of the claim
4 limitations required by claims 1 and 2 of
5 the '357 patent"; correct?

6 A. That is what I wrote.

7 Q. Okay. In this section, you do
8 not analyze how the person of ordinary
9 skill in the art would have changed the
10 Iafrate method in view of Kivioja;
11 correct?

12 MR. BEAVERS: Objection, form.

13 THE WITNESS: (Witness perusing
14 document.)

15 So I believe the section where I
16 would discuss this is in paragraph 133
17 around the potential combination of
18 Iafrate with Kivioja, but I do not
19 offer a separate opinion on the
20 combination of Kivioja and Iafrate or
21 the notion of combining.

22 BY MR. FLETCHER:

23 Q. I'd like that to just be clear.
24 You have analyzed the disclosure of
25 Kivioja and the disclosure of Iafrate;

1 correct?

2 A. I have.

3 Q. And you have not disclosed -- you
4 have not discussed or opined on the
5 motivation to combine Kivioja and Iafrate;
6 correct?

7 A. My read is that each of these
8 addresses separate -- separate problems
9 and I don't see a clear motivation to
10 combine.

11 Q. You don't see one discussed in
12 your declaration?

13 A. Sorry. I neither discuss it nor
14 do I see a clear motivation to combine
15 them at the time of the claimed invention.

16 Q. Are you offering a new opinion
17 today? I just want to be clear.

18 A. No, I don't speak to it in my
19 declaration.

20 Q. Okay, that's my thought. So just
21 to make sure the record is clear, in your
22 declaration you do not address the
23 motivation or not to combine Iafrate and
24 Kivioja; correct?

25 A. I do not offer a specific opinion

1 to that in the declaration.

2 Q. Let's turn to Exhibit 1013.

3 A. Okay.

4 Q. Dr. Bustamante, do you recognize
5 Exhibit 1013?

6 A. I do recognize this exhibit.

7 Q. What is Exhibit 1013?

8 A. It is the Bielas PCT publication
9 number WO 213123442.

10 Q. And you have read and analyzed
11 this Exhibit 1013 as part of your work in
12 this case; correct?

13 A. I have.

14 Q. And if you could please turn to
15 Figure 3B of Exhibit 1013, you analyze
16 this figure in your declaration; correct?

17 A. I do.

18 Q. And I'll direct you to paragraph
19 124 and it carries over to page 60 but you
20 reproduced this figure in your
21 declaration; correct?

22 A. That's correct.

23 Q. So is it fair to say that you
24 characterize the methodology disclosed in
25 Figure 3B of Exhibit 1013 in its teaching

1 the creation of consensus sequence?

2 A. (Witness perusing document.)

3 I write that Bielas group
4 sequences into families and that these
5 families are ultimately used to create a
6 consensus sequence.

7 Q. As you were analyzing it in
8 paragraph 124, what is a consensus
9 sequence?

10 A. It's a composite sequence
11 generated from overlapping nucleotide
12 sequences reflecting the most commonly
13 occurring nucleotide at each overlapping
14 position.

15 Q. And the methodology for
16 generating a consensus sequence is to
17 align the multiple sequences and select at
18 each position the most commonly occurring
19 nucleotide at that position; is that
20 correct?

21 A. They use their cypher approach to
22 group the sequences based on the
23 combination of the cypher sequences using
24 only the cypher sequences to group, the
25 combination of the cypher sequences, dual

1 barcoding, as it were.

2 Q. To make the consensus sequence,
3 the Bielas reference teaches comparing the
4 different target sequences in selecting
5 the most common nucleotide in each
6 position; correct?

7 A. After they have been grouped into
8 families and then based on -- one might
9 say it is the consensus sequence arising
10 from the set of families identified
11 through the dual cypher approach.

12 Q. And the grouping or clustering of
13 sequences is achieved using the molecular
14 barcodes; correct?

15 A. Using what they call their
16 cyphers, yes.

17 Q. The cyphers are molecular
18 barcodes?

19 MR. BEAVERS: Objection, form.

20 THE WITNESS: I would say that
21 they call them cyphers and we should
22 use the term they use.

23 BY MR. FLETCHER:

24 Q. But mapping them to the '357
25 patent as you did as part of your

1 analysis, you would agree that cyphers
2 meet the requirement of identifiers;
3 correct?

4 A. They would be identifiers, the
5 combination of them would be identifiers.

6 Q. So the sequences in Bielas are
7 then clustered using their identifiers;
8 correct?

9 A. The families are defined based on
10 the combination of cyphers and then the
11 consensus sequence is derived across the
12 families for those that have common cypher
13 combinations.

14 Q. So as shown, for example, on the
15 right-hand side of Figure 3B, we have a
16 collection of four pairs of sequences;
17 correct?

18 A. On the right-hand side, you have
19 eight sequences.

20 Q. Okay. And you notice how the top
21 of the figure there's two sequences
22 depicted?

23 A. I do.

24 Q. Do you understand that to be two
25 strands of a double-stranded sequence?

1 A. I would presume that that's what
2 that's meant to indicate.

3 Q. Okay. So then proceeding down
4 the figure, we see four pairs of sequences
5 on the right-hand side, correct, in cypher
6 family 2?

7 A. They may be pairs, they may be
8 eight different sequences. You know, we
9 could look at the legend to make sure
10 we're clear that they're -- that we're
11 talking about the same thing.

12 Q. It's on page 4 of Exhibit 1013.
13 Using the applications numbering using the
14 footer on the bottom, it's .0006.

15 A. Okay.

16 Q. So having refreshed on page 0006
17 of Exhibit 1013, you would agree that
18 these are double-stranded representations
19 in Figure 3B; correct?

20 A. I want to be accurate in
21 answering your question and ensure that
22 that is correct.

23 (Witness perusing document.)

24 So I do believe this is one of
25 the examples where they grouped into the

1 what we would call Watson and the Crick
2 strand to creating a consensus for each.
3 It's not clear from the figure so it could
4 be.

5 Q. If you go to page 006 where the
6 figure is described, the Exhibit 1013
7 indicates that the target nucleic acid
8 molecules were ligated into a cypher
9 vector library containing previously
10 catalogued dual double-stranded cyphers;
11 correct?

12 A. It says Figures 4A and B show the
13 cypher system can distinguish true
14 mutations from artifact mutations.

15 Q. Dr. Bustamante, I'm speaking
16 about 3B.

17 A. I'm sorry, I thought we were
18 talking about 4.

19 Q. We're looking at 3.

20 A. That explains the confusion. I
21 was on -- I apologize.

22 Q. No worries.

23 A. This explains the source of the
24 confusion. Thank you.

25 Q. For example, in the description

1 of Figure 3B on page 0006, the authors of
2 Exhibit 1013 indicate that all sequencing
3 reads having identical cypher pairs along
4 with their reverse complements were
5 grouped into pairs.

6 A. I completely -- I agree. I
7 apologize I was reading the wrong figure
8 legend.

9 Q. No worries. So returning to
10 Figure 3B.

11 A. That's why I wanted to
12 double-check and make sure we were
13 building consensus sequences for Watson
14 and Crick.

15 Q. We see in cypher family 2 a
16 representation of four different sequence
17 pairs; correct?

18 A. Correct.

19 Q. And these four sequence pairs
20 were grouped into a cypher family because
21 they have the same identifier sequences;
22 correct?

23 A. Or the reverse complement as the
24 case may be, yes.

25 Q. The next step in the sequence

1 process in pipeline disclosed in Figure 3B
2 is that the internal fragments are
3 compared to each other; correct?

4 A. Well, as we say here they create
5 a consensus sequence.

6 Q. And a consensus sequence is
7 created by comparing position by position
8 the various sequences in the family;
9 correct?

10 A. In this case, presumably you
11 create a consensus sequence for one strand
12 and a consensus sequence for the other
13 strand based on whether they have the
14 Watson or the Crick orientation of the
15 cyphers.

16 Q. And the end result of this
17 sequence processing shown in Figure 3B is
18 the sequence pair depicted at the bottom
19 of Figure 3B; correct?

20 A. Correct. And that's because the
21 method is meant to identify spurious
22 mutations, not duplicate sequencing reads.
23 The whole goal here is to identify sources
24 of errors and eliminate them.

25 Q. And you would agree that the

1 sequence pair number 1, sequence pair
2 number 3 and sequence pair number 4 in
3 cypher family 2 are identical; correct?

4 MR. BEAVERS: Objection, form.

5 THE WITNESS: I have no context
6 with which to say they are not
7 identical. It would seem based on the
8 figure as presented that there are no
9 differences across those sequences or
10 the representation of those sequences.

11 BY MR. FLETCHER:

12 Q. And the end result of this
13 processing is a single sequence; correct?

14 A. Well, this is where we're saying
15 there's a consensus sequence for one
16 strand and a consensus sequence for the
17 other and presumably those are compared
18 to.

19 Q. The final product of Figure 3B's
20 method is a single pair of sequences;
21 correct?

22 A. A single pair of sequences. A
23 single pair of consensus sequences, pair
24 of consensus sequences.

25 Q. Now, if we could please pull back

1 out from your pile what I previously
2 marked as Exhibit 1117, Claim Construction
3 - Scenario 4.

4 A. Yes.

5 Q. The --

6 A. Which version?

7 Q. Regular 4, vanilla 4, Exhibit
8 1117.

9 A. I have Exhibit 1117.

10 Q. Comparing Exhibit 1117 and the
11 right-hand side of Bielas Exhibit 1013
12 Figure 3B, we see the same type of input
13 information and the same type of output
14 information; correct?

15 A. I would not agree with that.

16 Q. Why do you disagree with that?

17 A. Because this method in Bielas is
18 using a dual cypher approach and it is
19 creating two consensus sequences, one for
20 the top strand and one for the bottom
21 strand.

22 Q. And the -- what's the
23 significance of using dual cyphers?

24 A. They claim in their PCT that
25 combinatorial possibilities of doing the

1 dual cyphers allows them to improve error
2 detection by relying simply on those dual
3 cyphers to identify parent of origin
4 molecules. So the original molecules are
5 defined as those that have identical dual
6 cyphers or the complement, if you're doing
7 the double-stranded approach.

8 Q. So in Exhibit 1013 the identical
9 identifier sequences are used to build a
10 family of sequences; correct?

11 A. Dual cyphers are used to create
12 families.

13 Q. And then the sequences of the
14 families are compared; correct?

15 A. The sequences of the families are
16 used to detect spurious mutations.

17 Q. And the end result is a single
18 sequence; correct?

19 A. The end result is a consensus
20 sequence or a pair of consensus sequences
21 in reverse complement.

22 Q. And the algorithm for generating
23 a consensus sequence and the algorithm for
24 detecting a duplicate are fundamentally
25 the same; right? Position by position,

1 two strings of information are compared
2 and checked to see whether they are the
3 same or different; correct?

4 MR. BEAVERS: Objection, form.

5 THE WITNESS: And I've stated
6 that the comparison to define
7 identical is an if and only if process
8 by which you are requiring the unique
9 identifiers to be identical and the
10 target sequences to be identical. And
11 once you reach a point they are not
12 identical, you proceed to not call
13 them duplicates.

14 BY MR. FLETCHER:

15 Q. And in Figure 3B of Exhibit 1013,
16 sequence pairs 1, 3 and 4 have identical
17 identifiers and identical target
18 sequences; correct?

19 MR. BEAVERS: Objection, form.

20 THE WITNESS: Sorry, can you
21 repeat your question?

22 BY MR. FLETCHER:

23 Q. In Figure 3B of Exhibit 1013,
24 sequence pairs number 1, 3 and 4 have
25 identical identifier sequences and

1 identical target sequences; correct?

2 MR. BEAVERS: Objection, form.

3 THE WITNESS: We're referring to
4 cypher family 2?

5 BY MR. FLETCHER:

6 Q. Correct.

7 A. Again, I don't have additional
8 context or reason to believe that they are
9 different. That's what is illustrated in
10 this figure to my understanding.

11 Q. And the algorithm being used in
12 the methods on 1013 generates a single
13 sequence from that information; correct?

14 A. In this schematic it generates an
15 identical representation of the sequence.
16 That's what's shown here.

17 Q. And it did so, to be fair, even
18 though it included in the data that it
19 processed sequence error number 2;
20 correct?

21 A. It did because the purpose of
22 this method is to identify spurious errors
23 in mutations. So the goal here was to
24 eliminate mutations such as the one in
25 sequence pair 2 as I believe you refer to

1 it.

2 Q. The algorithm for doing the
3 sequence comparison of generating a
4 consensus sequence involves the same
5 comparison of strings of information as
6 the algorithm for detecting a duplicate;
7 correct?

8 MR. BEAVERS: Objection, form.

9 THE WITNESS: To detect a
10 duplicate in the claimed invention you
11 compare the identifier, the unique
12 identifier and the target sequence for
13 pairs of sequences at a time and if
14 they are identical, you would label
15 them as duplicates.

16 BY MR. FLETCHER:

17 Q. And the algorithm for generating
18 a consensus sequence likewise compares the
19 sequence of an identifier and the sequence
20 of a target sequence to a second
21 identifier and target sequence?

22 A. It can. There are lots of ways
23 to create consensus sequence and lots of
24 decisions one might make about including
25 or not including quality, whether or not

1 the unique identifier is part of the
2 consensus sequence comparison or not.

3 Q. And in Exhibit 1013, Figure 3B,
4 the data processing pipeline starts by
5 comparing the identifier sequences to
6 build the family; correct?

7 A. It starts by defining the
8 families as those that have pairs of the
9 same cypher errors.

10 Q. And it then compares the
11 sequences of the inserts; correct?

12 A. It then processes the sequences
13 of the inserts to generate a consensus.

14 Q. And that process involves
15 comparing the sequences; correct?

16 A. It involves generating a
17 consensus sequence by the method.

18 Q. Of comparing sequences?

19 A. Of comparing positions in the
20 sequences.

21 Q. Dr. Bustamante, where are you
22 currently employed?

23 A. I'm a professor at the University
24 of Oklahoma in the College of Medicine.

25 Q. Are you still affiliated with

1 Stanford University?

2 A. I was an adjunct professor until
3 the end of August when I took this
4 position in Oklahoma.

5 Q. Are you still affiliated with the
6 University of Miami?

7 A. I have an entrepreneurial
8 residence position at the University of
9 Miami.

10 Q. Dr. Bustamante, do you know
11 Douglas Amorese?

12 A. I'm not sure I've ever met him
13 personally. I can't tell you that.

14 Q. Do you recall ever having worked
15 or collaborated with Dr. Amorese?

16 A. I don't believe so. To the best
17 of my knowledge, there may be a 500 author
18 paper somewhere but we certainly did not
19 work closely together at any time that I
20 can recollect.

21 Q. Before you reviewed any patents
22 in connection with your work with Tecan,
23 had you read anything by Dr. Amorese?

24 A. I read a lot of papers, to be
25 frank, so I can't tell you with certainty.

1 Probably but I can't tell you with
2 certainty.

3 Q. Let me ask it differently.
4 Sitting here today, you can't recall
5 having read a paper by Dr. Amorese?

6 A. That is correct, I don't recall
7 having reviewed a paper or similar but
8 I've been in the business for 25 years so
9 something might have crossed my desk at
10 some point.

11 Q. I have similar questions of a few
12 more people.

13 A. Okay.

14 Q. But maybe we'll not talk over
15 each other.

16 Do you know Jonathan Scolnick?

17 A. I similarly -- I'm familiar with
18 Scolnick. I'm not sure if we've met and I
19 don't believe we've coauthored any papers
20 together or I've been involved in
21 reviewing prior work, to the best of my
22 recollection.

23 Q. What about Ben Schroeder?

24 A. Same answer.

25 Q. What about Chris Armour?

1 A. Same answer.

2 Q. What about Nurith Kurn?

3 A. Same answer.

4 Q. Prior to your consulting on this
5 matter, had you ever heard of Tecan?

6 A. I had heard of Tecan.

7 Q. In what context have you heard of
8 Tecan?

9 A. They are a well-known
10 manufacturer of intermits and kits and
11 reagents used in molecular biology.

12 Q. Had you done any sponsored
13 research with Tecan?

14 A. Not to the best of my knowledge
15 or recollection.

16 Q. Prior to your consulting on this
17 case, had you ever heard of NuGEN?

18 A. I'm not sure I had, to be very
19 frank. I don't think I had.

20 Q. So you can't recall having ever
21 done a research collaboration with NuGEN?

22 A. I don't recall ever having done a
23 research collaboration with NuGEN. Again,
24 I'm coauthor on 250 papers. It doesn't
25 mean that at some point somebody didn't do

1 a project, but to the best of my
2 recollection I certainly don't recall
3 NuGEN ever sponsoring any project or
4 similar in my laboratory, any consulting
5 work prior to this.

6 Q. Dr. Bustamante, you've also had a
7 portion of your career focused on
8 investing and forming companies; correct?

9 A. That's correct.

10 Q. Have you ever been involved in
11 making any investments in NuGEN or Tecan?

12 A. Not to the best of my knowledge.
13 I was an advisor and was never on the
14 investment committee or involved with
15 overseeing investments except as a
16 scientific and technical advisor.

17 MR. FLETCHER: Why don't we take
18 a break.

19 (Recess taken from 2:53 p.m. to
20 3:08 p.m.)

21 EXAMINATION

22 BY MR. HOAGLAND-HANSON:

23 Q. So Dr. Bustamante, good
24 afternoon. You have in front of you
25 what's been previously marked as Tecan

1 Exhibit 2038 and this is your amended
2 declaration. Do you see that?

3 A. I do.

4 Q. I want to start with paragraph 5
5 of Exhibit 2038.

6 In paragraph 5, Dr. Bustamante,
7 you summarize the materials that you
8 considered in reaching your opinions. Do
9 you see that?

10 A. I do.

11 Q. And you considered the '012 and
12 the '108 patents, the file histories of
13 the patents and related patents, prior art
14 publications, two petitions for IPR, the
15 related institution decisions and two
16 declarations, the declaration of Eli Meyer
17 and the declaration of Peter Sims. Do you
18 see that?

19 A. I do.

20 Q. And it's fair to say that if you
21 didn't respond to an opinion that Dr. Sims
22 offered in your declaration, that you
23 didn't disagree with Dr. Sims?

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: I wouldn't make

1 that statement.

2 BY MR. HOAGLAND-HANSON:

3 Q. Okay. Are there other parts of
4 Dr. Sims' declaration that you disagree
5 with that aren't set forth in your
6 declaration?

7 A. I have discussed the elements of
8 Dr. Sims' declarations that were relevant
9 to the aspects of the IPR that I was asked
10 to respond to.

11 Q. Okay, and it's fair to say that
12 if you didn't respond to some aspect of
13 Dr. Sims' declarations, you either agreed
14 or didn't think it was relevant?

15 MR. BEAVERS: Objection, form.

16 THE WITNESS: Again, I wouldn't
17 make that statement. What I have put
18 in the declaration is in response to
19 what I was asked to respond to.

20 BY MR. HOAGLAND-HANSON:

21 Q. Did you only respond to the
22 portions of Dr. Sims' opinions that your
23 lawyers asked you to respond to?

24 A. I addressed the questions that I
25 was asked to address and relied on the

1 parts of the declaration that I felt were
2 relevant to those sections.

3 Q. Okay. What questions were you
4 asked to address?

5 MR. BEAVERS: So just as a
6 caution, you can answer generally but
7 not to reveal any privileged
8 information or communications.

9 THE WITNESS: As you're aware I'm
10 not a lawyer and I don't set the legal
11 strategy in any way, shape or form for
12 the client, and so I was asked to
13 opine on certain matters and those are
14 the matters that I opined on.

15 BY MR. HOAGLAND-HANSON:

16 Q. You list some references in
17 paragraph 5. In the next sentence you
18 say, "I have considered the prior art
19 relied on by IDT."

20 Do you see that?

21 A. I do.

22 Q. Okay. And then you list five
23 prior art references?

24 A. That's correct.

25 Q. It's fair to say you didn't

1 consider any of the other prior art
2 references at issue in this proceeding if
3 they are not listed in this paragraph; is
4 that right?

5 MR. BEAVERS: Objection, form.

6 THE WITNESS: Again, I wouldn't
7 agree with that statement. I'm simply
8 saying here that I have considered at
9 least the references cited here in
10 paragraph 5.

11 BY MR. HOAGLAND-HANSON:

12 Q. What other references did you
13 consider in reaching your opinions?

14 A. The references that are in my
15 declaration.

16 Q. Just so it's clear, the only
17 references you considered in reaching your
18 opinions are the ones that are cited in
19 your declaration?

20 A. At least the ones that are cited
21 in my declaration. I have also considered
22 additional materials that were provided as
23 part of the proceedings.

24 Q. Okay, where do you state what
25 those additional references were?

1 A. Throughout the declaration when I
2 rely on an exhibit, I cite to the exhibit.

3 Q. Okay. So I think I asked you to
4 be clear, the only references you
5 considered in reaching your opinions are
6 the ones that are cited in your
7 declaration; is that right?

8 A. At least the ones that are cited
9 in my declaration are the materials I
10 relied upon, as well as my knowledge as a
11 person of ordinary skill in the arts at
12 the time the patents were filed and have
13 priority date of January 26, 2012.

14 Q. Are you familiar with a section
15 of an expert report called a materials
16 considered section?

17 A. I have seen materials considered
18 sections in the past.

19 Q. You don't have such a materials
20 considered section in your declaration in
21 this case; right?

22 MR. BEAVERS: Objection, form.

23 THE WITNESS: I was not asked to
24 prepare a materials considered section
25 as part of this work and so one is not

1 provided here. To my understanding, I
2 don't believe there's a materials
3 considered section.

4 BY MR. HOAGLAND-HANSON:

5 Q. And so I just want to understand.
6 Are your opinions based on the references
7 that are cited plus your professional
8 experience or are your opinions based on
9 the references that are cited plus your
10 professional experience plus some other
11 references that you considered but didn't
12 cite in your declaration?

13 MR. BEAVERS: Objection, form.

14 THE WITNESS: I believe I have
15 cited where relevant material that is
16 either part of the material provided
17 or additional materials that I
18 identified as part of this work.

19 BY MR. HOAGLAND-HANSON:

20 Q. I want to look at the next
21 paragraph of Exhibit 2038, paragraph 6.

22 And here you say, "I have
23 considered IDT's allegations that the
24 challenged claims are anticipated or
25 rendered obvious by Meyer."

1 Do you see that?

2 A. I do.

3 Q. And then you say, "As explained
4 further below, I disagree with IDT's
5 assessment."

6 A. That is correct.

7 Q. It's fair to say that paragraph 6
8 is a summary of your opinions as to why
9 Meyer doesn't anticipate the claims?

10 A. I summarized some of my opinions
11 in paragraph 6.

12 Q. You say here, "The amplification
13 process of Meyer that IDT relies on for
14 its arguments does not result in the
15 enrichment of a target sequence"; right?

16 A. That is correct.

17 Q. And then you say, "Because Meyer
18 fails to disclose an amplification scheme
19 that enriches for a sequence of interest
20 over other sequences in a sequencing
21 library, I have concluded that Meyer fails
22 to anticipate the challenged claims";
23 correct?

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: Further on in the

1 paragraph that is what is written.

2 BY MR. HOAGLAND-HANSON:

3 Q. Okay. The absence of enrichment
4 of a target sequence and the absence of an
5 amplification scheme that enriches are the
6 only reasons why you say Meyer doesn't
7 anticipate the claims; right?

8 A. That's what I say in this
9 paragraph. That doesn't summarize all of
10 the opinions in my declaration.

11 Q. What are the other reasons why
12 Meyer doesn't anticipate the claims?

13 A. We can go through the declaration
14 in detail and address those questions.

15 Q. I guess I'm just trying to figure
16 out, you have another summary in paragraph
17 364 at the end of your declaration,
18 Exhibit 2038. Are you with me,
19 Dr. Bustamante?

20 A. I'm in paragraph 364.

21 Q. And you say, "In summation, Meyer
22 fails to disclose the claimed methods of
23 selectively amplifying for the
24 oligonucleotide extension product sequence
25 or its complement to enrich for a sequence

1 of interest, thus failing to anticipate or
2 render obvious the independent claims of
3 the '012 and '108 patents."

4 A. That is what I wrote.

5 Q. That's the same summary of your
6 opinion that's provided in paragraph 6?

7 MR. BEAVERS: Objection, form.

8 THE WITNESS: Both of those are
9 summaries of the opinions provided
10 throughout the report or detailed in
11 the report.

12 BY MR. HOAGLAND-HANSON:

13 Q. Okay. And I'm just trying to
14 figure out, Dr. Bustamante, if it's the
15 absence of selectively amplifying and
16 enriching for a sequence of interest that
17 are the reasons why Meyer doesn't
18 anticipate the claims or if there are some
19 other reasons.

20 A. And we can go through the report
21 in detail and understand why I have
22 written and documented my opinion that
23 Meyer fails to disclose the claimed
24 invention starting on page 126.

25 Q. Okay. So I see you're looking at

1 your table of contents and maybe this is
2 another place where the same issue comes
3 up. You have the headings for the
4 sections of your report and we're looking
5 at page 3 of Exhibit 2038; right?

6 A. I am looking at the table of
7 contents.

8 Q. And section Roman VIII is Meyer
9 fails to disclose the claimed inventions;
10 right?

11 A. Correct.

12 Q. And then you have subsections A,
13 B, C, D, E, F, G, H of that Roman VIII?

14 A. That's correct.

15 Q. And the headings are the
16 limitations that you say are absent from
17 Meyer?

18 MR. BEAVERS: Objection,
19 misstates the record.

20 BY MR. HOAGLAND-HANSON:

21 Q. Is that fair?

22 A. Section 8 lays out the scientific
23 arguments of why Meyer fails to disclose
24 the claimed invention and I'm happy to go
25 through it in detail with you.

1 Q. I'm just trying to figure out,
2 Dr. Bustamante, if you have any opinions
3 that there are other limitations in the
4 claims that are not taught by Meyer.

5 A. The opinions I'm prepared to
6 discuss today are the opinions that are in
7 my declaration.

8 Q. Dr. Bustamante, I'm going to hand
9 you what's been previously marked as IDT
10 Exhibit 1001. This, Dr. Bustamante, is
11 the '012 patent?

12 A. I believe this is the '012
13 patent.

14 Q. And I see you've flipped to
15 paragraph 117 of Exhibit 2038; is that
16 right?

17 A. I have gone to the section of my
18 report that speaks to the patented
19 invention.

20 Q. And in paragraph 117, you've
21 reproduced claim 1 of the '012 patent;
22 right?

23 A. Claim 1 of the '012 patent starts
24 in paragraph 117.

25 Q. You don't have any opinion,

1 Dr. Bustamante, that the step 1(a) here,
2 obtaining a nucleic acid fragment ligated
3 to a partial duplex adaptor, do you see
4 that, and then it goes on?

5 A. I do.

6 Q. That step of the claim doesn't
7 happen in the Meyer process?

8 MR. BEAVERS: Objection, form,
9 misstates the record.

10 THE WITNESS: We can go through
11 the Meyer reference and discuss this
12 in detail, if you wish.

13 BY MR. HOAGLAND-HANSON:

14 Q. I'm just asking for your opinion
15 as to whether this step happens in the
16 Meyer process or not.

17 Maybe I can help you out,
18 Dr. Bustamante. Could you go with me to
19 paragraph 283 of your declaration. And
20 you have a subsection here, subsection E,
21 do you see that?

22 A. I do.

23 Q. And you say, "As explained
24 above," in paragraph 283, "each limitation
25 of the independent claims of the '012

1 patent and '108 patent is conditional on
2 the presence of a 'sequence of interest,'
3 a 'nucleic acid fragment,' and/or an
4 amplification step that results in
5 enrichment of the sequence of interest.
6 Because Meyer fails to disclose those
7 claim elements, Meyer also fails to
8 disclose any of the limitations of the
9 independent claims."

10 A. That is what I wrote.

11 Q. I'm just trying to figure out,
12 Dr. Bustamante, it seems like what you're
13 saying is you disagree that any of the
14 chemistry is found here because these
15 three things aren't found; is that fair?

16 A. Again, we can discuss the Meyer
17 paper in the context of each of the
18 claimed elements and we can see where it
19 fails to disclose elements of the claim as
20 evident by what is actually in Meyer's,
21 including the sequence of interest, the
22 question around nucleic acid fragment and
23 the details of the amplification step
24 among others.

25 Q. Well, I just want to find out if

1 there are any others.

2 A. We can go through it in detail.

3 Q. Why don't you take your time and
4 find where there are any others.

5 A. (Witness perusing document.)

6 So the key arguments that I've
7 put forth is that at least fails to
8 disclose the amplifying of one or more of
9 the oligonucleotide extension products and
10 to enrich the nucleic acid sequence of
11 interest I believe is 1D of the '012,
12 largely because I don't believe that the
13 protocol selectively enriches or unique
14 elements of the CNA is actually a method
15 designed to do whole transcriptome
16 analysis so there's no enrichment as I lay
17 out. Furthermore, I believe that a
18 sequence of interest is an element of the
19 native molecule and the broken T adaptor
20 does not meet that criteria, and if any
21 enrichment occurs, it's occurring really
22 prior to fragmentation so the
23 fragmentation step is another element that
24 is not disclosed. So in my view, it does
25 not meet the key goal of the patent which

1 is to enrich for sequence of interest.

2 Q. So Dr. Bustamante, let me ask you
3 a hypothetical question.

4 If 3' end fragments in -- and you
5 know what I'm referring to when we talk
6 about the 3' end fragments; correct?

7 A. I understand what 3' end
8 fragments are and there are multiple
9 discussed in Meyer. We can speak to ways
10 that 3' ends are dealt with in Meyer.

11 Q. You would agree with me if the 3'
12 end fragments are of sequence of interest
13 as defined in the patents and the Meyer
14 method enriches for the 3' end fragments,
15 then Meyer anticipates the claim?

16 MR. BEAVERS: Objection, form,
17 incomplete hypothetical.

18 THE WITNESS: In my -- my read of
19 Meyer is consistent with Dr. Meyer's
20 declaration, that their goal was to do
21 full transcriptome analysis in a
22 non-model organism. So nowhere in
23 Meyer is there in my reading or
24 Dr. Meyer's position a goal to enrich
25 for 3' ends. It is not an accurate

1 representation of Meyer.

2 BY MR. HOAGLAND-HANSON:

3 Q. We're going to get to what you
4 think Meyer actually taught. I'm asking
5 you hypothetically, if the 3' end
6 sequences are a sequence of interest as
7 defined in the patent and the Meyer method
8 enriches for the 3' end fragments, you
9 agree that the claim is anticipated;
10 right?

11 MR. BEAVERS: Objection, form,
12 incomplete hypothetical.

13 THE WITNESS: I disagree. For
14 the claim to be anticipated, all
15 elements of the claim must occur in
16 the order specified in the claimed
17 invention and that would not
18 necessarily be the case even if 3' end
19 were target of interest. It would
20 have to disclose all elements of the
21 claimed invention.

22 BY MR. HOAGLAND-HANSON:

23 Q. What other elements of the claim
24 are missing if the 3' end fragments --
25 Dr. Bustamante, what other elements of the

1 claim are missing if the 3' end fragments
2 are a sequence of interest and the method
3 enriches for the 3' end fragment?

4 MR. BEAVERS: Objection, form,
5 incomplete hypothetical.

6 THE WITNESS: Sir, I don't have
7 any details on your hypothetical
8 scenario so we would need to flesh
9 that out and I can't provide an
10 opinion without understanding the
11 hypothetical that you have in mind.

12 BY MR. HOAGLAND-HANSON:

13 Q. What other information do you
14 need?

15 A. I need to understand the protocol
16 that you're discussing and how the
17 experiment was going to be carried out and
18 how the targeting was to happen and how
19 all of the elements would come together.
20 I don't read that at all in Meyer because
21 Meyer fails to disclose key elements of
22 the claimed invention.

23 Q. So I'm just positing a method
24 that's exactly the same as Meyer, right,
25 that the 3' end fragments are the sequence

1 of interest and Meyer does, in fact,
2 enrich for the 3' end fragments you agree
3 the claim anticipated?

4 MR. BEAVERS: Objection, form,
5 incomplete hypothetical.

6 THE WITNESS: Even a hypothetical
7 method can't be the same as Meyer
8 targeting the 3' ends, because that's
9 not what Meyer does.

10 BY MR. HOAGLAND-HANSON:

11 Q. You don't have an opinion sitting
12 here today that any other limitations are
13 missing?

14 MR. BEAVERS: Objection, form.

15 THE WITNESS: As I've stated, the
16 limitations I'm prepared to discuss
17 are the ones that are in my report and
18 we can go through those in detail.

19 BY MR. HOAGLAND-HANSON:

20 Q. So you agree that the proper
21 definition of a sequence of interest for
22 purposes of these patents is a
23 polynucleotide sequence of interest for
24 which enrichment is desired; is that
25 right?

1 MR. BEAVERS: Objection, form.

2 THE WITNESS: We can turn to --

3 there are multiple definitions of
4 target of interest and I believe this
5 is discussed and I respectfully
6 disagree with some of those
7 definitions so we can --

8 BY MR. HOAGLAND-HANSON:

9 Q. Can we go to paragraph 145 of
10 your declaration, Dr. Bustamante?

11 A. Sure.

12 Q. Are you with me, Dr. Bustamante?

13 A. I do.

14 Q. You agree that the proper
15 construction of sequence of interest is a
16 polynucleotide sequence of interest for
17 which enrichment is desired; right?

18 A. That is how the panel construed
19 it. I don't disagree with that
20 construction but I also believe that the
21 broken T primer site as indicated doesn't
22 fit deeply into that sequence of interest
23 definition.

24 Q. What's the difference between
25 agreeing with something and not

1 disagreeing with it?

2 A. The caveat that I've provided is
3 meant to put this into the context of the
4 Meyer paper.

5 Q. I'm just trying to find out are
6 you agreeing with the Board's construction
7 for purposes of the patents or disagreeing
8 with it?

9 A. As I said here, I don't disagree
10 with that definition. I don't believe
11 that the Meyer paper -- in the Meyer paper
12 the -- and we can talk about this when we
13 get to the Meyer paper, I don't believe
14 the broken T element of the adaptor meets
15 that criteria partly because it's not a
16 native sequence to the fragment.

17 Q. Okay. Not answering the question
18 that I'm asking. The question is: Do you
19 agree that the proper construction of
20 sequence of interest for purposes of the
21 '012 and '108 patent is a polynucleotide
22 sequence of interest for which enrichment
23 is desired, yes or no?

24 A. I understand that's how the panel
25 has construed it. I don't disagree with

1 that construction. I don't necessarily
2 agree that it applies to the Meyer
3 sequence that is under discussion.

4 Q. I'm not asking about the Meyer
5 reference.

6 A. Okay.

7 Q. I'm trying to figure out what you
8 think the '012 patent means. You agree
9 that sequence of interest is a term from
10 the '012 patent; right?

11 A. I agree with that.

12 Q. And for purposes of the '012
13 claim, you agree that it means a
14 polynucleotide sequence of interest for
15 which enrichment is desired?

16 MR. BEAVERS: Objection, asked
17 and answered.

18 THE WITNESS: I don't disagree
19 with that construction but it has to
20 be evaluated in the light of the
21 claimed invention and how enrichment
22 is carried out in the claimed
23 invention.

24 BY MR. HOAGLAND-HANSON:

25 Q. How is this definition changed by

1 the way in which enrichment is carried out
2 in the claimed invention?

3 A. Because enrichment is carried out
4 and defined in a very specific way in the
5 claimed invention, in the claims of the
6 invention.

7 Q. And how in your view does that
8 way that enrichment is carried out and
9 defined in the claimed invention limit the
10 scope of the sequence of interest?

11 MR. BEAVERS: Objection, form.

12 THE WITNESS: I think 145 speaks
13 for itself. I understand how the
14 panel construed the term sequence of
15 interest. I don't disagree with that
16 construction. I also have opinions
17 about whether or not parts of Meyer
18 meet that definition.

19 BY MR. HOAGLAND-HANSON:

20 Q. Dr. Bustamante, I'm just trying
21 to get to the bottom of what seems to me
22 to be a very lawyerly turn of phrase. And
23 as a lawyer I'm just trying to find out if
24 what you're saying is I agree with the
25 construction or if you're trying to say

1 something different.

2 A. I do not disagree with the
3 construction.

4 Q. What's the difference between not
5 disagreeing and agreeing?

6 A. As I've said, I don't disagree
7 with the construction.

8 Q. So you agree with the
9 construction?

10 A. I understand that that's the
11 panel's definition as they've construed
12 sequence of interest and I'm prepared to
13 evaluate the patent based on that
14 definition.

15 Q. Do you agree with it?

16 A. I agree that that's how the panel
17 has construed it and for the purposes of
18 evaluating the patent I will use that
19 definition.

20 Q. So you don't agree with it but
21 you understand you have to apply it; is
22 that right?

23 A. For the purpose of evaluating the
24 patent I understand this is how the panel
25 has construed the term and so in my

1 evaluating the patent, I am prepared to
2 use that definition in evaluating the
3 claims.

4 Q. Because that's different than
5 what you say in paragraph 145. You don't
6 say my lawyers have instructed me to apply
7 the Board's construction for purposes of
8 my analysis. Is that what paragraph 145
9 is meant to mean?

10 MR. BEAVERS: Objection, form,
11 argumentative.

12 THE WITNESS: I didn't mention
13 anything about lawyers in my response.
14 I'm saying that as laid out here for
15 these proceedings, the panel has
16 construed a definition of sequence of
17 interest to mean a polynucleotide
18 sequence of interest for which
19 enrichment is desired. I do not
20 disagree with that construction for
21 the purpose of evaluating this patent.

22 BY MR. HOAGLAND-HANSON:

23 Q. And you agree that the sequence
24 of interest is part of the nucleic acid
25 fragment; right?

1 A. We can turn to the claims and see
2 where sequence of interest appears in the
3 claimed invention.

4 Q. And Dr. Bustamante, you agree
5 that the sequence of interest is part of
6 the nucleic acid fragment; right?

7 A. So we see that sequence of
8 interest occurs in the preamble of claim 1
9 and it also appears in 1a, where the
10 nucleic acid fragment comprises a nucleic
11 acid sequence of interest. It's one
12 location where the sequence of interest
13 appears.

14 Q. Sequence of interest is part of
15 the nucleic acid fragment; right?

16 A. The nucleic acid fragment
17 comprises nucleic acid sequence of
18 interest.

19 Q. What's the difference between the
20 sequence of interest is part of the
21 nucleic acid fragment and the nucleic acid
22 fragment comprises the sequence of
23 interest?

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: My understanding is

1 that comprises means includes so it
2 includes a nucleic acid sequence of
3 interest.

4 BY MR. HOAGLAND-HANSON:

5 Q. The sequence of interest is a
6 known sequence in this method; right?

7 MR. BEAVERS: Objection, form.

8 THE WITNESS: That is -- in the
9 embodiments laid out here, the
10 sequence of interest is targeted by a
11 primer that is specific to that
12 sequence of interest and so we would
13 often know that sequence ahead of
14 time.

15 BY MR. HOAGLAND-HANSON:

16 Q. Okay. But the rest of the
17 fragment may or may not be known?

18 A. The rest of the fragment may or
19 may not be known. You at least would know
20 the sequence of interest in order to be
21 able to design the primer that faithfully
22 anneals to that fragment of interest, the
23 sequence of interest in the fragment.

24 Q. And you agree that one sequence
25 of interest might enrich for multiple

1 different fragment sequences?

2 A. Yes.

3 MR. BEAVERS: Objection, form.

4 THE WITNESS: Yes, you --

5 depending on how the primer is

6 designed, you may get multiple targets

7 that are primed off of a single

8 primer. Whether or not that meets the

9 definition of enrichment depends on

10 whether those elements are downstream

11 available for sequencing. So

12 enrichment here means in the context

13 of sequencing.

14 BY MR. HOAGLAND-HANSON:

15 Q. And you agree the sequence of

16 interest could be an artificial sequence;

17 right?

18 MR. BEAVERS: Objection, form.

19 THE WITNESS: I'm not sure I can

20 agree with that.

21 BY MR. HOAGLAND-HANSON:

22 Q. Why not?

23 A. Because in the plain and ordinary

24 meaning of fragment comprising a nucleic

25 acid, a nucleic acid fragment, you would

1 often be considering a sequence that is
2 native to that nucleic acid fragment.

3 Q. Can the sequence of interest be a
4 transposon?

5 A. That's an interesting question.
6 I would say it would be a complicated
7 experiment to sort out and often transpose
8 onto a native to or at least have been
9 incorporated into the genome if it's a
10 retrotransposon.

11 Q. Does the nucleic acid fragment
12 have to come from an organism?

13 A. Well, in many of the dependent
14 claims we're talking about nucleic acids
15 that are derived from organisms, such as
16 genomic DNA, cDNA, RNA. I guess you don't
17 need to limit the claimed invention to
18 nucleic acids that are derived from
19 organisms. They could be fully synthetic.
20 There's no specific limitation to that in
21 1 but at the time of this invention most
22 of the applications are targeted to DNA
23 that is derived from organisms.

24 Q. The sequence of interest can be,
25 doesn't have to be but it can be an

1 artificial sequence though; right?

2 MR. BEAVERS: Objection, form,
3 asked and answered.

4 THE WITNESS: In my reading, most
5 of the applications that would make
6 use of this would be targeting
7 multiplexing of multiple genes or
8 similar where you have sequence of
9 interests that are tied to a
10 biological question. So depending on
11 the biological question, one could
12 imagine scenarios. Now in my opinion,
13 there's nothing here that would
14 necessarily limit you to not consider
15 synthetic sequence, although most of
16 the applications would be to DNA
17 derived from organisms.

18 BY MR. HOAGLAND-HANSON:

19 Q. Can the sequence of interest be
20 an adaptor sequence that you add in a
21 preceding step?

22 A. I don't see anything in the
23 claims here that would prohibit you from
24 using an adaptor that you've added in a
25 prior step as long as there has been

1 fragmentation as part of 1a.

2 Q. And you agree the sequence could
3 be a random sequence; right?

4 A. It would be hard to target a
5 random sequence.

6 Q. Does the claim forbid it from
7 being a random sequence?

8 A. I don't read it as forbidding.

9 Q. Okay. You referred to
10 multiplexing earlier. You agree you can
11 use this method with multiple sequences of
12 interest at once; right?

13 A. I do agree with that.

14 Q. And you agree that the claimed
15 method can be used to enrich the entire
16 exome?

17 MR. BEAVERS: Objection, form.

18 THE WITNESS: Would you repeat
19 the question?

20 BY MR. HOAGLAND-HANSON:

21 Q. Sure. You agree that the claimed
22 method can be used to enrich the entire
23 exome?

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: The method does not

1 preclude targeting of the whole exome
2 but you would have to carry that out
3 as specified in claim 1 of the method
4 here or one of the dependent claims.

5 BY MR. HOAGLAND-HANSON:

6 Q. The claimed method can be used to
7 enrich for all known coding regions?

8 MR. BEAVERS: Objection, form.

9 THE WITNESS: Again, if the steps
10 are carried out as outlined in claim
11 1, I could imagine designing single
12 primer extension approaches that could
13 create a lot of coding sequence in an
14 organism or a pool.

15 BY MR. HOAGLAND-HANSON:

16 Q. The claimed methods aren't
17 limited to targeting just one gene or set
18 of genes; right?

19 A. There is nothing in my read of
20 this that says it's limited to a gene or a
21 set of genes.

22 Q. I want to look at the examples in
23 Exhibit 1001 and these start at column 31.

24 A. I'm at column 31.

25 Q. Example 1 is characterization of

1 the human oral microbiome by selective
2 enrichment of bacterial 16S Ribosomal DNA
3 sequences.

4 A. I see that.

5 Q. Are you familiar with what a 16S
6 Ribosomal sequence is?

7 A. I am.

8 Q. What is a 16S Ribosomal sequence?

9 A. The 16S Ribosomal sequence is a
10 component of the cellular machinery that
11 is common to many forms of life and is
12 often used as a way of identifying a
13 community of organisms through sequencing
14 of a combined sample of nucleic acids
15 derived from that community.

16 Q. And in the 16S Ribosomal RNA
17 region, there are highly conserved blocks
18 and there are highly variable blocks;
19 right?

20 A. That is a feature of the 16S.
21 You have elements that are highly
22 conserved and elements that vary that
23 allow you to identify the taxome that make
24 up the community being sequenced.

25 Q. So you talked about the 16S

1 Ribosomal sequence is used as a way of
2 sort of separating out bacteria basically.

3 A. Bacteria or other organisms. You
4 can create a map of a community based on
5 16S.

6 Q. And when you do that, what you're
7 interested in, what you're comparing are
8 the variable regions between the different
9 organisms; right?

10 A. You identify the community based
11 on known sequences from reference material
12 and then matching the sequencing data to
13 that reference material based on the
14 elements that differ among the so-called
15 reference taxonomic units.

16 Q. And in this particular method if
17 you look up at the top of column 32,
18 there's a method that's describing the
19 amplification step. Are you with me?

20 A. I am with you.

21 Q. At the end of that paragraph, it
22 says, "Oligonucleotides representing each
23 of the 18 highly conserved sequence blocks
24 identified across the 16S genomic loci are
25 synthesized and mixed in equal molecular

1 proportions."

2 A. I see that.

3 Q. When you're then analyzing the
4 sequencing data, the part that you're
5 interested in is not the highly conserved
6 sequence blocks; right?

7 A. Depends on what you mean the part
8 that you're interested in. Certainly in
9 the primer design phase you're interested
10 in the highly conserved regions.

11 Q. But when you're comparing between
12 the 16S sequences, what you want is the
13 part right next to that, the highly
14 variable region; right?

15 MR. BEAVERS: Objection, form.

16 THE WITNESS: As they lay out
17 here they use 40 bacterial species to
18 define the conserved regions and
19 presumably they're interested in
20 differentiating among the bacterial
21 species based on the variable portions
22 of the 16S.

23 BY MR. HOAGLAND-HANSON:

24 Q. And they know before they even
25 run the sequencing around what sequences

1 they are going to get back for the highly
2 conserved blocks; right?

3 MR. BEAVERS: Objection, form.

4 THE WITNESS: Well, they design
5 primers to the 16S conserved regions
6 in order to carry out the enrichment
7 and sequencing here.

8 BY MR. HOAGLAND-HANSON:

9 Q. And that's actually true any time
10 you do this method; right? The sequence
11 of interest is complementary to your
12 primers, right, so you always know what
13 the sequence of interest is going to be;
14 right?

15 MR. BEAVERS: Objection, form.

16 THE WITNESS: I'm not sure I
17 would agree with that because the
18 sequence of interest could be
19 downstream of where your primers are
20 designed and you may be interested in
21 variation in those regions as part of
22 the biological question being asked.

23 BY MR. HOAGLAND-HANSON:

24 Q. Can we go back to the claim?

25 A. Yes.

1 Q. Briefly? Okay. I guess because
2 I'm trying to understand your last answer.
3 Because in b, step b, right, this is the
4 part where the primer comes in; is that
5 right?

6 A. That is correct, so the primer is
7 binding to the sequence of interest in the
8 nucleic acid fragment.

9 Q. Or at least 10 bases of the
10 sequence --

11 A. At least 10 bases to be
12 complementary.

13 Q. And so at least those 10 bases of
14 the sequence of interest you always know;
15 right?

16 MR. BEAVERS: Objection, form.

17 THE WITNESS: You design the
18 primer so they bind to at least those
19 10 bases. If you have a mismatch it's
20 going to impact the efficiency claim.

21 BY MR. HOAGLAND-HANSON:

22 Q. You could have a bug where you
23 get some nonspecific primer binding but
24 that's not what we're going for here in
25 step B; right?

1 A. Usually you'd like very specific
2 binding.

3 Q. So you would agree with me that
4 when you're doing this method, you always
5 know what at least 10 of the bases of the
6 sequence of interest are going to be;
7 right?

8 MR. BEAVERS: Objection, form.

9 THE WITNESS: You design primers
10 to 10 bases that you want to target in
11 the nucleic acid fragment that's been
12 ligated to the partially fixed
13 adaptors that are then used for
14 sequencing. So the target here is
15 specific two fragments for which
16 you've ligated the adaptors that
17 contain the sequencing elements.

18 BY MR. HOAGLAND-HANSON:

19 Q. Okay, and what you're interested
20 in at the end, right, for example, in your
21 16S Ribosomal example, is the part of the
22 fragment that's next to the sequence of
23 interest?

24 MR. BEAVERS: Objection, form.

25 THE WITNESS: I think this is

1 a how long is a piece of string
2 question. So the nucleic acid
3 sequence of interest includes at least
4 the primer binding site and would
5 presumably be defined in this context
6 as the 16S region for which you've
7 designed primers as they lay out in
8 the patent, the notion of tiling
9 across a region of interest.

10 BY MR. HOAGLAND-HANSON:

11 Q. It's possible for the sequence of
12 interest here 20 be just the 10 bases that
13 are part of the primer, right,
14 complementary to the primer?

15 A. The sequence of interest would
16 contain at least those 10 bases.

17 Q. It could contain just those 10
18 bases; right?

19 A. Why would you do an extension
20 step if you only cared about those 10
21 bases?

22 Q. What if you care about what's
23 right next to it?

24 A. Then it includes more than just
25 those 10 bases, doesn't it?

1 Q. Isn't that the nucleic acid
2 fragment part?

3 A. The entire thing is the nucleic
4 acid fragment which may include much more
5 than the sequence of interest. If you're
6 targeting an exon, it could include the
7 exon, intron, regions you're not
8 interested in, for example.

9 Q. How do you figure out where the
10 sequence of interest begins and ends?

11 A. Depends on the experiment being
12 conducted and the biological question
13 being asked. So in this context, you're
14 designing the primers to at least 10 bases
15 in a region of interest and could be the
16 entire fragment that you're interested in,
17 it could be a portion of the fragment that
18 is a coding region, it could be a variable
19 region in the 16S as you've indicated.

20 Q. You're never interested in the
21 first 10 bases though because those you
22 already knew; right?

23 MR. BEAVERS: Objection, form.

24 THE WITNESS: Again, I think this
25 is a question that requires more

1 context in terms of the experiment
2 that we want to evaluate. But in
3 general, the 10 bases are used as the
4 guidepost on which to hang the primer
5 that will then sequence that will
6 extend into the region that you're
7 sequencing.

8 BY MR. HOAGLAND-HANSON:

9 Q. Let's just say I'm trying to use
10 this to look for novel ALK gene fusions.
11 Are you familiar with the concept of an
12 ALK gene fusion?

13 A. I am familiar with an ALK fusion.

14 Q. When I'm looking for an ALK
15 fusion, I'm not really interested in the
16 ALK gene, I'm interested in the gene next
17 to it; right?

18 MR. BEAVERS: Objection, form.

19 THE WITNESS: Depends on the
20 particular question you're asking. If
21 you're trying to decide whether to use
22 a targeted therapy or you're trying to
23 characterize in-depth a tumor, we can
24 imagine different ways it could be
25 used. Maybe you're interested in

1 mapping break points, maybe you're
2 interested in -- it's hard to know.
3 It's an incomplete question without
4 knowing the experiment you have in
5 mind.

6 BY MR. HOAGLAND-HANSON:

7 Q. If I am using a primer to the
8 first 10 bases of the -- complementary to
9 the first 10 base of the ALK gene, right,
10 just hypothetically?

11 A. Okay. I don't know if the
12 fusions -- on which part of the ALK. I'm
13 not sure that's the right way to detect an
14 ALK fusion so I don't want to speak out of
15 turn.

16 Q. Regardless, I'm going to know
17 those 10 bases of the ALK gene; right?
18 You're saying okay, if I'm looking for a
19 fusion maybe I'm looking for the back of
20 the sequence, right, towards the fusion
21 sighting.

22 A. It depends on the question you're
23 interested in. This approach here applies
24 in a myriad of settings.

25 Q. I guess I'm trying to figure out,

1 it's possible for the -- to your point,
2 right, the thing that I'm interested in
3 depends on the experimental design; right?

4 A. I would agree with that
5 statement.

6 Q. Okay. And so is it your
7 understanding that sequence of interest
8 for purposes of this claim depends on the
9 subjective intent of the person performing
10 the experiment?

11 MR. BEAVERS: Objection, scope,
12 form.

13 THE WITNESS: I wouldn't classify
14 it as subjective intent. You design
15 an experiment based on scientific
16 principles.

17 BY MR. HOAGLAND-HANSON:

18 Q. I guess is it in the eye of the
19 beholder. If I think this is a sequence
20 for which enrichment is desired but you,
21 Dr. Bustamante, disagree, am I practicing
22 the claim or not?

23 MR. BEAVERS: Same objection,
24 scope and form.

25 THE WITNESS: I would say

1 enrichment needs to occur through the
2 process laid out in claim 1 for it to
3 anticipate this claim. All of the
4 elements need to be there. So when we
5 think about a sequence of interest and
6 whether you're targeting a sequence of
7 interest, that's one component, but it
8 has to be carried through through all
9 of the elements here for it to enrich
10 in the way that's laid out in the
11 patent.

12 BY MR. HOAGLAND-HANSON:

13 Q. Okay. I guess in example 1 with
14 the 16S Ribosomal RNA, what's the sequence
15 of interest?

16 A. The 16S Ribosomal RNA as they lay
17 out.

18 Q. Including the highly conserved
19 sequence and the variable sequence?

20 A. They are components of how you
21 arrive at the composition of microbiome.
22 I think if you were to find it was missing
23 a highly conserved region, that could be
24 interesting, but I think to your point
25 most of what they'd like to do here is a

1 taxonomic characterization, and so those
2 primers that could be staggered throughout
3 the whole of the 16S enable the generation
4 of the sequence that is then combined to
5 arrive at a biological understanding of
6 the oral microbiome.

7 Q. So are the primer sequences to
8 the --

9 A. There's 18 highly --

10 Q. Sorry. I apologize.

11 A. No, no, please.

12 Q. It seems like you had more to
13 your answer.

14 A. I insist.

15 Q. So are the primer sequences to
16 the 18 highly conserved sequences part of
17 the sequence of interest or not?

18 A. We would need to understand
19 exactly how they are staggered because
20 they could overlap fragments and sequence
21 of interest. But I would say the goal
22 here is to characterize the 16S and in
23 each of these fragments, you're targeting
24 at least the segment, at least one segment
25 of a highly conserved sequence through the

1 amplification process.

2 Q. Is it just the use of that in the
3 amplification process that makes it a
4 sequence of interest?

5 MR. BEAVERS: Objection, form.

6 THE WITNESS: No, it involves the
7 primer extension component and then
8 enrichment through amplification for
9 sequencing.

10 BY MR. HOAGLAND-HANSON:

11 Q. Is a sequence of interest for
12 purposes of these claims something you
13 determine before you start or is it
14 something that you can only figure out if
15 you get the product at the end?

16 MR. BEAVERS: Objection, scope,
17 form.

18 THE WITNESS: In any carefully
19 designed experiment, especially a
20 sequencing experiment, you're
21 designing the experiment in such a
22 way, especially using this technology,
23 so you can target specific regions for
24 sequencing that you are interested in,
25 and your analysis of that sequence is

1 done in the context of a biological
2 question. So to your point around
3 what is predefined, well, it stands to
4 reason that the primers are
5 predefined, that's a fixed part of the
6 experiment as a controlled element of
7 the experiment. The sequencing reads
8 are then derived through the claimed
9 invention here and downstream
10 combination could include multiple
11 targets in addressing that biological
12 question. But the primers are a fixed
13 component of these, any one of these
14 designed experiments, and often
15 biological questions what's driving
16 which set of primers you would design
17 to use in this single primer extension
18 enrichment method.

19 BY MR. HOAGLAND-HANSON:

20 Q. I want to talk about example 3 in
21 column 34.

22 A. Okay.

23 MR. BEAVERS: Before we start we
24 have been going a little over an hour.
25 Could we take 5 minutes?

1 THE WITNESS: Sure, you could use
2 a break.

3 (Recess taken from 4:11 p.m. to
4 4:24 p.m.)

5 BY MR. HOAGLAND-HANSON:

6 Q. Okay, Dr. Bustamante, we were
7 just going to start talking about example
8 3 in column 34. Are you with me?

9 A. I'm in column 34.

10 Q. And the example 3 here is
11 characterization of transcriptional
12 activity of individual cells within a
13 population. Do you see that?

14 A. That's what it states.

15 Q. And if you go down to the second
16 full paragraph, there's a heading
17 Generation of Fragments With Ligated
18 Forward Adapters. Do you see that?

19 A. I do.

20 Q. And it says, "Components from the
21 NuGEN ovation ultra low library system are
22 used to generate next-generation
23 sequencing libraries from each of the
24 purified sample."

25 Do you see that?

1 A. I do.

2 Q. And that reference to the Ovation
3 system means that this is an embodiment of
4 the claims; right?

5 A. Presumably that's what we would
6 expect.

7 Q. If you go down another couple
8 paragraphs, there's a heading Primer
9 Extension.

10 A. I see that.

11 Q. And it says, "Libraries are
12 generated by introducing the reverse
13 adaptor attached to the 5' end of a random
14 hexamer."

15 A. I see that.

16 Q. And this is referring to the --
17 this random -- let me try a complete
18 sentence in English.

19 This reverse adaptor attached to
20 the 5' end of a random hexamer is the
21 oligonucleotide that's referred to in
22 steps B and C, right, of the claim?

23 MR. BEAVERS: Objection, form.

24 THE WITNESS: Give me a minute
25 here.

1 (Witness perusing document.)

2 If we go back to claim 1, and I'm
3 just going to ask you to repeat your
4 question, if you don't mind, to make
5 sure I give you an accurate answer.

6 BY MR. HOAGLAND-HANSON:

7 Q. Now that you've looked at that
8 I'll ask a summary question where I'm
9 getting. This example is saying the
10 sequence of interest can be a random
11 hexamer and whatever sequence is adjacent
12 to it; right?

13 MR. BEAVERS: Objection, form.

14 THE WITNESS: In this example,
15 they are using a random hexamer and as
16 it states here, the primer extension
17 step is performed via oligonucleotides
18 contained in a 3' random region, then
19 a 5' region that contains a portion of
20 the Illumina reverse adaptor sequence.
21 So it would appear that this would
22 extend when the random region is
23 matched.

24 BY MR. HOAGLAND-HANSON:

25 Q. So the sequence of interest here

1 is a random region and whatever is
2 adjacent to the random region?

3 MR. BEAVERS: Objection, form.

4 THE WITNESS: Whatever is ligated
5 onto the adaptor which would be the
6 fragment of interest. The fragment
7 that's ligated to the partial duplex
8 adaptor as described in 1B.

9 BY MR. HOAGLAND-HANSON:

10 Q. Just so I understand, the
11 sequence of interest here is the random
12 hexamer and then whatever else is in the
13 fragment?

14 MR. BEAVERS: Objection, form.

15 THE WITNESS: There's a 3' random
16 region and the 5' region that is the
17 Illumina reverse adaptor on the other
18 side.

19 BY MR. HOAGLAND-HANSON:

20 Q. That's within the
21 oligonucleotide; right?

22 A. That's the targeting primer.

23 Q. The sequence of interest then is
24 a complement of the random hexamer and
25 whatever else is in that fragment; right?

1 MR. BEAVERS: Objection, form.

2 THE WITNESS: You would have the
3 5' end of the random hexamer that's
4 ligated to the cDNA fragment in this
5 case. We can look at Figure 2 to be
6 clear.

7 BY MR. HOAGLAND-HANSON:

8 Q. I'm just trying to understand
9 what -- the sequence of interest is in the
10 fragment; right? It's in the duplex;
11 right?

12 A. It would be --

13 MR. BEAVERS: Objection, form.

14 THE WITNESS: The sequence of
15 interest would be the cDNA fragment
16 that is ligated to the adaptors.

17 BY MR. HOAGLAND-HANSON:

18 Q. Okay. And --

19 A. Which would include this random
20 hexamer.

21 Q. So let me just try one more time
22 because I think the record was a bit
23 messy.

24 The sequence of interest here in
25 example 3 is any fragment that has as the

1 complement the random hexamer; is that
2 right?

3 MR. BEAVERS: Objection, form.

4 THE WITNESS: I'm sorry, I was --
5 could you just repeat the question so
6 that I can give you an accurate
7 answer.

8 MR. HOAGLAND-HANSON: Sure, and
9 TJ, maybe if you could pause for a
10 second so I can get the question out.

11 MR. BEAVERS: Pause your answer.

12 BY MR. HOAGLAND-HANSON:

13 Q. The sequence of interest here in
14 example 3 is any fragment that has the
15 complement of the random hexamer; is that
16 right?

17 MR. BEAVERS: Objection, form.

18 THE WITNESS: I believe that is
19 accurate.

20 BY MR. HOAGLAND-HANSON:

21 Q. I want to talk about paragraph
22 277 of your declaration. It's your
23 opinion that the broken chain T primer is
24 not a sequence of interest; is that right?

25 A. My opinion is that a POSA would

1 not have applied the broken chain T primer
2 because they do not wish to enrich for the
3 broken T primer.

4 Q. You agree that Meyer -- let me
5 take a step back.

6 It's your opinion that the POSA
7 is not interested -- well, scratch that.
8 I'll try it one more time.

9 It's your opinion that Meyer is
10 not interested in sequencing the polyA
11 tail; right?

12 A. Meyer is interested in full cDNA
13 sequencing of the transcriptome of a
14 non-model organism. That is the purpose
15 of Meyer.

16 Q. So Meyer is interested in
17 sequencing the polyA tail?

18 A. The polyA tail is not an
19 interesting component of the full cDNA
20 transcriptome.

21 Q. You would agree with me that in
22 the Meyer method, they try to minimize the
23 amount of polyA tail they sequence; right?

24 A. Well, I think they are trying to
25 solve a problem with 454 homopolymer runs

1 first and foremost.

2 Q. You agree with me that Meyer is
3 interested in sequencing the 3' ends of
4 the mRNA; right?

5 A. They are interested in sequencing
6 the entire mRNA.

7 Q. Including the 3' ends?

8 A. The 3' end is part of the mRNA.
9 But they're interested in sequencing the
10 entire mRNA as the purpose of Meyer in a
11 non-model organism.

12 Q. In the Meyer method the broken
13 chain T primer sequence is always adjacent
14 to the 3' end; right?

15 A. Always is a strong word but in
16 general, that's where you expect it to be.

17 Q. In theory?

18 A. In theory. In their design that
19 is where they expect it to be.

20 Q. And you'd agree that that means
21 they are interested in the rest of the
22 sequence of the fragment containing the
23 broken chain T, just not the broken chain
24 T itself?

25 A. They are trying to deal with the

1 polyA tail problem in 454 and would like
2 to sequence the entire transcriptome.

3 Q. In terms of fragments, they are
4 trying to sequence the fragments that have
5 the broken chain T and the 3' end that's
6 adjacent to that; right?

7 A. Well, they don't fragment until
8 they have ligated their adaptors so and
9 they end up sequencing all the fragments
10 so not necessarily enriching for one class
11 of fragments over others.

12 Q. We'll get there. In the sauce,
13 once of the fragments they are trying to
14 sequence is the sequence that has the 3'
15 and the broken chain T; right?

16 MR. BEAVERS: Objection, form.

17 THE WITNESS: I would say one
18 class of fragments that they are going
19 to end up sequencing are those that
20 are on the 3' end ligated to the
21 broken T prime.

22 BY MR. HOAGLAND-HANSON:

23 Q. Okay.

24 A. Complement region.

25 Q. I want to go to paragraph 281 of

1 Exhibit 2038. And here you say,
2 "Expanding the sequence of interest to
3 apply to the sequence regions of the 3'
4 end fragment, however, destroys the
5 distinction that the patents make between
6 a 'nucleic acid fragment' and a 'sequence
7 of interest.' Instead, it would treat the
8 'sequence of interest' as the 'nucleic
9 acid fragment.'"

10 Do you see that?

11 A. I do.

12 Q. Can you explain why that's the
13 case?

14 A. Because the sequence of interest
15 is defined in the context of the design of
16 the protocol that's carried out as an
17 embodiment of the patent claims.

18 Q. If the sequence of interest can
19 be a random hexamer, why couldn't it be
20 the broken chain T primer?

21 A. The context of that experiment is
22 different than what's being discussed
23 here.

24 Q. How so?

25 A. In this case, the broken T primer

1 is a hack as it were for dealing with the
2 454 homopolymer problem.

3 Q. That's one thing it does but
4 doesn't it also demarcate the end of the
5 3' of the mRNA?

6 A. It is ligated onto the end of the
7 3' end.

8 Q. If you were interpreting the data
9 in Meyer without the broken chain T, how
10 would you know if you were at the 3' end
11 of a transcript?

12 A. You don't need the broken T
13 primer in transcriptome sequencing to know
14 you're at the end of the 3' end.

15 Q. You don't need it but it helps;
16 right?

17 A. In their design they use it to
18 get full cDNA sequences but there are
19 other ways of getting full cDNA sequences.

20 Q. But it also tells them when they
21 are interpreting data, this fragment had
22 the broken chain T, this must be the 3'
23 end; right?

24 A. Again, unless you had spurious
25 binding somewhere else, which we know will

1 happen at some small rate, but in general,
2 you would interpret and if you could
3 assemble the transcript and know that
4 you've got a stop cut out and so on, you
5 put that all together to decide if you've
6 reached the end of the fragment, that
7 you've reached the 3' end of the mRNA
8 transcript.

9 Q. The existence of the polyA tail
10 or the broken chain T, right, that's been
11 substituted for it is giving you some
12 biologically relevant information; right?

13 A. It is useful in context of the
14 full cDNA sequencing but again, you're not
15 looking to enrich for the 3' end of the
16 molecule. If anything Meyer puts in its
17 declaration. They are trying to deal with
18 overrepresentation of the 3' end, if
19 anything. They want to get an unbiased
20 view of the full cDNA repertoire of this
21 coral species that they are interested in.
22 That's the purpose of Meyer.

23 Q. I want to go back to example 3
24 briefly.

25 A. Okay.

1 Q. Example 3 starts from an RNA
2 sample. Do you see that?

3 A. It starts with total RNA present
4 in the lysate.

5 Q. Right. And then that RNA is
6 transcribed as cDNA; right?

7 A. That's what I read.

8 Q. Okay. And then it's fragmented,
9 the cDNA. Do you see that?

10 A. The next line 16 at column 34
11 says generation of fragments.

12 Q. Okay.

13 Claim 21, Dr. Bustamante, you
14 offer some opinions about this claim. Do
15 you remember that?

16 A. I do.

17 Q. And it's your opinion that if the
18 RNA is transcribed to cDNA prior to
19 fragmentation, that doesn't practice claim
20 21; right?

21 A. We can look at claim 21.

22 Let me just reference the section
23 in claim 21. I want to be accurate in my
24 answer.

25 Q. Sure, maybe I can direct you to

1 paragraph 287 of Exhibit 2038.

2 A. I was in the neighborhood.

3 Q. Just trying to make sure you make
4 your conference call.

5 A. I appreciate it.

6 Would you repeat your question
7 for me?

8 Q. Sure. Example 3 is not an
9 embodiment of claim 21?

10 MR. BEAVERS: Objection, form.

11 THE WITNESS: Well...

12 (Witness perusing document.)

13 It would appear here that the
14 cDNA is made from the total RNA
15 present in lysate. The
16 double-stranded cDNA is unpurified and
17 then the NuGEN system is used for
18 library preparation. So unless the
19 RNA is fragmented prior to reverse
20 transcription, then it couldn't meet
21 claim 21.

22 BY MR. HOAGLAND-HANSON:

23 Q. Okay, so example 3 does not meet
24 claim 21?

25 A. I'm not sure if they fragmented

1 the RNA before. They could have. I'm not
2 sure. If they did, then it would meet 21.
3 If they didn't, then it wouldn't. All
4 this says is generation of fragments with
5 ligated forward adaptors. So if they had
6 fragmented the RNA before they did the
7 cDNA preparation, then it would meet claim
8 21.

9 Q. Examples 1 and 2 don't start with
10 RNA; right?

11 A. In the beginning of example 1 it
12 says the microbial genomic DNA is isolated
13 in human saliva, so that to me would
14 suggest it's extracted DNA. And example 2
15 also begins with microbial generic DNA as
16 translated from human saliva.

17 Q. So you definitely agree with me
18 that example 1 and example 2 don't meet
19 claim 21?

20 A. Well, claim 21 is about RNA so
21 it's -- if they are starting with DNA they
22 are not starting with RNA.

23 Q. And example 3 doesn't refer to
24 any RNA ligation step prior to cDNA
25 transcription? I think I said the wrong

1 word. Example 3 doesn't refer to any RNA
2 fragmentation step prior to reverse
3 transcription; right?

4 A. It says kit reagents are used as
5 described by the manufacturer to generate
6 first and second strand cDNA. I do not
7 know from this alone whether or not that
8 includes a fragmentation component.

9 Q. The next paragraph is headed
10 generation of fragments with ligated
11 forward adaptors. Do you see that?

12 A. It does. This to me could mean
13 ligating forward adaptors onto fragments.

14 Q. Does it count if the RNA is
15 already fragmented but I don't -- well,
16 let me ask a better question.

17 Does it meet the limitation of
18 claim 21 if my sample of RNA is already
19 fragmented if I don't try to fragment it
20 further on purpose?

21 A. To be frank, my read of 21 would
22 include purposeful fragmentation.

23 Q. When you say include, it requires
24 purposeful fragmentation?

25 A. I think it requires a

1 fragmentation step that is done as part of
2 the preparation process.

3 Q. Okay.

4 A. On RNA to be clear.

5 Q. What I understand you to be
6 saying when you say a fragmentation step,
7 it means you take the RNA and you sonicate
8 it or you nebulize it but not like I just
9 have a bad post doc who doesn't know how
10 to prepare an RNA prep and so it ends up a
11 bit chunked up?

12 A. I would not consider an
13 inexperienced post doc a good
14 fragmentation system. It should be
15 purposeful fragmentation if we can call it
16 that.

17 MR. HOAGLAND-HANSON: I'm moving
18 on to a new topic and I know you have
19 your call coming up. Do you want to
20 break for the day today or do you want
21 to reconvene at 8 tomorrow?

22 THE WITNESS: I would appreciate
23 that. That's very kind of you.

24 (Time noted: 4:55 p.m.)

25

1 C E R T I F I C A T E .

2 STATE OF NEW YORK)

3 : ss.

4 COUNTY OF NASSAU)

5

6 I, CATHI IRISH, a Registered
Professional Reporter, Certified Realtime
7 Reporter, and Notary Public within and for
the State of New York, do hereby certify:

8 That DR. CARLOS BUSTAMANTE, the
witness whose deposition is hereinbefore
9 set forth, was duly sworn by me and that
such deposition is a true record of the
10 testimony given by the witness.

11 I further certify that I am not
12 related to any of the parties to this
13 action by blood or marriage, and that I am
14 in no way interested in the outcome of
15 this matter.

16 IN WITNESS WHEREOF, I have hereunto
17 set my hand this 22nd day of October,
18 2025.

19

20

21

22

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24

25


CATHI IRISH, RPR, CRR, CLVS

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1 Case: Integrated DNA Tech., Inc. vs Tecan Genomics, Inc.

2 Witness Name: Dr. Carlos Bustamante, Vol I

3 Deposition Date: 10/22/2025

4

5

6 Please be advised that the transcript in the above
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8 I do hereby acknowledge that I have read

9 and examined the foregoing pages

10 of the transcript of my deposition and that:

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12 () The same is a true, correct and

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16 attached Errata Sheet, the same is a true,

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1 Errata Sheet

2 NAME OF CASE: Integrated DNA Tech., Inc. vs Tecan Genomics, Inc.

3 DATE OF DEPOSITION: 10/22/2025

4 NAME OF WITNESS: Dr. Carlos Bustamante, Vol I

5 Reason Codes: 1. To clarify the record.

6 2. To conform to the facts.

7 3. To correct transcription errors.

8 Page _____ Line _____ Reason _____

9 From _____ to _____

10 Page _____ Line _____ Reason _____

11 From _____ to _____

12 Page _____ Line _____ Reason _____

13 From _____ to _____

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15 From _____ to _____

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22 DR. CARLOS BUSTAMANTE, VOL I

23

24

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