UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE PATENT TRIAL AND APPEAL BOARD

APOTEX INC. and APOTEX CORP.
Petitioners,

v.

AMGEN INC. and AMGEN MANUFACTURING LIMITED
Patent Owners

Case IPR2016-01542
Patent 8,952,138

PATENT OWNER RESPONSE

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

TABLE OF CONTENTS

TABLE OF AUTHORITIES

Page(s)

Cases

EXHIBIT LIST

Exhibit Number	Exhibit
2001	Declaration of Richard C. Willson, Ph.D.
2002	C.V. of Richard C. Willson, Ph.D.
2003	Findings of Fact and Conclusions of Law, *Amgen Inc. et al. v. Apotex Inc. et al.*, Case no. 0:15-cv-61631-JIC, Dkt. 267, entered September 6, 2016
2004	Partial Findings Regarding Apotex's Assertion of Invalidity of the '138 Patent, *Amgen Inc. et al. v. Apotex Inc. et al.*, Case no. 0:15-cv-61631-JIC, Dkt. 245, ordered July 14, 2016
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2006	Lehninger, "Principles of Biochemistry" (1982), excerpted
2007	Declaration of Richard C. Willson, Ph.D. Regarding Claim Construction of Shultz et al., U.S. Patent No.

Exhibit Number	Exhibit
	8,952,138, *Amgen Inc. et al. v. Apotex Inc. et al.*, Case no. 0:15-cv-61631-JIC, Dkt. 77-4, filed December 11, 2015
2008	Rebuttal Declaration of Richard C. Willson, Ph.D. Regarding Claim Construction of Shultz et al., U.S. Patent No. 8,952,138, *Amgen Inc. et al. v. Apotex Inc. et al.*, Case no. 0:15-cv-61631-JIC, Dkt. 83-1, filed January 8, 2016
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Exhibit Number	Exhibit
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2013	UniProt Mouse IL-31, http://www.uniprot.org/uniprot/Q6EAL8 (last visited 11/21/2016)
2014	IUPAC, https://goldbook.iupac.org/D01514.html (last visited (11/21/2016)
2015	Berg et al., "Biochemistry," 2002 (5th edition), excerpted
2016	Schrödel et al., "Characterization of the Aggregates Formed During Recombinant Protein Expression in Bacteria," BMC Biochemistry, 6:10 (2005)
2017	European Patent Application No. 0657466
2018	Slide
2019	May 8, 2017, Deposition Transcript of Anne S. Robinson, Ph.D.
2020	Second Declaration of Richard C. Willson, Ph.D.

Exhibit Number	Exhibit
2021	Declaration of Roger A. Hart, Ph.D.
2022	Presentation entitled "High concentration refolding" (February 2009) [Filed as "**Parties and Board only**"]
2023	Metadata for Exhibit 2022
2024	Presentation entitled "High concentration refolding of AMG 745 and AMG 386" (September 2008) [Filed as "**Parties and Board only**"]
2025	Metadata for Exhibit 2024
2026	Padhi et al., "Pharmacological Inhibition of Myostatin and Changes in Lean Body Mass and Lower Extremity Muscle Size in Patients Receiving Androgen Deprivation Therapy for Prostate Cancer," J. Clin. Endocrinol. Metab., 99(10), pp. E1967-E1975 (2014)
2027	International Patent Application No. WO2014144903
2028	Transcript of Bench Trial held on 7/14/2016, *Amgen Inc. et al. v. Apotex Inc. et al.*, Case no. 0:15-cv-61631-JIC, Dkt. 250, filed July 18, 2016

Exhibit Number	Exhibit
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2033	Maachupalli-Reddy et al., "Effect of Inclusion Body Contaminants on the Oxidative Renaturation of Hen Egg White Lysozyme," Biotechnol. Prog., 13, pp. 144-150 (1997)
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Exhibit Number	Exhibit
	(1999)
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Exhibit Number	Exhibit
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2044	Tobacman et al., "The Kinetics of Action Nucleation and Polymerization," The Journal of Biological Chemistry, 258(5), pp. 3207-3214 (1983)
2045	Darby et al., "Refolding of Bovine Pancreatic Trypsin Inhibitor *via* Non-native Disulphide Intermediates," J. Mol. Biol., 249, pp. 463-477 (1995)
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Exhibit Number	Exhibit
	human lysozyme variants underlying amyloid fibrillogenesis," Nature, 385, pp. 787-793 (February 27, 1997)
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2049	Radford et al., "The folding of hen lysozyme involves partially structured intermediates and multiple pathways," Nature, 358, pp. 302-307 (July 23, 1992)
2050	Tatsumi et al., "Denatured State of Ovalbumin in High Concentrations of Urea as Evaluated by Disulfide Rearrangement Analysis," The Journal of Biological Chemistry, 269(45), pp. 28062-28067 (1994)
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Exhibit Number	Exhibit
	(1993)
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I. Introduction

U.S. Patent No. 8,952,138 ("the '138 Patent") is directed to a novel and efficient protein refolding method based on control of redox conditions; with reductant and oxidant ("redox") reagents, disulfide bonds are formed and reshuffled to refold mis-folded proteins. In a substantial departure from the trial-and-error approach of the prior art, that novel method surprisingly and unexpectedly led to a more rational design of refolding recombinant proteins expressed in non-mammalian expression systems, *e.g.* bacteria.

Notably, the inventors defined a unique equation for thiol-pair ratio ($\frac{[reductant]^2}{[oxidant]}$) that accurately reflects the complex, redox chemistry of disulfide bond formation in proteins. By identifying a relationship not known in the prior art between that unique equation and the redox buffer strength equation ($2[oxidant] + [reductant]$), the '138 Patent provides greater predictability in identifying optimal conditions for refolding proteins; in a departure from the prior art, the '138 Patent does so even under high protein concentrations, anaerobic conditions, and for complex proteins (*e.g.* antibodies, multimeric proteins, and Fc-protein conjugates).

Amgen Inc. and Amgen Manufacturing Limited (together "Patent Owners" or "Amgen") respectfully disagree with the Board's institution of *inter partes* review of the '138 Patent based on the alleged obviousness of: (1) Claims

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

1-11 and 13-24 over *Schlegl* (EX1003) and *Hevehan* (EX1004); and (2) Claim 12 over *Schlegl* (EX1003), *Hevehan* (EX1004), and *Hakim* (EX1006). *Apotex Inc. et al. v. Amgen Inc. et al.*, IPR2016-01542, Paper 10 (P.T.A.B. Feb. 17, 2017) ("Institution Decision") at 34.

Both Grounds suffer from fundamental deficiencies. As Dr. Robinson, Petitioners' expert, admitted at deposition, *Schlegl* and *Hevehan* do not teach the thiol-pair ratio ($\frac{[reductant]^2}{[oxidant]}$) and redox buffer strength ($2[oxidant] + [reductant]$) equations. And yet, both equations are necessary to calculate the claimed "final thiol-pair ratio" ("TPR") and "redox buffer strength" ("RBS") values. In performing her obviousness analysis, Dr. Robinson could only apply impermissible hindsight by applying the equations of the '138 Patent to the prior art. In any event, a POSITA would not have been motivated to combine *Schlegl* and *Hevehan*. They teach fundamentally different and incompatible refolding methods: *Schlegl*'s method of refolding is a mechanical approach at extremely dilute protein concentrations whereas *Hevehan*'s method is a chemical approach at high protein concentrations. A POSITA would also understand that *Schlegl* and *Hevehan*'s methods for refolding purified, model proteins are not applicable to nor predictive of refolding recombinant proteins expressed in non-mammalian expression systems, as required by the claims of the '138 Patent. A key distinction between (1) purified, model proteins and (2) proteins expressed recombinantly in

non-mammalian expression systems (*e.g.*, proteins in inclusion bodies): host-cell contaminants. It is significant that published work by the very same lab responsible for *Hevehan* makes clear that methods for refolding the former are not applicable to methods for refolding the latter; host-cell contaminants confound and interfere with protein refolds.

Even assuming, *arguendo*, that a POSITA would combine *Schlegl* and *Hevehan*, that combination does not render the challenged claims obvious. As Dr. Robinson admitted at deposition, *Hevehan* teaches a refold buffer with zero reductant; it follows that the TPR value is necessarily zero in *Hevehan*. And Petitioners' position (that the Board agreed with) that a POSITA would apply *Hevehan*'s redox conditions to *Schlegl*'s methods necessarily leads to a combination with a TPR value of zero; that value falls outside the scope of all of the claimed TPR values, including the broadest range of sole independent Claim 1.

Nor would there be a reasonable expectation that the combination of *Schlegl* and *Hevehan* would lead to methods that could successfully refold proteins expressed in a non-mammalian expression system. As already noted, *Schlegl* and *Hevehan*—with their purified, model proteins—are not applicable to refolding recombinant proteins expressed in non-mammalian expression systems, as required by the claims of the '138 Patent. Furthermore, a POSITA would also understand that *Schlegl*'s sole example of its refolding method used bovine α-lactalbumin ("α-

LA"). Because that unique protein refolds readily without redox chemicals, it is unpredictive of refolding other proteins. Moreover, the *Hevehan* model has been soundly refuted. Given that it was known prior to *Schlegl* that *Hevehan* could not even accurately predict refolding of its own purified, model protein, hen egg white lysozyme ("HEWL"), a POSITA would not have applied *Hevehan* to refolding methods for any other proteins with any reasonable expectation of success.

As for Claims 9-12 and 18 (which depend from Claim 1), there are additional reasons why those claims are not rendered obvious by the asserted prior art combinations. For Claims 9-12, as Dr. Robinson readily admitted at deposition, *Schlegl* and *Hevehan* do not teach refolding antibodies (Claim 9), complex proteins (Claim 10), multimeric proteins (Claim 11) or Fc-protein conjugates (Claim 12). Conclusory assertions aside, Petitioners submit no empirical evidence that a POSITA would have had a reasonable expectation of success of refolding any of those classes of proteins with the methods taught in *Schlegl* and *Hevehan*.

Nor does *Hakim* cure the deficiencies of *Schlegl* and *Hevehan* in relation to the Fc-protein conjugates of Claim 12. *Hakim* is not prior art. At least Claim 12 and Claims 7 (recombinant protein), 10 (complex protein), and 11 (multimeric protein) of the '138 Patent invention were reduced to practice, at the latest, on February 26, 2009, and predate *Hakim*'s publication date of May 1, 2009. In any event, even if *Hakim* qualified as prior art, it provides no details about its

refolding methodology. Without that critical information, there is no way for a POSITA to know whether the methods of *Schlegl* and *Hevehan* are even compatible to refolding *Hakim*'s Fc-protein conjugate—there is no motivation to combine with a reasonable expectation of success.

As for Claim 18, that claim depends from Claim 1 (which references "incubating the refold mixture") and further requires that such "incubation is performed under non-aerobic conditions." But there is no evidence that the combination of *Schlegl* and *Hevehan* teaches this limitation. Petitioners do not rely on *Schlegl* for this limitation—nor can they, since *Schlegl* only discloses refolding tanks for incubation under aerobic conditions. As for *Hevehan*, Petitioners can only cite to a passage that—as Dr. Robinson readily admitted at deposition—bears no relationship whatsoever to the incubation of the refold mixture, as required by Claim 18. What Petitioners notably do not cite: a different *Hevehan* passage, teaching that its refolds were done aerobically.

In view of these fundamental deficiencies, both instituted Grounds should be denied in their entirety.

II. The '138 Patent Invention

A. Prior to the '138 Patent

The '138 Patent is an improved methodology for refolding recombinant proteins expressed in non-mammalian cell culture systems (*e.g.*

bacteria) at high concentration. EX1001 at 1:11-12, 2:17-30, 4:20-32, 4:55-58, 10:9-16, 12:40-49; EX2001 at ¶58[1]. As reflected by its title, "Refolding Proteins Using a Chemically Controlled Redox State," the '138 Patent is a redox chemistry-based method; redox chemicals assist in forming and reshuffling disulfide bonds for protein refolding.

The goal of protein refolding is to maximize yield of properly folded protein species. EX2020 at ¶20; EX2021 at ¶14. Because refolding recombinant proteins (particularly those with multiple disulfide bonds) into their proper three-dimensional configuration is complex, it is challenging to achieve that goal. EX2021 at ¶14. Prior to the '138 Patent, those skilled in the art needed to manipulate a large number of variables—through trial-and-error—to determine methods for refolding proteins. *Id.*

Prior to the '138 Patent, the primary approach was to refold proteins at dilute protein concentrations. EX2001 at ¶57; EX1001 at 1:54. This is because unfolded or improperly folded proteins are "sticky" and tend to clump together or aggregate—*e.g.*, the exposed hydrophobic regions stick together (similar to how

[1] Except for patent and patent application Exhibits, declaration Exhibits (EX1002, EX2001, EX2020, EX2021), and deposition and trial testimony Exhibits (EX2019, EX2028), all cites herein refer to the page numbers added by Petitioners, Amgen, or the Board at the bottom of each Exhibit or Paper.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

oils clump together in water because oil and water do not mix). EX2021 at ¶15. But, as noted in the '138 Patent, refolding proteins at dilute concentrations at industrial scale requires huge refolding tanks and housing facilities; it is a costly approach. EX2001 at ¶57; EX1001 at 1:57-60.

Attempting to refold at high protein concentrations exacerbates the aggregation problem: since the protein molecules are in much closer proximity, the likelihood that they will bump into each other and stick together is increased. EX2021 at ¶15. Rather than properly refolding into their biologically active configurations, the proteins aggregate. *Id.* Such aggregation interferes with obtaining good yields of properly refolded recombinant proteins. *Id.* Prior to the '138 Patent, refolding at high protein concentrations at industrial scale was, thus, both costly and inefficient, to the extent even possible. EX1001 at 1:52-60, 2:17-22.

Prior to the '138 Patent, there was no rationale for selecting redox conditions. EX2021 at ¶¶14-15; EX1003 at [0073]-[0075] (providing no reasoning for choosing its redox conditions); EX2019 at 46:8-18 ("no specific rationale given in the Schlegl patent for those specific concentrations of" α-LA); EX1004 at 5 (randomly selecting concentrations of redox chemicals); EX1006 (no disclosure of its refolding methodology). Although practitioners occasionally referred to simple ratios of reductant and oxidant concentrations (*i.e.*, [reductant] / [oxidant]), merely

varying the relative amounts of reductant and oxidant generated random results; trial-and-error was necessary to identify acceptable reductant and oxidant concentrations for protein refolding. EX2021 at ¶19; EX2020 at ¶¶27-30; EX2030 at 2; EX2031 at 3; EX2032 at 2 ("for a researcher working with a novel protein, finding the most suitable conditions for expression, solubilization, and refolding of proteins a priori can be a relatively random process.") (emphasis added).

There was a need for the rational design of refolding proteins using redox chemicals: for an efficient method that could predictably refold proteins, especially at high protein concentrations and for more complex proteins (*e.g.* multimeric proteins such as antibodies and Fc-protein conjugates). With such a method, greater amounts of biologically active proteins could be produced at industrial scale at a given time, saving both time and money.

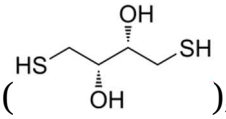
B. The Novel '138 Patent Method Surprisingly and Unexpectedly Led to a More Rational Design of Refolding Proteins

The inventors of the '138 Patent invented a novel and efficient protein refolding method at high protein concentrations through control of redox conditions. EX2021 at ¶17; EX2001 at ¶¶58-59.

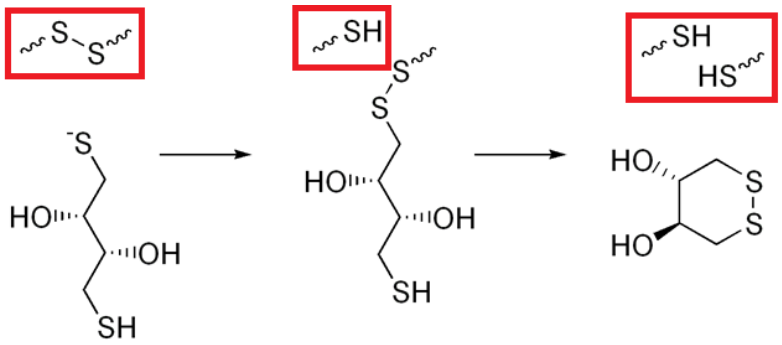
Notably, one of the inventors (Dr. Roger Hart), defined a unique equation for thiol-pair ratio ($\frac{[reductant]^2}{[oxidant]}$). EX2021 at ¶¶17-24. Dr. Hart derived that unique equation $\frac{[reductant]^2}{[oxidant]}$ based on his substantial work with various redox

chemicals (*e.g.*, DTT and cysteine/cystine) and his understanding of the oxidation-reduction chemistry of disulfide bond formation. *Id.* at ¶¶21-22.

Prior to the '138 Patent, Dr. Hart had studied the reductant, DTT

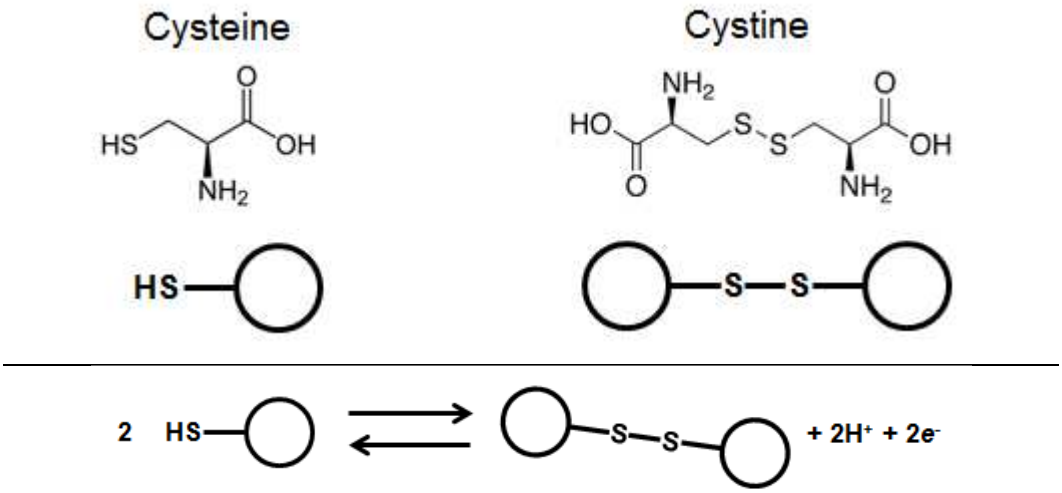


(), which breaks disulfide bonds. *Id.* at ¶21. The following is a typical redox reaction (oxidation-reduction reaction) involving DTT (DTT and its derivatives are in black, and the oxidized species and its derivatives are in the red box):



Id. Notably, DTT is an irreversible reductant. *Id.* DTT is completely oxidized (used up) after reducing (breaking) disulfide bonds. *Id.* This makes DTT a good reagent for denaturing (unfolding) proteins. *Id.* But it makes DTT a poor reagent for refolding proteins because it does not permit "reshuffling" of disulfide bonds. *Id.* Once oxidized, DTT is unavailable to act as a reductant for breaking any additional (incorrectly formed) disulfide bonds. *Id.*

Dr. Hart also studied the reductant cysteine, which is involved in disulfide bond formation in proteins:



Id. at ¶22. Notably, cysteine is not irreversibly oxidized like DTT. *Id.* Cysteine is in equilibrium with cystine, such that disulfide bonds can be "reshuffled" until the correct disulfide bonds are formed and the protein is properly refolded. *Id.* at ¶¶22-23.

After conducting a series of experiments, Dr. Hart derived a relationship between the concentration of reductant squared and the concentration of oxidant that applied in the context of disulfide bond formation in proteins. *Id.* at ¶24. The two molecules of cysteine (the reductant) and the one molecule of cystine (the oxidant) lead to exponents of 2 and 1, respectively, in the equation $\frac{[reductant]^2}{[oxidant]}$. *Id.* That formula accurately reflects the complex chemistry of disulfide bond formation—whereas the simple ratio of [reductant] / [oxidant] associated with the trial-and-error approach of the prior art did not. *Id.* at ¶¶22-24.

The inventors of the ’138 Patent identified a relationship between that unique equation $\frac{[reductant]^2}{[oxidant]}$ and the RBS equation $(2[oxidant] + [reductant])$ that was not known in the prior art. *Id.* at ¶26; EX2020 at ¶¶31-33. That discovery is disclosed in the ’138 Patent:

> As described herein, the relationship between thiol buffer strength and redox thiol-pair ratio has been investigated and optimized in order to provide a reproducible method of refolding proteins at concentrations of 2.0 g/L, and higher on a variety of scales. A mathematical formula was deduced to allow the precise calculation of the ratios and strengths of individual redox couple components to achieve matrices of buffer thiol-pair ratio and buffer thiol strength. Once this relationship was established, it was possible to systematically demonstrate that thiol buffer strength and the thiol-pair ratio interact to define the distribution of resulting product-related species in a refolding reaction.

EX1001 at 4:35-45 (emphasis added), Figs. 1a-1f; EX2020 at ¶¶17-19. The relationship between TPR and RBS surprisingly and unexpectedly provided greater predictability in identifying optimal conditions for refolding proteins at high protein concentrations. EX2021 at ¶26; EX2020 at ¶20. Specifically, [redacted] EX2021 at ¶26.

[redacted]

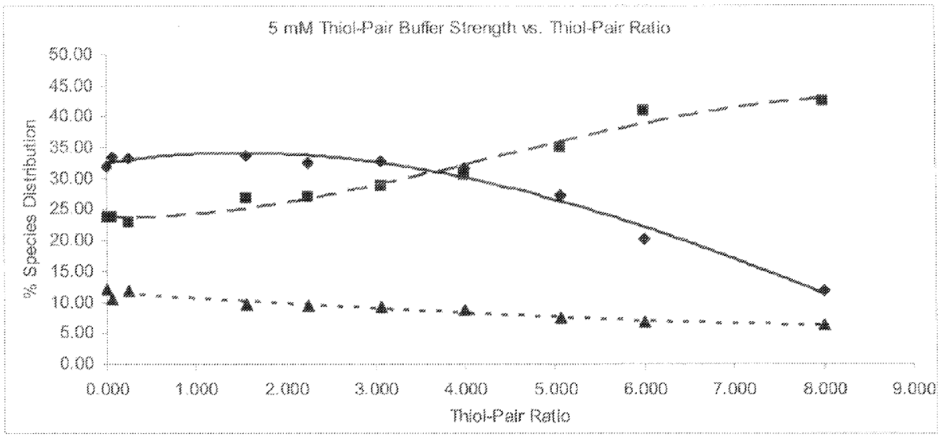
EX2024 at 8[2]; EX2022 at 23; EX2021 at ¶26. That TPR and RBS relationship clearly identified the optimal refolding condition [redacted] for a particular protein concentration:

[2] [redacted] which is the same as TPR in the '138 Patent; both refer to the same equation $\frac{[reductant]^2}{[oxidant]}$. [redacted]

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

[redacted]

EX2022 at 21; EX2021 at ¶26. That TPR and RBS relationship also clearly indicated which redox conditions resulted in diminishing or lower protein yields. EX2021 at ¶26. For example, in Figure 1a of the '138 Patent, at an RBS value of 5 mM, there are quickly diminishing yields of properly refolded protein (solid line) and increasing yields of incorrectly folded protein (dashed line) at TPR values greater than 4:



EX1001 at Figure 1a; EX2021 at ¶26.

As noted in the ’138 Patent, that TPR and RBS relationship “allows for the optimization of the yield of a desired folded protein form” and enhanced efficiency of refolding proteins at significantly higher protein concentrations than in the prior art. EX1001 at 9:11-13; EX2001 at ¶¶58-60. The inventors applied their novel method to efficiently and predictably refold numerous proteins, including complex and multimeric proteins, at high protein concentrations. EX2021 at ¶27.

III. Person of Skill in the Art

The Board agreed with Amgen that a POSITA requires a “graduate level of education and experience . . . due to the sophistication of the area of protein refolding.” Institution Decision at 7. The Board determined that a POSITA “would have an advanced degree in biochemistry with an engineering component and significant experience in protein production, including refolding.” *Id.*

IV. Claim Construction

Claim 1 of the ’138 Patent reads as follows:

> 1. A method of refolding a protein expressed in a non-mammalian expression system and present in a volume at a concentration of 2.0 g/L or greater comprising:
> (a) contacting the protein with a refold buffer comprising
> a redox component comprising

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

a final thiol-pair ratio having a range of 0.001 to 100 and

a redox buffer strength of 2 mM or greater

and one or more of:

(i) a denaturant;

(ii) an aggregation suppressor; and

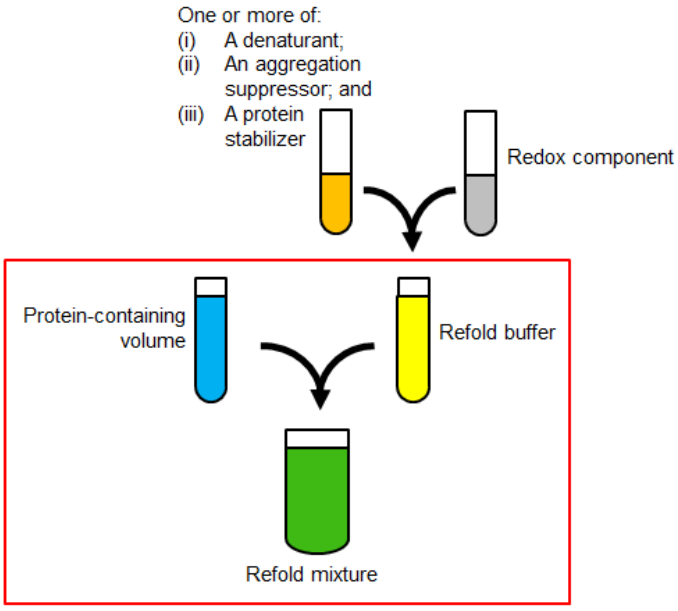
(iii) a protein stabilizer;

to form a refold mixture;

(b) incubating the refold mixture; and

(c) isolating the protein from the refold mixture.

The highlighted terms are color-coded to match the illustration below, and illustrates the elements that make up step (a) of Claim 1, the "contacting" step (the "contacting" step is represented by the red box). Petitioners' expert, Dr. Robinson, agreed that the below illustration (including the five volumes but without the red box) was a "fair representation of the process of the '138 Patent." EX2028 at 101:6-16; EX2018.



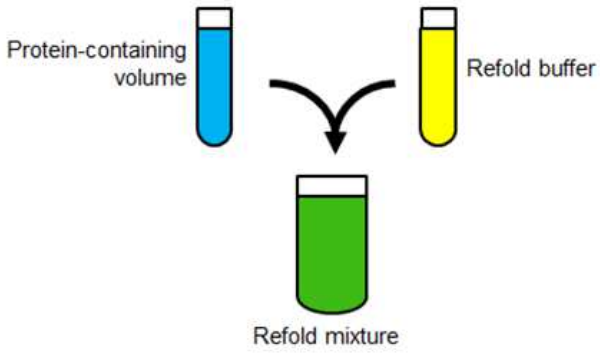
PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

The Board construed five terms in the '138 Patent: (1) "protein" (Claim 1); (2) "buffer thiol-pair ratio"; (3) "thiol-pair buffer strength"; (4) "refold mixture" (Claim 1); and (5) "complex protein" (Claim 10). Institution Decision at 9-10.

A. Construed Terms

1. "refold mixture"

The Board construed "refold mixture" (Claim 1) to mean "a mixture formed from contacting [1] the protein with [2] the refold buffer." *Id.* at 10 (citation omitted). There is no dispute that the protein is "in a separate volume" from the refold buffer. EX2019 at 20:24-21:7; EX2020 at ¶7; EX2001 at ¶72. At deposition, Dr. Robinson agreed that the below depicts the Board's construction of "refold mixture":



EX2019 at 115:3-20, 116:5-15. And Dr. Robinson testified that when "combining two different volumes, [] the final mixture will be generally larger than each individual solution." *Id.* at 116:24-117:9. It follows that the volume of the refold

mixture is greater than the refold buffer volume and protein-containing volume, individually. *Id.* at 119:3-12; EX2001 at ¶72.[3]

2. "complex protein"

The Board adopted the specification's definition of "complex protein" (Claim 10):

> The protein can be a complex protein, i.e., a protein that (a) is larger than 20,000 MW, or comprises greater than 250 amino acid residues, and (b) comprises two or more disulfide bonds in its native form

Institution Decision at 10 (emphasis in original); EX1001 at 12:58-61. Accordingly, a "complex protein" is either (1) larger than 20,000 MW with two or more disulfide bonds in its native form or (2) comprises greater than 250 amino acid residues with two or more disulfide bonds in its native form. EX2020 at ¶9.

3. "protein"

The Board appears to have adopted the specification's definition of "protein." Institution Decision at 9. Thus, Amgen understands the Board's construction of "protein" to mean "any chain of at least five naturally or non-

[3] The Board rejected Amgen's and the District Court's construction that the refold mixture has a "'high protein concentration' . . . at or above about 1 g/L protein." Institution Decision at 10 (citation omitted). Amgen respectfully disagrees. *See* Amgen's Preliminary Response ("POPR") at 13-16; EX2001 at ¶¶73-76.

naturally occurring amino acids linked by peptide bonds."

4. "final thiol-pair ratio"

Claim 1 recites a "final thiol-pair ratio." EX1001 at Claim 1. The Board construed "buffer thiol-pair ratio," which is not recited by any claim.

For the purposes of this proceeding post-institution, Amgen applies the Board's construction of "buffer thiol-pair ratio" to the claim term "final thiol-pair ratio," which is recited in Claim 1; TPR, thus, means $\frac{[reductant]^2}{[oxidant]}$, where the concentrations are determined in the refold buffer[4]. Institution Decision at 9.

[4] Amgen respectfully disagrees with the Board's construction. The parties agreed that the concentrations in $\frac{[reductant]^2}{[oxidant]}$ are determined in the redox component. Petition at 24; POPR at 11-12. That agreed-upon construction is supported by the claim language, which requires "a refold buffer comprising a redox component comprising a final thiol-pair ratio having a range of 0.001 to 100 and a redox buffer strength of 2 mM or greater and one or more of" three chemicals. EX1001 at Claim 1 (emphasis added). The plain language makes clear that the refold buffer comprises (1) a redox component and (2) "one or more of" three chemicals; the TPR and RBS are referring back to (and thus, part of) the redox component—"a redox component comprising a final thiol-pair ratio . . . and a redox buffer strength . . . ." *Id.* The Board's construction rewrites the claim as either: (1) "a refold

In contrast, Dr. Robinson, Apotex's expert, takes the position that the Board's construction is irrelevant to this proceeding. At deposition, she insisted that "the board has not construed the term 'final thiol-pair ratio,'" and that the Board instead construed a term "buffer thiol-pair ratio" that does not appear in any claims. EX2019 at 12:19-13:5, 13:23-14:5. Dr. Robinson apparently agrees that TPR values are determined by the equation $\frac{[reductant]^2}{[oxidant]}$, but she now has no idea which volume to apply when calculating TPR values. *Id.* at 14:24-15:10, 15:19-16:5; EX1002 at ¶79. And yet, volume matters. At deposition, Dr. Robinson agreed that $\frac{[reductant]^2}{[oxidant]}$ is "volume-dependent" and that "if you have different volumes, you will get different results." EX2019 at 14:24-15:10, 128:17-129:2.

It is readily apparent why Dr. Robinson rejects applying the Board's construction of "buffer thiol-pair ratio" to the claim term "final thiol-pair ratio." As described below, *Hevehan* falls outside of Claim 1, which requires TPR values "having a range of 0.001 to 100." Because there is no reductant in *Hevehan*'s

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buffer comprising a redox component [wherein the refold buffer further] compris[es]~~ing~~ a final thiol-pair ratio having a range of 0.001 to 100 and a redox buffer strength of 2 mM or greater" or (2) "a refold buffer comprising ~~a redox component comprising~~ a final thiol-pair ratio having a range of 0.001 to 100 and a redox buffer strength of 2 mM or greater."

refold buffer, the TPR value = $\frac{[reductant]^2}{[oxidant]} = \frac{0^2}{[oxidant]}$ = zero in *Hevehan* based on the Board's construction. Dr. Robinson now claims that TPR is vague with respect to which volume the TPR equation applies. EX2019 at 14:24-15:10, 15:19-16:5. That position directly contradicts her prior, adamant opinion that TPR is measured in relation to a specific volume. EX2029 at 21-22. The Board should not countenance Dr. Robinson's and Apotex's continually-shifting positions as to this key claim term.

5. "redox buffer strength"

Claim 1 recites a "redox buffer strength." EX1001, Claim 1. The Board construed "thiol-pair buffer strength," which is not recited in any claim.

For purposes of this proceeding post-institution, Amgen applies the Board's construction of "thiol-pair buffer strength" to the claim term "redox buffer strength," which is recited in Claim 1; RBS, thus, means $2[oxidant] + [reductant]$. Institution Decision at 10. Amgen also interprets the Board's construction to require that the reductant and oxidant concentrations are determined in the refold buffer, just like the $\frac{[reductant]^2}{[oxidant]}$ equation[5].

[5] Amgen respectfully disagrees with the Board's construction. The parties agreed that the concentrations in $2[oxidant] + [reductant]$ are determined in the redox

Dr. Robinson, Apotex's expert, again takes the position that the Board's construction is irrelevant to this proceeding. At deposition, she insisted that "PTAB has not construed redox buffer strength" and that the Board instead construed a term "thiol-pair buffer strength" that does not appear in any claims. EX2019 at 19:2-8, 17:6-20. Dr. Robinson apparently agrees that RBS values are determined by the equation $2[oxidant] + [reductant]$, but twice she now claims to have no idea which volume to apply when calculating RBS values. *Id.* at 19:2-8; EX1002 at ¶79. That position directly contradicts her prior, adamant opinion that TPR is measured in relation to a specific volume. EX2029 at 23-24. The Board should not countenance Dr. Robinson's and Apotex's continually-shifting positions as to this key claim term.

B. Additional Claim Term to be Construed

1. "non-mammalian expression system"

Claim 1 recites "non-mammalian expression system." As in the POPR, Amgen respectfully proposes a construction for "non-mammalian expression system" that is taken directly from the definition in the specification:

> [T]he term "non-mammalian expression system" means a system for expressing proteins in cells derived from an organism other than a

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component. Petition at 25; POPR at 11-12. As discussed above in footnote 4, that agreed-upon construction is supported by the claim language.

mammal, including but not limited to, prokaryotes, including bacteria such as *E. coli* and yeast.

EX1001 at 4:63-67; EX2001 at ¶69; POPR at 17-18. That construction satisfies the broadest reasonable interpretation in light of the specification.

V. The Challenged Claims of the '138 Patent Are Not Obvious in View of the Combined Prior Art

The Board instituted review of the '138 Patent based on two Grounds: that (1) Claims 1-11 and 13-24 are rendered obvious over *Schlegl* and *Hevehan* and (2) Claim 12 is rendered obvious over *Schlegl*, *Hevehan*, and *Hakim*. For the reasons articulated below, those Grounds should be rejected because they suffer from fundamental deficiencies.

A. *Schlegl* and *Hevehan*, Alone or in Combination, Do Not Render Claims 1-11 and 13-24 Obvious

1. *Schlegl* and *Hevehan*, Alone or in Combination, Do Not Teach the TPR and RBS Equations

Based on Amgen's understanding of the Board's claim constructions, the equation ($\frac{[reductant]^2}{[oxidant]}$) is required to calculate TPR values. EX1001 at Claims 1-2. Likewise, the equation ($2[oxidant] + [reductant]$) is required to calculate RBS values. *Id.* at Claims 1, 3.

It is undisputed (and Dr. Robinson admits) that *Schlegl* and *Hevehan* do not teach the critical TPR and RBS equations. EX2019 at 40:12-21, 67:19-68:8; EX2020 at ¶¶18, 20. Without those equations, a POSITA cannot calculate

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

TPR and RBS values and determine whether those values fall within the claimed ranges of the ’138 Patent. It is readily apparent that Dr. Robinson applied impermissible hindsight in performing her obviousness assessment. EX2020 at ¶26. As she admitted at deposition, in order to calculate TPR and RBS values purportedly based on *Schlegl* and *Hevehan*’s disclosure, she applied TPR and RBS equations from the ’138 Patent. EX2019 at 43:23-44:23, 45:8-17, 68:11-69:2 (“I took the concentrations of DTT and GSSG disclosed in Hevehan and put them in the equation 1 from the '138 patent.”).

Because *Schlegl* and *Hevehan*, either alone or in combination, do not teach the threshold TPR and RBS equations required to calculate TPR and RBS values, *Schlegl* and *Hevehan* cannot teach the claimed TPR and RBS values required by Claim 1. Because Claims 2-11 and 13-24 depend from Claim 1 and share the same material limitations, those dependent claims are also not rendered obvious by *Schlegl* and *Hevehan*, either alone or in combination.

2. A POSITA Would Not be Motivated to Combine *Schlegl* and *Hevehan*

(a) *Schlegl* and *Hevehan* are Fundamentally Different and Incompatible Protein Refolding Methods

Schlegl and *Hevehan* are fundamentally different and incompatible approaches to protein refolding. *Schlegl* teaches a mechanical approach to achieve protein refolding at extremely dilute protein concentrations. POPR at 19-20, 33-

34; EX2001 at ¶¶93-99, 111-113; EX2020 at 23, n.7. To achieve "conditions that approximate ideal mixing," *Schlegl* combines a stream containing solubilized (unfolded) protein at a low flow rate with a refolding buffer stream at a very high flow rate. POPR at 19; EX2001 at ¶94; *see* EX1003 at [0023], [0024], [0033], [0037]. Under such conditions, refolding takes place at very low protein concentrations. POPR at 19; EX2001 at ¶95. According to *Schlegl*,

> By maintaining a very high flow rate of the refolding buffer and a low flow rate of the feed stream containing the unfolded protein, the method of the invention provides very high local dilution rates; preferred dilution rates range from 1:5 to 1:5000 and from 1:10 to 1:10000.
>
> * * *
>
> In the process of the invention, the actual protein concentration immediately after mixing is much lower as compared to conventional refolding methods.

EX1003 at [0033] (emphasis added), [0039] (emphasis added); EX2020 at ¶¶48-50. In sheer contrast, *Hevehan*'s method is a chemical approach (focused on denaturant and oxidant, but not reductant, in the refold buffer) to achieve protein refolding at high protein concentrations. POPR at 33; EX2001 at ¶¶107, 111.

It is illogical for a POSITA to combine *Schlegl*'s method of refolding at extremely dilute protein concentrations with *Hevehan*'s method of refolding at

high protein concentrations. EX2020 at ¶¶37-38. In *Schlegl*, protein aggregation is avoided by physically separating the protein molecules by dilution. POPR at 33; EX2001 at ¶112. In *Hevehan*, refolding proteins at high concentrations necessarily reduces or eliminates such physical separation; chemicals are necessary to avoid aggregation and to achieve proper refolding. *Id.*; EX1004 at 1 (". . . low recovery of correctly folded protein is often due to aggregation . . . The most direct means of minimizing aggregation is by decreasing protein concentration."); EX1003 at [0008] ("The higher the protein concentration, the higher the risk of intermolecular mis-folding, and vice versa."). *Hevehan* primarily relies on controlling the amount of denaturant (GdmCl) in the refold buffer in order to minimize protein aggregation, but not redox chemicals as in the '138 Patent. POPR at 33-34; EX2001 at ¶112; EX1004 at 2 ("In particular, addition of solubilizing agents [denaturant] in nondenaturing concentrations to the renaturation buffer seemed to be most effective at decelerating the rate of aggregation."); EX2020 at ¶23.

The Board proffers a reason why a POSITA would combine *Schlegl* and *Hevehan*:

> Schlegl's dilution approach itself suggests customizing the refolding buffer to be used for a particular protein. [sic] *Id.* at [0036], [sic] as well as the addition of refolding additives such as redox systems. *Id.* at [0041]. Hevehan optimizes those redox systems. Ex. 1004, 2.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

Institution Decision at 15 (emphasis added). The Board cites to *Schlegl*'s [0036] and [0041], which explicitly state that redox systems are "optional[]." EX1003 at [0036], [0041]; EX2020 at ¶21.

Amgen respectfully disagrees. *Hevehan* does not optimize *Schlegl*'s "optional[]" redox systems of [0036] and [0041]. EX2020 at ¶46. Instead, *Hevehan* teaches that redox systems appropriate for refolding at low protein concentrations are inappropriate when refolding at high protein concentrations. *Id.* at ¶22. *Hevehan* reports redox conditions for refolding protein at low protein concentrations of "0.01-0.1 mg/mL." *Id.* at ¶¶22, 46; EX1004 at 5. And yet, *Hevehan* did not adopt those reported conditions for its high protein concentration refolds because they "might not be appropriate when folding a protein at 1 mg/mL or higher concentrations." *Id.* Instead, *Hevehan* resorted to trial-and-error studies: selecting random concentrations of DTT (the reductant) in the protein-containing volume and GSSG (the oxidant) in the refold buffer.[6] EX2020 at ¶46. Based on

[6] Dr. Robinson testified that *Hevehan* chose the concentrations of DTT and GSSG based on the teachings of "Saxena and Wetlaufer, 1970." EX2019 at 71:3-72:5. Dr. Robinson is incorrect. *Hevehan* discloses that "Saxena and Wetlaufer, 1970" teaches concentrations of "0.004-0.4 mM" of GSSG. EX1004 at 5. Critically, *Hevehan* did not use those concentrations; she used between 4-13 mM of GSSG, which is 10 to 3,250 times more GSSG. *Id.*

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

the explicit teachings of *Hevehan*, a POSITA would likewise reject the optional redox conditions of *Schlegl*. Redox conditions for the extremely dilute refolding method of *Schlegl* would be viewed as "not [] appropriate" for the high concentration refolding method of *Hevehan*. EX2020 at ¶47. And just as in *Hevehan*, a POSITA would resort to trial-and-error studies to find appropriate redox conditions. *Id*. Indeed, *Hevehan* teaches that refold conditions that work at one protein concentration will not necessarily work at another. EX1004 at 5; EX2020 at ¶¶22, 46.

Moreover, as discussed in Amgen's POPR, a POSITA would see no benefit to combining *Schlegl* and *Hevehan*'s fundamentally different teachings. Adding *Hevehan*'s denaturant and oxidant chemicals to *Schlegl*'s dilute refolding method would have been viewed as making *Schlegl*'s process more costly and complicated. POPR at 34.

(b) The Methods of Refolding Pure, Model Proteins Disclosed in *Schlegl* and *Hevehan* are Not Applicable to Refolding Proteins Made in Non-Mammalian Expression Systems

Dr. Robinson asserted (and the Board accepted) that the refolding methods of *Hevehan* and *Schlegl* are "just as applicable to the refolding of proteins in inclusion bodies as to the proteins in denatured native proteins." Institution Decision at 13 (citation omitted). There is no merit to Dr. Robinson's contention. Literature (from the lab associated with *Hevehan* that Dr. Robinson admits is

authoritative) underscores the fact that the two protein systems are fundamentally different with respect to the challenges they pose for refolding.

As an initial matter, Claim 1 of the '138 Patent recites a method for refolding proteins expressed in "non-mammalian expression systems." There is no dispute that neither *Hevehan* nor *Schlegl* refolded such proteins. The HEWL used in *Hevehan* was not made in a non-mammalian expression system. It was "purchased" as a "purified protein," meaning the HEWL contained nominal, if any, contaminants. EX2019 at 72:6-14, 73:20-74:2 ("I would expect that [the HEWL used in *Hevehan* is] relatively pure"); EX2020 at ¶80. Even if *Hevehan*'s HEWL had been collected from its natural source, hen eggs, the HEWL was not made in a non-mammalian expression system; as Dr. Robinson acknowledged, a POSITA "wouldn't generally think of going to a hen cell for a non-mammalian system." EX2019 at 87:13-18; EX2020 at ¶¶78, 80. And Dr. Robinson repeatedly testified that the α-LA protein used in *Schlegl* was not made in a non-mammalian expression system. EX2019 at 46:23-48:4. Even if *Schlegl*'s α-LA had been obtained from its natural source, cow's milk, such α-LA would not be made in a non-mammalian expression system, since a cow is a mammal. *Id.* at 34:13-19; EX2020 at ¶79.

Critically, the pure, model proteins of *Hevehan* and *Schlegl* are fundamentally different than proteins made in non-mammalian expression systems,

as required by Claim 1 of the '138 Patent. EX2020 at ¶¶42, 84. Proteins made recombinantly in non-mammalian expression systems can be contaminated by other materials in host cells. *Id.* For example, proteins made in *E. coli* aggregate inside the host cell in so-called inclusion bodies; those insoluble aggregates include host-cell contaminants, such as "DNA, ribosomal RNA, phospholipids, lipopolysaccharides, and other proteins." EX2033 at Abstract; EX2019 at 24:25-26:8 ("in the inclusion body, there's protein, there's lipids, there's sometimes nucleic acid"), 27:13-28:1, 28:2-9 (typically, 20-65% of the inclusion body comprises contaminants); EX2020 at ¶42. The type and amount of host-cell contaminants vary depending on the protein being expressed and the conditions used. EX2020 at ¶42.

Notably, such host-cell contaminants—which are associated with proteins expressed in non-mammalian expression systems in inclusion bodies, but are absent from pure, model proteins—negatively impact refolded protein yields. *Id.*; EX2034 at 2 (showing that inclusion bodies isolated from *E. coli* expressing β-lactamase contained 35 to 95% pure protein depending on the conditions used). The same lab responsible for *Hevehan*—which Dr. Robinson agreed was an "authoritative one in relation to refolding of proteins"—studied this very issue. EX2019 at 66:23-67:18 (Dr. Robinson also testified that De Bernardez Clark's "lab at Tufts or some of the papers that she's published are considered classics in the

field."); EX2020 at ¶41. *Hevehan*'s colleagues investigated the negative impact that typical contaminants found in inclusion bodies had on the refold rate and yield of the very same pure, model protein as in *Hevehan*, HEWL. EX2033 at Title, Abstract, 3 (describing study of the "effect of typical contaminants in inclusion body preparations . . . on renaturation rate and yield of [HEWL]" at dilute protein concentrations (maximum of 0.1 mg/mL)); EX2020 at ¶43.

Hevehan's colleagues unequivocally demonstrated that even adding a single contaminating protein—*e.g.*, a common *E. coli* protein known as β-galactosidase—at low protein concentrations (0.1 mg/mL), significantly and adversely affected yield of refolded protein. EX2020 at ¶43. It decreased by 40% to 50%:

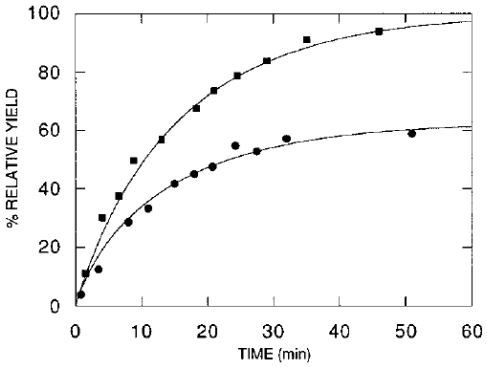


Figure 8. Effect of β-galactosidase on the kinetics of the oxidative renaturation of lysozyme. Renaturation was initiated by rapidly diluting a solution containing reduced−denatured lysozyme and β-galactosidase into renaturation buffer. Final conditions were 0.1 mg/mL lysozyme, 2 mM DTT, 5 mM GSSG, and 0.5 M GdmCl in 50 mM tris-HCl and 1 mM EDTA (pH 8, 22 °C), and (■) 0.0 mg/mL β-galactosidase and (●) 0.60 mg/mL β-galactosidase.

EX2033 at Figure 8; EX2020 at ¶43. Other contaminants (such as plasmid DNA, lipopolysaccharide, phosphatidylethanolamine) likewise negatively affected

protein yield. EX2033 at Figures 5-7; EX2020 at ¶43. The cumulative effect of all such contaminants present in inclusion bodies: there is dramatically less yield of refolded HEWL from inclusion bodies than from purified HEWL, as in *Hevehan*. EX2020 at ¶43. Contrary to Petitioners' and Dr. Robinson's positions, the very same lab responsible for *Hevehan* published that methods for refolding denatured pure, model proteins are not "just as applicable" to methods for refolding proteins in inclusion bodies; contaminants present in inclusion bodies confound and interfere with protein refolds. *Id.* at ¶¶43, 85-87.

Furthermore, it was known that protocols that can optimize refolding pure, model proteins (*e.g.*, HEWL and α-LA) are not predictive of results obtained when refolding recombinant proteins made in non-mammalian expression systems. *Id.* at ¶¶43, 86-87. For example, in one study, the authors found that "the efficiency of β-lactamase refolding was inversely proportional to the level of contaminants present in the inclusion body preparation." EX2034 at 2 (emphasis added); EX2020 at ¶43; *see also* EX2035 at 1-2.

Thus, the refolding methods for pure, model proteins as in *Hevehan* and *Schlegl* are not predictive of successfully refolding proteins made in non-mammalian expression systems. EX2020 at ¶¶44, 81-84. Petitioners have not met their burden; there is no evidence that the specific methods of *Hevehan* and *Schlegl*—based on pure, model proteins without contaminants—can successfully

refold proteins made from non-mammalian expression systems, with contaminants found in inclusion bodies.

The combination of *Schlegl* and *Hevehan* does not teach or suggest refolding of proteins "expressed in a non-mammalian expression system," as required by Claim 1. Because Claims 2-11 and 13-24 depend from Claim 1 and share the same material limitation, those dependent claims are also not rendered obvious by *Schlegl* and *Hevehan*, either alone or in combination.

3. Assuming, *Arguendo*, That a POSITA Would Combine *Schlegl* and *Hevehan*, That Combination Does Not Render Obvious Any Claim of the '138 Patent

(a) The Combination of *Schlegl* and *Hevehan* Does Not Teach the Claimed TPR Limitation

Even if a POSITA would combine *Schlegl* and *Hevehan*, that combination does not result in the claimed invention of Claim 1. The TPR value of that combination is zero, and falls outside the claimed TPR ranges. EX1001 at Claims 1, 3-24 ("0.001 to 100"), Claim 2 (lowest TPR value is "0.05").

Amgen understands that the Petitioners, Dr. Robinson, and the Board contend that a POSITA would use *Hevehan*'s redox conditions in *Schlegl*'s method to optimize *Schlegl*'s optional redox systems. Petition at 40 ("It follows that a POSITA in 2009 would have known that the teachings of Hevehan apply to the dilution refolding methods for refolding taught in Schlegl"); EX1002 at ¶114 ("Thus, in my view, it follows that one of ordinary skill in 2009 would have known

that the teachings of Hevehan apply to the dilution refolding methods for refolding the protein bovine α-lactalbumin as taught by Schlegl"); Institution decision at 15 ("Schlegl's dilution approach itself suggests customizing the refold buffer . . . Hevehan optimizes those redox systems".); EX2020 at ¶53.

But *Hevehan* explicitly teaches that there is no reductant in the refold buffer. EX2020 at ¶54. *Hevehan* teaches two volumes: a protein-containing volume and a refold buffer (called the renaturation media). EX2019 at 74:20-75:3; EX2020 at ¶55; EX1004 at 2-3. *Hevehan*'s protein-containing volume contains, in relevant part, HEWL (the protein) and DTT (which Dr. Robinson identifies as the reductant). EX1004 at 2; EX1002 at ¶68, fn. 5; EX2020 at ¶55. *Hevehan*'s refold buffer contains Tris-HCl, EDTA, GSSG (the oxidant), and possibly some GdmCl and L-arginine—none of which are reductants. EX1004 at 2-3; EX2019 at 75:4-25 (GdmCl and L-arginine are not redox chemicals); EX2020 at ¶55 (Tris-HCl and EDTA are not redox chemicals).

Critically, *Hevehan* explicitly teaches that the reductant is not necessary in the refold (renaturation) buffer:

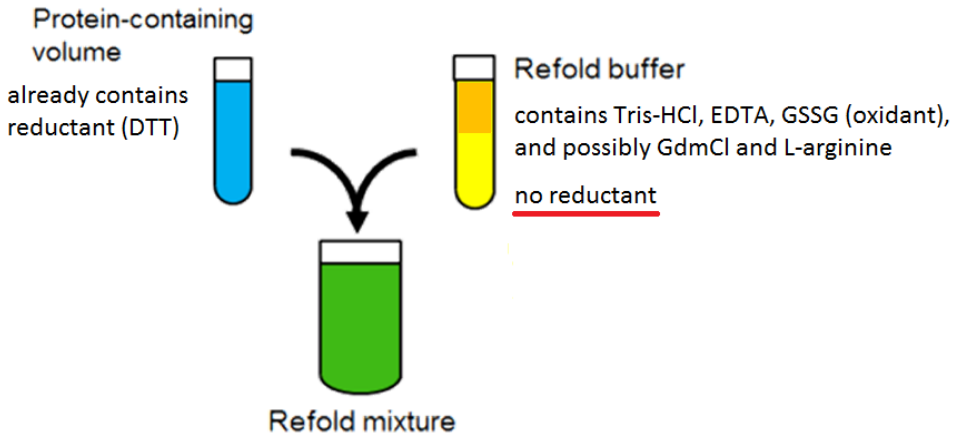
> Addition of GSSG's reducing partner, GSH, to the renaturation system was not necessary due to the DTT carried over from the denatured [protein] solution.

EX1004 at 3; EX2019 at 77:8-16. And Dr. Robinson admitted at deposition that there is no DTT reductant in the refold buffer. EX2019 at 76:1-5; EX2020 at ¶56.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

Thus, in *Hevehan*, a protein-containing volume containing a reductant (DTT) is contacted with a refold buffer containing only an oxidant, GSSG, but not a reductant (GSSG's partner GSH or DTT)[7]. EX2001 at ¶109.

A diagram of *Hevehan* is depicted below:



EX2020 at ¶55.

Hevehan's reliance on the DTT "carried over from the denatured [protein] solution" reflects the fact that *Hevehan* did not appreciate the significance of carefully controlling redox chemicals in a refold buffer. EX2020 at ¶23. Some DTT added to the solubilization/protein-containing volume will be irreversibly consumed to denature (unfold) the protein; it will be unavailable as a reductant in

[7] The De Bernardez Clark lab preferred no reductant in the refold buffer, because it reduced the use of costly redox-chemicals such as GSSG. EX2020 at ¶57; EX2033 at 2-3; EX1020 at 4. Based on *Hevehan*'s teachings, a POSITA would eliminate reductant from the refold buffer as a cost-saving measure.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

the refold mixture. *Id.* That amount was neither measured nor calculated in *Hevehan*—meaning that there was an unknown, uncontrolled amount of reductant carried over to the refold mixture. *Id.*

It follows that a POSITA would not arrive at Claim 1 of the '138 Patent by using *Hevehan*'s redox conditions in *Schlegl*'s method to optimize *Schlegl*'s optional redox systems. EX2020 at ¶¶59-63. As discussed above, Amgen understands that the Board construed "final thiol-pair ratio" as $\frac{[reductant]^2}{[oxidant]}$, wherein the concentrations are calculated in the refold buffer. *Id.* Because that combination has no reductant in the refold buffer, the TPR value must be zero (*i.e.*, $\left(\frac{[reductant]^2}{[oxidant]}\right) = \left(\frac{[0]^2}{[oxidant]}\right) = 0$), which falls outside of the claimed range of "0.001 to 100." EX1001 at Claim 1; EX2020 at ¶¶61-63. Because Claims 2-11 and 13-24 depend from Claim 1 and share the same material limitation, those dependent claims are also not rendered obvious by the combination of *Schlegl* and *Hevehan*.

(b) The Combination of *Schlegl* and *Hevehan* Does Not Provide a Reasonable Expectation of Success When Refolding Proteins Expressed in a Non-Mammalian Expression System

As discussed in section V.A.2.(b), the methods of refolding pure, model proteins of *Schlegl* and *Hevehan* are not applicable to refolding proteins made in non-mammalian expression systems. Assuming, *arguendo*, that *Schlegl*

and *Hevehan*'s methods can be applied to refolding protein in non-mammalian expression systems, there is no reasonable expectation of success that those methods—tested on pure, model proteins—would successfully refold proteins expressed in non-mammalian expression systems. EX2020 at ¶64.

(i) A POSITA Understands That Redox Chemicals Do Not Play a Role in *Schlegl*'s Refolding Method

The only protein studied in *Schlegl*'s sole example is α-LA. EX1003 at [0073]; EX2020 at ¶65. *Schlegl* explicitly discloses that α-LA is a "model protein" with an "oxidative pathway [that] is well characterized." EX2020 at ¶65; EX2038 at 1; EX2036 at 1. *Schlegl*'s α-LA is most likely a purified protein, and not one made in a non-mammalian expression system. EX2020 at ¶79.

As of the priority date of the '138 Patent, a POSITA knew that α-LA was not difficult to refold. EX2020 at ¶66. A POSITA also knew that calcium drives refolding of α-LA. *Id.* In a process referred to as ligand-assisted refolding, calcium (the ligand) forms a complex with α-LA; because calcium binds "very tightly to the native [α-LA] confirmation," it promotes refolding of α-LA to its native structure. EX2039 at 17-18; EX2020 at ¶66; EX2031 at 3. At deposition, Dr. Robinson acknowledged that α-LA "<u>needs</u> calcium" to refold. EX2019 at 35:20-36:4 (emphasis added). And *Schlegl* itself acknowledged the significance of calcium to refolding α-LA: it teaches that α-LA "has an additional calcium-

binding site, which increases the stability of the native protein" (*see* EX1003 at [0073]) and "prevent[s] off-pathway reactions" (*see* EX2031 at 3). EX2020 at ¶66.

There is no evidence that the optional redox chemicals disclosed in *Schlegl* had any impact on refolding α-LA. *Schlegl* instead takes advantage of a ligand-assisted refolding method—which is distinct from redox chemical-based methods that promote proper disulfide bond formation—with calcium acting as the ligand. EX2020 at ¶67. First, to denature α-LA, *Schlegl* adds EDTA to remove calcium stabilizing the native α-LA; but there is no evidence that 1 mM EDTA is enough to denature all of the α-LA—such that there could be some retained, properly folded α-LA. EX1003 at [0074]; EX2020 at ¶67; EX2040 at 1 ("adding 3.5 mM EDTA to remove bound Ca^{2+}"). *Schlegl* then adds 5 mM of $CaCl_2$ (which includes calcium ions, the ligand) to the refold buffer to drive refolding of the subset of α-LA that was denatured with EDTA. EX1003 at [0075]; EX2020 at ¶67.

Indeed, redox chemicals should have <u>little to no</u> effect on refolding α-LA. The purpose of redox chemicals is to form and reshuffle disulfide bonds to make properly refolded proteins. EX2020 at ¶68. But, well before *Schlegl*, it was known that disulfide bond formation contributed very little, if any, to successful refolds of α-LA.

> It is interesting to note that an α-LA mutant in which all eight cysteines were mutated to alanine, was nearly as compact as wild-type α-LA at acidic pH . . . . Overall, the architecture of the protein fold of α-LA is determined by the polypeptide sequence itself and not as a result of disulfide bond cross-linking.

EX2038 at 4; EX2020 at ¶68; EX2036 at 1 ("The α-lactalbumin molten globule state is shown largely to result from nonspecific hydrophobic collapse, to be devoid of cooperative or specific tertiary interactions, and not to be stabilized substantially by the native or rearranged disulfide bonds.") (Emphasis added).

Schlegl has only one example of its protein refolding method. In that sole example, *Schlegl* applies its method to just one, unique protein—purified α-LA; that protein uses calcium ions and does not require redox chemicals to reshuffle disulfide bonds in order to refold. EX2020 at ¶69. A POSITA would not find that sole example predictive of refolding other proteins. *Id.* A POSITA would have no reasonable expectation of success in applying *Schlegl*'s method to refolding proteins made in non-mammalian expression systems with redox chemicals, as required by Claims 1-11 and 13-24 of the '138 Patent.

(ii) A POSITA Understands That *Hevehan* has Been Refuted and Applies to Only Hen Egg White Lysozyme

There would have been no motivation for a POSITA to combine *Hevehan* with *Schlegl*. *Hevehan* is a study of the reaction pathways for refolding a pure, model protein, HEWL. EX1004 at 6-8, Figure 7; EX2020 at ¶70. *Hevehan*

is a "quantitative analysis of rates leading to both reactivation [refolding] and aggregation" of HEWL and reports on a kinetic model for HEWL refolding, including refolding and aggregation rate constants. EX2020 at ¶71. *Schlegl* teaches protein refolding at the industry scale, *e.g.*, by disclosing refolding tanks. EX1003 at Figures 1-3. And yet, it was known prior to *Schlegl* that *Hevehan*'s kinetics model was fundamentally flawed and with limited to no applicability to the industry scale. EX2020 at ¶72. Buswell et al., made the following pointed criticisms of *Hevehan*:

- *Hevehan* is not predictive of fed-batch refolding processes, which Dr. Robinson acknowledged at deposition are disclosed in *Schlegl*. EX2019 at 40:7-11. *Hevehan*'s kinetic model is simplistic and assumes first-order kinetics for the refolding pathway and third-order kinetics for the aggregation pathway. EX1004 at Abstract; EX2020 at ¶72. But Buswell determined that *Hevehan*'s scheme of "competing first- and third-order reaction [] for lysozyme is shown to <u>not predict fed-batch lysozyme refolding</u> when the model is parameterized using independent bath experiments, even when variations in chemical compositions during the fed-batch experiment are accounted for." EX2042 at Abstract (emphasis added); EX2020 at ¶72.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

- *Hevehan* is not predictive of yield. Actual experimental data shows that the *Hevehan* model overestimates protein yield by 50%, even at low protein concentrations. EX2042 at 8, Figure 6; EX2020 at ¶72.

Dr. Willson, Amgen's expert, identifies additional flaws with *Hevehan*'s model:

- *Hevehan*'s kinetics model incorrectly assumes that each step is irreversible and proceeds in only one direction. EX2020 at ¶74; EX1004 at 8, Figure 7. But the folding of many proteins includes reversible steps where a species can revert back to its precursor. EX2020 at ¶74; EX2045 at 2, Fig. 1 (depicting the folding pathway of BPTI, with arrows indicating that the steps are reversible).
- *Hevehan*'s assumption that the aggregation pathway follows third-order kinetics does not apply to all protein aggregation pathways. *See generally* EX2043 (noting that protein aggregation processes can involve multiple steps, and different protein aggregation processes can follow different mechanisms); see also EX2044 at Abstract (describing actin nucleation and polymerization kinetics); EX2020 at ¶73.
- *Hevehan* incorrectly assumes that only proteins in the intermediate state (between folded and unfolded) aggregate. EX2020 at ¶74. But there are numerous examples of proteins for which aggregation can start from the folded or unfolded state. EX2020 at ¶74; EX2046 at 1 (noting that the

folded monomer of transthyretin "rapidly undergoes partial denaturation and self assembles into amyloid [a protein aggregate] when subjected to a mild denaturation stress"); EX2047 at 1, 6, Fig. 7 (describing the unfolding and aggregation of two naturally occurring human lysozyme variants); EX2048 at 1 (describing aggregation of interleukin 1β as occurring from an unfolded state, as opposed to solely from an intermediate state). Even properly folded HEWL can form aggregates. EX2020 at ¶74; EX2042 at 1.

- *Hevehan* incorrectly assumes that there is a single pathway for converting one protein state (*e.g.* folded) to another (*e.g.*, unfolded). EX2020 at ¶75; EX1004 at 8, Figure 7. But generally (including when refolding HEWL) multiple, parallel pathways convert one protein state to another. EX2020 at ¶75; EX2049 at 1 (concluding that folding of lysozyme "is not a simple sequential assembly process but involves parallel alternative pathways, some of which may involve substantial reorganization steps"); EX2039 at 9, 15, 19 (noting that hen lysozyme "appears to follow a parallel-channel mechanism," and more generally, "folding is likely to be a complex combination of both multiple and unique pathways").

Given that it was known prior to *Schlegl* that *Hevehan* does not accurately predict refolding of its own model protein, HEWL, a POSITA would not have applied *Hevehan*'s teachings to refolding any other proteins. EX2020 at

¶76. A POSITA would have no reasonable expectation of success that the *Hevehan* method would be successful for a protein made in non-mammalian expression systems, as required by Claims 1-11 and 13-24 of the '138 Patent.

4. *Schlegl* and *Hevehan*, Alone or in Combination, Do Not Render Claims 9-11 Obvious

In addition to the reasons stated above with respect to Claim 1, there are additional reasons why *Schlegl* and *Hevehan*, alone or in combination, do not render obvious dependent Claims 9-11.

Claim 10 recites "[t]he method of claim 1, wherein the protein is a complex protein." EX1001 at Claim 10. As discussed above in section IV.A.2., the Board has construed "complex protein" to mean either (1) larger than 20,000 MW with two or more disulfide bonds in its native form or (2) comprises greater than 250 amino acid residues with two or more disulfide bonds in its native form. EX2020 at ¶¶9, 90. Claims 9 and 11 further limit the "protein" in Claim 1 to "an antibody" and "a multimeric protein," respectively. EX1001 at Claims 9, 11.

Schlegl refolded only one protein, α-LA; *Hevehan* refolded only one protein, HEWL. As discussed in Amgen's POPR (at 43-48), neither *Schlegl* nor *Hevehan* teaches or suggests "a complex protein" (Claim 10), "an antibody" (Claim 9), and/or "a multimeric protein" (Claim 11). There is no dispute on this point: Dr. Robinson readily admitted at her deposition that α-LA and HEWL are

not "complex protein[s]" (EX2019 at 92:7-15, 90:10-25); not "antibod[ies]" (*id.* at 65:5-10); and not "multimeric protein[s]" (*id.* at 65:11-15).

Notably, Petitioners point to no empirical evidence that a POSITA would have had a reasonable expectation of success of refolding "complex protein[s]," "antibody[ies]," or "multimeric protein[s]" using the methods taught in *Schlegl* and *Hevehan*. EX2020 at ¶¶94, 97.

Petitioners find significant *Schlegl*'s broad overstatement that its methods can be applied to "any protein, protein fragment or peptide that requires refolding upon recombinant expression in order to obtain such protein in its biologically active form." EX1003 at [0031]. Nothing in *Schlegl* supports that blanket statement. EX2020 at ¶94. In fact, a POSITA knows that refolding complex proteins with many disulfide bonds—such as complex proteins, antibodies, and multimeric proteins—is "extremely difficult." EX2051 at 3; EX2020 at ¶98. Dr. Robinson testified that "another class of proteins that are challenging to refold are things with more than one disulfide bond because of the challenges of forming cross-disulfide or mixed-disulfide bonds." EX2019 at 60:6-10 (emphasis added). Based on math alone, more disulfide bonds means more possibilities of mis-matched disulfide bonds resulting in more mis-folded protein species. EX2020 at ¶98; EX2052 at 6, Table 1 (*e.g.*, a protein with 2 disulfide bonds has 3 possible disulfide bond patterns, one of which is correct, whereas a

protein with 4 disulfide bonds has 105 possible disulfide bond patterns, only one of which is correct).

Dr. Robinson, on re-direct, attempted to bolster her testimony as to *Schlegl*'s disclosure in relation to Claim 10. In response to Petitioners' questioning, she unequivocally testified that *Schlegl* teaches a "complex protein" (Claim 10), ovalbumin in [0010]. EX2019 at 153:23-154:22 ("Ovalbumin is a complex molecule by the definition of the '138 Patent"). Despite assessing ovalbumin "[o]ver the weekend" immediately prior to her Monday deposition, she could not recall the number of disulfide bonds in ovalbumin on re-cross-examination. EX2019 at 155:2-14. And yet, ovalbumin has only <u>one</u> disulfide bond. EX2020 at 52, n.24; EX2050 at Abstract. It is <u>not</u> a "complex protein," which requires two or more disulfide bonds. *Id.* Apparently realizing this error after-the-fact, Petitioners informed Amgen over a week later that Dr. Robinson's testimony is "incorrect" and "[h]er testimony in response to the question [posed by Petitioners' attorney whether ovalbumin is a complex protein by the definition of the '138 Patent] is no." EX2054.

In any event, there is no suggestion in *Schlegl* that any proteins apart from α-LA were refolded with the disclosed methodology. In [0010], *Schlegl* merely identifies examples of proteins that are known to have a "burst phase" during refolding: "[i]mmediately after initiation of the folding reaction, the

unfolded protein collapses and a partly structured intermediate is formed." EX1003 at [0010]; EX2020 at 52, n.24.

Because *Schlegl* and *Hevehan* do not teach refolding "complex protein[s]," "antibody[ies]," or "multimeric protein[s]," Petitioners cite only *Hakim* for their bald assertion that "a POSITA "would immediately recognize that the methods taught by Schlegl could be applied" to the complex proteins, antibodies, and multimeric proteins of Claims 9-11. Institution Decision at 25-26 (citing EX1002 at ¶145 and EX1006 at 2).

As an initial matter, there is no merit to Petitioners' *Hakim*-based contentions. As discussed *infra* at section V.B.1., *Hakim* is not prior art to the '138 Patent; as of February 26, 2009, the inventors had reduced to practice refolding AMG 745, prior to *Hakim*'s publication date of May 1, 2009. AMG 745 is a "complex protein" per Claim 10; it has 510 amino acids, a theoretical molecular mass of 57,099 Daltons, 2 interchain disulfide bonds, and 3 intrachain disulfide bonds in each polypeptide chain. EX2021 at ¶37; EX2027 at [0312]; EX2020 at ¶99. AMG 745 is a "multimeric protein" per Claim 11; it has "2 identical polypeptide chains, which are covalently linked through disulfide bonds." *Id.*

Even assuming, *arguendo*, that *Hakim* qualifies as prior art, *Hakim* provides no rationale for applying the refold methods of *Schlegl* and *Hevehan* to complex proteins (Claim 10), antibodies (Claim 9) or multimeric proteins (Claim

11). EX2020 at ¶96. *Hakim* lacks a critical disclosure; it provides no details about its refolding methodology. *Id.*; EX1006 at 3 (stating only that "Refolding was initiated after mixing 50 mg of heavy chain and 50 mg of light chain inclusion bodies protein and reducing the mixture with 1,4-dithioerythritol (DTE)."); *id.* at 7 ("The inclusion bodies were completely solubilized in 6 M guanidine hydrochloride, 50 mM Tris (HCl) pH 8.0, 20 mM EDTA, mixed, reduced and refolded essentially as described."). (And, as Dr. Robinson readily admits (*see* EX2019 at 83:10-21), *Hakim* does not teach the TPR and RBS equations.) Without details as to *Hakim*'s refolding methods, there is no way for a POSITA to determine whether the methods of *Schlegl* or *Hevehan* are even compatible with *Hakim*'s protein. *Id*. As Dr. Robinson testified at deposition, "all proteins are unique." EX2019 at 59:12.

For the additional reasons above, *Schlegl* and *Hevehan*, either alone or in combination, do not render obvious Claims 9-11 of the '138 Patent. And *Hakim* does not support Petitioners' conclusory statement that a POSITA "would immediately recognize" that *Schlegl*'s methods could apply to such proteins. Institution Decision at 25-26 (citing EX1002 at ¶145 and EX1006 at 2).

5. *Schlegl* and *Hevehan*, Alone or in Combination, Do Not Render Claim 18 Obvious

Claim 18 recites "[t]he method of claim 1, wherein the incubation is performed under non-aerobic conditions," *i.e.*, conditions without oxygen.

EX2020 at ¶101. Claim 1 recites "incubating the refold mixture"; it follows that Claim 18's incubation under non-aerobic conditions is performed after the refold mixture is made. In addition to the reasons stated above with respect to Claim 1, there are more reasons why *Schlegl* and *Hevehan*, alone or in combination, do not render obvious dependent Claim 18.

Redox chemicals play a critical role in anaerobic refolding conditions. Oxygen is an oxidant and aids in protein refolding. EX2019 at 56:5-17; EX2020 at ¶102. But as refold volumes increase (such as in industrial settings), it becomes increasingly more difficult to ensure that oxygen will dissolve in the refold mixture to aid in refolding protein; the refold mixture becomes increasingly more anaerobic and redox chemicals are necessary to compensate for increasingly less oxygen (that would otherwise contribute to refolding protein). EX2020 at ¶103.

Petitioners incorrectly assert that the combination of *Schlegl* and *Hevehan* teaches that "incubation is performed under non-aerobic conditions." Petitioners do not rely on *Schlegl* for this limitation. At deposition, Dr. Robinson testified that *Schlegl* is "silent on the presence or absence of oxygen." EX2019 at 54:20-55:2. In fact, *Schlegl*'s figures make abundantly clear that the refolding tanks are open to air, *i.e.*, under aerobic conditions. EX1003 at Figures 1-3. EX2020 at ¶104. In support of their contention, Petitioners cite to a passage in *Hevehan* under the heading "Materials" that states "[s]olutions of reduced DTT

were prepared immediately prior to each experiment to minimize air oxidation." Petition at 55 (emphasis added); EX1004 at 2; EX2020 at ¶105. At deposition, Dr. Robinson agreed that the *Hevehan* passage "teach[es] preparing solutions of reduced DTT to minimize DTT from air oxidation."[8] EX2019 at 82:17-20 (emphasis added); EX2020 at ¶106. As indicated by the *Hevehan* passage's reference to "immediately prior to each experiment," this passage teaches a preliminary step, taken before denaturing (unfolding) protein to make the protein-containing volume. EX1004 at 2; EX2020 at ¶107. Critically, this passage bears no relationship to incubation, let alone incubating under anaerobic conditions; incubation refers to a much later step of protein refolding, after the refold mixture is made. EX2020 at ¶107; EX1001 at Claim 1 ("incubating the refold mixture"). Indeed, Dr. Robinson agreed at deposition that the *Hevehan* passage is "not teaching minimizing air oxidation in the context of protein refolding." EX2019 at 82:21-24 (emphasis added); EX2020 at ¶108.

[8] It is important to minimize oxidation of DTT, a reductant. Not only does DTT oxidize quickly, but it also oxidizes irreversibly: once it oxidizes, it is no longer available as a reductant. EX2021 at ¶21; EX2019 at 82:4 ("DTT is highly reactive").

In any event, *Hevehan* acknowledges that its refolding method is exposed to air. *Hevehan* teaches that "air oxidation plays a minimal role in these experiments." EX1004 at 5 (emphasis added); EX2020 at ¶104.

The Board appears to rely on Exhibits 1020 and 1028 (both of which mention "fermentation") as support for why the combination of *Schlegl* and *Hevehan* teaches incubating the refold mixture at non-aerobic conditions. Institution Decision at 29-30. Amgen respectfully disagrees. Exhibits 1020 and 1028 discuss fermentation processes for producing the recombinant protein in the non-mammalian host cell; those fermentation processes occur well before incubating the refold mixture to refold mis-folded recombinant protein, as required by Claim 18. EX2020 at ¶110; EX1020 at 3 (fermentation mentioned under the heading "Inclusion body isolation, purification and solubilization," which is before any step in Figure 1); EX1028 at 6 (fermentation discussed under the heading "*E. coli* expression systems and pathways" and disclosed as "aerobic fermentation process").[9]

[9] To the extent the Board is relying on the following passage as reflecting incubation of the refold mixture at non-aerobic conditions, Amgen respectfully disagrees: "solubilization solution to prevent metal-catalyzed air oxidation of cysteines." EX1028 at 3. The solubilization solution is the protein-containing volume; it is not the refold mixture. EX2020 at ¶111.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

There is no evidence that the combination of *Schlegl* and *Hevehan* teaches Claim 18. EX2020 at ¶112. Petitioner cannot and does not articulate how that combination bears any relevance to anaerobic incubation of the refold mixture.

B. *Schlegl*, *Hevehan*, and *Hakim*, Alone or in Combination, Do Not Render Claim 12 Obvious

1. *Hakim* is Not Prior Art to the '138 Patent

Claim 12 recites "[t]he method of claim 1, wherein the protein is an Fc-protein conjugate." An Fc-protein conjugate is a protein comprising an Fc region[10] joined to another protein. Petitioners do not assert that *Schlegl* or *Hevehan* teach methods for refolding an Fc-protein conjugate. Petition at 56-58. And Dr. Robinson readily admits that the proteins refolded in *Schlegl* (α-LA) and *Hevehan* (HEWL) are not Fc-protein conjugates. EX2019 at 65:16-20. Instead, Petitioners rely on *Hakim* for its teaching of Fc-protein conjugates.

Petitioners obviousness contention is flawed: *Hakim* is not prior art. Amgen's inventors fully reduced to practice the invention of Claim 12 as of at least February 26, 2009, which predates *Hakim*'s publication date of May 1, 2009.

Amgen can antedate and strike *Hakim* as prior art by showing, before May 1, 2009 (*Hakim*'s publication date), (1) performance of a process that met all

[10] An antibody, also known as an immunoglobulin (Ig), is a Y-shaped protein. The Fc region is the "tail end" or bottom part of the Y.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

the limitations of the claim; (2) that the invention would work for its intended purpose, and (3) sufficient evidence to corroborate the inventor's testimony. *Medichem, S.A. v. Rolabo, S.L.*, 437 F.3d 1157, 1169 (Fed. Cir. 2006); *Green Cross Corp. v. Shire Human Genetic Therapeutics, Inc.*, IPR2016-00258, Paper 89, 11 (P.T.A.B. Mar. 22, 2017). And yet, "no similar condition of 'corroboration' is imposed on . . . any documentary or physical evidence, as a condition for its serving as evidence of reduction to practice." *Medichem*, 437 F.3d at 1169. For actual reduction to practice, there must also be "contemporaneous appreciation of the invention at issue by the inventor." *Cooper v. Goldfarb*, 154 F.3d 1321 (Fed. Cir. 1998). But recognition of the invention "does not need to . . . [be] 'in the same terms as those recited' in the patent claims." *Fox Grp., Inc. v. Cree, Inc.*, 700 F.3d 1300, 1305 (Fed. Cir. 2012) (quoting *Dow Chem. Co. v. Astro-Valcour, Inc.*, 267 F.3d 1334, 1341 (Fed.Cir.2001)). Showing reduction to practice prior to the critical date does not require a showing of conception, which is "only [necessary] if the alleged prior inventor had not successfully reduced the invention to practice before the critical date of the patent-at-issue." *Fox*, 700 F.3d at 1304.

(a) Documentary Evidence Establishes That the '138 Patent Inventors Actually Reduced the Method of Claim 12 to Practice by at Least February 26, 2009

Every limitation of Claim 12 is reflected in a powerpoint presentation [redacted] that identifies the three '138 Patent

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

inventors (and others) as co-authors, and was last modified on February 26, 2009 ("Feb. 2009 Presentation"). EX2022 at 1; EX2023 at 1 ("DateModified" is "Feb 26, 2009 16:06:01"); EX2021 at ¶33. A more detailed description of the method of Claim 12 is reflected in an earlier powerpoint presentation [redacted] that identifies three authors, one of which is a '138 Patent inventor (Nick Keener), and was dated September 16, 2008, but was last modified on September 15, 2008 ("Sept. 2008 Presentation"). EX2024 at 1; EX2025 at 1 ("DateModified" is "Sept 15 2008 16:50:06"); EX2021 at ¶33.

(i) "A method of refolding [an Fc-protein conjugate]"

AMG 745 is an Fc-protein conjugate, as required by Claim 12. It comprises "a human Fc at the N terminus and a myostatin-neutralizing bioactive peptide at the C terminus." EX2026 at 2, 6 ("AMG 745 is a novel antimyostatin peptibody. A peptibody represents the component peptide (the pepti) and the Fc portion of an immunoglobulin in an overall structure that resembles an antibody . . ."); EX2021 at ¶36. AMG 745 resembles an antibody:

[redacted]

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

EX2022 at 16; EX2021 at ¶36.

(ii) "[An Fc-protein conjugate] expressed in a non-mammalian expression system"

AMG 745 was manufactured in a non-mammalian expression system, as required by Claim 12. The Feb. 2009 Presentation describes [redacted]

[redacted]

EX2022 at 2; EX2021 at ¶38. The first step is to make AMG 745 [redacted] *Id.* As discussed above, recombinant proteins made in non-mammalian expression systems [redacted] form inclusion bodies; the inclusion bodies containing AMG 745 were [redacted] *Id.* The Feb. 2009 Presentation describes the denaturation/solubilization of AMG 745 in [redacted]

EX2024 at 5 (emphasis added); EX2022 at 24; EX2021 at ¶39.

(iii) "[An Fc-protein conjugate] present in a volume at a concentration of 2.0 g/L or greater comprising: (a) contacting the protein with a refold buffer . . . to form a refold mixture;"

The Feb. 2009 Presentation discloses that a volume containing protein (AMG 745) was contacted with a refold buffer to form a refold mixture, as required by Claim 12. The volume containing [redacted] of AMG 745 (which meets the limitation of an Fc-protein conjugate "present in a volume at a concentration of 2.0 g/L or greater comprising," as required by Claim 12) was contacted with a refold buffer [redacted] to form a refold mixture [redacted], as required by Claim 12:

EX2024 at 5 (emphasis added); EX2022 at 24; EX2021 at ¶39.

(iv) "a refold buffer comprising a redox component . . . and one or more of: (i) a denaturant; (ii) an aggregation suppressor; and (iii) a protein stabilizer;"

The Feb. 2009 Presentation shows a refold buffer comprising redox components and at least an aggregation suppressor, as required by Claim 12. The refold buffer [redacted] comprises an oxidant [redacted] and a reductant [redacted]:

[redacted]

EX2022 at 20 (emphasis added), 18; EX2021 at ¶40. The refold buffer further comprises an [redacted], which is an aggregation suppressor. EX1001 at Claims 1, 12, 15-16 (listing [redacted] as an "aggregation suppressor"). The Feb. 2009 Presentation discloses [redacted]

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

[redacted]

EX2022 at 4 (emphasis added); EX2021 at ¶41. The Feb. 2009 Presentation also discloses [redacted]

EX2022 at 25 [redacted] (emphasis added); EX2024 at 12-13; EX2021 at ¶41. There is no [redacted] in the protein-containing volume; thus, when the protein-containing volume contacted the refold buffer to form the refold mixture, the [redacted] must have come from the refold buffer. EX2022 at 4 (emphasis added); EX2021 at ¶41.

(v) ". . . comprising a final thiol-pair ratio having a range of 0.001 to 100 and a redox buffer strength of 2 mM or greater . . ."

The Feb. 2009 Presentation describes TPR and RBS values that fall within the ranges claimed in Claim 12. The TPR and RBS values were calculated using the TPR and RBS equations ($\frac{[reductant]^2}{[oxidant]}$ and $2[oxidant] + [reductant]$, respectively) taught in the '138 Patent:

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

EX2022 at 19; EX2021 at ¶40. The TPR values were [redacted], which is within the claimed TPR range of "0.001 to 100," and the RBS values were [redacted], which is within the claimed RBS range of "2 mM or greater":

EX2022 at 21; *see also* EX2024 at 7-11; EX2021 at ¶40.

(vi) "(b) incubating the refold mixture; and (c) isolating the protein from the refold mixture."

The Feb. 2009 Presentation discloses the final two steps of Claim 12. The refold mixture incubates the refold mixture containing AMG 745 for [redacted]

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

[redacted]

EX2022 at 25 *see also* EX2024 at 12-13; EX2021 at ¶42. Then the protein is isolated using [redacted]

[redacted]

EX2024 at 5; EX2022 at 24; EX2021 at ¶43.

(b) The Inventors Contemporaneously Appreciated the '138 Patent Invention

As discussed above in section II, prior to the '138 Patent invention, optimizing redox conditions was done by trial-and-error. Dr. Roger Hart, one of the co-inventors, defined a unique equation for TPR ($\frac{[reductant]^2}{[oxidant]}$). EX2021 at ¶18. By taking the relationship between TPR (as defined by the inventors) and RBS

(defined as $2[oxidant] + [reductant]$) into account, the '138 Patent inventors surprisingly and unexpectedly provided greater predictability in identifying optimal conditions for refolding proteins at high protein concentrations. *Id.* at ¶44. In particular, as of September 2008, the inventors discovered and appreciated that

EX2024 at 8; EX2021 at ¶44. Specifically,

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

EX2024 at 11; EX2021 at ¶44. That same appreciation appears again in the Feb. 2009 Presentation. EX2022 at 23; EX2021 at ¶44.

2. *Hakim* Does Not Cure the Deficiencies of *Schlegl* and *Hevehan*

Even if the Board deems *Hakim* to be prior art to the '138 Patent, the combination of *Schlegl*, *Hevehan*, and *Hakim* does not render Claim 12 obvious. As discussed above in sections V.A.1. to V.A.3, *Schlegl* and *Hevehan* do not render obvious Claim 1; *Hakim* cannot cure the deficiencies of *Schlegl* and *Hevehan*.

As discussed above in section V.A.1., neither *Schlegl* nor *Hevehan* teaches the TPR and RBS equations ($\frac{[reductant]^2}{[oxidant]}$ and $2[oxidant] + [reductant]$, respectively). As Dr. Robinson readily admitted at deposition, neither does *Hakim*.

EX2019 at 83:10-21; EX2020 at ¶117. *Hakim* also provides no details regarding its refolding methodology; without that critical information, a POSITA does not know whether the methods of *Schlegl* or *Hevehan* are applicable to refolding *Hakim*'s protein. EX1006 at 3, 7; EX2020 at ¶117. As Dr. Robinson testified at deposition, "all proteins are unique." EX2019 at 59:12. And a POSITA knows that refolding complex proteins with many disulfide bonds, such as Fc-protein conjugates, is "extremely difficult." EX2051 at 3; EX2020 at ¶118; *see also* EX2019 at 60:6-10.

Further, a POSITA would not combine the narrow teachings of *Schlegl* and/or *Hevehan* with *Hakim*. As discussed above in section V.A.3.(b), a POSITA understands that *Schlegl*'s refolding of α-LA is not predictive of refolding other proteins (including a complex, multimeric protein like Fc-protein conjugates), and *Hevehan*'s refolding method has not only been soundly refuted but also applies just to HEWL. In view of the fact that *Schlegl* and *Hevehan* would not be viewed by POSITAs as applicable to refolding complex, multimeric proteins made in non-mammalian expression systems (*see* section V.A.3.(b)), a POSITA would have had no reasonable expectation of success refolding Fc-protein conjugates made in non-mammalian expression systems with the limited and/or flawed methodologies disclosed in *Schlegl* and *Hevehan*. EX2020 at ¶117.

PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

For the additional reasons above, *Schlegl*, *Hevehan*, and *Hakim*, either alone or in combination, do not render obvious Claim 12 of the ’138 Patent.

VI. Conclusion

For the reasons set forth above, Amgen requests that the Board deny Apotex’s Petition in its entirety.

Dated: May 22, 2017

Respectfully submitted,

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CERTIFICATE OF COMPLIANCE

Pursuant to 37 C.F.R. § 42.24(d), the undersigned certifies that the foregoing PATENT OWNER RESPONSE complies with the type-volume limitation of 37 C.F.R. § 42.24(b)(2) because it contains 11,922 words as determined by the Microsoft® Office Word 2010 word-processing system used to prepare the brief, excluding the parts of the brief exempted by 37 C.F.R. § 42.24(a)-(b).

Dated: May 22, 2017

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PROTECTIVE ORDER MATERIAL – SUBJECT TO PROTECTIVE ORDER

CERTIFICATE OF SERVICE

Pursuant to 37 C.F.R. § 42.6(e), the undersigned certifies that on May 22, 2017, a true and correct copy of the foregoing PATENT OWNER RESPONSE, along with all exhibits supporting and filed with the Patent Owner Response, was served by email on the following counsel of record for Apotex Inc. and Apotex Corp.:

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