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Transcript of Bradley L. Pentelute, Ph.D.

Date: September 15, 2025

Case: Alamar Biosciences, Inc. -v- Olink Proteomics AB (PTAB)

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1 UNITED STATES PATENT AND TRADEMARK OFFICE

2
3 BEFORE THE PATENT TRIAL AND APPEAL BOARD

4
5 ALAMAR BIOSCIENCES, INC.,

6 Petitioner,

7 v.

8 OLINK PROTEOMICS AB,

9 Patent Owner.

10
11 Case No. IPR2024-01353

12 U.S. Patent No. 7,883,848

13
14 Oral Deposition of
15 BRADLEY L. PENTELUTE, Ph.D.

16 Denver, Colorado

17 Monday, September 15, 2025

18 8:57 a.m. MDT

19
20
21
22 Job No.: 600004

23 Pages: 1 - 206

24 Reported By: Lisa A. Knight

25 Notary Public, RDR, CRR, RSA

1 The oral deposition of
2 BRADLEY L. PENTELUTE, Ph.D., held at:

3
4
5 Denver Airport Marriott at Gateway Park
6 16455 East 40th Circle
7 Aurora, Colorado 80011

8
9
10 Pursuant to Notice, before Lisa A.
11 Knight, Realtime Diplomate Reporter, Certified
12 Realtime Reporter, Realtime Systems
13 Administrator, and Notary Public in and for the
14 State of Colorado.

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A P P E A R A N C E S

ALSO PRESENT:

PAMELA MANCINI, Ph.D., Sterne Kessler
(*via realtime feed)

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Conducted on September 15, 2025

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23 *STENOGRAPHER'S NOTE: All quotations from
24 exhibits are reflected in the manner in which
25 they were read into the record and do not
necessarily indicate an exact
quote from the document.

1 PROCEEDINGS

2 (September 15, 2025, at 8:57 a.m.)

3 BRADLEY L. PENTELUTE, Ph.D.,
4 having been first duly sworn to state
5 the whole truth, testified as follows:

6 EXAMINATION

7 BY MR. PLATT:

8 Q. Good morning. My name is
9 Christian Platt. I represent Alamar
10 Biosciences. Also with me on the line is
11 Laura Vining, also from Jones Day,
12 representing Alamar Biosciences, the
13 petitioner.

14 Could you state and spell your
15 full name.

16 A. Yeah. Bradley Lether
17 Pentelute. And then B-r-a-d-l-e-y; Lether,
18 L-e-t-h-e-r; Pentelute, P-e-n-t-e-l-u-t-e.

19 Q. And what is your current
20 position?

21 A. I'm a professor at MIT, in the
22 Department of Chemistry. And also I'm on
23 sabbatical this past 12-plus months. And so
24 I'm also interim CEO at a company I founded
25 called Amide Technologies.

1 Q. And where do you currently
2 live, just generally?

3 A. Boston area, yeah.

4 Q. Okay. And you understand
5 you've taken an oath to tell the truth today;
6 correct?

7 A. I do.

8 Q. Is there any reason why you
9 can't give complete, accurate, and truthful
10 testimony today?

11 A. No reason.

12 Q. Have you ever given sworn
13 testimony today -- sorry. Strike that.

14 Have you ever given sworn
15 testimony prior to today, like in a
16 deposition, in trial?

17 A. Yes, I have.

18 Q. And when was that?

19 A. Let's see. In the -- this
20 year, I did a deposition. It was in the
21 spring. And also it went to a judge trial in
22 Boston early summer, if I remember correctly.

23 Q. And did you testify at that
24 bench trial?

25 A. I did.

1 Q. And what was the name of that
2 case?

3 A. It was a case against Radius
4 Health and Orbicular.

5 Q. Could you spell that last name?

6 A. Yeah. O-b-i-c-u-l-a-r [sic].

7 Q. And what was the scope,
8 generally, of your testimony in that case?

9 A. Yeah, it was about Radius
10 Health's patents regarding a drug product
11 called Tymlos that they actually use to treat
12 osteoporosis.

13 Q. And did you give testimony as
14 an expert witness in that case?

15 A. I did.

16 Q. And did you prepare an expert
17 report in advance of that?

18 A. I did.

19 Q. And before the Radius Health
20 case, had you ever been deposed or given
21 sworn testimony before?

22 A. Yes. I had done two other
23 depositions.

24 Q. And what were those?

25 A. One was about methods of

1 manufacturing an oligonucleotide referred to
2 as a PMO, phosphorodiamidate morpholino
3 oligomer. And that was regarding, you know,
4 methods of manufacturing for Sarepta
5 Therapeutics and Nippon Shinyaku.

6 Q. And what was your role in that
7 case?

8 A. Expert.

9 Q. Okay. And what is the status
10 of that case?

11 A. I think it's finished.

12 Q. Do you know if that case went
13 to trial?

14 A. I don't think that case went to
15 trial.

16 Q. And you provided an expert
17 report in that case as well?

18 A. I did.

19 Q. And then you mentioned one
20 other case.

21 A. Yeah. The other case was about
22 a test for nonmaternal DNA, to understand
23 newborn health. And it was Ravgen, was one
24 of -- was one of the parties, and I think
25 Natera.

1 Q. And you gave -- were you an
2 expert witness in that case?

3 A. Yes, I was.

4 Q. And do you know if that case
5 went to trial?

6 A. I think it did, but I wasn't
7 involved. And it's finished. That was a few
8 years ago.

9 Q. On the Radius Health case, just
10 going backwards, what attorneys did you work
11 with on that case?

12 A. I always say their name wrong,
13 but it's C-h-o-a-t [sic]. "Coat." "Chote."

14 Q. Choate Hall?

15 A. Yeah. They're in Boston.

16 Q. And who were they representing?

17 A. Radius.

18 Q. And then the Sarepta case, who
19 were -- what attorneys were you working with
20 on that case?

21 A. I think it's Fenwick & West, if
22 I remember right. But I could be wrong.

23 Q. And do you recall who they were
24 representing?

25 A. Sarepta.

1 Q. And the Ravgen-Natera case?

2 A. Yeah. I was on the Natera
3 side, and it was WilmerHale.

4 Q. So you've done depositions
5 before, so you will have heard some of these
6 questions that have been asked. You've
7 probably been asked these before.

8 So you understand I'm going to
9 be asking you a series of questions today.
10 Correct?

11 A. I do.

12 Q. And if you need clarification,
13 will you ask me for clarification?

14 A. I will.

15 Q. And if you don't ask for
16 clarification, can we assume that you
17 understood the question?

18 A. Yes.

19 Q. Now you understand that counsel
20 for Olink may object to some of my questions
21 today. Do you understand that?

22 A. I do.

23 Q. And do you understand that
24 unless you're specifically instructed to not
25 answer my question, that you need to answer

1 my question?

2 A. I do.

3 Q. And you understand that you're
4 giving testimony today for a PTAB proceeding?

5 A. I do.

6 Q. And you understand that your
7 testimony here today is as if we were in the
8 courtroom?

9 A. I do.

10 Q. You also understand that your
11 testimony here is going to be filed publicly.
12 Do you understand that?

13 A. I do.

14 Q. Okay. You understand that your
15 colleagues and students could review your
16 deposition testimony?

17 A. I do.

18 Q. Now for the benefit of the
19 stenographer, you know, we're going to be
20 asking a series -- you know, going through a
21 series of questions and answers today, so
22 I would just ask that we try not to speak
23 over each other.

24 Is that fair?

25 A. It is.

1 Q. And you understand that it's
2 important to avoid giving nonverbal answers,
3 like uh-huh or huh-uh or shaking or nodding
4 your head?

5 A. I do.

6 Q. Now generally speaking we'll go
7 about an hour and take a break, but if, at
8 any time, that we need -- that you feel like
9 you need to take a break, just let us know,
10 and we can. The only thing I ask is that you
11 answer the question that's pending.

12 Is that fair?

13 A. Okay.

14 Q. Now it looks like you have a
15 document in front of you. What did you bring
16 with you?

17 A. This is my declaration.

18 Q. Okay. And is there any
19 notations or markings in it?

20 A. No.

21 Q. So let's go through a little
22 bit of terminology that we'll be using today.

23 You understand that this
24 deposition pertains to your declaration and
25 an inter partes review proceeding for U.S.

1 Patent No. 7,883,848?

2 A. I do.

3 Q. Now if I say "the '848 patent,"
4 you understand that I'm referring to the
5 patent with the number 7,883,848? It's also
6 on the cover of your declaration.

7 A. Yep. That's correct.

8 Q. Now if I refer to "Olink" or
9 "patent owner," you'll understand that I'm
10 referring to the patent owner, Olink
11 Proteomics AB?

12 A. Yes.

13 Q. And if I refer to "Alamar" or
14 "the petitioner," you'll understand I'm
15 referring to Alamar Biosciences. Is that
16 fair?

17 A. Correct.

18 Q. Now in your declaration, you
19 offer opinions from the perspective of a
20 person of skill in the art. Is that right?

21 A. I do.

22 Q. Now if I refer to "a person of
23 ordinary skill in the art," "a person of
24 ordinary skill," "a POSA" -- that's a
25 P-O-S-A -- will you understand I'm referring

1 to someone of ordinary skill in the art as of
2 July 2005?

3 A. I do.

4 Q. And if I don't specify
5 otherwise, will you assume that I'm asking
6 about your opinions from the perspective of a
7 person of ordinary skill in the art as of
8 July 2005?

9 A. I do.

10 Q. And will you answer my
11 questions from the perspective of a person of
12 skill in the art?

13 A. I will.

14 Q. So when did you first become
15 aware of this inter partes review proceeding?

16 A. I think in the spring of this
17 year.

18 Q. And how did you learn about it?

19 A. I received an e-mail from a
20 firm looking for experts with certain
21 background. And I responded to the e-mail
22 and met with them.

23 Q. And what firm was that?

24 A. It's called Patexia.

25 Q. Is that an expert witness

1 search firm?

2 A. Yes.

3 Q. And how long have you been
4 working with Patexia?

5 A. Since then. This is my first
6 time working with them.

7 Q. And then what happened after
8 you first were contacted by Patexia?

9 A. We had a virtual call to
10 discuss the general idea of the case and my
11 background.

12 Q. And who was that call with?

13 A. I don't remember. Maybe --
14 I think the person's name was Mosha, if
15 I remember correctly.

16 Q. At that point had you met with
17 any of the lawyers from Sterne Kessler yet?

18 A. No.

19 Q. So what happened after you met
20 with Mosha at Patexia?

21 A. I next met -- we had a call
22 with all of us, Sterne Kessler and Patexia.

23 Q. And who was on from Sterne
24 Kessler?

25 A. If I remember, David and Adil.

1 Q. And had you ever met David or
2 Adil prior to that call?

3 A. No, I haven't.

4 Q. And did you work with any of
5 the lawyers at Sterne Kessler prior to that
6 call?

7 A. So I've filed patents at MIT,
8 and MIT organizes who files those patents on
9 behalf of MIT, which I'm involved in because
10 I'm a coinventor. And in some cases, lawyers
11 from Sterne Kessler have filed those
12 provisional patents.

13 Q. And do you recall who at Sterne
14 Kessler you worked with on filing those
15 patents?

16 A. No, I don't.

17 Q. So what happened after you had
18 this virtual call with Sterne Kessler and
19 Patexia?

20 A. We decided to move forward. So
21 I went ahead and was retained as an expert.
22 And then started learning about the case.

23 Q. And what materials were you
24 provided?

25 MR. HOLMAN: I just want to

1 caution the witness not to disclose
2 the substance of any communications
3 with counsel.

4 Aside from that, you can
5 answer.

6 A. Materials that I cite in my
7 declaration.

8 BY MR. PLATT:

9 Q. Did you review any other
10 documents not cited in your declaration?

11 A. I don't believe so.

12 Q. So what did you do to prepare
13 for your deposition today?

14 A. Read all the documents. I've
15 thought about the content within these
16 documents. Discussed the materials with my
17 counsel. And, yeah, as a high-level, that's
18 what I did.

19 Q. And so did you meet with
20 counsel to prepare for the deposition?

21 A. Yes.

22 Q. And when did you meet with
23 them?

24 A. Periodically since we started
25 to work together in the spring. And then we

1 had planned a deposition, I think, three
2 weeks ago approximately. And we met in
3 person and were preparing for the meeting and
4 the deposition in Washington, D.C., that
5 ended up getting rescheduled to now.

6 Q. And I apologize to everybody
7 for that. It's outside of my control. So
8 I appreciate everybody's flexibility in being
9 able to reschedule this. So thank you for
10 that.

11 A. I hope you're healthy.

12 Q. So how long did you meet in
13 Washington, D.C., during that first -- about
14 three weeks ago?

15 A. I think we met for two days
16 before the scheduled deposition. And then
17 since the deposition was cancelled, we met in
18 the morning.

19 Q. Okay. When you say you met in
20 the morning, was that yesterday or today
21 or --

22 A. Oh, that was back when the --
23 during the reschedule in Washington, D.C.

24 Q. Okay. And then have you met
25 again since it was rescheduled?

1 A. Yes.

2 Q. And when did you meet?

3 A. We met a few times on Zoom, and
4 then we met yesterday.

5 Q. And how long did you meet
6 yesterday?

7 A. I think maybe six hours.

8 Q. And then how many hours did you
9 meet in Washington, D.C., in person?

10 A. So we had met in person for
11 two days, approximately eight hours each day.
12 And then a morning, the morning of the
13 originally scheduled deposition, so three,
14 four hours.

15 Q. And were there any other
16 lawyers present besides David and Adil?

17 A. One of their colleagues has
18 been participating. I think she's attending
19 law school and a colleague. Her name, if
20 I remember right, is Maria or something -- I
21 haven't met her in person, so -- I'm bad with
22 names. But, yeah, she's attended some of the
23 meetings, or actually most of them.

24 Q. Before you were contacted by
25 Patexia, had you ever worked with Olink

1 before?

2 A. No, I haven't.

3 Q. Had you worked with Thermo
4 Fisher, other than purchasing their equipment
5 maybe or using their equipment?

6 A. [REDACTED]
7 [REDACTED]
8 [REDACTED]
9 [REDACTED]
10 [REDACTED]
11 [REDACTED]
12 [REDACTED]
13 [REDACTED]
14 [REDACTED] [REDACTED]
15 [REDACTED]
16 [REDACTED] [REDACTED]
17 [REDACTED] [REDACTED]
18 [REDACTED]
19 [REDACTED] [REDACTED]
20 [REDACTED] [REDACTED]
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23 [REDACTED] [REDACTED]
24 [REDACTED]
25 [REDACTED] [REDACTED]

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Q. And do you know own any stock
or options in Thermo Fisher?

A. No, I don't.

Q. And have you ever worked with
Alamar Biosciences?

A. No, I haven't.

Q. I'm going to hand you what's
been previously marked as Exhibit 2010, which
is -- I'll just ask you: What is
Exhibit 2010?

A. It's my CV.

(Previously marked
Deposition Exhibit 2010 was
tendered to the witness.)

Q. And was this current at the
time that you submitted it to the Patent
Office?

A. (Document[s] reviewed.)

Yeah, it looks mostly current.
I think the one thing that's missing is my
interim CEO role at Amide Technologies.

Q. And when did you start your
sabbatical?

1 A. Last summer. So July of 2024.

2 Q. And when does that sabbatical
3 end?

4 A. January of 2026.

5 Q. And what are your obligations
6 to MIT during this sabbatical period?

7 A. So I still manage and run my
8 research group. So that's mentoring graduate
9 students and postdoctoral, publishing papers,
10 and filing patents. So the research
11 enterprise still needs to be maintained.

12 Teaching responsibilities are
13 reduced.

14 Q. And what is your -- so in terms
15 of hourly obligations to MIT, what are those
16 when you're on sabbatical?

17 A. It's not -- it's not defined in
18 terms of hourly. It's generally just thought
19 about having flexibility to do things other
20 than full-time teaching.

21 Q. When you are on sabbatical, how
22 much time can you spend on non-MIT-related
23 activities?

24 A. Oh, as much as you think is
25 responsible.

1 Q. And do you still have to clear
2 your consulting activities with your
3 department head?

4 A. Yes.

5 Q. And did you do that with
6 respect to your interim CEO role with Amide?

7 A. Yes.

8 Q. And did you clear your work
9 on -- as an expert witness in this PTAB
10 proceeding with your department head?

11 A. Yeah, so something like this,
12 it's just considered what's called our OPA,
13 outside professional activities. And we're
14 allowed one full day every week to do things
15 like expert witness and so forth.

16 I think that -- I have to
17 double-check, but this doesn't necessarily
18 fall under something like having it
19 cleared -- of being an interim CEO, for
20 instance, or being a cofounder of a company.
21 But it's generally cleared by MIT to act as
22 an expert witness.

23 Q. But you didn't get approval
24 from your department head to act as an expert
25 witness in this PTAB proceeding?

1 A. We don't need to.

2 Q. I understand that you don't
3 need to. But did you get approval?

4 A. We don't need -- by default, we
5 have approval to be an expert witness.

6 Q. So it was -- but after being
7 retained by Patexia, you did not obtain
8 separate approval from your department head
9 to act as an expert witness in this IPR
10 proceeding?

11 A. We have approval by default, by
12 being professors at MIT, to act as an expert
13 witness. There's no formality to where
14 it's -- that says you have to go get approval
15 to be an expert witness. We're already
16 granted approval to do this.

17 Q. So that's why you didn't ask
18 for approval?

19 A. Exactly. There's no approval
20 process. We're already granted approval.

21 Q. So if you look at the last
22 page of your CV --

23 A. Okay.

24 Q. -- you'll see there's a listing
25 of Companies Started.

1 A. Yes.

2 Q. And the first one there is --
3 actually the first one there is Amide
4 Technologies. That's what we've been talking
5 about?

6 A. Correct.

7 Q. And how much time have you
8 spent working with Amide Technologies during
9 your sabbatical?

10 A. It certainly varies, depending
11 on what's needed. I would say generally
12 sometimes a day a week, sometimes two days a
13 week. Certainly not five days a week, so...

14 Q. And you'll see also that of the
15 companies that are still active, there's New
16 Frontier Bio.

17 A. Yes.

18 Q. And what is your commitment to
19 New Frontier Bio?

20 A. So I'm a scientific cofounder,
21 and so if there are questions that come up
22 regarding the science that I can contribute
23 to, I'll participate on a Zoom call. The
24 commitment is -- really varies, but over the
25 past multiple months, it's minimal.

1 Q. And what about Decoy
2 Therapeutics, same question?

3 A. Yeah. So similar level of
4 involvement. I'm a scientific advisor.
5 Certainly provide scientific input, as
6 needed. And my contributions are, you know,
7 ad hoc. It depends on when a science
8 question comes up that's relevant to me.

9 Q. Now do you -- as part of your
10 research work at MIT, do you receive grants?

11 A. I do.

12 Q. And what grants do you
13 receive -- what grants have you received that
14 are currently ongoing?

15 A. I have some grants from the
16 National Institutes of Health. I have a
17 grant from the Foundation. And I have a
18 number of grants from industry.

19 Q. With respect to your grants
20 from the National Institutes of Health, have
21 those been cut back, withheld under the
22 current administration?

23 A. There's been delays. At this
24 stage, withheld, no.

25 Q. And have any changes with

1 respect to grant funding affected your
2 research laboratory?

3 A. Yes, generally research at MIT
4 and, as we hear about, at many universities
5 has taken a big change. There's
6 university-wide budget cuts, there's hiring
7 freezes. And certainly there's significantly
8 reduced mechanisms to apply for additional
9 grants and funding.

10 Q. And how has that affected your
11 lab?

12 A. At this moment, we're still
13 able to continue and do research. We slowed
14 down hiring, but mainly it was being really
15 careful because of potential limited funds.

16 Q. And when you say "slowed down
17 hiring," who would you be hiring?

18 A. So I hire postdoctoral
19 researchers -- right? -- and graduate
20 students.

21 Q. Now your opinions in this case,
22 they're not MIT's positions; right?

23 A. Correct.

24 Q. And you're being paid for your
25 opinions; is that right?

1 A. Yes.

2 Q. And how much are you being
3 paid?

4 A. \$500 per hour.

5 Q. And is that the Patexia rate?

6 A. I don't know what they charge.

7 Q. And how much time have you
8 spent so far on this proceeding?

9 A. So multiple tens of hours.
10 I would say maybe approaching 40 hours.

11 Q. 40 hours total?

12 A. Total.

13 Q. Including through your
14 preparation time and all the way up until
15 yesterday?

16 A. Yeah, I think that's --
17 I haven't ran an exact tally, but it's
18 around, you know, that number.

19 Q. And of that time, how much time
20 was spent preparing your declaration?

21 A. I don't know the exact number,
22 but, you know, a fair amount of time.
23 Probably, I don't know, 15 hours, 10 hours.
24 I don't know the exact number.

25 Q. And that would have included

1 your preparation work as well as in terms of
2 reviewing documents?

3 A. Yeah. Understanding aspects of
4 the materials and understanding Dr. Nelson's
5 report. There was a lot of exhibits and
6 things that I needed to think through and
7 then bring it together in my declaration.

8 Q. And while you're at -- while
9 you're on sabbatical from MIT, do you have a
10 reduced salary?

11 A. Yes.

12 Q. And what is that reduced
13 salary?

14 A. It's half.

15 Q. And what would that be?

16 A. [REDACTED]
17 [REDACTED]
18 [REDACTED]
19 [REDACTED]
20 [REDACTED]
21 [REDACTED]
22 [REDACTED]
23 [REDACTED]
24 [REDACTED]
25 [REDACTED]

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Q. Okay. So going back to the first page of your CV. So you received your BS in chemistry from the University of Southern California; is that right?

A. Correct.

Q. I have to ask, were you at the Rose Bowl?

A. I was studying.

Q. All right. Hook 'em. Okay. Great game.

And then you received your master's degree in chemistry from the University of Chicago in 2004?

A. Correct.

Q. Now what did you do after you received your master's in 2004?

A. So I was entered into the Ph.D. program at the University of Chicago. And so when you complete your classes and so forth in your first two years, you're -- providing everything is going well, you get your

1 master's. But I continued there, as planned,
2 to do my Ph.D.

3 Q. Okay. So you just continued.
4 You didn't have a break?

5 A. No.

6 Q. Okay. And then when did you
7 join MIT?

8 A. In 2011.

9 Q. And you obtained the position
10 of full professor in 2021?

11 A. Correct.

12 Q. Now you aren't a lawyer; right?

13 A. Correct.

14 Q. You're not a patent agent;
15 right?

16 A. Correct.

17 Q. And you're not an expert on
18 patent law issues; is that right?

19 A. That is correct.

20 Q. So looking at your CV, if we go
21 to page 6 and we look at your listing of
22 publications, these are listed in reverse
23 chronological order. Is that correct?

24 A. (Document[s] reviewed.)

25 Yes.

1 Q. And if we go all the way to
2 page 18, your earliest publication from MIT
3 was in 2012; is that right?

4 A. Let's see.
5 (Document[s] reviewed.)

6 That is correct.

7 Q. And then if we look at your
8 publications prior to MIT, those are also in
9 reverse chronological order. Is that right?

10 A. Yes.

11 Q. And if we turn to page 20, your
12 earliest publication is from 2006. Is that
13 right?

14 A. That's correct.

15 Q. And your CV doesn't list any
16 other publications that you authored prior to
17 2006; is that right?

18 A. (Document[s] reviewed.)
19 That's correct.

20 Q. Have you ever written an
21 article about a cis-reactive cell?

22 A. No, I haven't.

23 Q. When was the first time you've
24 heard the term "cis-reactive cell"?

25 A. When reviewing this case.

1 Q. Had you heard the term
2 "cis-reactive cell" before this case?

3 A. I've certainly heard of words
4 that resemble forming higher-order complexes
5 that involve oligonucleotides, and have heard
6 the word "cis" as opposed to "trans." And
7 I've heard the word "cell," but I don't
8 clearly remember a CRC being described in
9 terms of how this case does.

10 Q. Okay. You didn't come across
11 publications, for example, scientific
12 publications that use the term "cis-reactive
13 cell"; right?

14 MR. HOLMAN: Objection to form.

15 A. I don't remember.

16 BY MR. PLATT:

17 Q. The term "cis-reactive cell,"
18 that's not something that you use in your
19 ordinary research; is that right?

20 A. I haven't used it in my
21 ordinary research.

22 Q. And when was the first time you
23 heard this concept of a detectable trace?

24 MR. HOLMAN: Objection.

25 Foundation.

1 A. Generally, a detectable
2 trace -- it's difficult for me to pinpoint
3 when I've heard that for the first time.
4 I've certainly read about detectable tracing
5 in the '848 patent.

6 BY MR. PLATT:

7 Q. But prior to that, had you used
8 that term "detectable trace" in your
9 research?

10 A. I'm not sure. I don't
11 remember.

12 Q. So what is your area of
13 expertise, Dr. Pentelute?

14 A. So I'm a chemical biologist.
15 What that means is that I understand
16 chemistry of molecules related to biology.
17 I use tools and techniques taken from
18 chemistry and biology to understand typically
19 biological processes.

20 I invent methods to do this,
21 and I also use the methods to understand
22 various biological processes.

23 Q. So are you familiar with
24 chemical synthesis techniques?

25 A. Yes.

1 Q. And what about designing
2 nucleic acids?

3 A. Yes.

4 Q. And what's your experience with
5 high-throughput assays?

6 A. Quite extensive.

7 Q. And what generally -- what
8 kinds of high-throughput assays are you
9 familiar with?

10 A. Working with large libraries of
11 molecules and synthesizing them, figuring out
12 what molecules in these large libraries have
13 the function that we're searching for, and
14 being able to understand what one of those
15 molecules in those large libraries actually
16 have that particular function.

17 And then taking those new
18 molecules and using them to probe and
19 understand biology.

20 Q. And do you have experience with
21 Sanger sequencing?

22 A. I'm certainly aware of Sanger
23 sequencing. I've read papers that use Sanger
24 sequencing, especially being a leader in
25 peptide chemistry. So experience from the

1 literature and understanding how it's done.

2 But since the '90s, largely use
3 mass spectrometry instead of Sanger
4 sequencing. And that's what I use.

5 Q. And what is Sanger sequencing?

6 A. It's an ability to map a
7 molecule.

8 Q. And by mapping a molecule,
9 would that include mapping the nucleotides
10 and oligonucleotide?

11 A. Correct.

12 Q. And are you familiar with
13 sequencing by synthesis?

14 A. I am.

15 Q. And what is that?

16 A. That's the ability -- when you
17 actually end up doing polymerase reactions
18 synthesis, you're actually incorporating
19 probes that allow you to sequence.

20 Q. So I'm going to hand you what's
21 been marked as Exhibit 1042.

22 (Pentelute Exhibit 1042
23 marked for identification and
24 attached to transcript.)

25 Q. Do you recognize Exhibit 1042?

1 A. Yes.

2 Q. And what is Exhibit 1042?

3 A. It's the course syllabus for
4 2025 at MIT or -- what do they call this?
5 Course 5: Chemistry, Fall 2025.

6 Q. And these are the chemistry
7 courses that are being taught in the fall of
8 2025?

9 A. Yes.

10 Q. If we go to the fourth page in,
11 you'll see there's a class with the number
12 5.111, Principles of Chemical Science.

13 A. Correct.

14 Q. And then you'll see it refers
15 to -- I'll just ask you: What is the
16 description of the class?

17 A. Yeah, it's introduction to
18 chemistry, with emphasis on basic principles
19 of atomic and molecular electronic structure,
20 thermodynamics, acid-base and redox
21 equilibria, chemical kinetics, and catalysis.
22 Introduction to the chemistry of biological,
23 inorganic, and organic molecules.

24 Q. And you'll see there under
25 Fall, it says B. Pentelute?

1 A. Yes.

2 Q. And is that you?

3 A. It is.

4 Q. And you're going to be teaching
5 this class this fall?

6 A. Yeah, I teach this part of the
7 class, because I'm in a reduced teaching
8 load.

9 Q. Is that the first time you'll
10 have taught the Principles of Chemical
11 Science?

12 A. No, it's not.

13 Q. How many times have you taught
14 that class?

15 A. I don't remember exactly. I've
16 been a professor at MIT for 15 years, but
17 I would think I've probably taught this
18 around five times.

19 Q. And how often do you teach
20 classes versus, you know -- how many classes
21 do you normally teach in a semester?

22 A. Yeah, so it really depends on
23 the dynamics, at the time, of the department:
24 what teaching is needed, what your load is
25 for committees, and many other things. But

1 some professors will teach an entire semester
2 of a class or even two semesters of a class
3 throughout the academic year.

4 And so it varies. I think by
5 and large, it tries to be that every
6 professor teaches at least an equivalent of a
7 whole course, but it could be split up into
8 two separate half-courses, for instance.

9 Q. You're not just exclusively a
10 research professor; right?

11 A. No.

12 Q. And so when you teach, do you
13 have TAs teach for you or do you lecture
14 yourself?

15 A. Yeah, so the requirement in the
16 chemistry department is that all professors
17 give the lectures. Now there are TAs, and
18 they teach discussion sections that are
19 separate from the main lectures.

20 Q. And how do you -- just
21 generally how do you conduct your lectures?

22 Do you have a PowerPoint
23 presentation that you give? Do you diagram
24 things on chalkboards, whiteboards?

25 You know, what generally is

1 your approach to teaching a class?

2 A. It depends on the class. In
3 particular, this class, 5.111, is -- all MIT
4 undergraduates need to take a chemistry
5 class. And I do this class largely on the
6 chalkboard for the entire 50 minutes.
7 However, I do teach them biological systems,
8 in light of what I said my expertise is. And
9 in some cases, then, I'll use PowerPoints.

10 Q. So let's go over some basic
11 science questions so we can make sure we're
12 on the same page. What is a mole?

13 A. A mole refers to a number of
14 molecules. In particular, 6 times 10 to the
15 power of 23 molecules equals a mole.

16 Q. And what is molar
17 concentration?

18 A. That's the number of moles
19 divided by a volume, typically in liters.

20 Q. And how does a millimolar
21 concentration compare to a molar
22 concentration?

23 A. So as the word implies, it's a
24 thousand times less.

25 Q. How does having a lower molar

1 concentration affect chemistry?

2 A. It's hard to generalize.

3 There's -- it's -- it depends on what you're
4 looking at. Certainly there's less
5 molecules.

6 Q. And less molecules, when you're
7 dealing with enzymes, can result in less
8 activity; is that right?

9 MR. HOLMAN: Objection to form.

10 A. It really depends on the
11 system. Right?

12 If you're at saturating
13 conditions, even with a reduced
14 concentration, you still have the same level
15 of activity.

16 BY MR. PLATT:

17 Q. So let me ask you a question.
18 If you have a 1 millimolar concentration of
19 something in a 10 mls solution -- aqueous
20 solution, if you add in 90 mls more of that
21 solution, the new molar concentration would
22 be 0.1 millimolar. Is that right?

23 A. It would be reduced by a factor
24 of 10.

25 Q. So if it was 1 millimolar, then

1 it would be .1 millimolar?

2 A. Yeah.

3 Q. So in your class, do you teach
4 students what nucleic acid is?

5 A. Yes. I have sections of --
6 mentions of how proteins are made in cells,
7 which follows the central dogma of biology,
8 from DNA to RNA to protein. And that
9 involves an oligonucleotide.

10 Q. And what is a nucleic acid?

11 A. It's an oligo polymer, a
12 precision polymer that has essentially bases
13 connected by phosphate backbones, and, in
14 between, a ribose or a deoxyribose ring.

15 Q. And what are the bases in DNA?

16 A. So you have adenine, thymine,
17 guanine, and -- let's see. You also have
18 uracils, thymine, guanine, and adenine.

19 Q. And are those abbreviated A, C,
20 T, and G?

21 A. Yes.

22 Q. And then U is?

23 A. Uracil.

24 Q. And that's an RNA?

25 A. Correct.

1 Q. Now you also mentioned
2 oligonucleotide. What do you mean by an
3 oligonucleotide?

4 A. Oh, that's a string of nucleic
5 acids.

6 Q. And nucleic acids can be
7 double-stranded? Is that right?

8 A. They can be double-stranded.

9 Q. And nucleic acids can be
10 single-stranded?

11 A. Yes.

12 Q. What is the -- can you explain
13 the difference between the double-stranded
14 DNA and the single-stranded DNA?

15 A. Yeah, so if you have a 5 prime
16 to 3 prime oligonucleotide and you go ahead
17 and make or have a 3 prime to 5 prime, that's
18 the complement. They can base stack and form
19 a duplex, which can be referred to as
20 double-stranded DNA or double-stranded RNA.

21 Q. And when you say that they
22 complement, is that also referred to as
23 hybridization?

24 A. Yes.

25 Q. And how do nucleic acids

1 hybridize?

2 A. So they hydrogen-bond with
3 their bases.

4 Q. And certain nucleic acids
5 hybridize with another nucleic acid because
6 they have affinity for each other. Is that
7 right?

8 A. It's the side chains or the
9 bases have atoms arranged correctly. For
10 instance, A will pair with T, and G will pair
11 with C. And it's essentially
12 hydrogen-bonding interactions between those
13 bases within the oligonucleotides.

14 Q. And is that what causes the
15 oligonucleotides to have affinity for each
16 other?

17 A. Yes.

18 Q. Now when you have a
19 double-stranded DNA that has complementary
20 region, can you increase the affinity between
21 those two nucleic acid strands by increasing
22 the length of the complementary region?

23 MR. HOLMAN: Objection. Form.

24 A. So there's a number of
25 different ways to increase affinity.

1 Depending on the circumstance, if you
2 increase the length, it could increase the
3 affinity, but it depends on the design.

4 BY MR. PLATT:

5 Q. How so?

6 A. Well, you need to make sure
7 that your base pairing is complementary. So
8 if you're increasing the length, are you
9 still maintaining that complementary
10 interaction?

11 And then if you're adding more
12 GCs, for instance, that gives you three
13 hydrogen bonds as opposed to more ATs; it
14 gives you two. And that has to -- that
15 alters or varies the degree of potential
16 affinity increase.

17 Q. So a higher number of GCs will
18 increase affinity more than a higher
19 concentration of ATs?

20 A. Correct.

21 Q. Have you heard the term
22 "spatial affinity"?

23 A. Spatial affinity. Seems like
24 an open-ended term. I'd like to know a
25 little bit more about what you're referring

1 to.

2 Q. Would you agree that nucleic
3 acids that are in spatial proximity to one
4 another -- to single-stranded nucleic acids
5 that are in spatial proximity are more likely
6 to hybridize the nucleic acids that are not
7 in spatial proximity?

8 A. I see.

9 MR. HOLMAN: Objection to form.

10 A. So depending on the situation
11 of how all this works, you could template two
12 nucleotides to be close in space, but they
13 may not necessarily have increased affinity,
14 depending on the design.

15 However, there could be
16 situations when you could, by chance,
17 increase their affinity by one another.

18 BY MR. PLATT:

19 Q. So are there situations where
20 you may have a longer single-stranded
21 oligonucleotide and a shorter single-stranded
22 oligonucleotide that hybridizes with part of
23 it?

24 MR. HOLMAN: Objection to form.

25 A. So there are many possibilities

1 in the world of nucleotides. I'd have to
2 understand exactly what you're saying.

3 But short oligonucleotides can
4 hybridize with long oligonucleotides.

5 BY MR. PLATT:

6 Q. And are there -- are you
7 familiar with using polymerase to extend one
8 of those strands?

9 A. Yes.

10 Q. And what is -- how does
11 polymerase work in DNA?

12 A. Yeah, so it's called a
13 polymerase chain reaction. And polymerase is
14 referring to an enzyme. And essentially if
15 you take a double-stranded piece of DNA and
16 you design primers that recognize 5 prime and
17 3 prime, short pieces of oligonucleotides
18 that recognize the existing piece of
19 double-stranded DNA, you go ahead and heat
20 that up.

21 And what you do is the
22 annealing breaks the association of the DNA
23 duplex. You cool it down, and those primers
24 are added in excess. So they will, in fact,
25 now hybridize.

1 And then what happens is Nobel
2 Prize chemistry, is then you use the
3 polymerase, which will march along at those
4 primer sites and fill in to produce copies of
5 those double-stranded DNA that was often
6 there at a much lower amount.

7 Q. And in PCR, it's thermal
8 cycling to increase the amount of nucleic
9 acid?

10 A. That's why it's called a
11 polymerase chain reaction, is because it's a
12 chain reaction. And it exponentially
13 increases the amount of DNA, depending on the
14 number of thermal cycles that are ran.

15 Q. And prior to PCR being
16 invented, was there a way to do just the
17 polymerase piece of it to -- for gap filling,
18 or other ways of addressing a nucleic acid
19 that may only have -- you know, one strand is
20 longer than the other strand?

21 MR. HOLMAN: Objection to form.

22 A. So if I remember right, PCR was
23 invented or discovered by Mullis in the
24 1980s. And the aha moment was realizing that
25 a polymerase would be adapted to do the

1 polymerase chain reaction.

2 Before that, there were many
3 techniques that were used to modify DNA.

4 BY MR. PLATT:

5 Q. And are you familiar with
6 primer extension reactions?

7 A. Yes.

8 Q. And what is that?

9 A. That's essentially when you add
10 a primer to a PC DNA and you use that to
11 extend off of the DNA.

12 Q. And what do you use to do that?

13 A. You still use a polymerase and
14 you use a primer and a PC DNA.

15 Q. And where do you get that
16 polymerase?

17 A. You know, they're catalog
18 reagents. New England Bio Labs, for
19 instance, will sell them.

20 Q. Okay. Can you also make the
21 polymerase?

22 A. Yes.

23 Q. And how would you make the
24 polymerase?

25 A. So if you're doing a

1 recombinant expression, say from an E. coli
2 host, you would isolate the DNA that encodes
3 for that polymerase. Or nowadays, you would
4 order it from a plasmid or DNA manufacturer.
5 And then you would have that gene
6 incorporated into what's called a plasmid.

7 The important thing about a
8 plasmid, it has a selection marker on it.
9 And so you'll transfect that into a, say,
10 E-coli BL21 (DE) star. And you'll go ahead
11 and select for the bacteria that only took in
12 that plasmid by virtue of an antibiotic
13 selection.

14 And then you'll have bacteria
15 that now can produce that particular protein.
16 And you'll actually ferment it. And you'll
17 isolate the enzyme from E. coli through lysis
18 and follow on by purification.

19 Q. And when you make that
20 polymerase, you would then separate that out?
21 Is that right?

22 A. Yeah, so a really nice trick is
23 you use what's called affinity chromatography
24 nowadays. In the past, you could use
25 ammonium sulphate precipitation or other

1 tools.

2 But nowadays you use what's
3 called His-tag affinity purification, if your
4 enzyme is still functioning with that
5 His-tag. Because it may alter its function.

6 But you'll use a nickel NTA
7 column to capture the His-tag, purify it,
8 elute it with imidazole. And then in some
9 cases people will remove that affinity tag.
10 So hopefully you'll get the native, wild-type
11 enzyme.

12 Q. And then you could put that
13 polymerase in a buffer? Is that right?

14 A. Yes.

15 Q. And you could design that
16 buffer to be such that it would work best in
17 the reaction that you're doing? Is that
18 right?

19 A. So every reaction needs the
20 correct conditions. It needs to have the
21 right pH, the right ions, the right amount of
22 salt.

23 Yeah, so a biochemist would set
24 up a set of conditions that allow that enzyme
25 to be functional.

1 Q. You wouldn't want to do a
2 reaction, for example, that wouldn't work
3 because you used the wrong buffer conditions,
4 for example?

5 MR. HOLMAN: Objection to form.

6 A. So it depends on the context.
7 So if you're trying to
8 understand the structure and function of an
9 enzyme, one would test a number of different
10 conditions. And if they learned that pH 4
11 didn't work, that's useful scientific
12 information.

13 If the goal was more a
14 synthetic biology approach. You wouldn't
15 want to know conditions where your enzyme was
16 able to turn over.

17 BY MR. PLATT:

18 Q. So for example, if you had it
19 in, like you said, a pH of 4 and that caused
20 problems with your reaction, you would not do
21 it in a pH of 4?

22 A. You would change the pH.

23 Q. And if, for example, you had
24 something in your buffer that would cause
25 problems with your reaction, you would adjust

1 your buffer so that it wouldn't cause that.

2 A. So it's a spectrum. It depends
3 on what the goal is.

4 So you may have an additive or
5 something in a buffer that doesn't allow
6 maximal performance, but it turns out to be
7 important for something else. And so in that
8 case, you may tolerate a condition that's not
9 as efficient for the chemistry perhaps, but
10 important for a different variable.

11 Generally speaking, it's
12 multiple variables that have to be understood
13 in order to do, you know, the set of
14 chemistry or the goal of the experiment.

15 Q. And you would try and balance
16 those conditions to achieve the result that
17 you were trying to obtain as a result of the
18 reaction that you were doing with that
19 enzyme?

20 A. Yeah, you would want to take a
21 world view of what you're trying to
22 accomplish and adjust the parameters so that
23 the system is working as you desired.

24 MR. PLATT: We've been going a
25 little over an hour. Now would be

1 good for a break.

2 MR. HOLMAN: Yeah.

3 (Recess: 10:00 a.m. to
4 10:10 a.m.)

5 BY MR. PLATT:

6 Q. So before the break, we were
7 talking about polymerases, and you mentioned
8 that you could purchase them. Is that right?

9 A. Yes.

10 Q. How do you purchase polymerase?

11 A. You go to a catalog or a
12 website and you find a polymerase that you
13 want to buy. And if you're at MIT, we do
14 this through what's called a punch-out
15 vendor, which is more or less a catalog, like
16 a Sigma-Aldrich catalog, that you say, "I
17 want 10 micrograms of this particular catalog
18 number or this SKU," and the punch-out vendor
19 system automatically issues a PO and the
20 enzyme shows up at some specified delivery
21 date.

22 Q. And in issuing orders, are you
23 able to request custom buffers, for example?

24 A. So most catalog products will
25 have predetermined buffers.

1 Q. But if you wanted to order
2 polymerase with a custom buffer, could you do
3 that?

4 A. So most of the time, no,
5 because the vendors that sell enzymes have --
6 make a bucket of it, and that's it. That's
7 what you get.

8 Now, you know, you could call a
9 service provider and you would have to work
10 with them to figure out some mechanism to
11 change the eventual buffer.

12 Q. But you could do that, it would
13 just be probably more expensive?

14 MR. HOLMAN: Objection to form.

15 A. Yeah, I mean, it's a
16 hypothetical. You don't know if it would
17 work.

18 BY MR. PLATT:

19 Q. But if you wanted to order a
20 polymerase with a custom buffer, that's
21 something you could order from a service
22 provider?

23 A. I would say it depends on the
24 context.

25 So if you picked the wrong

1 buffer, the polymerase may not even be
2 soluble. So then you can't get it.

3 Q. But if somebody wanted to order
4 a polymerase with a custom buffer that they
5 knew would work, that's something that they
6 could order?

7 A. I'm sure there's a service
8 provider somewhere where you could devise
9 some strategy to get a protein in a buffer.

10 Q. And a protein like an enzyme
11 like polymerase; right?

12 A. Yeah.

13 Q. So we were talking about
14 oligonucleotides that are hybridized
15 together.

16 Do those oligonucleotides stay
17 hybridized -- do they only stay hybridized
18 unless sort of reaction conditions change?

19 MR. HOLMAN: Objection to form.
20 Foundation.

21 A. It depends. Are they
22 oligonucleotides that are a million years
23 old? Right? Are they -- under what
24 conditions? All chemistry depends on the
25 conditions.

1 BY MR. PLATT:

2 Q. How does pH dehybridize DNA?

3 A. So pH, if you make it more
4 acidic, eventually you'll protonate
5 everything. And when you start to protonate
6 things, it's going to disturb how it can
7 interact with one another, with the sense and
8 antisense strand.

9 If you go to the other end of
10 the spectrum, say basic pH, you'll start to
11 deprotonate. Deprotonating means nitrogen is
12 there, or on or part of the base, the
13 hydrogen comes off the nitrogen.

14 And you're going to start to
15 cause side reactions and various detrimental
16 effects. And that will be how it actually
17 comes apart in response to changes in pH.

18 Q. Taking, for example, a pH of
19 7.5, that wouldn't dehybridize the DNA?

20 MR. HOLMAN: Objection to form.

21 A. It really depends on the
22 conditions. What else is in there? Right?
23 What type of buffer is being used? What
24 other ions are present?

25 I can't generalize because

1 there's many ways to have pH 7.5. But what
2 else is there? How is that pH being
3 maintained? What is the salt concentration?

4 BY MR. PLATT:

5 Q. And we were talking earlier
6 about PCR. Can PCR be used on single-
7 stranded DNA?

8 A. No.

9 Q. And why do you say that?

10 A. Because the way the polymerases
11 work, they need to start from a
12 double-stranded PC DNA.

13 Q. Let me ask it this way. Can
14 PCR work with single-stranded DNA and
15 appropriate primers?

16 MR. HOLMAN: Objection. Form.

17 A. So in the context as I walked
18 through the PCR reaction, you take
19 double-stranded DNA, you heat it up. And if
20 you do that in the presence of primers, you
21 actually -- when you cool it down, it will
22 anneal with those primers, now forming that
23 double-stranded DNA at one region. And
24 another region of that PC DNA will be
25 single-stranded.

1 BY MR. PLATT:

2 Q. Could you start that process
3 with a single-stranded DNA, along with the
4 appropriate primers and polymerase and other
5 reaction conditions?

6 A. Yes.

7 Q. And then are you familiar with
8 microarray analysis?

9 A. I am.

10 Q. And what is that?

11 A. That's a tool that's used to
12 detect the presence of particular
13 oligonucleotides. So typically it's a glass
14 surface that has covalently attached pieces
15 of DNA in a specific array. And then if you
16 have DNA in solution that has a fluorophore
17 on it, for instance, it can hybridize.

18 And if you see it associate
19 with this glass surface, you actually have a
20 method of detection of a particular piece of
21 DNA that was in solution with a microarray.

22 Q. And does a microarray analyze
23 single-stranded DNA?

24 A. It can.

25 Q. It can also analyze

1 double-stranded DNA?

2 A. You have to have the right
3 conditions, but you can detect pieces
4 ultimately that were part of double-stranded.
5 But for a hybridization to take place, it
6 will end up being double-stranded on a piece
7 of microarray.

8 Q. And that's because what's on
9 the microarray is single-stranded, and you're
10 hybridizing it with whatever the sample is
11 that's single-stranded?

12 A. In the microarray we're talking
13 about. There are other types of microarrays,
14 but often the first base microarray is a
15 single-stranded DNA attached.

16 Q. Going back to Exhibit 1042, the
17 class list.

18 A. Yes.

19 Q. And if you go to page 8, you'll
20 see there's class 5.383, Fast-Flow Peptide
21 and Protein Synthesis.

22 Do you see that?

23 A. Yes. Let me get there. Hold
24 on a second.

25 (Pause.)

1 Okay.

2 Q. And is this a class that you
3 are teaching?

4 A. So it's a laboratory class that
5 I designed. It's on protein and peptide
6 synthesis.

7 Q. How long have you taught this
8 class?

9 A. So since this is one class that
10 I helped to design, it's been -- I've taught
11 it more or less maybe 12 times out of my
12 15 years at MIT.

13 Q. And is this a fair description
14 of the class that you're teaching?

15 A. (Document[s] reviewed.)

16 Yes.

17 Q. And one of the things that you
18 teach in this class is disulfide bond
19 formation. Is that right?

20 A. That is correct.

21 Q. What is disulfide bond
22 formation?

23 A. So it's a reaction between a
24 sulfur, often within amino acids, such as
25 cystine, or in thiols.

1 Generally speaking in
2 chemistry, that when you have a sulfur atom
3 attached to another sulfur atom, it's called
4 a disulfide bond. And those are found in
5 proteins and other types of molecules.

6 Q. And are those also used as
7 tools in research?

8 A. Yes.

9 Q. And how are they used as tools
10 in sort of peptide research, peptide nucleic
11 acid research?

12 A. Broadly speaking, the sulfur
13 moiety is what we call a nucleophile. It's a
14 really nice nucleophile.

15 And so it's used broadly as a
16 bioconjugation handle. So if you have an
17 oligonucleotide or a protein and you want to
18 attach a biotin or a fluorophore, it's often
19 done at that sulfur atom.

20 Q. And are you also familiar with
21 breaking disulfide bonds?

22 A. Yes.

23 Q. And how are disulfide bonds
24 broken?

25 A. Primarily with a reductant.

1 Q. And what is that?

2 A. So a reductant is a molecule
3 that provides electrons.

4 Q. And what are some examples of
5 reductants that are used with breaking
6 disulfide bonds?

7 A. So they can be thiols, for
8 instance. So the simplest one is cysteine,
9 which is a thiol-containing amino acid. It
10 could be glutathione.

11 DTT is another example. TCEP
12 is another example. There are a lot of
13 different molecules that can provide
14 electrons that will reduce the disulfide,
15 which refers to as breaking it.

16 Q. And how do those different
17 reducing agents that you referred to, how do
18 they differ amongst each other in breaking
19 disulfide bonds?

20 A. It really depends on the
21 context again. It's a -- first off, you have
22 to understand: What disulfide bond are you
23 going to reduce or break? Right?

24 Then the next questions are:
25 Under what conditions? What's the pH?

1 What's the concentration? Then the next
2 level is: Okay, well, what reducing agent
3 are you going to use?

4 So some of them are based on
5 thiol, which have a certain set ability to
6 donate electrons. Others are based on
7 phosphines, such as TCEP, which has a
8 different set of properties.

9 So it really depends on the
10 context in which you're aiming to do the
11 reduction or disulfide bond breakage.

12 (Pentelute Exhibit 1043
13 marked for identification and
14 attached to transcript.)

15 Q. I'm going to hand you what's
16 been marked as Exhibit 1043, which is U.S.
17 Patent No. 11,014,961.

18 Do you recognize Exhibit 1043?

19 A. I do.

20 Q. And this is a patent that
21 you're an inventor on; is that right?

22 A. That's correct.

23 Q. And it's called Self-Labeling
24 Miniproteins and Conjugates Comprising Them.
25 Is that right?

1 A. That's correct.

2 Q. What does this patent relate
3 to?

4 A. I didn't hear --

5 Q. What does this patent relate
6 to? What idea did you come up with?

7 A. Oh, yeah --

8 MR. HOLMAN: Objection. Form.

9 A. I tried to understand in the
10 world of a biopolymer, such as a peptide, if
11 there is some type of higher-order structure,
12 three-dimensional confirmation that we're
13 referring to as a mini protein that's capable
14 of promoting a chemical reaction that we
15 refer to as an SnAr reaction. It's a
16 reaction at a thiol within a cysteine
17 residue.

18 BY MR. PLATT:

19 Q. And how would that be used?

20 A. So as I mentioned, generally
21 speaking, chemical biologists, biochemists,
22 chemists need tools to attach small
23 molecules, often, onto proteins. And so if
24 you have a reaction that's selective at a
25 cysteine residue, then you can attach a small

1 molecule to that particular cysteine residue,
2 perhaps within the context of a protein or
3 some other type of molecule.

4 Q. So if you look at -- go to
5 column 2. It's about -- it's after all the
6 figures. And there's a discussion there
7 under Figure 4. It says -- and you mentioned
8 selection a second ago.

9 It says, "Figure 4 shows
10 selection scheme with the capture agent (CA)
11 and library design." And then it says, "An
12 mRNA display selection scheme and embodiment
13 of the water-soluble CA is shown as a cartoon
14 and chemical structure."

15 And if you could take a look at
16 Figure 4. Can you describe what Figure 4 is
17 showing from a selection standpoint?

18 A. So --

19 Q. Maybe just ask generally: What
20 is selection?

21 A. Yeah. So what we're doing is
22 we're taking a library of mRNA that encodes
23 for a peptide. So the mRNA is shown as the
24 block solid line, and then the peptide that's
25 attached to the RNA is shown as circles. And

1 those are the amino acids.

2 And ultimately what we're doing
3 is we're taking this probe, this
4 perfluoroarene. This is the molecule down in
5 the bottom right panel with the fluorine
6 atoms. And it has a biotin.

7 Now we have a ten to the power
8 of 13 molecules in the test tube right now.
9 And we're asking the question: How many of
10 these molecules are capable of reacting with
11 this fluorine-containing molecule that I'm
12 showing you, the one with Fs on it? So are
13 there any molecules there that are able to
14 react at cysteine? And if so, can I capture
15 and detect that?

16 So the selection step is
17 whether or not we're able to form a reaction
18 with any one of these 10 to the power of 13.
19 And that is part of the molecule that's
20 designed in this perfluoroarene moiety.

21 Q. So part of the selection is the
22 reaction taking place?

23 A. The selection has to do with
24 whether or not there's a matched pair. Are
25 there any molecules that are folded correctly

1 and have the right confirmation to then
2 actually interact with this probe, and
3 consequently you have a chemical reaction.

4 So it's the matched pair
5 phenomenon that provides the selection step.

6 Q. So you can have -- when you're
7 in -- then it says "Streptavidin pull down"
8 and "Biotin." So what is that?

9 A. So that's the detection step.
10 So we're able to detect the molecules that
11 got biotin because of the selection. And
12 then we can go to NGS sequencing, in this
13 case, to understand what biopolymer actually
14 ended up facilitating that matched pair.

15 Q. So part of the selection can be
16 a preexisting reaction step that makes the
17 compound sufficient such that the biotin can
18 be captured?

19 MR. HOLMAN: Objection to form.

20 A. Yeah, this experiment is really
21 about the fact that it's the probe, the
22 molecule that reacts, matched with the
23 polypeptide chain and adopting the right
24 interaction that then consequently gets the
25 chemistry to take place. That's the

1 selection step.

2 Now getting rid of everything
3 else enriching for or detecting what took
4 place is what the biotin is used for.

5 BY MR. PLATT:

6 Q. But just generally when you're
7 doing selection, part of that can include
8 reactions that take place prior to, for
9 example, this capture step of biotin and
10 streptavidin?

11 MR. HOLMAN: Objection. Form.

12 A. Yeah, I don't understand what
13 you mean.

14 BY MR. PLATT:

15 Q. Well, you mentioned that you're
16 doing these matched pairs and that that's
17 what is then being, you know -- the matched
18 pairs, is that a proxy for something? Or
19 what is it?

20 A. So this is a discovery
21 experiment. So we had to make 10 to the
22 power of 13 molecules. That's what's in the
23 beaker, test tube.

24 Q. Okay.

25 A. So we have no idea what one of

1 these molecules are capable of adopting a
2 confirmation that would then allow for an
3 interaction to match the two so that then a
4 bond forming at this thiol step will take
5 place.

6 So it's really the matched
7 pairs. It's all three parts coming together
8 that enable for the selection step to take
9 place.

10 BY MR. PLATT:

11 Q. So the selection can be a
12 process?

13 A. Well, in this case, it's a
14 number of things that are taking place.

15 Q. In order to select for those
16 matching pairs; is that right?

17 MR. HOLMAN: Objection. Form.

18 A. So you have, again, the power
19 of -- 10 to the power 13. We don't know what
20 any of these molecules are doing. And we
21 don't know if any of them can interact with
22 this perfluoroarene to form this complex.

23 When the complex forms, there's
24 one more step, and it's a bond-forming
25 reaction that takes place, which we didn't

1 know if that could even take place.

2 And that's the selection step
3 in this experiment.

4 BY MR. PLATT:

5 Q. When you say "selection step,"
6 that can include a process; is that right?

7 MR. HOLMAN: Objection to form.

8 A. Yeah, I don't know what you
9 mean by "process."

10 BY MR. PLATT:

11 Q. Well, it's a series -- you
12 can -- in a selection step, you can have
13 certain activities that happen beforehand in
14 order to facilitate that selection. Is that
15 right?

16 MR. HOLMAN: Objection to form.

17 A. I mean, the key step in this
18 experiment is having everything in the test
19 tube and selecting for the matched pair.
20 Everything is just mixed in the test tube.

21 BY MR. PLATT:

22 Q. So if you could turn to
23 column 15. You'll see -- if you go down to
24 about line 28 or so, it's talking about -- it
25 begins with "In another aspect, the

1 disclosure relates to a conjugate comprising
2 many of the proteins," and it goes on.

3 So what is a conjugate?

4 A. Let's see if I defined it here.

5 (Document[s] reviewed.)

6 MR. HOLMAN: Objection.

7 Relevance.

8 A. (Document[s] reviewed.)

9 I haven't looked at this patent
10 in a while.

11 (Document[s] reviewed.)

12 So we don't define the word
13 "conjugate" in the patent, at least in
14 column 17 and 18.

15 So a conjugate is a general
16 chemical term that refers to attaching A and
17 B molecules together by a covalent bond or a
18 noncovalent.

19 BY MR. PLATT:

20 Q. You'll see further in, it
21 refers to an antibody.

22 A. And you're on column 15 still?

23 Q. Column 15, yeah. Sorry.

24 My question is: What's an
25 antibody?

1 MR. HOLMAN: Objection.

2 Relevance.

3 A. Okay. So an antibody is a
4 molecule classically produced by our immune
5 system. It is also produced as therapies in
6 the factories.

7 It's a molecule that has
8 properties that recognize what's called an
9 antigen. It also has an FC region that has a
10 whole 'nother set of functional properties.

11 But an antibody is a protein
12 molecule.

13 BY MR. PLATT:

14 Q. And you'll see, if you go in
15 further, it says "In some embodiments, the
16 conjugation reaction occurs under mild
17 protein-compatible conditions."

18 And then it says "In some
19 embodiments, the conjugation reaction further
20 comprised a reducing agent, such as" -- I'll
21 abbreviate here -- "DTT or TCEP."

22 Do you see that?

23 A. I do.

24 Q. Is that the reducing agents you
25 were talking about earlier?

1 A. It's two of them. I listed a
2 number of additional reducing agents.

3 Q. And what's -- what are some of
4 the differences between DTT and TCEP?

5 A. Yeah, so DTT -- 1,2
6 dithiothreitol -- is a thiol-based reducing
7 agent. So it's using the electrons from the
8 sulfur atom to reduce the disulfide bond.

9 Whereas TCEP is a
10 phosphine-based reducing agent; so not sulfur
11 S, but P for phosphate. And so it's still --
12 ultimately is able to, under the right
13 conditions, provide electrons to reduce a
14 bond.

15 Q. And what are the right
16 conditions in order for it to reduce a bond?

17 A. As I mentioned, every example
18 determines the conditions. The conditions
19 aren't generalized. You need to understand
20 the pH, the concentrations, the salt, what is
21 it that you're trying to reduce.

22 Q. Do you know what the molar
23 concentration of DTT is needed to break a
24 disulfide bond?

25 A. Again, a range is needed. And

1 it depends on the disulfide bond. It could
2 be micromolar. It could be millimolar. It
3 could be thousands of times, in terms of the
4 spectrum, in terms of -- it depends on what
5 you're trying to reduce.

6 Q. So in the same conditions, you
7 know, looking at TCEP and DTT, is TCEP a
8 stronger reducing agent?

9 MR. HOLMAN: Objection to form.

10 A. It depends on what you're
11 trying to reduce, and it depends on even the
12 confirmation.

13 So is the disulfide bond locked
14 into a protein confirmation? Is it even
15 accessible? The physical properties of TCEP
16 versus DTT would matter in that case.

17 So you can't generalize, in
18 terms of how strong of a reductant it is. It
19 also depends on the pH, for instance. If you
20 change the pHs, it changes the reducing
21 potential.

22 BY MR. PLATT:

23 Q. So you mentioned the range of
24 concentration of DTT could break disulfide
25 bonds, you know, when you're working with

1 general buffered conditions used in
2 biological experiments.

3 What level of molar
4 concentration of DTT would break a disulfide
5 bond?

6 MR. HOLMAN: Objection to form.

7 A. As I said, it really depends.
8 Is it just a single protein in a test tube?
9 And at what pH? Or is it a cell lysate with
10 a bunch of proteins that you're working up to
11 reduce for a proteomic experiment?

12 The conditions really matter.
13 So the range is -- varies by a thousandfold.

14 BY MR. PLATT:

15 Q. Okay. So would 50 millimolar
16 of DTT reliably and measurably break
17 disulfide bonds?

18 MR. HOLMAN: Objection to form.

19 A. As I mentioned, under what
20 conditions?

21 BY MR. PLATT:

22 Q. You don't know?

23 A. Yeah, I don't. It depends on
24 the pH. It depends on the protein. It
25 depends on what are you trying to reduce.

1 Q. What about 10 millimolar?

2 MR. HOLMAN: Objection to form.

3 A. Same thing. It depends on
4 the -- what you're trying to reduce.

5 BY MR. PLATT:

6 Q. What's the lowest concentration
7 of DTT that you have seen reliably and
8 measurably break disulfide bonds?

9 A. Yeah, I don't remember.

10 Q. What is the lowest
11 concentration of DTT that you can remember
12 that's reliably and measurably broken
13 disulfide bonds?

14 A. So I did an experiment with a
15 protein. This is a specific set of
16 conditions. This is what -- it's -- every
17 reaction matters.

18 I'm not giving a general
19 answer, but I did reduce a disulfide bond in
20 a protein called EETI-II at pH 7.8 with a
21 reductant, such as DTT, approximately --
22 I don't remember the exact number, but
23 probably 1 millimolar concentration.

24 Q. And is that the lowest
25 concentration of DTT that you recall using

1 that reliably and measurably broke a
2 disulfide bond?

3 MR. HOLMAN: Objection to form.

4 A. I don't remember.

5 BY MR. PLATT:

6 Q. Are you aware of any other
7 situation where a lower amount -- sorry.
8 Strike that.

9 Are you aware of any other
10 experiments where a lower concentration of
11 DTT has been used to break disulfide bonds?

12 A. I mean, I'm sure there are.
13 But like I said, I want to be able to give
14 you specific examples because all the
15 parameters matter. So I don't walk around
16 and memorize what concentration of DTT is the
17 lowest concentration to break one disulfide
18 bond.

19 BY MR. PLATT:

20 Q. Sitting here today, you can't
21 recall an experiment where a lower
22 concentration of less than 1 millimolar of
23 DTT was used to break disulfide bonds? Is
24 that right?

25 MR. HOLMAN: Objection to form.

1 A. I'm sure I've done experiments
2 where I've -- yeah, I can't remember.

3 BY MR. PLATT:

4 Q. What is the -- hold on one
5 second.

6 (Pause.)

7 What is the lowest
8 concentration of TCEP that you can recall
9 being used to break disulfide bonds?

10 MR. HOLMAN: Objection. Form.

11 A. Yeah, the question is way too
12 broad. There are thousands of disulfide
13 bonds. The conditions matter. I can't give
14 you a concentration.

15 BY MR. PLATT:

16 Q. But what's the lowest
17 concentration of TCEP that you've seen break
18 disulfide bonds?

19 A. I don't remember the number.

20 And there's so many different
21 -- I mean, there are thousands of disulfide
22 bonds that even people in my research group
23 have been able to reduce it. And I don't --
24 I can't give you a number.

25 Q. And are you familiar with DTT

1 being used to prevent oxidation?

2 MR. HOLMAN: Objection.

3 Foundation.

4 A. Can you refer to what you mean
5 by "oxidation"?

6 BY MR. PLATT:

7 Q. With respect to enzymes.

8 A. Well, oxidation means a lot of
9 things, so -- and enzymes means a lot of
10 things, so...

11 Q. Are you familiar with --
12 I understand that means a lot of things. So
13 my question would be: How have you become
14 familiar with DTT being used for
15 oxidization -- oxidation? Sorry.

16 MR. HOLMAN: Objection to form.

17 A. From first principles, DTT is a
18 reductant. That means it has electrons.

19 From first principles,
20 oxidation means loss of electrons. So if you
21 have a reducing agent and an oxidation --
22 oxidizing agent, you can offset those two
23 sets of chemistry.

24 Specifically in regard to
25 enzymes, it's a question that means many

1 things that aren't clear based on what you're
2 asking me.

3 BY MR. PLATT:

4 Q. I'm going to show you what's
5 previously been marked as Exhibit 1001.

6 (Previously marked
7 Deposition Exhibit 1001 was
8 tendered to the witness.)

9 Q. And this is U.S. Patent No.
10 7,883,848.

11 You've analyzed the '848
12 patent; correct?

13 A. Correct.

14 Q. And you analyzed it closely; is
15 that right?

16 A. I have.

17 Q. And you've analyzed the
18 specification all the way through to the
19 claims; is that right?

20 A. Yes.

21 Q. So if you could turn to the
22 second-to-the-last page, column 41.

23 A. Okay.

24 Q. The third-to-the-last page
25 maybe, column 41, you'll see claim 1 there?

1 A. I do.

2 Q. Okay. So let's take a look at
3 some of the language of claim 1.

4 Now is it your understanding
5 that interactors are formed by coupling each
6 molecule of interest with at least one
7 nucleic acid moiety?

8 A. (Document[s] reviewed.)

9 So in claim 1(a), it says
10 "forming a plurality of interactors by
11 coupling each molecule of interest with at
12 least one nucleic acid moiety."

13 And if you go to the definition
14 of interactor, which you need to know in
15 order to understand the claim -- so "As used
16 herein" -- this is column 9, line 33 in the
17 patent. "As used herein, the term interactor
18 refers to at least one MOI coupled with at
19 least one NAM."

20 And then the NAM is important.
21 That's part of the interactor part when
22 connected -- coupled to MOI, so...

23 Q. And how are the interactors
24 bound together to form the cis-reactive cell?

25 A. So there's a number of ways in

1 which interactors can end up forming a
2 cis-reactive cell.

3 So in 1(b), it walks through
4 some of the possibilities. "Forming a
5 plurality of cis-reactive cells wherein a
6 cis-reactive cell comprises at least two
7 interactors bound in proximity to one another
8 by an associated oligonucleotide formed from
9 the association [sic] between at least two
10 nucleic acid moieties, wherein the associated
11 oligonucleotide comprises at least two
12 identification elements derived from at least
13 two nucleic acid moieties."

14 Q. So in forming a CRC, are they
15 at least two interactors bound in proximity
16 to one another by an associated
17 oligonucleotide?

18 A. What -- it says "interactors
19 bound in proximity to one another by an
20 associated oligonucleotide formed from the
21 association [sic] between at least two
22 nucleic acid moieties."

23 Q. So in claim 1, what is it that
24 binds the two interactors in proximity to
25 each other?

1 MR. HOLMAN: Objection to form.

2 A. It says that there are at least
3 two interactors bound in proximity to one
4 another by an associated oligonucleotide
5 formed from the association [sic] between two
6 nucleic acid moieties.

7 BY MR. PLATT:

8 Q. And is it your understanding
9 that that's a requirement of the claim?

10 Or do you think that
11 interactors can be bound in proximity to one
12 another through some alternative way?

13 MR. HOLMAN: Objection to form.

14 A. So I'm not a lawyer. But
15 reading the claim as written, saying two
16 interactors bound in proximity to one another
17 by an associated oligonucleotide formed from
18 the associated oligonucleotide between at
19 least two nucleic acid moieties.

20 BY MR. PLATT:

21 Q. And what is your understanding
22 of how that would work?

23 A. So it's detailed in one of the
24 figures. Go to Figure 1.

25 So in -- Figure 1 is walking

1 through essentially the library morphology
2 with molecules of interest associated with a
3 defined NAM -- so nucleic acid moiety.

4 And the library's allowed to
5 interact by affinity through the
6 co-localization of MOI and the respective
7 NAMs. That's, for instance, in B.

8 C is a different topology where
9 they can interact. So "The association
10 function of the NAM is activated. And NAM is
11 brought into proximity by affinity between
12 their respective MOIs and are associated to
13 for a novel associated oligonucleotide
14 comprising nucleic acid from both of the
15 originating NAMs." So you can see that being
16 diagrammed in C.

17 And then in D is "Selective
18 amplification of the different NAM
19 combinations. NAMs which are not associated
20 will not yield amplification products." So
21 this is a different way in which they can
22 associate, as shown in D.

23 Q. So you relied on Figure 1. And
24 it sounds like you were reading from the
25 description of Figure 1 in column 6 on

1 page 54 as part of your opinions that you
2 rendered in this case.

3 A. There was also a Figure 2,
4 which is important too.

5 Q. And why did you rely on
6 Figure 1?

7 A. It's just one way, as it
8 describes a description of an embodiment for
9 interaction detection.

10 Q. And you understand that claim 1
11 relates to functional interactions and not
12 affinity interactions?

13 A. It does.

14 So in Figure 2, it goes through
15 functional interactions.

16 Q. So here, back to my original
17 question, which is: Are you aware of
18 cis-reactive cells within the context of
19 claim 1 being formed by two interactors that
20 are bound in proximity to one another by
21 something other than an associated
22 oligonucleotide formed from the association
23 between at least two nucleic acid moieties?

24 MR. HOLMAN: Objection to form.

25 A. (Document[s] reviewed.)

1 Can you ask me the question one
2 more time?

3 BY MR. PLATT:

4 Q. Sure.

5 Are you aware of cis-reactive
6 cells, again, within the context of claim 1,
7 being formed by two interactors that are
8 bound in proximity to one another by
9 something other than an associated
10 oligonucleotide formed from the association
11 between these two nucleic acid moieties?

12 MR. HOLMAN: Objection. Form.

13 A. So I know in Figure 5, they
14 talk through -- the '848 patent describes
15 many possibilities of forming CRCs. We
16 walked through them.

17 BY MR. PLATT:

18 Q. In Figure 5 -- and this is
19 described on column 7 -- is nonlimiting
20 examples of association of NAMs. Is that
21 right?

22 A. Yeah. I need to look at --
23 I lost my place.

24 (Document[s] reviewed.)

25 Q. As we look at this, it might be

1 easiest logistically here if you can keep
2 claim 1 handy --

3 A. Okay.

4 Q. -- as we talk through Figure 5.

5 A. All right.

6 Q. And you'll see that claim 1
7 refers to -- it says "the association between
8 at least two nucleic acid moieties." Right?

9 A. It says that, yes.

10 Q. And is that what Figure 5 is
11 depicting?

12 A. Yeah, let me double-check.
13 Before association, after association.

14 (Document[s] reviewed.)

15 So Figure 5, nonlimiting
16 examples of association of NAMs. So if
17 arrows indicate nucleic acid 3 prime ends,
18 Figure 5(a) through (d) illustrate ligation
19 approaches.

20 The structure of 5(e) can be
21 associated with either ligation or
22 polymerization, with a polymerase comprising
23 of 5 prime exonuclease activity.

24 And 5(f) can be associated with
25 polymerization. 5(g) illustrates association

1 by introduction of a functional group which
2 reacts to create the native nucleic acid.

3 Reactive functions of 5(h) are
4 used which associate the nucleic acids by a
5 link separate from the nucleic acid.

6 So in Figure 5, there was a
7 chemical reaction that's bringing these two
8 interactors together.

9 Q. So you believe the depiction in
10 5(g) and 5(h) depict an associated
11 oligonucleotide formed from the association
12 between at least two nucleic acid moieties?

13 MR. HOLMAN: Objection. Form.

14 A. Well, I have two interactors.
15 Interactors have MOI and a NAM. NAMS have
16 the properties, as defined by the patent.

17 And the two are being brought
18 together in this depiction by a chemical
19 reaction.

20 BY MR. PLATT:

21 Q. And you consider that to be an
22 association between these two nucleic acid
23 moieties?

24 A. There are two nucleic acid
25 moieties there in G. There are two

1 interactors.

2 Q. So one question, to get our
3 bearings straight on claim 1. This
4 association between at least two nucleic acid
5 moieties that takes place prior to step (c),
6 which is subjecting the plurality of
7 cis-reactive cells to conditions which
8 stimulate the desired functional interaction
9 having a detectable trace. Is that right?

10 A. (Document[s] reviewed.)

11 Step (c), that's what it says.

12 Q. And the association between at
13 least two nucleic acid moieties that take
14 place prior to subjecting the plurality of
15 cis-reactive cells to conditions which
16 stimulate a desired functional interaction
17 having a detectable trace; right?

18 A. So (b) comes before (c).

19 And (b) does discuss forming
20 the plurality of CRCs that has at least two
21 interactors.

22 And then (c) is subjecting to
23 conditions which stimulate the desired
24 functional interaction having a detectable
25 trace.

1 Q. So looking back at Figure 5 and
2 the discussion of Figure 5 on column 7. One
3 example of association of NAMs is using
4 ligation; correct?

5 A. Nonlimiting examples of
6 association by NAMs, arrows indicate nucleic
7 acid 3 prime -- that's right -- can be
8 associated either ligation or polymerization.

9 And 5(g) illustrates
10 association by introduction of a functional
11 group which reacts to create a native nucleic
12 acid.

13 So ligation.

14 Q. And so ligation is one of the
15 ways of associating the NAMs; correct?

16 MR. HOLMAN: Objection. Form.

17 A. So in this figure, they walk
18 through using a chemical reaction to bring
19 two interactors together.

20 BY MR. PLATT:

21 Q. Is that 5(g) and 5(h)?

22 A. 5(g). Let me double-check what
23 5(h) is.

24 (Document[s] reviewed.)

25 So 5(h), as written in the

1 patent, reactive functions are used which
2 associate the nucleic acids by a link
3 separate from nucleic acid.

4 Q. Looking at Figure 5,
5 Figure 5(a) through (d) show association via
6 ligation; correct?

7 A. (Document[s] reviewed.)

8 Well, they have specifically
9 written an (a), (b), (c), (d), (e), (f).
10 They actually have ligation written or
11 ligation or polymerization.

12 Q. And that's in 5(e); right?

13 A. 5(e), ligation or
14 polymerization. (f) is polymerization.

15 Q. So one example of association
16 of NAMs is through ligation in Figures 5(a)
17 through (e). Is that fair?

18 A. As written in the figure.

19 Q. And then Figures 5(e) and 5(f)
20 show association with polymerization;
21 correct?

22 A. (E) is ligation or
23 polymerization; and (f), specifically
24 polymerization.

25 Q. In claim 1, can the ligation

1 take place after the plurality of
2 cis-reactive cells are subjected to
3 conditions which stimulate a desired
4 functional interaction having a detectable
5 trace?

6 MR. HOLMAN: Objection. Form.

7 A. So what section of claim 1 did
8 you just read?

9 BY MR. PLATT:

10 Q. I was looking at step (c).

11 A. (c).

12 (Document[s] reviewed.)

13 Q. My question is: Does
14 association between at least two nucleic acid
15 moieties using ligation need to take place
16 prior to step (c)?

17 MR. HOLMAN: Objection. Form.

18 Scope.

19 A. (Document[s] reviewed.)

20 So from my understanding of the
21 claims, the steps as outlined are followed.
22 And a ligation step is referenced later in
23 the claim set.

24 It says -- so claim 16 says a
25 method of detecting functional interactions

1 between at least two molecules of interest as
2 recited in claim 1, wherein the association
3 is via ligation.

4 BY MR. PLATT:

5 Q. And that association via
6 ligation, that's what's -- that's the
7 association that's being referred to in
8 step (b) of claim 1? Is that right?

9 A. (Document[s] reviewed.)

10 So, again, I'm not a patent
11 attorney, but it looks like it could be. I'm
12 not entirely sure. But it does talk about
13 association in step (b).

14 And then in claim 16, detecting
15 functional interactions between at least two
16 molecules of interest wherein association is
17 via ligation, according to claim 1.

18 Q. So that's -- when claim 16 is
19 discussing the association via ligation,
20 that's the association that's discussed in
21 step (b) of claim 1 -- or further narrowing
22 that association? Is that right?

23 MR. HOLMAN: Objection. Form.

24 A. Well, claim 1 is -- means many
25 different types of associations. And one

1 type of association can be ligation.

2 BY MR. PLATT:

3 Q. And that would take place in
4 step (b); correct?

5 A. (Document[s] reviewed.)

6 So in column 17 of the patent,
7 line 25, "Association may also be
8 accomplished via chemical rather than
9 enzymatic ligation.

10 "Chemical ligation may be
11 performed by providing nucleic acids with the
12 reactive groups on the ends which allow
13 association by a chemical reaction.

14 "The chemical reaction may
15 result in a nucleic acid made of native
16 composition which can serve as, for example,
17 the polymerization substrate or nucleic acid
18 derivative which does not serve as a
19 polymerization substrate. Two illustrative
20 approaches examples for chemical association
21 are illustrated in 5(g) and (h)"-- which is
22 what we're talking about.

23 "Other nonlimiting examples of
24 NAM association approaches which could be
25 used are gap fill polymerization and

1 subsequent ligation and gap ligation as
2 described by Lizardi (1998) or invader
3 cleavage, both of which are fully
4 incorporated herein by reference. In
5 specific embodiments, the nucleic acid is DNA
6 but other nucleic acids may also be used."

7 So in the association, as
8 described in 17, can be chemical
9 association -- chemical ligation, is what
10 it's referred to as.

11 Q. And then my question was, when
12 we're talking about association via ligation,
13 that step needs to take place prior to
14 step (c) in claim 1; is that right?

15 MR. HOLMAN: Objection. Form.

16 A. I think there's -- make sure
17 we're talking about -- I'm talking about
18 chemical ligation. There's also
19 ligation-based discrimination.

20 Are you talking about chemical
21 ligation?

22 BY MR. PLATT:

23 Q. My question is with respect to
24 the association via ligation described in
25 claim 16, that that association via ligation

1 needs to take place prior to step (c) in
2 claim 1.

3 MR. HOLMAN: Objection. Form.

4 A. (Document[s] reviewed.)

5 No, I don't.

6 MR. HOLMAN: Counsel, we've
7 been going for over an hour now. Are
8 you ready to take a break?

9 MR. PLATT: Yeah. Let me just
10 ask one question on that.

11 BY MR. PLATT:

12 Q. What association in claim 16 do
13 you believe it is referring to in claim 1?

14 MR. HOLMAN: Objection. Form.

15 A. (Document[s] reviewed.)

16 BY MR. PLATT:

17 Q. Let me just rephrase that.

18 What association in claim 1 do
19 you believe claim 16 is referring to when it
20 uses the word "association"?

21 A. So the word "ligation" --
22 "chemical ligation" is important. I was
23 referring to chemical ligation.

24 And 16, it's a ligation after
25 carrying out all the steps of claim 1, which

1 is (a) through (e), to detect functional
2 interactions between two molecules of
3 interest.

4 Q. So you read claim 16 to be an
5 additional step?

6 A. Yes. It's a method of
7 detecting functional interactions between two
8 molecules, yeah, of interest as recited by
9 claim 1.

10 Claim 1 is all steps (a)
11 through (e), wherein association is via
12 ligation.

13 Q. So you read claim 16 to be an
14 additional step after steps 1(a) through (e)?

15 MR. HOLMAN: Objection. Form.

16 A. I read it as being a dependent
17 claim of claim 1.

18 BY MR. PLATT:

19 Q. You don't know if it needs to
20 take place after?

21 MR. HOLMAN: Objection to form.

22 A. There's a lot of different
23 possibilities in this patent. So I would
24 carry out these steps and then do claim 16.

25 MR. PLATT: Okay. We can take

1 a break.

2 (Recess: 11:26 a.m. to

3 11:38 a.m.)

4 BY MR. PLATT:

5 Q. So if you could turn back to
6 Exhibit 1001, the patent. And we'll go to
7 column 44, claim 17. And you'll see this one
8 says "The method of detecting functional
9 interactions between at least two molecules
10 of interest as recited in claim 1, wherein
11 association is via polymerization."

12 A. I do.

13 Q. Is that the association that's
14 taking place in step (b) of claim 1?

15 A. (Document[s] reviewed.)

16 So there's two parts in this
17 claim which actually talks about detection of
18 functional interactions between two molecules
19 of interest wherein an association is via
20 polymerization.

21 So in step (b), this is the
22 detection step.

23 Q. Were you done with your answer?

24 A. Let me look at this for a
25 minute.

1 (Document[s] reviewed.)

2 Can you ask me the question
3 again?

4 Q. Sure.

5 The association via
6 polymerization referenced in claim 17, that
7 is referring to the association taking place
8 in step (b) of claim 1; is that right?

9 A. (Document[s] reviewed.)

10 So in the summary of invention
11 in column 5 -- I actually walked through
12 this. It says, on line 40, "One embodiment
13 is directed to a method of detecting an
14 affinity interaction between at least two
15 molecules of interest.

16 "The method comprises forming
17 plurality of interactors by coupling each
18 molecule of interest with at least one
19 nucleic acid moiety comprising of an
20 identification sequence element and an
21 association element; promoting an association
22 between at least two nucleic acid moieties to
23 form a plurality of unique associated
24 oligonucleotides; promoting an association
25 between at least two nucleic acid" -- I'm

1 sorry. I'll reread that.

2 So (b), "Promoting an
3 association between at least two nucleic acid
4 moieties to form a plurality of unique
5 associations of oligonucleotides, wherein
6 each nucleic acid moiety may form more than
7 one unique associated oligonucleotide, and
8 wherein each unique associated
9 oligonucleotide comprises of at least two
10 identification sequence elements derived from
11 at least two nucleic acid moieties."

12 And then "Selecting the
13 plurality of associated oligonucleotides; and
14 (d) subjecting the selected associated
15 oligonucleotides to an analysis that permits
16 detection of at least two identification
17 sequence elements."

18 So the association appears to
19 be, according to claim 1, the association as
20 described in claim 1(b).

21 Q. So why don't we turn back and
22 look at Figure 5 again.

23 A. Okay.

24 Q. You'll see on the top, it
25 says -- sorry -- the left-hand side is Before

1 Association.

2 Do you see that?

3 A. Yes.

4 Q. And then on the right-hand
5 side, it has After Association.

6 Do you see that?

7 A. I do.

8 Q. So what do the circles
9 represent in Figure 5?

10 A. So from my understanding,
11 there's the molecules of interest.

12 Q. And the lines with the arrows
13 on them, those represent single-stranded
14 nucleic acid?

15 A. They're NAMs.

16 Q. And you understand the NAMs to
17 be single-stranded nucleic acid?

18 A. Yes.

19 Q. And looking at Figure 5(a), is
20 that a single-stranded nucleic acid shown on
21 the top that's hybridizing with two NAMs
22 shown in Figure 5(a)?

23 A. So there is a -- what's
24 referred to as an association element, which
25 is hybridizing to the two NAMs.

1 Q. And if we look at Figure 5(f),
2 the one that says "polymerization" in the
3 middle --

4 A. Yes.

5 Q. -- what is shown in the Before
6 Association side?

7 A. It's showing that the
8 interactors, a region of the single-stranded
9 DNA is hybridized.

10 Q. And then what takes place with
11 the polymerization?

12 A. End up elongating the remaining
13 single-stranded pieces of the NAMs.

14 Q. And looking at the left-hand
15 side, Before Association, of Figure 5(f), the
16 left-hand NAM has an identification sequence
17 on it; correct?

18 A. The definition of the NAMs can
19 have identification sequence.

20 Q. And the right-hand NAM shown on
21 the left-hand side of Figure 4, that also has
22 an identification sequence on it too;
23 correct?

24 A. You're still on Figure 5?

25 Q. Yes, I am.

1 A. And you said -- you're talking
2 about Figure 5(f)?

3 Q. Correct.

4 A. Ask me again, please.

5 Q. So the left-hand NAM has an
6 identification sequence; right?

7 A. Correct.

8 Q. And the right-hand NAM has
9 another identification sequence; right?

10 A. It still has an identification
11 sequence.

12 Q. And so when you go to the
13 right-hand side, After Association, do each
14 of those strands have both of the
15 identification sequences?

16 MR. HOLMAN: Objection. Form.

17 A. I have to double-check and make
18 sure, as described in the specification. But
19 let's see how -- let me just double-check how
20 these are defined.

21 Nonlimiting examples of
22 association of NAMs. So these are NAMs. And
23 according to the patent, NAMs have
24 identification sequences.

25 So before and after

1 polymerization, they still have their
2 identification sequence.

3 BY MR. PLATT:

4 Q. So you see on the right-hand
5 side of Figure 5(f), there's now a
6 double-stranded nucleic acid between the two
7 molecules of interest? Is that right?

8 A. I do.

9 Q. And do each of those strands
10 include both identification sequences?

11 A. If they had NAMs, as defined by
12 the patent, they will have identification
13 sequences.

14 Q. And it's the polymerization
15 that takes place that encodes the second
16 identification sequence onto the other
17 nucleic acid which is being polymerized?

18 A. In this CRC.

19 Q. Is that right?

20 A. Yes.

21 Q. Now when you look at -- let me
22 look at -- when you look at Figure 8,
23 Figure 8 shows an association element; is
24 that right?

25 A. So there's an association

1 element in the middle of the construct as
2 drawn in Figure 8, yes. It's labeled
3 Association Element.

4 Q. And what is labeled Tag Motif,
5 that is also referred to as the
6 identification sequence in the '848 patent;
7 is that right?

8 A. (Document[s] reviewed.)
9 I don't remember where they
10 defined tag motif.

11 Q. All right. Let's go back to
12 claim 1 --

13 A. Okay.

14 Q. -- column 41 of Exhibit 1001.

15 A. Okay.

16 Q. And you'll see at the end of
17 step (b), it says "wherein the associated
18 oligonucleotide comprises at least two
19 identification elements derived from the at
20 least two nucleic acid moieties."

21 Do you see that?

22 A. I do.

23 Q. Okay. Is it your understanding
24 that the associated oligonucleotide must be
25 double stranded?

1 A. No. Actually if you look at
2 the NAM definition, it can be single
3 stranded.

4 Q. The associated oligonucleotide
5 can be single stranded?

6 A. Let me double-check the
7 definition of a NAM.

8 (Document[s] reviewed.)

9 So nucleic acid moiety is
10 column 10, line 60.

11 Q. Okay.

12 A. "The NAM may comprise
13 single-stranded DNA, double-stranded DNA,
14 single-stranded RNA, double-stranded RNA, or
15 a combinations thereof. It may include any
16 type of modified nucleotides or nucleotide
17 analogues or other types of molecules which
18 can be incorporated into nucleic acids."

19 Q. So when we look at the
20 associated oligonucleotide in claim 1, that
21 can be either single stranded or double
22 stranded?

23 A. According to the definition of
24 a NAM in claim 1, an interactor is an MOI
25 connected to a NAM.

1 Q. So my question was a little bit
2 different.

3 When you look at the last part
4 of claim 1(b), where it says "wherein the
5 associated oligonucleotide comprises at least
6 two identification elements derived from the
7 at least two nucleic acid moieties," is it
8 your view that that associated
9 oligonucleotide must be double stranded?

10 A. Let me look at the definitions.

11 (Document[s] reviewed.)

12 So can you ask me the question
13 again?

14 Q. Sure.

15 So when you look at the last
16 section of step (b) of claim 1, and it refers
17 to the "associated oligonucleotide comprises
18 at least two identification elements derived
19 from the at least two nucleic acid moieties,"
20 that's referring to double-stranded DNA?

21 MR. HOLMAN: Objection. Form.

22 A. It really depends on the case.

23 There's many examples, as
24 pointed out -- or possibilities, as pointed
25 out in the specification about how the

1 associated oligonucleotides can form these
2 CRCs.

3 BY MR. PLATT:

4 Q. And is one of those ways using
5 a single-stranded associated oligonucleotide?

6 A. It depends on what's being --
7 what assay is being ran. It may have
8 components of double-stranded. It may have
9 components of single-stranded.

10 Q. So the associated
11 oligonucleotide referenced in step (b) of
12 claim 1 can be single stranded or double
13 strand, as long as it comprises of at least
14 two identification elements derived from the
15 at least two nucleic acid moieties?

16 A. So when these associate, they
17 may have parts of single and parts of double
18 stranded.

19 Q. Could it also just be a
20 single-stranded associated oligonucleotide?

21 A. I'd have to understand the
22 context for the experiment. For association,
23 you need some level of hybridization.

24 Q. So if you could take a look at
25 column 8, referring to Figure 8. The

1 Figure 8 that we were looking at earlier, it
2 says that's an illustration of a
3 single-stranded associated oligonucleotide.

4 A. (Document[s] reviewed.)

5 I see this figure, yes.

6 Q. And based on looking at that
7 figure and the discussion of Figure 8, do you
8 believe that the associated oligonucleotide
9 referenced in step (b) of claim 1 can be
10 single stranded as long as it comprises at
11 least two identification elements derived
12 from the at least two nucleic acid moieties?

13 A. (Document[s] reviewed.)

14 And so they're doing -- in
15 Figure 8, they're doing a restricted enzyme
16 digest and then hybridizing it.

17 So the figure, "Illustration of
18 a single-stranded associated oligonucleotide
19 or single-stranded amplification product
20 thereof. Two separate single-stranded
21 oligonucleotides comprising of type IIS
22 restriction enzyme recognition sites are
23 hybridized to the general sequence flanking
24 the identification elements.

25 "The restriction enzyme

1 cleavage occur between the identification
2 element and the general sequence.
3 Figure 8(b) illustrates the single-stranded
4 DNA produced by restriction enzyme cleavage
5 with identification elements in the 3 prime
6 to 5 prime."

7 Let me double-check and see
8 exactly what was done here with this
9 restriction enzyme, because sometimes you get
10 hybridization.

11 (Document[s] reviewed.)

12 So if you look on column 29,
13 where they actually describe what's going on
14 in Figure 8, it's discussing using the
15 single-stranded amplification product that
16 may comprise of an association sequence in
17 the middle, flanked by two identification
18 elements, which, in turn, are flanked by a
19 general sequence, Figure 8.

20 "The general sequence may be
21 designed to comprise the recognition sequence
22 for restriction enzyme, more specifically a
23 type IIS restriction, endonuclease, and in a
24 more specific example comprises of a type IIS
25 restriction enzyme, MlyI.

1 "In one specific embodiment of
2 generating a single-stranded nucleic acid
3 comprising identification elements in 3 prime
4 and 5 prime ends, the nucleic acid may be
5 designed so the restriction enzyme cleavage
6 occur right between the general sequence
7 contacted by the hybridizing oligonucleotide
8 and the single-stranded identification
9 element.

10 "In the very specific
11 embodiment, the restriction enzyme comprises
12 MlyI. This results in single-stranded
13 nucleic acid with a general sequence in the
14 middle of the identification."

15 So from my understanding of
16 this, there is double-stranded, and it's
17 getting cleaved and generated a single
18 strand.

19 Q. And this is in the section
20 discussing the detection of associated
21 oligonucleotides using microarray analysis?

22 A. So let's see. Example 4. So
23 I was just reading from column 29. It's
24 walking through example 4. Enhancement for
25 microarray analysis is in this section.

1 So how do you prepare a sample
2 for microarray analysis.

3 Q. And so when you're doing that,
4 you can have -- an associated oligonucleotide
5 could be single stranded?

6 A. So eventually what ends up
7 happening is you turn it into single stranded
8 so that you can actually end up detecting it
9 by microarray.

10 Q. And is that still an associated
11 oligonucleotide as referenced in step (b) of
12 claim 1?

13 MR. HOLMAN: Objection. Form.

14 A. (Document[s] reviewed.)

15 Well, it has the NAM still
16 associated and the MOI.

17 BY MR. PLATT:

18 Q. So the associated oligonucleotide
19 in step (b) of claim 1 can be single stranded?

20 A. So this is -- this figure that
21 we're discussing right now, Figure 8, is
22 talking about how to do the microarray
23 analysis in preparing this sample.

24 So from my understanding of
25 Figure 8, they have a CRC. Now they're

1 processing it to do the microarray analysis.

2 Q. So in that case, it's no longer
3 a CRC because the associated oligonucleotide
4 is single stranded?

5 A. I didn't say that. I don't
6 understand all that cut sites that are being
7 described here.

8 But you're still analyzing a
9 CRC.

10 Q. So the question is: Can you
11 have a single-stranded associated
12 oligonucleotide that comprises at least two
13 identification elements derived from the at
14 least two nucleic acid moieties?

15 MR. HOLMAN: Objection. Form.
16 Scope.

17 A. I mean, as the patent walks
18 through, there's a lot of possibilities.

19 BY MR. PLATT:

20 Q. Do you know or you don't know?

21 A. I mean, parts of the CRC can
22 be -- can have single-stranded DNA.

23 Q. My question is with respect to
24 the associated oligonucleotide here. It's
25 referenced in step (b). And it says

1 "associated oligonucleotide comprises of at
2 least two identification elements derived
3 from the at least two nucleic acid moieties."

4 And my question is: Can that
5 associated oligonucleotide be single stranded
6 or does it have to be double stranded or can
7 it be either one?

8 MR. HOLMAN: Objection. Form.

9 A. So you can have both double
10 strand and single stranded.

11 BY MR. PLATT:

12 Q. At the same time?

13 A. Yes.

14 Q. And does it cease becoming an
15 associated oligonucleotide when you perform
16 the microarray analysis?

17 A. Ask me again.

18 Q. Sure.

19 Does the associated
20 oligonucleotide stop being an associated
21 oligonucleotide when you conduct the
22 microarray analysis?

23 MR. HOLMAN: Objection. Form.

24 A. It depends on how the
25 microarray analysis is done.

1 BY MR. PLATT:

2 Q. With respect to example 4 in
3 the '848 patent, does the associated
4 oligonucleotide stop being an associated
5 oligonucleotide when microarray analysis is
6 performed?

7 A. It's still an associated
8 oligonucleotide.

9 Q. Is that because an associated
10 oligonucleotide can be single stranded as
11 long as it comprises at least two
12 identification elements derived from the at
13 least two nucleic acid moieties?

14 MR. HOLMAN: Objection. Form.

15 A. So the molecules that are being
16 selected for will still have those NAM
17 properties.

18 BY MR. PLATT:

19 Q. And so an associated
20 oligonucleotide that is single stranded would
21 still be an associated oligonucleotide if
22 it's derived from those NAM properties?

23 A. So by definition, a NAM has an
24 association element and an identification
25 element. And so it's still a NAM. And

1 that's what's being detected -- or part of
2 the microarray.

3 Q. So I understand that. But my
4 question is: When you're looking at step (b)
5 of claim 1, this whole stepwise process that
6 goes through it -- right? -- and it's talking
7 about, you know, forming the cis-reactive
8 cells -- right? -- and then it's talking
9 about subjecting it to this trace and then
10 it's selecting for it and subjecting to it to
11 detection, what my question is: In that
12 step (b), when it's talking about the
13 associated oligonucleotide, is it your
14 opinion that that associated oligonucleotide
15 is double sided -- double stranded or single
16 stranded or can it be either one?

17 MR. HOLMAN: Objection to form.

18 BY MR. PLATT:

19 Q. If you don't know, that's okay
20 too.

21 A. I'm just going to remind myself
22 in the actual examples of what was done in
23 the patent.

24 (Document[s] reviewed.)

25 So from my understanding,

1 they're largely double stranded.

2 Q. But the associated
3 oligonucleotide could also be single
4 stranded?

5 A. In step (b), largely double
6 stranded.

7 Q. So when you're conducting the
8 microarray analysis in example 4, it does an
9 analysis of DNA that is no longer an
10 associated oligonucleotide?

11 MR. HOLMAN: Objection. Form.

12 A. Still associated
13 oligonucleotide.

14 BY MR. PLATT:

15 Q. Even though it's single
16 stranded?

17 A. Parts of it are going to be
18 double stranded. Let me check. Let me read
19 the experiment and make sure I'm correct.

20 (Document[s] reviewed.)

21 Let me make an important point
22 here, which is, step 1(b) is largely double
23 stranded. Step (e) is when we're talking
24 about microarrays and identification of two
25 sequence elements.

1 Are you referring to sequence
2 elements for detection on microarray?

3 Q. So I'm talking about the
4 associated oligonucleotide referenced in
5 step (b).

6 And the question is: Does that
7 associated oligonucleotide need to be double
8 stranded?

9 Does it need to be single
10 stranded?

11 Can it be either one? Or do
12 you not know?

13 MR. HOLMAN: Objection. Form.

14 A. So largely, most cases, it's
15 double stranded.

16 BY MR. PLATT:

17 Q. But in some cases, it can be
18 single stranded. For example, when you're
19 doing microarray analyses.

20 A. Well, that's not step (b).
21 Microarray analysis is not done in step (b).

22 Q. Okay. So then in step (b), the
23 associated oligonucleotide is double
24 stranded?

25 A. For the most part.

1 Q. Why do you say "for the most
2 part"?

3 A. Because a lot of it -- examples
4 in the patent, I don't know if every single
5 example was all double stranded or not.

6 (Pause.)

7 A. Can we grab lunch or keep
8 going?

9 Q. I just had a couple more
10 questions and then we can take a break.

11 A. Okay.

12 Q. But in your take, when given
13 all the examples in the patent and the
14 discussion of single-stranded associated
15 oligonucleotides, are you saying that the
16 associated oligonucleotides could also be
17 single stranded in column 1?

18 A. Let me walk through the
19 examples here.

20 (Document[s] reviewed.)

21 So right in the beginning,
22 example 1 says double-stranded DNA on
23 column 25.

24 And then when I said partially,
25 if you look further down, at some stage, the

1 complete function is sticky end. So that's
2 why I use the word "largely double stranded."

3 Q. I guess, you know, looking back
4 at claim 1, and we were talking about how
5 step (b) also says subjecting the associated
6 oligonucleotides and talking about that being
7 the detection step.

8 Is it your view that the
9 associated oligonucleotides in step (e) are
10 different from the associated
11 oligonucleotides referenced in step (b)?

12 A. Well, they're going to have at
13 least two identification sequence elements.
14 So they're going to have that. That's what
15 it says. But they're actually -- they don't
16 have to be the NAM, nucleic acid moiety.

17 Q. So when we look at step (b)
18 and look at associated oligonucleotides in
19 step (e), that can be single stranded?

20 A. Well, there's a variety of
21 methods that are used to identify the
22 sequence elements. Right?

23 When they do the ligation
24 reaction with the thiols, for instance, they
25 work with double-stranded DNA but still do

1 microarray.

2 I mean, the example we walked
3 through, they started with double stranded
4 and then rejiggered it to get it into a
5 format that would be compatible with the
6 microarray. But it still has a sequence
7 element.

8 Q. And then in that situation, the
9 associated oligonucleotides were single
10 stranded for the microarray analysis?

11 A. Not 100 percent. Part of it
12 became suitable for microarray, which would
13 need to hybridize with an antisense strand.

14 Q. And it would -- and what was --
15 the associated oligonucleotide that was used
16 for that analysis was largely single
17 stranded?

18 A. I don't know about "largely."
19 But there was some work that was done to get
20 it suitable for a hybridization with a
21 microarray.

22 Q. So is it your view that if
23 it's -- that a single-stranded
24 oligonucleotide cannot be an associated
25 oligonucleotide?

1 MR. HOLMAN: Objection --

2 sorry.

3 BY MR. PLATT:

4 Q. Is that your opinion?

5 MR. HOLMAN: Objection to form.

6 A. My opinion is what I said
7 earlier, which is the step (b) in the NAM is
8 largely double-stranded DNA oligonucleotides.

9 BY MR. PLATT:

10 Q. My question is a little bit
11 different, which is: Is it your opinion that
12 a single-stranded oligonucleotide cannot be
13 an associated oligonucleotide within the
14 meaning of claim 1?

15 MR. HOLMAN: Objection to form.

16 A. There's a lot of examples in
17 this patent.

18 (Document[s] reviewed.)

19 So the associated
20 oligonucleotides form the CRCs. And those
21 are largely double stranded pieces of DNA.

22 BY MR. PLATT:

23 Q. But my question is different.
24 I just want to know: Is it your opinion that
25 a single-stranded oligonucleotide cannot be

1 an associated oligonucleotide, as used in
2 claim 1?

3 MR. HOLMAN: Objection to form.

4 A. (Document[s] reviewed.)

5 BY MR. PLATT:

6 Q. And so -- and while you're
7 thinking about that, let me just ask a
8 different question, which is: Looking at
9 Figure 8, it refers to an illustration of a
10 single-stranded associated oligonucleotide.
11 This is column 8, Figure 17.

12 So my question for you is: Can
13 you have a single-stranded associated
14 oligonucleotide?

15 A. I guess from my understanding,
16 after reading Figure 8 and actual
17 experimental description, this is not for
18 step (b).

19 Q. Whether it's step (b) or
20 step (d).

21 A. So I need to look to see if
22 there's an example of a single-stranded CRC
23 that's being formed in step (b), which I'm
24 not aware of, so...

25 Q. Let's focus on step (d), the

1 reference to associated oligonucleotide

2 -- I'm sorry -- step (d). Strike that.

3 So looking at -- step (e)

4 references the associated oligonucleotides.

5 The patent in column 8 discussing Figure 8

6 discusses an illustration of a

7 single-stranded associated oligonucleotide.

8 And my question is: Can an

9 associated oligonucleotide be single

10 stranded? Or is the patent wrong?

11 MR. HOLMAN: Objection to form.

12 A. Associated -- as defined in

13 (b)?

14 Where's the definition of

15 "associated oligonucleotide" in the patent?

16 Let me make sure I understand the definition.

17 (Document[s] reviewed.)

18 So it defines, in column 9,

19 page 16, line 45, as used herein, the term

20 "association of NAM" -- so let's think about

21 what this means -- refers to "an association

22 between the NAM components of interactors,

23 which results in a molecule containing

24 nucleic acid sequences originating from

25 associated NAM molecules.

1 "This combination of NAMs is
2 referred to herein as 'associated
3 oligonucleotide,' and, in specific
4 embodiments, comprises the identification
5 sequences for two component NAMs. The
6 association may be covalent or noncovalent
7 and occurs between at least two NAMs. In
8 some embodiments, the association occurs
9 pairwise between NAMs."

10 So it doesn't say whether or
11 not it can be single- or double-stranded DNA.

12 MR. PLATT: Okay. Why don't we
13 take lunch.

14 (Recess: 12:37 p.m. to
15 1:08 p.m.)

16 BY MR. PLATT:

17 Q. So, Dr. Pentelute, if you could
18 look back at Exhibit 1001 again, the '848
19 patent.

20 A. Okay.

21 Q. We'll go to claim 1 again. And
22 so one thing we've -- we've talked about
23 molecules of interest.

24 What are molecules of interest?

25 A. Let's go back to the definition

1 here. So it's column 8, close to line 55.
2 "As used herein, the term 'molecule of
3 interest' refers to a molecule about which
4 interactive information is desired. A MOI is
5 coupled with at least one NAM.

6 "In one embodiment, each MOI is
7 coupled to a single unique NAM. Examples of
8 suitable MOIs include, but are not limited
9 to, whole proteins, protein domains,
10 peptides, peptoids, functional enzymes, low
11 molecular weight compounds, polymers,
12 non-proteinaceous cellular products, virus
13 particles, cells, metabolites, lipids,
14 carbohydrates, nucleic acids, inorganic
15 compounds, or combinations thereof."

16 So that's what's being listed
17 as MOIs in the '848 patent.

18 Q. And it says "a molecule about
19 which interactive information is desired."

20 What is "interactive
21 information"?

22 A. So as the context of what's
23 described in the '848 patent, it's about
24 molecules in some way or another interacting
25 with each other.

1 Q. And what is the information
2 that is desired with respect to molecules of
3 interest?

4 A. So the information that you
5 desire, as discussed in this patent, there
6 are many, many, many aspects that you desire
7 information on: functional interactions,
8 chemical reactions, enzyme small-molecule
9 interactions.

10 Those are the -- some of the
11 examples.

12 Q. Now in that -- strike that.

13 And you mentioned functional
14 interactions. That's also used in claim 1.

15 What is a functional
16 interaction?

17 A. It's also defined in the patent
18 too, if I remember correctly.

19 (Document[s] reviewed.)

20 Okay. So on 15, line 10,
21 Functional Interactions. "Another embodiment
22 of the present invention is directed to a
23 method of detecting functional interactions
24 between at least two molecules of interest.

25 "The method comprises of

1 forming a plurality of interactors by
2 coupling each molecule of interest with a
3 nucleic acid moiety, the nucleic acid moiety
4 comprising an identification sequence element
5 and an association element, wherein the
6 affinity exists between the nucleic acid
7 moieties;

8 "Forming the plurality of
9 cis-reactive cells wherein the cis-reactive
10 cell comprises at least two interactor bound
11 in proximity to one another by an associated
12 oligonucleotide formed from affinity between
13 at least two nucleic acid moieties, wherein
14 the associated oligonucleotide comprises of
15 at least two identification elements derived
16 from at least two nucleic acid moieties;

17 "Subjecting the plurality of
18 cis-reactive cells to conditions which
19 stimulate a desired functional interaction
20 having a detectable trace."

21 Selecting all cis-reactive
22 cells and subjecting them -- the associated
23 oligonucleotide from the cis-reactive cells
24 -- to analysis that permits detection of at
25 least the two identification sequencing.

1 So then it says "Functional
2 interactions are defined as interactions
3 between molecules where the nature of at
4 least one of the partners which participate
5 in the interaction deviates from the nature
6 of the partner where the other participates
7 [sic] of the functional interaction is/are
8 present in conditions permitting the
9 functional interaction."

10 They may include affinity
11 interactions. And then there's a list of
12 what the functional interactions are included
13 in this specification.

14 Q. Now if you could look at
15 column 5 --

16 A. Yep.

17 Q. -- around line 14 or so.

18 And would you agree with me
19 that the information desired about the
20 functional interaction is information not
21 previously known?

22 MR. HOLMAN: Objection. Form.

23 A. So I'm reading column 5,
24 line 15?

25 ///

1 BY MR. PLATT:

2 Q. Correct.

3 A. So it says "There is a need in
4 the art for methods capable of investigating
5 affinity and/or functional interactions
6 within libraries of molecules wherein such
7 interactions are not previously established
8 to exist, and methods for quantification of
9 any detected interactions."

10 So that's what this says.

11 Q. So in terms of detecting
12 functional interactions, what you're looking
13 for is information about that interaction
14 that's not previously established or exists?

15 MR. HOLMAN: Objection to form.

16 A. I mean, there's a lot of
17 reasons why you want to detect functional
18 interactions. It could be that they're not
19 yet known. It could be that they're known
20 and you want to understand it in that
21 context.

22 Every assay has a certain set
23 of conditions to where you want to measure a
24 functional interaction, even though it might
25 be known.

1 So I wouldn't say -- I would
2 say that generally you need to be able to
3 measure functional interactions.

4 BY MR. PLATT:

5 Q. My question is within the
6 context of claim 1 of the '848 patent.

7 Are you saying that it's a
8 method of detecting functional interactions
9 where those interactions are already
10 previously established to exist?

11 A. Well, it's a method that works
12 generally that can detect interactions; and
13 it can quantify them based on these CRCs.

14 Q. And my question is: Is it a
15 method of detecting functional interactions
16 where such interactions are not previously
17 established to exist?

18 MR. HOLMAN: Objection. Form.

19 A. It's a broad technology. It
20 can detect any interaction using this format.

21 BY MR. PLATT:

22 Q. So that would -- in your view,
23 that would include methods that utilize
24 molecules with predefined affinities?

25 MR. HOLMAN: Objection.

1 Foundation.

2 A. So this claim 1 can detect many
3 types of interactions. Interactions can be
4 quite broad, as described in the patent.

5 BY MR. PLATT:

6 Q. And you would include
7 interactions based on predefined affinities?

8 MR. HOLMAN: Objection.

9 Foundation.

10 A. As I've already said, it's a
11 broad platform. It can measure many types of
12 interactions.

13 BY MR. PLATT:

14 Q. Including those that are known.
15 Is that your position?

16 A. It's applicable to be able to
17 measure many interactions.

18 Q. Including those that are known?

19 A. I mean, as shown in this
20 patent, they show that it can measure many
21 types of interactions. "Platform" is quite
22 general.

23 Q. So is that a "yes," you would
24 agree that it -- that the claimed method in
25 your view can utilize molecules with

1 predefined affinities?

2 MR. HOLMAN: Objection. Form.

3 Foundation.

4 A. So let me look through the
5 examples and see if they help.

6 (Document[s] reviewed.)

7 So if you read in column 19,
8 page -- around 60, and it's going through --
9 again, continuing on, the functional
10 interactions. And it's walking through the
11 other possibilities of selecting for positive
12 CRCs in the case of interactions which
13 includes addition of a group by one member of
14 the interaction to another member, such as a
15 kinase or glycation [sic] interaction between
16 an enzyme and a substrate may be accomplished
17 by affinity purification of the CRCs with
18 respect to the added group.

19 "In the specific example of a
20 kinase interaction, the library CRCs can be
21 affinity purified with phosphate-specific
22 antibody. The specificity can be controlled
23 to a phosphorylated amino acid, peptide
24 motif, or a whole protein. CRCs which do not
25 contain the modification to be discarded."

1 So there are many, many
2 possibilities. If you read further down,
3 line 10 on column 20, as nonlimiting
4 examples, "...this type of selection would be
5 suitable for the case of phosphatase
6 activity, glycosylation/deglycosylation,
7 ubiquitinylation/deubiquitinylation, and for
8 any other interactions which serves to add or
9 remove molecules as part of the interaction."

10 And the list goes on. There's
11 a lot of applications of this technology of
12 measuring functional interactions.

13 Q. So you mentioned in reading
14 that affinity purification.

15 A. Yes.

16 Q. What is that?

17 A. They're probably defining it.

18 Let's make sure we're following what the
19 patent says.

20 Affinity interaction --

21 Q. Not affinity interaction,
22 affinity purification.

23 A. Okay.

24 Q. I was just wondering what that
25 is.

1 A. (Document[s] reviewed.)

2 So it doesn't specifically
3 define "affinity purification."

4 But, generally speaking,
5 there's a lot of different tools for
6 purifying by affinity. One example could be
7 using a molecule that would selectively bind
8 or have affinity to a particular cell, like a
9 resin, an agarose resin. And that way, you
10 could actually have that binding interaction
11 enabling you to purify your sample.

12 Q. So I think we were looking at
13 your other patent -- well, I think we are
14 looking at one of your patents. But one
15 patent we looked at -- there are a lot out
16 there. Thank you for your contribution to
17 our society, as the patent lawyers say that.

18 But in your patent, do you
19 recall you were talking about using biotin?

20 A. Yep.

21 Q. Is that an example of affinity
22 purification, using biotin with streptavidin?

23 A. That would be a capture step,
24 is what I would refer to it in the case of my
25 patent.

1 Q. Can that also be used for
2 the -- you know, biotin and streptavidin used
3 for affinity purification?

4 A. So the biotin-streptavidin
5 complex is driven by affinity. And you do
6 capture it because of the high affinity.

7 And the molecules that have
8 biotin will be captured to the resin, and the
9 other molecules that don't have biotin will
10 not be captured.

11 So there is an enrichment step
12 or purification step.

13 Q. And there's different examples
14 in the '848 patent about that affinity
15 purification. I think you were walking
16 through some of those.

17 A. Well, they describe how a
18 person skilled in the art could do this.

19 We need to walk through the --
20 I think the examples and make sure that they
21 do do that.

22 (Document[s] reviewed.)

23 I'm looking at example 1 on
24 column 25.

25 (Document[s] reviewed.)

1 So there is a biotin capture
2 step in example 1.

3 Q. So going back, we were talking
4 about functional interactions on column 15.
5 And there was -- you had read that section
6 about -- you know, it starts at about
7 line 30. I won't read it --

8 A. Okay.

9 Q. -- but it refers there to the
10 nature of at least one of the partners which
11 participates in the interaction, deviates
12 from the nature of the partner when the other
13 participants of the functional interaction
14 is/are present and conditions permitting the
15 functional interaction.

16 And what is that? What is that
17 talking about?

18 A. (Document[s] reviewed.)

19 Q. Like, does there need to be
20 some change to at least one of the molecules
21 of interest in a functional interaction?

22 MR. HOLMAN: Objection. Form.

23 A. So the way I view this is
24 according to how claim 1 is written, which is
25 that you have two MOIs connected to NAMs.

1 Right? And you have them come together. And
2 if there's a functional interaction, you'll
3 form this complex, this cis-reactive cell.

4 And that's where saying the
5 interaction deviates because this whole
6 complex that's formed is the deviation.
7 That's the way I understand this.

8 And it says from the nature of
9 that partner when the other participants of
10 the functional interaction is or are present.

11 So it's really about forming
12 the CRC.

13 BY MR. PLATT:

14 Q. Turn back to claim 1.

15 A. Claim 9?

16 Q. Claim 1.

17 A. Claim 1. Okay.

18 Q. You'll see in step (d), it says
19 "selecting all cis-reactive cells exhibiting
20 the detectable trace."

21 Do you see that?

22 A. I do.

23 Q. Why is the selection step done?

24 A. Why is it there?

25 Can you repeat the question?

1 Q. Yeah. Why do you need a
2 selection step in this process in claim 1?

3 MR. HOLMAN: Objection. Form.

4 A. So if you think about what's
5 being done here, you're doing a MOI-NAM,
6 MOI-NAM.

7 So you have at least two
8 different MOIs and NAMs. Okay. But you have
9 many more than that. So you have X number
10 and Y number. And mix everything together.
11 So let's refer to it as the combinatorial
12 nature of this patent.

13 Now if you were just to skip
14 the detectable trace and selecting, you would
15 have very high background. You wouldn't be
16 able to measure/understand the cis-reactive
17 cells that have that detectable trace.

18 BY MR. PLATT:

19 Q. So if you look at column 19.
20 This is at about line 40 or so. And it's
21 talking about detecting functional
22 interactions in -- is this what you're
23 referring to?

24 A. What line number?

25 Q. This is around line 40.

1 A. 40. Functional Interactions.

2 So I can read this. Let's see.

3 (Document[s] reviewed.)

4 Q. Is that what you're referring
5 to, on the selection part, about it being you
6 want it to -- you have to detect functional
7 interactions in a pool of CRCs? The CRCs
8 which display functional interactions must be
9 selected for detection and/or analysis?

10 A. With a detectable trace, yeah.

11 Q. And is that because prior to
12 that selection step, you're going to have a
13 pool of CRCs, some of which had a functional
14 interaction and some of which didn't?

15 MR. HOLMAN: Objection. Form.

16 A. So that's, you know, a hard to
17 generalize question.

18 It depends on what one of these
19 examples that they're actually probing and
20 investigating with CRCs. This patent lists a
21 lot of different possibilities. Right?

22 So in some instances, that
23 could be part of it.

24 BY MR. PLATT:

25 Q. What do you mean?

1 A. Well, depending on the
2 experiment that's being done, you may have a
3 background. It could be many different
4 things. And the goal is to select the select
5 for the CRCs that are there, but also provide
6 a detectable trace.

7 Q. So you want to go through this
8 process of selecting the CRCs with a
9 detectable trace from the other CRCs that did
10 not have a detectable trace?

11 A. You do need a detectable trace.

12 Q. But the point of the process is
13 to be able to select for the CRCs with a
14 detectable trace from the background CRCs?

15 MR. HOLMAN: Objection. Form.

16 A. Not just background CRCs, but
17 there's also many interactors that are
18 present too.

19 BY MR. PLATT:

20 Q. Okay.

21 A. Yeah. So there are a lot of
22 things in solution that are in that test
23 tube. And you want to be able to get those
24 cis-reactive cells that give you a readout.
25 And the readout is the detectable trace.

1 Q. And that's the process of
2 selection that you want to do?

3 A. Yes.

4 Q. Okay. So now when you look at
5 dependent claim that's number 4, I think --
6 sorry -- dependent claim 2, and you'll see it
7 says "the method of detecting functional
8 interactions between at least two molecules
9 of interest as recited in claim 1, wherein
10 selecting all cis-reactive cells exhibiting
11 the detectable trace is accomplished via
12 selective separation or selective detection
13 of the cis-reactive cells."

14 Do you see that?

15 A. Yes.

16 Q. So selective separation and
17 selected detections are methods of selecting
18 cis-reactive cells that exhibit the
19 detectable trace; is that right?

20 MR. HOLMAN: Objection. Form.

21 A. Let's see. The claims -- "the
22 method of detecting functional interactions
23 between at least two molecules of interest,
24 as recited in claim 1, wherein selecting all
25 cis-reactive cells exhibiting the detectable

1 trace is accomplished via selective
2 separation."

3 That's what it says. Selective
4 separation -- or selective detection of the
5 CRCs.

6 BY MR. PLATT:

7 Q. And are those two different
8 ways of selecting CRCs exhibiting the
9 detectable trace?

10 A. Yes. So it says "In claim 1,
11 wherein selecting all cis-reactive cells
12 exhibiting the detectable trace."

13 Q. And what is "selective
14 separation"?

15 A. So selective separation is a
16 general phrase that enables one to separate
17 everything that's in the mixture in a way
18 that would then provide the selective part,
19 which is -- the molecules of interest are
20 actually captured.

21 So separation is just another
22 word for ultimately you get a purification
23 step.

24 Q. And what about selective
25 detection?

1 A. Yeah, that could just be that.
2 Okay. All these CRCs that now have a
3 detectable trace, you actually have an assay
4 that tells you they're there.

5 Q. And if we look at claim 19.
6 And you'll see this is the back page on the
7 correction, it says that it's actually
8 referring to claim 2 there, not claim 12.

9 A. Changed 12 to 2, column 44.

10 Q. So my question is: You'll see
11 claim 19, it says "the method of detecting
12 functional interactions between at least two
13 molecules of interest as recited in claim 2"?

14 A. Oh, I see.

15 Q. "Wherein the selected detection
16 comprises labeling the cis-reactive cells
17 with an affinity reagent."

18 So what is that referring to?

19 A. So the method of detecting
20 functional interactions between at least two
21 molecules of interest -- so MOIs -- as
22 recited in claim 12 -- or claim 2, which we
23 just went through -- wherein the selective
24 detection comprises labeling cis-reactive
25 cells within an affinity reagent.

1 So that's referring to
2 essentially adding on a what we call affinity
3 handle onto the CRCs.

4 Q. So what is an affinity handle?

5 A. So it's, like -- for instance,
6 a biotin could be an affinity handle.

7 Q. Okay. I'm going to hand you
8 what's previously been marked as
9 Exhibit 1010.

10 (Previously marked
11 Deposition Exhibit 1010 was
12 tendered to the witness.)

13 Q. This is an article published by
14 Kanan.

15 And do you know Matthew Kanan?

16 A. No, I don't.

17 Q. Okay. Because I'll try and
18 figure out how to correctly pronounce his
19 last name.

20 A. "CAN-uhn." We say Cannon.

21 Q. "CAN-uhn." "KAY-nan."

22 And it's -- Exhibit 1010 is a
23 letter to Nature entitled Reaction Discovery
24 Enabled by DNA-Templated Synthesis and
25 In Vitro Selection.

1 And you analyzed Kanan in your
2 analysis for this case; right?

3 A. I did.

4 Q. And if you turn to the second
5 page. And if you look at Figure 1(b).

6 You're familiar with
7 Figure 1(b); correct?

8 A. (Document[s] reviewed.)

9 Yes.

10 Q. And Figure 1(b) is showing --
11 what is your understanding of the pool A and
12 pool B substrates and oligonucleotides shown
13 in Figure 1(a) and (b) of Kanan?

14 A. Okay. So I can walk you
15 through this.

16 Figure 1(a) is describing
17 molecules that are defined as A. These are
18 small molecules. And then they're covalently
19 attached to a piece of DNA. That DNA has a
20 region for annealing to B -- and we'll get to
21 that in a minute, what is B -- and then a
22 coding region for A.

23 Then if you look at pool B,
24 it's small molecule B1, B2, B to M. And then
25 there's a region between B and that sulfur

1 which has a biotin, then a sulfur, another
2 sulfur that connects via disulfide to what's
3 called a coding region for B1 and so on.

4 So each one of these pool B
5 molecules will have a unique coding region.
6 And 1(b) did -- an experiment is being done
7 where pools A and pool B are being mixed
8 together under a certain set of conditions.
9 And then some downstream steps are done to
10 eventually identify a productive chemical
11 reaction between A and B.

12 Q. And if we look at Figure 1(b),
13 that is showing the reaction selection and
14 analysis process in Kanan?

15 A. Yeah, so there's two sets.
16 First step is the reaction. You can see
17 there's -- (b) has now a bond connected to A
18 on the first line of the figure.

19 The second line is -- there's
20 been a reduction step to cleave the disulfide
21 and a washing. So you'll see that
22 essentially there's a piece that's being
23 removed. The black moiety in the middle
24 panel of 1(b) has that molecule A-12 on it,
25 for instance, but all of the other ones are

1 being washed away. And what's being retained
2 are the ones that have biotin.

3 And it's interacting with an
4 avidin that's on a little sphere, which is
5 a -- typically an agonist B. And then
6 there's some downstream steps to actually
7 understand what PCR microarray, what that
8 A -- or that oligonucleotide that was
9 captured actually is.

10 Q. So if you look at the -- after
11 it says, you know, it's -- in the top part of
12 Figure 1(b), it says "Pool A plus pool B."
13 And then it says "One set of reaction
14 conditions," and there's an arrow to the
15 right.

16 A. Correct.

17 Q. And then what is shown in the
18 right is under those reaction conditions, you
19 would then have -- in the plot, you would
20 have non-bond-forming combinations, you would
21 have bond-forming combinations, and then you
22 would have other pool A and pool B members.

23 Is that right?

24 A. So you would definitely have
25 other pool A and other pool B members. You

1 would have all of them. Everything put into
2 the pot.

3 Q. Right.

4 A. And then a subset of those
5 would be the bond-forming combination.
6 Right. Because presumably all other pool A
7 and pool B are going to be able to form the
8 non-bond-forming combination. That's just
9 for an illustration.

10 Q. But if we look at after the
11 reaction, there are two types of combinations
12 that exist in the pot. There's the
13 non-bond-forming combinations and the
14 bond-forming combinations.

15 Is that right?

16 A. For the large part, yes.

17 Q. And then if we go down to the
18 next line, it says "Cleave disulfide."

19 Do you see that?

20 A. I do.

21 Q. And let's talk a little bit
22 about how this cleavage works. So right
23 before the cleavage, there's two sets of
24 combinations. There's the non-bond-forming
25 combinations and the bond-forming

1 combinations that we talked about. Right?

2 A. It's what's written on the
3 figure.

4 Q. And both -- prior to the --
5 cleaving the disulfide, both the
6 non-bond-forming combinations and the
7 bond-forming combinations are linked to
8 biotin. Is that right?

9 A. As part of the complexes that
10 are formed -- they successfully form. Even
11 though there's no bond that's been formed,
12 they still have biotin.

13 Q. Now, if you were to put
14 avidin-linked beads in at this time before
15 cleaving the disulfide bonds, both the
16 non-bond-forming combinations and the
17 bond-forming combinations would be captured.
18 Is that right?

19 MR. HOLMAN: Objection to form.

20 A. So there is -- everything with
21 biotin that's within that pot will be
22 captured if enough streptavidin or avidin
23 beads are added.

24 BY MR. PLATT:

25 Q. Including both non-bond-forming

1 combinations and bond-forming combinations.

2 It's not going to discriminate between the

3 two?

4 A. If biotin is there, it's going
5 to be captured.

6 Q. Whether the biotin's connected
7 to one of these non-bond-forming combinations
8 or connected to the bond-forming
9 combinations?

10 MR. HOLMAN: Objection. Form.

11 A. Yes. And there also -- as
12 shown here, there's other pool A and pool B
13 members that are called out in the figure.
14 So pool B members do have biotin still, but
15 pool A members do not.

16 BY MR. PLATT:

17 Q. So then we're just talking
18 about combinations. The combinations that
19 are going to be captured by the streptavidin
20 would be those combinations that have biotin,
21 whether it's non-bond-forming or
22 bond-forming?

23 A. So biotin will be captured by
24 streptavidin or avidin. So every biotin
25 molecule will be captured that's in the pot.

1 Q. And it's not going to matter if
2 it's non-bond-forming?

3 MR. HOLMAN: Objection to form.

4 BY MR. PLATT:

5 Q. It's still captured; right?

6 MR. HOLMAN: Objection to form.

7 A. So we have to be really
8 careful. It does matter. You're asking me a
9 hypothetical, which is not what was done in
10 this experiment.

11 What was done in this
12 experiment is that after essentially
13 subjecting pool A and pool B together, mixing
14 everything together and then proceeding down
15 with a series of steps, which includes
16 disulfide bond cleavage, then they are
17 capturing the biotin group.

18 That's the experiment that's
19 being outlined here.

20 BY MR. PLATT:

21 Q. Right. I understand that.

22 But I'm just trying to
23 understand what is going to happen with
24 the -- you know, if you didn't do the
25 cleavage, if it hadn't happened yet, the

1 avidin beads are going to end up capturing
2 any of these combinations that have biotin on
3 it, whether it's bond-forming or
4 non-bond-forming.

5 MR. HOLMAN: Objection. Form.

6 A. Well, generally speaking,
7 avidin beads will bind to biotin.

8 BY MR. PLATT:

9 Q. But would non-bond-forming
10 combinations also be captured?

11 MR. HOLMAN: Objection. Form.

12 A. I mean, in this experiment,
13 it's not what was done. So I can't --
14 I mean, biotin does bind streptavidin and
15 avidin. But that's not what they did in this
16 experiment. And there's no evidence to talk
17 about what would the outcome be if they did
18 what you're suggesting.

19 BY MR. PLATT:

20 Q. So you don't know.

21 A. Well, it is an experiment. But
22 I am saying biotin does bind to avidin.

23 Q. Right.

24 So my question for you is: If
25 you put in avidin beads prior to cleaving the

1 disulfide bonds, would non-bond-forming
2 combinations with biotin on it bind to the
3 avidin?

4 MR. HOLMAN: Objection. Form.

5 BY MR. PLATT:

6 Q. If you don't know, that's okay.
7 I mean --

8 A. I mean, the experiment is not
9 done here. I'd have to -- do you have the
10 supporting information? We can look back at
11 the conditions.

12 Q. So I'm going to hand you what's
13 previously been marked as Exhibit 1013.

14 (Previously marked
15 Deposition Exhibit 1013 was
16 tendered to the witness.)

17 A. Thank you.

18 Q. This is the Kanan supplementary
19 information.

20 A. (Document[s] reviewed.)

21 So the question about whether
22 or not everything in the mixture would end up
23 being captured by avidin, so they don't do
24 the experiment. And they don't tell me if
25 they've added enough avidin to be able to

1 capture everything in the solution. So

2 I don't know.

3 Q. Okay.

4 A. Yeah.

5 Q. But in Kanan -- we were talking

6 about the experiments done in Kanan -- that

7 avidin-linked beads are used after the

8 cleavage of the disulfide bond. Correct?

9 A. That's shown in Figure 1(b),

10 yes. Let's be specific. It says

11 "Cleavage [sic] disulfide, select for binding

12 to avidin-linked beads."

13 That's what's written in the

14 figure.

15 Q. And so what is in the -- what's

16 in the pot after the disulfide bonds are

17 cleaved?

18 A. So I have it both in my

19 declaration and we also have it in the

20 supporting information. We can go through

21 what's in the pot.

22 (Document[s] reviewed.)

23 So there's a reference to

24 biotin oligonucleotide. There's a universal

25 clamp oligonucleotide. I'm reading on

1 Exhibit 1013, page 8.

2 The selection has five
3 components of pools A and pools B: universal
4 clamp, biotinylated internal standard, and
5 metal complex for reagents being tested. So
6 then there's DNA that gets added of 168
7 oligonucleotides, 144 additional ones. And
8 then there's a whole series of buffers that
9 end up getting into the pot. Right? So
10 there's MOPS, sodium chloride, pH 7, copper,
11 sodium ascorbate, additional MOPS. There's
12 EDC. There's a number of different things,
13 palladium and palladium (II).

14 That's what's in the pot.

15 Q. So let me narrow it down a
16 little bit.

17 What's in the pot with respect
18 to the pool A and the pool B members?

19 MR. HOLMAN: Objection. Form.

20 A. So pool A members, as I walked
21 through already as described in A -- 1(a),
22 they're oligonucleotides that have annealing
23 region to B, and coding region for A. And
24 then the pool B just has the coding region
25 for B-1. And those have a biotin in pool B

1 to -- whereas pool A does not. And then when
2 you mix them together, they're in a buffer at
3 a particular temperature and a particular
4 concentration.

5 BY MR. PLATT:

6 Q. So when the avidin-linked beads
7 are used, the only material that's captured
8 are going to be the bond-forming combinations
9 and the B substrates that do not contain any
10 nucleic acid? Is that correct?

11 A. Yes. The pool B
12 oligonucleotide will be removed because the
13 disulfide bond cleavage. So you will still
14 capture biotin B-1. And you will capture the
15 constructs that formed the bond between B and
16 A.

17 Q. So when we look at what's
18 captured in these beads in Kanan's selection
19 process, the only nucleic acid that is
20 captured is ones related -- representing the
21 bond-forming combinations?

22 A. Yes. So the only nucleic acid
23 that's captured is pool A, nucleic acids. So
24 one piece of it. Nothing to do with pool B.
25 There's not nucleic acid from pool B being

1 captured. It gets -- it has no biotin.

2 And then everything that didn't
3 form the bond-forming reaction gets washed
4 away.

5 Q. So if we look at Exhibit 1013,
6 the supplementary information. So if you
7 look about a third of the way down, and
8 you'll see it says, "After one hour" --

9 A. On what page?

10 Q. On page 8. Sorry. That would
11 help.

12 So about one-third of the way
13 down, a couple sentences into the last
14 paragraph there. And it says, "After one
15 hour at 25 degrees C, this solution was added
16 to a 20 picomolar-binding capacity aliquot of
17 streptavidin-linked magnetic particles (Roche
18 Biosciences) suspended in 30 microliters of
19 .1 molar sodium chloride, 10 millimolar Tris,
20 1 millimolar EDTA, pH 7.5 (loading buffer)."

21 Based on your experience, was
22 this loading buffer dehybridized DNA?

23 MR. HOLMAN: Objection. Form.

24 A. So let's see. "To the eluant
25 was added 150 microliters of 0.1 molar

1 aqueous solution of Tris phosphine
2 hydrochloride in 1 molar of sodium phosphate,
3 pH 8, to effect disulfide cleavage."

4 Correct. That's what it says.

5 After one hour at 25C, the
6 solution was added. The binding aliquot of
7 streptavidin-linked magnetic particles
8 suspended in 30 microliters of 0.1 sodium
9 chloride and 1 millimolar EDTA. That's the
10 loading buffer.

11 So this will start to
12 essentially break apart pieces of DNA, the
13 duplex DNA. There's no magnesium, for
14 instance. That's required to have a strong
15 duplex formation. And there's not a lot of
16 salt either, sodium chloride.

17 And that has to do with the
18 fact that DNA in the duplex form is
19 negatively charged phosphates binding the
20 negatively charged phosphates. So you have a
21 lot of charge-charge propulsion. So if you
22 don't have the right ions in the solution,
23 the duplex DNA will start to break apart.

24 And the additional parts of
25 this are actually additional steps that will

1 break apart duplex DNA.

2 BY MR. PLATT:

3 Q. Okay. And this section, these
4 paragraphs that we're looking at, this is
5 part of Kanan's selection procedure? Is that
6 right?

7 These paragraphs here,
8 two-thirds of the page down?

9 A. Yep. So starting on page 7, it
10 says "Selection for bond formation." So
11 that's where it starts.

12 But the key part of the
13 selection is the reaction discovery
14 experiments and selection procedure, and
15 that's the section that we're discussing
16 right now.

17 Q. Okay.

18 THE DEPONENT: If we're
19 shifting topics, is there a way to do
20 a bio break?

21 MR. PLATT: Yeah. Yeah. Sure.

22 (Recess: 2:10 p.m. to
23 2:16 p.m.)

24 BY MR. PLATT:

25 Q. I'm going to hand you what's

1 been previously marked as Exhibit 1011.

2 (Previously marked

3 Deposition Exhibit 1011 was

4 tendered to the witness.)

5 Q. This is Patent Application

6 Publication Number U.S. 2004/0014090, also

7 what we referred to as Neri in the IPR

8 petition.

9 Dr. Pentelute, you analyzed the

10 Neri reference; correct?

11 A. Yes.

12 Q. And if you could turn to

13 Figure 5 of the Neri reference. And you'll

14 see on the left-hand side, there's this

15 figure of p and q and the self-assembly

16 sequences. Do you see that?

17 A. I do.

18 Q. You'll recall that Dr. Nelson

19 also included this figure, this portion in

20 Figure 5, in his declaration.

21 Do you recall that?

22 A. I think so.

23 Q. And if we look at sort of the

24 left-hand nucleic acid, the one that has the

25 p attached to it.

1 Do you see that?

2 A. Yes.

3 Q. And can we refer to that as the
4 A strand?

5 A. Okay.

6 Q. And you'll see then to the
7 right-hand side of that, connected to q, is
8 the B strand. Is that right?

9 A. Right.

10 Q. Okay. So if we look at the top
11 part where it is -- those black lines exist.

12 Do you see that?

13 A. I do.

14 Q. What are these ladders that are
15 being depicted there?

16 A. Let's see the patent to find
17 it, before I say anything.

18 (Document[s] reviewed.)

19 So it's mentioning in Figure 5
20 caption that "One method of this EASCHEL
21 decoding: The oligonucleotides of
22 sub-library A bear chemical entities at 3
23 prime end."

24 Towards the 3 prime extremity,
25 the DNA sequence is designed to hybridize to

1 the DNA sequence at the 5 prime extremity of
2 the oligonucleotides of sub-library B.

3 So hybridization is often shown
4 with dashed lines. Is that what you're
5 asking me about, what these dashed lines
6 were?

7 Q. Correct.

8 A. Hybridization.

9 Q. And where were you reading
10 from?

11 A. Figure 5 caption -- it doesn't
12 have a column. Column -- I guess 4, page 4.
13 Is that right? It doesn't have a
14 column labeling number, but it's page 4 of
15 18, either way you look at it.

16 Q. Paragraph 29?

17 A. I don't have labeling on
18 paragraphs on my -- is that correct?

19 Q. Is that on page 4? The number
20 is on the front of the paragraph.

21 A. Oh. Paragraph 29. Oh, yeah,
22 thanks. Okay. It's not line number.

23 Q. And so what is that -- so in
24 looking at Figure 5, what is that? What does
25 the black section represent on the A strand?

1 A. You're asking me to consider
2 the whole complex; right? A and B; right?

3 There's no dashed lines in the
4 region called d-spacer. Is that what you're
5 asking me to pay attention to?

6 Q. What I'm wondering is what is
7 the -- you'll see that in the discussion in
8 paragraph 29, it talks about the
9 hybridization region. Do you see that?

10 A. Um-hmm. Okay. So "The
11 hybridization region is interrupted by a
12 small segment. In sub-library A, this small
13 segment is conveniently composed of
14 phosphodiester backbone without bases (termed
15 d-spacer)."

16 That's what it is.

17 Q. Okay. And so when we look at
18 the hybridization region on strand A, that's
19 the black region above the d-spacer and the
20 black region below the d-spacer. Is that
21 right?

22 A. That's correct.

23 Q. And that is shown then
24 hybridizing with the hybridization region on
25 the B strand; correct?

1 A. Parts of the A strand are
2 hybridizing with parts of the B strand.

3 Q. And on the B strand, the
4 hybridization region is interrupted by code
5 B; is that right?

6 A. Correct.

7 Q. And above code B in strand B,
8 it's -- the black portion is hybridizing with
9 the hybridization -- part of the
10 hybridization region on the A strand; right?

11 A. That's correct.

12 Q. And part of the hybridization
13 region below code B in the B strand is
14 hybridizing with the hybridization -- part of
15 the hybridization region on the A strand?

16 A. That's correct.

17 Q. That's what's being shown in
18 these dotted lines between the black regions?

19 A. Yes.

20 Q. Now I notice there's only four
21 lines drawn.

22 Does that mean anything to you?

23 Does that mean that there's
24 only four nucleic acids involved in that top
25 portion of hybridization?

1 A. Typically it means that they're
2 hybridized.

3 Q. Is it meant to be numerical and
4 perfect, in terms of the number of nucleic
5 acids?

6 A. In this patent, no.

7 Q. And is that -- I mean, when
8 people are diagramming nucleic acids, is
9 it -- I guess for lack of a better word, that
10 you referred to it in one of your patents as
11 a cartoon.

12 A. Yeah.

13 Q. I mean, very scientific
14 cartoons.

15 But when generating cartoons of
16 nucleic acids, are these hybridization
17 regions supposed to have perfect information
18 in them?

19 MR. HOLMAN: Object to the
20 form.

21 A. So the important part is the
22 caption. And that's why I read the caption
23 before I answered your question, to make sure
24 that the dashed lines were indicating
25 hybridization.

1 So when you read the caption,
2 it actually specifically says "designed to
3 hybridize DNA sequences." So a POSA would
4 read this and say, okay, these are hybridized
5 except for the region in the d-spacer.

6 BY MR. PLATT:

7 Q. All right. So you understand
8 Dr. Nelson, in illustrating the combination
9 of Kanan and Neri, generated a cartoon of
10 that?

11 Do you recall that?

12 A. Yes, I have some of the
13 cartoons in my declaration.

14 Q. So let me show you what's been
15 marked as Exhibit 1044.

16 (Pentelute Exhibit 1044
17 marked for identification and
18 attached to transcript.)

19 Q. This is a call-out from
20 Dr. Nelson's declaration, Figure 2. You'll
21 see it shows pool A and pool B cartoons.

22 Do you see that?

23 A. I do.

24 Q. Okay. And we were talking
25 about the d-spacer on it, in Neri. Right?

1 Figure 5? And it showed that there was no
2 hybridization.

3 A. Correct.

4 Q. But you see on there on that
5 figure, it shows on -- in the red area, it
6 shows these ladders there?

7 A. Yes.

8 Q. Do you see that?

9 A. Yes.

10 Q. Okay. So based on your
11 understanding of Neri, should there be those
12 little ladder illustrations on there?

13 MR. HOLMAN: Objection. Form.

14 A. I mean, I can't interpret what
15 Dr. Nelson was trying to portray in his
16 cartoon.

17 BY MR. PLATT:

18 Q. If you were portraying a
19 d-spacer, would you include those red lines
20 on there?

21 MR. HOLMAN: Objection. Form.

22 Scope. Relevance.

23 A. Yeah, I just -- I mean, I --
24 the d-spacer is a term that's used in the
25 Neri patent, but d-spacer, according to

1 Nelson, could have meant something else. And
2 it could have meant -- I can't interpret what
3 he was implying.

4 BY MR. PLATT:

5 Q. Sure.

6 I'm just asking based on your
7 understanding. Should a d-spacer, should it
8 include those little red diagrams there?

9 MR. HOLMAN: Objection. Form.
10 Scope.

11 A. Yeah, I just -- when I looked
12 at this, I didn't naturally, all of a sudden,
13 assume anything other than what Dr. Nelson
14 had put together. And then I read what Neri
15 put together.

16 In science, the reason why we
17 have figure captions is -- I read with you
18 that figure -- is because without defining
19 it, people don't know what it is.

20 BY MR. PLATT:

21 Q. Okay. But my question for you
22 is a little bit different.

23 And you reviewed Dr. Nelson's
24 transcript relating to his discussion of his
25 figures. Correct?

1 A. Yes.

2 Q. Okay. And you understood that
3 he testified that those little red lines
4 shouldn't have been on there?

5 MR. HOLMAN: Objection.

6 Foundation.

7 A. In what context? See,
8 I comment on this --

9 BY MR. PLATT:

10 Q. On the d-spacer.

11 MR. HOLMAN: Same objection.

12 A. (Document[s] reviewed.)

13 Where did he say in his
14 declaration that he meant to have the hatches
15 in the d-spacer -- not to have those in the
16 d-spacer?

17 BY MR. PLATT:

18 Q. You don't recall reading that?

19 A. I think there was some
20 confusion from Dr. Nelson about this.

21 Q. My question for you, though,
22 is: If you were portraying the d-spacer in a
23 cartoon, would you have included the hatches?

24 MR. HOLMAN: Objection. Form.

25 Scope. Relevance.

1 A. So it's not my report, so
2 I can't -- I didn't write a report, as
3 Dr. Nelson did, describing a d-spacer.

4 BY MR. PLATT:

5 Q. You understand a d-spacer
6 doesn't have any bases; right?

7 A. As defined, it's a polybasic
8 side without bases.

9 Q. And you understand that
10 Dr. Nelson testified that the small vertical
11 lines representing bases should not be there?

12 MR. HOLMAN: Objection.

13 Foundation.

14 A. (Document[s] reviewed.)

15 So in my declaration on
16 paragraph 140, page 76, I comment that at
17 Dr. Nelson's deposition -- this is in my
18 declaration.

19 "Dr. Nelson initially confirmed
20 that, as presented, regions of his figures
21 that show vertical lines from one but not
22 both strands of his construct, for example,
23 d-spacer region, indicate no hybridization
24 between strands. Accordingly, when there are
25 regions of his figures showing vertical lines

1 from one strand, but not both, there is no
2 hybridization. This is how the critical
3 region is presented. Accordingly, as
4 Dr. Nelson presents it in his declaration,
5 there is no hybridization in this region at
6 issue."

7 And then I annotated this in
8 the figure below, which is shown on page 76,
9 where he says that there's no issue here
10 without these hatches for hybridization.

11 BY MR. PLATT:

12 Q. I'm going to show you this.
13 Let me hand you what's been marked as
14 Exhibit 1045.

15 (Pentelute Exhibit 1045
16 marked for identification and
17 attached to transcript.)

18 MR. HOLMAN: Do you have a copy
19 of that for us?

20 MR. PLATT: I don't.

21 MR. HOLMAN: I just want to
22 object to Exhibit 1045 under FRE 401,
23 901, 1002, 1003, and 702.

24 MR. PLATT: Okay. We're going
25 to serve you with a picture of it

1 right now.

2 (Pause.)

3 MR. HOLMAN: While you're doing
4 that, I'd also like to raise an
5 objection to Exhibit 1044 under
6 FRE 401, 901, 1002, and 1003.

7 (Pause.)

8 MR. PLATT: I guess you'll be
9 getting a copy of that momentarily.
10 But you're more than welcome to look
11 over Dr. Pentelute's shoulder.

12 MR. HOLMAN: Let's please wait
13 until we get the exhibit before you
14 start on the questioning.

15 MR. PLATT: If you want to
16 wait, we can go off the record.

17 MR. HOLMAN: Okay. Let's just
18 go off the record then.

19 (Recess: 2:38 p.m. to
20 2:40 p.m.)

21 BY MR. PLATT:

22 Q. So, Dr. Pentelute, if
23 Dr. Nelson had meant not to include ladders
24 on the d-spacer, is Exhibit 1045 what it
25 should look like?

1 MR. HOLMAN: Objection.

2 Foundation. Form. Scope.

3 A. Has anything been changed in
4 pool A?

5 I can't tell, is that Wite-Out
6 up here too on this, or no?

7 BY MR. PLATT:

8 Q. No.

9 A. Okay. Just a glare.

10 So this third Wite-Out on the
11 hatches for the d-spacer?

12 Q. Correct.

13 A. You know, I don't -- I mean, as
14 we just went through the Figure 5 caption and
15 it described what the d-spacer was, I don't
16 know how -- maybe Dr. Nelson came up with a
17 new version of a d-spacer.

18 Q. Are you familiar with him
19 testifying that the d-spacer did not include
20 bases on it?

21 A. I think there was some
22 confusion there. That's why I was trying to
23 double-check that's exactly what he said in
24 the context.

25 Q. My question for you is: If he

1 testified that there weren't bases and that
2 there should not have been the small vertical
3 lines representing bases on the d-spacer, is
4 that what's shown in 1045?

5 A. That's correct. The hatches on
6 the d-spacer now have Wite-Out on them.

7 Q. Okay. Now you also prepared
8 one of your own figures; correct?

9 A. I have.

10 Q. And I will mark this as
11 Exhibit 1046.

12 (Pentelute Exhibit 1046
13 marked for identification and
14 attached to transcript.)

15 Q. This one, we're going to -- I
16 will have to keep up with this one.

17 So as we look at this, you'll
18 see -- when we looked at Neri, Neri included
19 hybridization lines on both sides of the
20 d-spacer. Is that right?

21 A. Let me double-check.

22 Q. Look at Figure 5.

23 A. So he has his d-spacer for
24 strand A. He had hybridization hatches on
25 oligonucleotide A on both sides of the

1 d-spacer.

2 Q. Right. So that would be -- on
3 yours, if it included that, it would look
4 like that. Is that right?

5 MR. HOLMAN: Objection. Form.
6 Foundation. Relevance.

7 A. Well, this is different than
8 what's in Neri. Right? He doesn't go by --
9 BY MR. PLATT:

10 Q. I understand.

11 But my question is: If you
12 were using the hybridization region of what
13 was described in Figure 5 of Neri, it would
14 show hybridization on both sides of the
15 d-spacer; correct?

16 MR. HOLMAN: Objection. Form.

17 A. Taken out of context of what is
18 shown in Neri, but this is a subset of what's
19 drawn in Neri's Figure 5.

20 BY MR. PLATT:

21 Q. And then if you were to do
22 polymerization on the A strand that's shown
23 here in 1046, would it polymerize this way,
24 in this format?

25 MR. HOLMAN: Objection. Form.

1 Scope. Foundation.

2 A. I don't know. It depends on
3 what's actually being done for polymerization
4 reactions.

5 BY MR. PLATT:

6 Q. But if a polymerization
7 reaction was done correctly using the
8 appropriate reaction conditions, the
9 appropriate primers, would it polymerize this
10 way through coding region B?

11 MR. HOLMAN: Objection. Form.

12 A. I'm not entirely clear why
13 someone would do that experiment.

14 BY MR. PLATT:

15 Q. If somebody did polymerization
16 and they used the appropriate primers and
17 wanted to extend it through the coding
18 region, is this what it would look like on
19 Exhibit 1046?

20 MR. HOLMAN: Objection. Form.

21 A. That is so hypothetical. Like,
22 there's a lot of things that you have to
23 think about for this polymerization, let
24 alone -- I don't even know how long the
25 coding region for B is. So how -- what is

1 the primer design? Where would the primer
2 go? How would this be actually done to do a
3 polymerization reaction?

4 BY MR. PLATT:

5 Q. Well, let's assume that all
6 those parameters are taken care of. A person
7 of skill in the art understands how to use
8 primers, they understand how to use the
9 polymerase, they understand what appropriate
10 buffers to use, and how to conduct it with a
11 coding region that's long enough to be
12 extended, and somebody did a primer extension
13 reaction.

14 Would that extend the DNA of
15 strand A through this coding region for B?

16 MR. HOLMAN: Objection. Form.
17 Scope. And foundation.

18 A. Yeah, I mean, the problem is is
19 that a lot of what you're drawing here can
20 oligomerize when you do the denaturation step
21 and then they're reannealing, to where you
22 can have many possible outcomes.

23 And so it's not clear how well
24 that's actually going to work.

25 BY MR. PLATT:

1 Q. I understand.

2 My question is a little bit
3 different than that, okay, which is: Looking
4 at this construct, okay, a person of skill in
5 the art that's skilled in polymerization,
6 that uses the appropriate primers and desires
7 to do primer extension reaction on the A
8 strand and successfully did a primer
9 extension reaction, would this be a fair
10 cartoon for showing that extension?

11 MR. HOLMAN: Objection. Form.
12 Scope. Foundation.

13 A. I've already mentioned, look at
14 the number of variables here. Right? The
15 first thing you have to think about is: I'm
16 going to add in a primer. What are the
17 primers? Where are they binding? And then
18 you have to denature this whole thing, and
19 then you have to anneal it.

20 But if you look, there's pieces
21 in here that can self-anneal in different
22 directions. And so all of a sudden you're
23 going to have these, I call them, phantom
24 constructs, where it's not clear what's going
25 to happen if you're going to do a

1 polymerization that you're proposing might
2 happen.

3 But as a POSA, I can't --
4 I don't know what's going to happen. I have
5 to sit down and think about a lot of
6 parameters. There's a lot of parameters
7 here.

8 BY MR. PLATT:

9 Q. Well, Dr. Pentelute, we'll take
10 a little detour here for a second.

11 You're not a POSA, are you?

12 A. I'm a POSA.

13 Q. Well, let's take a look at your
14 definition of a POSA.

15 A. Okay.

16 Q. Okay. First off, why don't you
17 show me where you state in your declaration
18 where you are a POSA.

19 A. Sure. If you go to page 40.
20 So do you want me to read through that --

21 Q. Sure.

22 You say a POSA for the '848
23 patent, as of July 2005, would have had an
24 advanced degree, such as a Ph.D. in
25 chemistry, organic chemistry, biochemistry,

1 biotechnology, molecular biology, or related
2 discipline, with about two years of
3 experience in laboratory research involving
4 small molecule protein nucleic acid chemistry
5 and hybridization techniques.

6 Now, Dr. Pentelute, you did not
7 have a Ph.D. in July 2005, did you?

8 A. That's correct. I was an
9 advanced graduate student, a Ph.D. student.

10 Q. Okay. So let's go back to
11 Exhibit 1046. And if you don't know if this
12 is what polymerization would look like,
13 that's fine.

14 Is that your answer, that you
15 don't know that if a person of skill in the
16 art who was doing a primer extension reaction
17 on Exhibit 1046 and extended that through
18 this coding region, that this is what it
19 would look like in a cartoon? You just don't
20 know?

21 MR. HOLMAN: Objection. Form.
22 Scope.

23 A. Well, as I mentioned, there's a
24 lot of variables here. And you're asking me
25 to on the fly decide whether or not a PCR

1 reaction would work when I don't even know
2 what primer is being designed, I don't know
3 what -- the temperatures are being used for
4 the polymerization reaction. I don't know
5 what polymerase is being used.

6 Moreover, this molecule, like I
7 said, has a lot of features in it that could
8 cause off-pathway polymerization reactions to
9 occur.

10 BY MR. PLATT:

11 Q. Okay. So you don't know?

12 MR. HOLMAN: Objection. Form.

13 A. So if the polymerization
14 reaction, as you're opining on, it's not
15 clear if it's going to work.

16 BY MR. PLATT:

17 Q. That's even though -- let me
18 draw this out so we finish it out through the
19 end here, with the other black section that's
20 at the end of this.

21 So assuming that the person of
22 skill in the art understands all those
23 parameters that you talked about and can
24 address them -- including pH, buffer
25 conditions, primers -- and they desire to do

1 a primer extension reaction on this, you
2 don't know whether it would look like it,
3 what's depicted in Exhibit 1046. Is that
4 right?

5 MR. HOLMAN: Objection. Form.
6 Scope. Foundation.

7 A. This is a hypothetical that
8 you're throwing at me. And I haven't --
9 I need to sit down and think through all the
10 variables and consider the experiment. It's
11 important.

12 BY MR. PLATT:

13 Q. Okay. So you can't answer that
14 question sitting here today?

15 MR. HOLMAN: Objection. Form.

16 A. So I don't know if it's going
17 to work.

18 MR. HOLMAN: I'll just note for
19 the record that all the markings on
20 Exhibit 1046 are counsel's own
21 markings, not the expert's.

22 BY MR. PLATT:

23 Q. So why don't we take a look --
24 go back to the patent, if you can, the '848
25 patent, Exhibit 1001. Why don't we take a

1 look at Figure 5 there.

2 So if we look at Figure 5(f),
3 okay, and somebody was doing a primer
4 extension reaction on that, it's your
5 understanding that the '848 patent provides
6 sufficient details in order for someone to do
7 primer extension reactions on what is
8 depicted in 5(f) on the left to get
9 polymerization?

10 MR. HOLMAN: Objection. Form.

11 A. So Figure 5(f) has a
12 polymerization extension reaction, as an
13 example. So let me remind myself. I know we
14 talked about this earlier.

15 (Document[s] reviewed.)

16 So they certainly do PCR
17 reactions. The primers are listed in the
18 '848 patent. And the conditions are given in
19 some of these examples.

20 BY MR. PLATT:

21 Q. So a person of skill in the art
22 would understand how to do the primer
23 extension reactions as necessary to conduct
24 the polymerization in Figure 5(f)? Is that
25 your testimony?

1 A. Generally speaking, there's a
2 lot of PCR reactions that are provided here.
3 But specific examples and conditions and
4 specific primers as shown in the
5 specification of doing a polymerase chain
6 reaction to find a specific example for the
7 polymerase example that you're talking --
8 chain extension that you're talking about.

9 (Document[s] reviewed.)

10 There are examples of
11 polymerase chain reactions in this patent.
12 And the conditions are described.

13 Q. And that would allow a person
14 of skill in the art to conduct the reactions
15 in Figure 5(f)?

16 A. The cartoon in figure 5(f)
17 showing a PCR reaction, yes.

18 Q. Okay.

19 (Previously marked
20 Deposition Exhibit 1005 was
21 tendered to the witness.)

22 Q. So I'm going to hand you what's
23 previously been marked as Exhibit 1005.
24 This is the declaration of Dr. Norman Nelson.

25 And this is something you

1 looked at in your analysis; correct?

2 A. Correct.

3 Q. And why don't you take a look
4 at page 118. And we'll go through to
5 page 119.

6 A. (Document[s] reviewed.)
7 Okay.

8 Q. Okay. So if we look at
9 page 119, on the right-hand side of the
10 figure that's shown there, and it's
11 referencing Exhibit 1003, Figure 1, do you
12 understand Exhibit 1003 to be the Baez
13 reference?

14 A. Yes. '809 patent.

15 Q. And that was a patent that you
16 looked at in performing your analysis;
17 correct?

18 A. Correct.

19 Q. Now if we look at that diagram
20 on the right, you will see there that it
21 says -- it's annotated. It says "partial and
22 color added."

23 Do you see that?

24 A. Um-hmm.

25 Q. Okay. And on the left-hand

1 side, you'll see that there's an antibody
2 colored in green. Is that right?

3 MR. HOLMAN: Objection. Form.

4 A. Yeah, I haven't seen the
5 caption to this figure.

6 BY MR. PLATT:

7 Q. Do you see that there's an
8 antibody on the left colored green?

9 A. Oh, I see a molecule that's
10 green. Is that the same molecule as on
11 the -- in the Baez figure?

12 Q. Show you what's been previously
13 marked as Exhibit 1003.

14 (Previously marked
15 Deposition Exhibit 1003 was
16 tendered to the witness.)

17 Q. This is the Baez patent.

18 Do you understand, looking at
19 Figure 1 of the Baez patent, whether what is
20 depicted on the left-hand side is an antibody
21 which is shown in green by Dr. Nelson in his
22 declaration?

23 MR. HOLMAN: Objection. Form.

24 Foundation.

25 A. (Document[s] reviewed.)

1 So in the Baez, which is
2 Figure 1, Exhibit 1003, is it a figure
3 captioned specifically called an antibody?
4 Let me double-check.

5 Figure 1 -- so it's the Baez
6 patent. Column 5, line 45, "Figure 1 is a
7 diagram which illustrates overlap approach
8 for joining a nucleic acid-labels of the
9 analyte bound reporter conjugates to form
10 analyte-specific-amplicon in a heterogenous
11 format."

12 Strange. They don't define
13 that as an antibody.

14 (Document[s] reviewed.)

15 I see. So if you go to
16 column 7, they're calling these binding
17 pair, "includes any of the class of
18 immune-type binding pairs, such as
19 antibody/antigen or hapten/anti-hapten
20 systems...nonimmuno-type binding pairs, such
21 as biotin/avidin."

22 So, you know, they're calling
23 it -- these could be antibodies. And it
24 looks like an antibody.

25 BY MR. PLATT:

1 Q. And what you're saying, it
2 looks like an antibody that is what
3 Dr. Nelson colored green on page --

4 A. Has he altered Figure 1 of the
5 patent, I guess? Is that what's happened?

6 It's been -- is that what it
7 means, "partial color added"?

8 Q. Correct. Do you see that?

9 A. Oh. I didn't know what
10 "partial" meant.

11 So he's altered the figure?

12 Q. (Nodded head up and down.)

13 A. Okay. Okay.

14 Q. You see it's an excerpt from
15 Figure 1. Do you see that?

16 A. Oh, that's why partial.
17 Partial excerpt, it should say, and color
18 added. Okay.

19 Q. And you see on the -- and so on
20 the left-hand side, Dr. Nelson has colored
21 the antibody green?

22 Do you see that?

23 MR. HOLMAN: Objection. Form.
24 Foundation.

25 A. Well, now he's jumping into

1 some new graphic. So we need to look at this
2 graphic -- or let's see what he is actually
3 saying about Exhibit 1008 in his --

4 BY MR. PLATT:

5 Q. My question is just about the
6 color.

7 Do you see that he's --

8 A. Well, there's two colors in
9 this figure that are green.

10 Q. I'm talking about just the one
11 on the right-hand side.

12 A. Okay.

13 Q. And so the right-hand side,
14 where it says Exhibit 1003, Figure 1 (partial
15 and color added).

16 A. Yes.

17 Q. Okay. The depiction on the
18 left, Dr. Nelson colored the antibody green;
19 correct?

20 MR. HOLMAN: Objection. Form.

21 A. I don't know that he colored
22 the antibody green. All I know is he took a
23 figure from the Baez patent and changed it to
24 be green and yellow.

25 ///

1 BY MR. PLATT:

2 Q. And one antibody is green and
3 the other antibody was yellow; correct?

4 A. Yes.

5 Q. And then there are these
6 reporter tags that are shown under where it
7 says D1, and that's colored blue.

8 Do you see that?

9 A. I do.

10 Q. And then on the other antibody,
11 there's a reporter tag labeled D2 that's
12 colored red. Do you see that?

13 A. Yeah, I do -- did you say left?
14 You were talking about the right panel;
15 correct?

16 Q. Yes. To be clear, for the
17 whole -- for the record, we're talking
18 about -- there are two items shown in the
19 right-hand panel.

20 A. And we're talking about the
21 right-hand panel.

22 Q. Correct.

23 A. Okay. Okay.

24 Q. That doesn't change your
25 answers, does it?

1 A. No. The right-hand panel shows
2 a molecule that's green attached to a blue
3 molecule and a molecule that's yellow
4 attached to a red molecule.

5 Q. Okay. So let me show you
6 another exhibit.

7 (Pentelute Exhibit 1047
8 marked for identification and
9 attached to transcript.)

10 Q. So I'm going to show you what's
11 been marked as Exhibit 1047. And you'll see
12 1047 is an article called Expanding Small RNA
13 Experience by Thomas Tuschl?

14 A. I do.

15 Q. Did you review any part of
16 Exhibit 1047 as part of your analysis?

17 A. I don't believe so. I don't
18 reference it in my declaration.

19 Q. What about if you look to the
20 last page, the top part?

21 Did you review all the way
22 through this, including this top part of
23 page 448?

24 A. It was a fan block.

25 Q. I'm talking about the Tuschl

1 reference.

2 A. I don't reference this in my
3 declaration.

4 I've seen a lot of documents,
5 but I don't specifically remember seeing this
6 Tuschl reference.

7 Q. Okay. Did you read all the way
8 through it?

9 Do you recall reading these
10 other sections of it, including what's shown
11 on page 448?

12 A. This is talking about siRNA and
13 antisense. Yeah, I haven't seen this before.

14 Q. Okay. So I'm going to show you
15 what's been marked previously as
16 Exhibit 1012.

17 (Previously marked
18 Deposition Exhibit 1012 was
19 tendered to the witness.)

20 MR. HOLMAN: Before we move on
21 with questions, I want to object to
22 Exhibit 1047 under FRE 401.

23 BY MR. PLATT:

24 Q. Do you have Exhibit 1012 in
25 front of you?

1 A. I do.

2 Q. Okay. And Exhibit 1012, you
3 reference in your declaration. Is that
4 right?

5 A. I do.

6 Q. And did you review all of 1012?

7 A. I read it, yes.

8 Q. Including the top part --

9 A. No.

10 Q. -- on the first page?

11 A. No, I didn't read that.

12 Q. Okay. So you read the part of
13 1012 that was relevant to your analysis?

14 A. Yes.

15 MR. HOLMAN: Objection. Form.

16 MR. PLATT: Why don't we take a
17 break and see -- let me see where
18 I am.

19 MR. HOLMAN: Okay.

20 (Recess: 3:11 p.m. to

21 3:21 p.m.)

22 BY MR. PLATT:

23 Q. So, Dr. Pentelute, you prepared
24 a declaration in this case; right?

25 A. I did.

1 Q. Who did you work with to
2 prepare that declaration?

3 A. My counsel.

4 Q. And who was that?

5 A. David and Adil, and then one of
6 their colleagues, as I mentioned. I feel bad
7 I didn't meet her in person. She was on
8 Zoom. But I think it was Maria or something.
9 I don't remember her first name.

10 Q. And who wrote your declaration?

11 MR. HOLMAN: Objection to the
12 extent it calls for privileged
13 information.

14 I caution the witness not to
15 divulge the substance of any
16 communications with counsel.

17 I'm going to instruct you not
18 to answer.

19 BY MR. PLATT:

20 Q. Are you going to follow that
21 instruction?

22 A. Yes.

23 Q. What portion of your
24 declaration did you write?

25 MR. HOLMAN: Same objection.

1 Instruct the witness not to answer.

2 Privileged information.

3 BY MR. PLATT:

4 Q. Did you speak with anyone
5 besides attorneys in preparing the opinions
6 expressed in your declaration?

7 A. No.

8 Q. How many hours did you spend
9 preparing your declaration?

10 A. As I mentioned earlier,
11 multiple tens of hours. So I haven't added
12 up exactly, but time understanding, reading,
13 and evaluating documents and preparing the
14 declaration, it's 40 hours. It could be
15 more; it could be a little bit less.

16 Q. I think you gave me that same
17 estimate when we were talking about leading
18 all the way up until today.

19 A. Yeah.

20 Q. I'm just doing the math on two
21 eight-hour days of preparation, plus another
22 five hours of preparation, plus preparation
23 yesterday. Make that 20-plus hours of
24 preparation for deposition.

25 Do you think you spent 20 hours

1 or so on preparing your declaration?

2 A. Yeah, I haven't -- I'm
3 approximating. I sat down and added all
4 the hours up, especially with what we're
5 doing now, I wasn't necessarily factoring
6 that in right now.

7 Q. Okay. And have you been paid
8 for the work you've done?

9 A. I think in part.

10 Q. And do you recall how big the
11 last check was you got?

12 A. I think 10, 15 hours or so --

13 Q. Okay.

14 A. -- of work.

15 Q. And I said the "last check."
16 But, you know, cumulatively, do you recall
17 how much you've been paid so far to date?

18 A. I think it's either 10 or
19 20 hours. I don't remember.

20 Q. But you don't know if you've
21 been paid \$5,000 or \$10,000?

22 A. I have given you the
23 approximate. I don't know if it's -- it's
24 definitely not -- it's definitely less than
25 20 hours I've been paid for.

1 Q. All right. During the course
2 of preparing for your deposition, anything
3 leading up to this time, did you become aware
4 of any mistakes in your declaration?

5 A. Nothing major. I think there
6 might have been a typo here or there, but
7 nothing that would change my declaration.

8 Q. And just for the record, when
9 we're talking about your declaration, it's
10 Exhibit 2008.

11 (Previously marked
12 Deposition Exhibit 2008 was
13 tendered to the witness.)

14 Q. Did you do any independent
15 searching for any of the exhibits cited in
16 your declaration?

17 A. No. I read the exhibits that
18 were given to me by the lawyers. I might
19 have not been able to find a PDF file, so
20 I might have searched for a patent online.
21 But I just stuck to the exhibits that I was
22 provided.

23 Q. Did you read every word of
24 every document that you were provided?

25 A. I certainly read all of them.

1 I might have missed "and," "or," or certain
2 subparts of a sentence. But I've gone
3 through them. I spent a fair amount of time
4 looking at these and seeing what's in these
5 documents.

6 Q. Including the 500-page-plus
7 prosecution history?

8 A. Oh. So a lot of that, I don't
9 understand because it's legalese.

10 Q. Right.

11 A. So I have looked through it,
12 gone through it, but I don't understand a lot
13 of the stuff that they're talking about.

14 Q. So maybe you skipped over the
15 stuff that wasn't relevant to your analysis?

16 MR. HOLMAN: Objection. Form.

17 A. Yeah, I would read it, but it
18 would go fast because I don't understand what
19 I'm reading.

20 MR. PLATT: Okay. We don't
21 have any further questions at this
22 time. We pass the witness.

23 MR. HOLMAN: All right. Let's
24 go off the record.

25 (Recess: 3:29 p.m. to

1 3:41 p.m.)

2 MR. HOLMAN: Dr. Pentelute, we
3 have no more questions for you. So
4 thank you for your time. This
5 deposition is over.

6 THE STENOGRAPHER: Counsel, can
7 you place on the stenographic record
8 your transcript orders, please;
9 whether or not you would like to order
10 the rough draft, and if you would like
11 standard or expedited delivery of the
12 final.

13 MR. PLATT: The final Thursday.
14 And a rough.

15 MR. HOLMAN: No rush on our
16 end. So we're fine with regular
17 final.

18 THE STENOGRAPHER: Do you want
19 the rough draft tonight?

20 MR. HOLMAN: Sure. That would
21 be great.

22 THE STENOGRAPHER: Thank you.

23 (Time noted: 3:42 p.m.)
24
25

CERTIFICATE OF SHORTHAND REPORTER

I, LISA A. KNIGHT, Registered
Diplomate Reporter and Certified Realtime
Reporter, do hereby certify:

That BRADLEY L. PENTELUTE, Ph.D.,
the witness whose deposition is hereinbefore set
forth, was given an acknowledgement before the
commencement of such deposition and that such
deposition was taken before me and is a true
record of the testimony given by such witness.

I further certify that the adverse
party, Olink Proteomics AB, was represented by
counsel at the deposition.

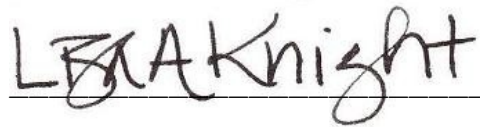
I further certify that the deposition
of BRADLEY L. PENTELUTE, Ph.D., occurred at the
location of the Denver Airport Marriott at
Gateway Park, 16455 East 40th Circle, Aurora,
Colorado 80011, on Monday, September 15, 2025,
commencing at 8:57 a.m. to 3:42 p.m.

I further certify that inspection,
reading and signing of said deposition was not
requested on the record.

I further certify that I am not
related to any of the parties to this action by
blood or marriage, I am not employed by or an

1 attorney to any of the parties to this action,
2 and that I am in no way interested, financially
3 or otherwise, in the outcome of this matter.

4 IN WITNESS WHEREOF, I have hereunto
5 set my hand this 17th day of September, 2025.

6
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8 
9

10 LISA A. KNIGHT

11 NCRA Certified Realtime Reporter

12 NCRA Registered Diplomate Reporter

13 NCRA Realtime Systems Administrator

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