UNITED STATES DISTRICT COURT
DISTRICT OF MASSACHUSETTS

AMGEN, INC.,

Plaintiff,

v.

F. HOFFMANN-LA ROCHE, LTD.,
ROCHE DIAGNOSTICS GMBH, and
HOFFMANN-LA ROCHE, INC.

Defendants.

Civil Action No. 05-CV-12237 WGY

DECLARATION OF DR. EDWARD EVERETT HARLOW, JR. IN SUPPORT OF DEFENDANTS' MOTION FOR SUMMARY JUDGMENT THAT THE CLAIMS OF PATENTS-IN-SUIT ARE INVALID FOR DOUBLE PATENTING OVER AMGEN '016 PATENT

I, Dr. Edward Everett Harlow, Jr., declare under penalty of perjury that:

1. I am Professor and Chair of the Department of Biological Chemistry and Molecular Pharmacology at Harvard Medical School. My responsibilities at Harvard Medical School include: (i) running the administration of a large basic science research and teaching department that currently has 31 faculty members, (ii) teaching graduate and medical students in courses around my own research interests, and (iii) running my own research lab where we study the basic differences between normal and cancer cells. The research group for which I am responsible consists of approximately 40 individuals including graduate students, post-doctoral fellows, clinical fellows, technicians, and senior research scientists. Since 1974, when I graduated from college, the focus of my scientific research has been on various aspects of basic cancer research. I am also an Associate Director of the Dana Farber/Harvard Cancer Center,

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where I am responsible for planning and evaluation for a large multi-institution research consortium of that unites approximately 900 research laboratories within the Harvard community with interests in cancer.

2. I earned a B.S. in Microbiology from the University of Oklahoma in 1974. In 1978, I received an M.S., also from the University of Oklahoma. In 1982, I earned a Ph.D. at Imperial Cancer Research Fund Laboratories, London, England, with my degree from Kings College, University of London. Here, I was part of a team of scientists that successfully cloned the mouse tumor suppressor gene now known as p53. We were the second team to successfully clone this gene. During this time, I became aware of and used many of the methods described in the work for the patents discussed in this declaration. Later, in 1985, while a staff scientist at Cold Spring Harbor, I became the first to clone and express the human p53 gene.

3. Since graduating from college in 1974, I have held many positions as a researcher, scientist, and teacher. Among these positions, I was Senior Staff Scientist at Cold Spring Harbor Laboratory in Cold Spring Harbor, NY, from 1988 to 1991. I was also Adjunct Assistant Professor in the Department of Microbiology at SUNY Stony Brook and, as stated above, am currently a professor and department chair at Harvard Medical School. I was the Scientific Director of the Massachusetts General Hospital Cancer Center from 1990 to 2002. From July 1996 to October 1998, I was Associate Director for Science Policy at the National Cancer Institute of the National Institutes of Health. I have also held a number of visiting professorships and named lecturer positions, including at Columbia University, Massachusetts Institute of Technology, University of California San Francisco, Baylor University, and Memorial Sloan-Kettering Cancer Center. Since 1992 I have been an American Cancer Society Research Professor.

4. I have been fortunate to receive many honors and awards for scientific achievement. Among these, I was elected to the National Academy of Sciences in 1993. In 1996, I was elected a Fellow of the American Academy of Arts and Sciences. In 1999, I was elected to the Institute of Medicine. I also received the Alfred P. Sloan, Jr. Prize from the General Motors Cancer Research Foundation in 1995, the Dickson Prize in Medicine from the University of Pittsburgh in 1996, and the Bristol Myers-Squib Award for Distinguished Achievement in Cancer Research in 1991.

5. I hold a number of positions on scientific advisory, award, and review committees. For example, I am a member of the Board on Life Sciences for the National Research Council. I currently sit of the External Advisory Board of several cancer centers including the University of California at San Francisco, Stanford University, and New York University. I am a member of or chair of several award committees including the Jane Coffin Childs Memorial Fund for Medical Research and the Smith Family Medical Foundation. I currently am a member of several scientific advisory boards for biotechnology companies. Currently I chair the Scientific Advisory Board of 3V Biosciences in Zurich, and I am a member of the Scientific Advisory Board of Alnylam Pharmaceuticals in Boston. In the past I have chaired or been a member of several advisory boards for such companies as Onyx Pharmaceuticals and Genomics Collaborative.

6. I am the author or co-author of a number of scientific articles and textbooks. I have had articles published in such peer-reviewed journals as *Nature*, *Science*, *Cell, Genes and Development, Journal of Virology*, *Molecular Cell Biology*, *Oncogene*, and the *Proceedings of the National Academy of Science U.S.A.*, among others. I also co-authored *Antibodies*: *A Laboratory Manual*, first published by Cold Spring Harbor Laboratory Press in 1988. This is a

basic laboratory manual on immunology and immunochemical methods. A revised edition of *Antibodies* was published in 1998.

7. A more complete list of my educational and professional experience, honors, and publications is found in my Curriculum Vitae attached as Exhibit 1 to this declaration.

8. In forming my below-stated opinions, I have relied upon my education, training, experience, and documents cited in this declaration. My knowledge and experience is further informed by numerous articles, books and journals that I have read over the years which have informed my understanding of the field of cell or molecular biology.

Vertebrate cells, in particular mammalian cells, and more particularly CHO cells were typical cells used to synthesize proteins for human use

9. As of 1983, three major classes of host cells were typically used by those skilled in the art to produce recombinant proteins: bacterial cells, such as *E. coli*, yeast cells, such as *Saccharomyces cerevisiae*, and cells derived from mammals, such as Chinese Hamster Ovary (CHO) cells.

10. It was also known at that time and before, that CHO cells were a cell line that proliferates under suitable nutrient conditions. Suitable nutrient conditions vary according to the cell type, but there were numerous published papers and other references, such as those supplied by the companies who sold the nutrient medium (e.g., Gibco, and others) that provided standard recipes for the cell media that could be used to grow standard cells of interest. The recipes provided in the various papers and manuals included recipes comprising the minimal required nutrients for a given cell type (minimal media), media enriched with certain ingredients (rich media) and other variations useful for particular cell growth conditions. Of course, all cells need suitable nutrient conditions to proliferate. While suitable nutrient conditions for a bacterial cell

are fairly flexible, and typically comprise a powdered medium dissolved in water that contains some simple salts and sugars, a mammalian cell culture requires a richer medium, as was well known in 1983. Suitable nutrient conditions for mammalian cells typically use media based on a much richer recipe, and one that also includes some quantity of mammalian sera, typically from newborn or fetal calf, to keep the cells reproducing in culture. By 1983, the conditions for growing any of these cells were easily available, both from the manufacturers of the media ingredients, and from a variety of published articles and manuals.

11. Further it was known at that time, CHO cells, like other host cells derived from mammals suitable for expression of foreign DNA, could produce properly glycosylated polypeptides, also known as glycoproteins.[1]

12. It was well known in the art that production of glycoproteins could be accomplished with mammalian derived cell lines but not bacterial cells because bacterial cells typically lack the enzymes required for glycosylation of proteins.[2] CHO cells were well known by 1983 to express foreign glycoproteins having their known *in vivo* biological activity.[3]

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[1] *See e.g.,* Ex. 2, Li et al., "Biosynthesis of lipid-linked oligosaccharides," J. BIOL. CHEM., 254: 1600-1605 (1979); Ex. 3, Scahill, et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983); Ex. 4, Singer et al., "Characterization of a porcine genomic clone encoding a major histocompatibility antigen: Expression in mouse L cells," PROC. NAT'L ACAD. SCI. 79:1403-1407 (1982); Ex. 5, Haynes and Weissman, "Constitutive, long-term production of human interferons by hamster cells containing multiple copies of a cloned interferon gene," NUCL. ACIDS RES., 11: 687-706 (1983).

[2] *See* Ex. 6, Sherwood and Goldwasser, "A radioimmunoassay for erythropoietin," BLOOD, 54: 885-893 (1979).

[3] *See e.g.,* Ex. 7, EPO 0 093 619 (Goeddel et al, 1983); Ex. 3, Scahill et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983); Ex. 5, Haynes et al, "Constitutive, long-term production of human interferons by hamster cells containing multiple copies of a cloned interferon gene," NUCL. ACIDS RES., 11: 687-706 (1983). See also, Ex. 8, EPO 0 117 059, 0 117 060 (Levinson et al, 1984); Ex. 9, McCormick et al., "Inducible expression of amplified human beta interferon genes in CHO cells," MOL. & CELL. BIOL., 4: 166-172 (1984); Ex. 10, Devos et al., "Purification of recombinant glycosylated human gamma interferon expressed in transformed Chinese hamster ovary cells," J. INTERFERON RES., 4: 461-468 (1984).

13. Moreover, by 1983, it was well understood that CHO cells were especially useful host cells. In addition to their ability to express a wide variety of foreign polypeptides, it was known at the time that foreign DNA could be stably integrated into the CHO host cells to produce stably transfected cell lines.[4] It was known that CHO cells could express and secrete exogenous proteins, if the correct signals for secretion were included in the coding regions of the foreign protein. It was also known that these secreted proteins were glycosylated. Furthermore, at that time, there was a reasonable expectation that the secreted proteins would have their expected biochemical activities.

14. The use of CHO cells as host cells was well-known prior to the conception and reduction to practice of the claims-in-suit.[5]

Scientists routinely sought amplification of protein production through use of promoter DNA and amplified marker gene and DHFR in suitable nutrient conditions

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[4] *See e.g.,* Ex. 7, EPO 0 093 619 (Goeddel et al, 1983); Ex. 2, Li et al, "Biosynthesis of Lipid-linked Oligosaccharides," J. BIOL. CHEM., 254: 1600-1605 (1979); Ex. 3, Scahill et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983); Ex. 5, Haynes et al, "Constitutive, long-term production of human interferons by hamster cells containing multiple copies of a cloned interferon gene," NUCL. ACIDS RES., 11: 687-706 (1983). See also, Ex. 10, Devos et al., "Purification of recombinant glycosylated human gamma interferon expressed in transformed Chinese hamster ovary cells," J. INTERFERON RES., 4: 461-468 (1984); Ex. 8, EPO 0 117 059, 0 117 060 (Levinson et al, 1984); Ex. 9, McCormick et al., "Inducible expression of amplified human beta interferon genes in CHO cells," MOL. & CELL. BIOL., 4: 166-172 (1984).

[5] *See e.g.,* Ex. 7, EPO 0 093 619 (Goeddel et al, 1983); Ex. 2, Li et al, "Biosynthesis of Lipid-linked Oligosaccharides," J. BIOL. CHEM., 254: 1600-1605 (1979); Ex. 3, Scahill et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983); Ex. 5, Haynes et al, "Constitutive, long-term production of human interferons by hamster cells containing multiple copies of a cloned interferon gene," NUCL. ACIDS RES., 11: 687-706 (1983). See also, Ex. 8, EPO 0 117 059, 0 117 060 (Levinson et al, 1984); Ex. 10, Devos et al., "Purification of recombinant glycosylated human gamma interferon expressed in transformed Chinese hamster ovary cells," J. INTERFERON RES., 4: 461-468 (1984); Ex. 9, McCormick et al., "Inducible expression of amplified human beta interferon genes in CHO cells," MOL. & CELL. BIOL., 4: 166-172 (1984).

15. DNA that carries the coding sequence for a protein of interest requires additional sequences including a promoter in order to express the corresponding polypeptide in a host cell.

16. The promoter is the controlling region that directs the host cell transcriptional machinery to produce a mRNA corresponding to the coding sequence in the vector. It must be operably linked to the DNA encoding the desired polypeptide in order to express the polypeptide.

17. As of 1983, the promoters of choice were specially selected promoters that were known to express large quantities of the desired mRNA in the host cell.

18. Some of the first promoters to be found and characterized were viral promoters. In general, promoters isolated from mammalian cells or from viruses that grow in mammalian cells will work effectively in most mammalian cells. Bacterial promoters were also used to express proteins in bacterial cells, and yeast promoters were used to express proteins in yeast cells. Thus, many promoters from various mammalian sources, typically non-human, were likely to work in CHO cells. The use of promoter DNA to direct the synthesis of foreign mRNAs within a host cell was well known in the art by 1983.[6]

19. By 1983, it was known how to insert a coding region for a protein of interest behind an active and efficient promoter capable of expressing in mammalian cells large quantities of mRNA from the inserted gene. The preferred method to introduce the promoter–coding region construct into a host cell at this time was transfection. The transfected DNA entered the cell and was inserted into the host cell DNA. There it was replicated along with the host DNA at every cell division and thus was maintained in each daughter cell in the population.

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[6] *See* Ex. 11, U.S. Pat. Nos. 4,766,075 (to Goeddel); Ex. 12, 4,356,270 (to Itakura); *see also* Ex. 13, U.S. Pat. Nos. 4,264,731 (to Shine); Ex. 14, 4,273,875 (to Manis); Ex. 15, 4,293,652 (to Cohen); Ex. 7, EPO 0 093 619 (Goeddel et al, 1983).

20. Even better, if one linked the promoter-coding region construct of interest to another gene, a selectable marker gene such as dihydrofolate reductase (DHFR), there were ways to manipulate the host cells into making even more target protein. After the DNA containing the two genes are transfected into cells, the DNA is inserted into the host chromosomal DNA, commonly in one location and commonly with only few copies linked together. Then the cells harboring the foreign DNA are grown in low concentrations of a drug that inhibits cells without the selectable marker gene. In this case, the selectable marker gene is present; it encodes a protein that blocks the action of the drug. When cells are then grown in slightly higher concentrations of the drug, only a minor population of the cells that happen to have more copies of the foreign DNA (and thus make more of the selectable drug resistance marker) can grow. This process is repeated with increasing drug concentration, and the resulting cells eventually have a large number of copies of the foreign DNA. Although there was no selection of the original promoter-coding region of interest, the copy number of this DNA construct is also amplified along with the drug resistance. The outcome is a population of cells that have very high numbers of the gene of interest.

21. The end result of using a marker gene is a host cell that (1) has amplified DNA (i.e., multiple copies of the DNA vector having the desired gene for the target protein), (2) has a promoter that makes abundant copies of the target protein, and (3) expresses an amplifiable marker (DHFR), which prevents the cell line from losing the desired gene. Thus, resultant cells frequently synthesize high quantities of the target protein. All this was known by 1983.

22. The use of DHFR with CHO cells was known prior to 1983.[7]

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[7] Ex. 16, Srinivasan and Lewis, "Transfer of the Dihydrofolate Reductase Gene into Mammalian Cells Using Metaphase Chromosomes or Purified DNA," INTRODUCTION OF MACROMOLECULES INTO VIABLE MAMMALIAN CELLS, 27-45 (1980); Ex. 17, Milbrandt et al., "Organization of a Chinese Hamster Ovary Dihydrofolate Reductase

Recombinant proteins were typically sought after for use in pharmaceutical compositions

23. After production and purification of any therapeutic substance, including small molecule drugs, natural proteins and recombinant proteins, these substances must be formulated into pharmaceutical compositions suitable for administration into a human.

24. Known formulations in 1983 included a variety of known diluents, adjuvants and carriers.

ERYTHROPOIETIN HAD BEEN EXTENSIVELY STUDIED AS OF 1983

25. One of ordinary skill in the art in December 1983 would have understood the following, as articulated in Judge Young's opinion in *Amgen, Inc. v. Hoechst Marion Roussel, Inc.*, 339 F. Supp. 2d 202, 214 (D. Mass 2004):

> EPO is a naturally occurring hormone that controls erythropoiesis, the production of red blood cells in bone marrow. … Erythropoiesis occurs continuously to offset cell destruction…. It enables a sufficient (but not excessive) amount of red blood cells to be available in the blood to provide tissue oxygenation…Hemoglobin is the protein in the red blood cells that actually transports the oxygen. … The amount of hemoglobin correlates to the amount of oxygen…. Hematocrit, which indicates the relative proportion of red blood cells to the total volume of blood, measures the ability of the blood to supply oxygen to the body…. Thus, generally an increase or decrease in hematocrit equates with an increase or decrease in hematocrit equates with an increase or decrease in the ability to supply oxygen to the body…. Under normal conditions, a person has a hematocrit of about forty-five to fifty, which means forty-five to fifty percent of the blood is made up of red blood cells.
>
> EPO is produced in the kidney and liver. Therefore, patients with chronic renal failure lack normal levels of EPO and suffer from anemia… Introduction of additional EPO into the patient's body can increase a patient's hematocrit level

Gene Identified by Phenotypic Rescue," MOL. CELL BIOL. 3: 1266-1273 (1983); Ex. 18, Nunberg et al., "Amplified dihydrofolate reductase genes are localized to a homogeneously staining region of a single chromosome in a methotrexate-resistant Chinese hamster ovary cell line," PROC. NAT'L ACAD. SCI., 75**:** 5553-5556 (1978); *See also,* Ex. 19, U.S. Pat. No. 4399216 (Axel et al., filed 1980); Ex. 20, Urlaub and Chasin, "Isolation of Chinese hamster cell mutants deficient in dihydrofolate reductase activity," PROC. NAT'L ACAD. SCI., 77: 4216-4220 (1980); Ex. 21, Schimke et al. "Gene amplification and drug resistance in cultured murine cells," SCIENCE, 202: 1051-1055 (1978).

> and sustain it at or near normal levels…. In other words, the blood is able to provide a steady supply of sufficient oxygen to the tissues….

26. A procedure for isolating human EPO from urine was published as of 1977.[8] This procedure was in fact cited in the patents-in-suit[9] as the procedure used to isolate urinary EPO (uEPO).

27. As far back as the 1960s, EPO had been associated with the *in vivo* biological activity of stimulating of reticulocytes and red blood cell production.

28. Because it became clear that the production of EPO from human urine would be economically unfeasible, when recombinant DNA technology became available, various groups set out to clone the EPO gene and use the cloned EPO gene to express and purify EPO in a suitable cell line. Because EPO is naturally made in the kidney, kidney damage, such as the type that creates the necessity for dialysis, was known to cause low serum levels of EPO and resultant anemia.

29. By 1983 there were various *in vitro* and *in vivo* assays known[10] for detecting EPO in various cellular fractions, fluids and media.

30. Some of these assays relied on antibody-protein interactions (e.g., the *in vitro* RIA referred to in the patents-in-suit), among other things, to quickly estimate levels of materials immunoreactive with EPO antibodies. The RIA measures the amount of antibody reactive

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[8] *See* Ex. 22, Miyake, Goldwasser et al, "Purification of Human Erythropoietin," J. BIOL. CHEM., 252: 5558-5564 (1977).

[9] *See, e.g.,* Lin '008, col. 9, lines 13-21 (referencing an Amgen patent application to Egrie for a mouse-mouse hybridoma cell line that produces mAb to hEPO made from a 20-aa EPO sequence) and the Lin 561024 application, p. 17, lines 1-6 referencing the same Amgen patent application.

[10] *See* Ex. 23, Goldwasser et al., "An assay for erythropoietin *in vitro* at the milliunit level," ENDOCRINOLOGY*,* 97: 315-323 (1975); Ex. 6, Sherwood and Goldwasser, "A radioimmunoassay for erythropoietin," BLOOD, 54: 885-893 (1979); Ex. 24, Cotes and Bangham, "Bio-assay of erythropoietin in mice made polycythæmic by exposure to air at a reduced pressure," NATURE*,* 191: 1065-1067 (1961); Ex. 25, Hammond et al., "Production, utilization and excretion of erythropoietin," ANN. N.Y. ACAD. SCI., 169: 516-527 (1968).

material present in a sample, and *in vivo* activity assays were used to determine the activity of the produced EPO compared to isolated human uEPO. These *in vivo* assays could be used to confirm the EPO-containing fractions in the EPO purification methods described by the patents-in-suit, and to assess the amount, and relative activity, of the isolated and purified EPO.[11]

31. In addition, hybridoma cell lines producing mAbs to EPO were known at least by 1982, such as Goldwasser's report of a rat-mouse hybridoma cell line that produced monoclonal antibody directed to human EPO.[12]

Mammalian cells were generally preferred for seeking glycosylation of expressed proteins

32. At least since 1979, it was known that EPO was glycosylated (i.e., containing sugar) when isolated from naturally occurring sources and that the asialo form was not active.[13]

33. In the case of EPO, it was known by 1979 that naturally occurring isolated human EPO was glycosylated.[14]

34. It had been well understood from before 1983 that a cell system that would produce a glycosylated EPO would be needed to allow the production and purification of biologically active EPO. It was also known that the most likely source of glycosylated EPO would be from mammalian cells. Other expression systems such as yeast cells might succeed, but mammalian cell culture would be highly likely to produce appropriately glycosylated proteins. Other secreted glycoproteins had been successfully produced and purified at this time. These proteins were known to be active. Among the glycosylated and active proteins produced

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[11] *See* Lin '008, col. 33, lines 30-51, particularly lines 44-51 "Radioimmunoassay activity for the isolates …; in vitro assay activity ranged from …; and in vivo assay activity ranged from …"

[12] *See* Ex. 26, Weiss, et al., "Characterization of a monoclonal antibody to human erythropoietin," BIOCHEMISTRY, 79: 5465-5469 (1982).

[13] *See* Ex. 6, Sherwood and Goldwasser, "A radioimmunoassay for erythropoietin," BLOOD, 54: 885-893 (1979).

[14] *See* Ex. 6, Sherwood and Goldwasser, "A radioimmunoassay for erythropoietin," BLOOD, 54: 885-893 (1979).

by 1983 were some of the interferons,[15] immunoglobulins,[16] tPA[17] and other glycosylated proteins. It would have been reasonable to expect that in making biologically active recombinant EPO (rEPO), one would prefer to use mammalian cells to achieve desired glycosylation.

35. The investigative and development activity surrounding EPO in the 1970s and early 1980s was directed ultimately to an EPO treatment for humans.[18] Goldwasser and Miyake, for example, focused on isolating and investigating human EPO. And the goal of the various groups looking to develop a recombinant EPO in the early 1980s was precisely to develop a recombinant human EPO.

ORDINARY SKILL IN THE ART

36. Throughout this declaration, I refer to one of ordinary skill in the art and what that person would know or understand from reading the patents and patent claims at issue. By this I mean a person with an advanced degree, such as a Ph.D. in biochemistry, immunology, genetics, molecular biology, medicine or related scientific fields, and two years or more of post-graduate or professional laboratory experience. I believe I can speak to what this person of ordinary skill

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[15] Ex. 3, Scahill, et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983); Ex. 27, Gray and Goeddel, "Cloning and Expression of Murine Immune Interferon cDNA," PROC. NAT'L. ACAD. SCI., 80:5842-5846 (1983); Ex. 28, Taniguchi et al., "Construction and Identification of a Bacterial Plasmid Containing the Human Fibroblast Interferon Gene Sequence," PROC. JAPAN ACAD., 55: 464-469 (1979); Ex. 5, Haynes and Weissman, "Constitutive, Long-Term Production of Human Interferons by Hamster Cells Containing Multiple Copies of a Cloned Interferon Gene," NUCL. ACIDS RES., 11: 687-706 (1983). See also, Ex. 9, McCormick et al., "Inducible expression of amplified human beta interferon genes in CHO cells," MOL. & CELL. BIOL., 4: 166-172 (1984).

[16] *See* Ex. 29, Oi et al., "Immunoglobulin gene expression in transformed lymphoid cells," PROC. NAT'L. ACAD. SCI. 80:825-829 (1983); Ex. 30, Neuberger, "Expression and regulation of immunoglobulin heavy chain gene transfected into lymphoid cells," EMBO J. 2:1373-1378 (1983); Ex. 31, Sidman, "Differing requirements for glycosylation in the secretion of related glycoproteins is determined neither by the producing cell nor by the relative number of oligosaccharide units," J. BIOL. CHEM., 256: 9374-9376 (1981)*; see also,* Ex. 32, Seidman et al., "Antibody Diversity," SCIENCE, 202: 11 (1978).

[17] *See* Ex. 11, US 4,766,075 to Goeddel et al.

[18] See Background section of patents-in-suit, e.g. '933 col. 5:39 – col. 6:59.

would have known in 1983, when the original patent applications were filed, because, at that time, I had been working since 1974 with the then-current methods for molecular biology, including my work on the cloning of the p53 gene. Indeed, I was working in the early 1980's at two of the leading institutions in the field of molecular biology, the Imperial Cancer Research Fund Laboratories in London, England and Cold Spring Harbor Laboratory in Cold Spring Harbor, New York. Accordingly, I am well aware of the range of knowledge and understanding by young scientists and veterans during the relevant time period. My testimony set forth herein is thus based on my own substantial experience and knowledge in the field and on my opinion of how one of ordinary skill in the art of cloning technology would view and understand the patents and prior art.

'016 PATENT CLAIM 10

37. Generally speaking, claim 10 of the '016 patent is directed to production of purified recombinant EPO. The process begins with culturing mammalian cells transfected with the EPO gene sequence in a suitable nutrient environment to express recombinant EPO that is purified through a series of steps.

38. The language of Claim 10 from the '016 patent reads as follows:

> A process for the efficient recovery of recombinant erythropoietin from a mammalian cell culture supernatant fluid, said process comprising the following steps in sequence:
>
> (1) subjecting the fluid to ion exchange chromatographic separation at about pH 7.9, thereby to selectively bind erythropoietin in said sample to a DEAE agarose cationic resin;
>
> (2) stabilizing materials bound to said resin against degradation by acid activated proteases through treatment with urea;
>
> (3) selectively eluting bound materials having a pKa greater than that of erythropoietin by treatment with aqueous acid at a pH of about 4.3.

> (4) selectively eluting erythropoietin by treatment with an aqueous salt at a pH of about 7.0;
>
> (5) subjecting erythropoietin-containing eluent fractions to reverse phase liquid chromatographic separation involving an immobilized C4 resin, thereby to selectively bind erythropoietin in said fluid to said resin;
>
> (6) selectively eluting bound erythropoietin from said resin with an aqueous ethanol solution of about 60 percent at a pH of about 7.0; and,
>
> (7) isolating erythropoietin-containing fractions of the eluent.

39. For claim 10, I considered two ways to define the meaning of recombinant erythropoietin. In the first, I examined the '016 patent itself and found a concise statement on how to practice the '016 invention:

> Practice of the present invention is believed to be suitably illustrated by the following examples practiced on pooled CHO cell supernatants prepared in the manner described in Example 10 of the aforementioned U.S. patent application Ser. No. 675,298 [now the '008 patent].

'016 patent, at col. 4, lines 33-39. Example 10 set forth in the patents-in-suit (set forth at col. 26, line 35 – col. 30, line 29 of the '008 patent, for example), concludes:

> Glycoprotein products provided by the present invention are thus comprehensive of products having a primary structural conformation sufficiently duplicative of that of a naturally occurring erythropoietin to allow possession of one or more of the biological properties thereof….

'008 patent, col. 30, lines 22-27. The person of ordinary skill in the art would understand from the '016 patent that the starting material, a mammalian cell culture supernatant fluid containing recombinant erythropoietin used in claim 10, describes the CHO cell supernatant product containing glycosylated, biologically active recombinant erythropoietin, as obtained through Example 10.

40. Second, I also considered the meaning of recombinant erythropoietin to a person of ordinary skill in the art in 1983. The person of ordinary skill in the art would have the

understanding of recombinant DNA technology as described above. In the following paragraphs, I discuss how the person of ordinary skill in the art would understand the claims-in-suit.

41. I also considered other terms in claim 10. Supernatant is well known in the art to be the liquid/fluid from media from the cell culture. Any cells, cellular debris, membranes, insoluble proteins, etc. may be removed by methods such as centrifugation or filtration of the cell media.

42. The ‘016 patent claims the recovery of recombinant EPO from the culture of mammalian host cells. The steps taught and claimed in the ‘016 patent allow one of ordinary skill in the art to provide acceptable EPO for pharmaceutical purposes.

43. Any solid portions of the mammalian host cells (e.g., disturbed cells, cellular debris, etc.) are removed using techniques so well known in the prior art that the techniques are not described in any detail in either the ‘016 patent or the patents-in-suit. The remaining liquid – the supernatant – includes the recombinant EPO (rEPO) as well as many other molecules. Since it is highly undesirable to deliver to a human many of these other molecules (e.g., DNA fragments, mRNA fragments and proteins endogenous to the mammalian host cell), the rEPO is isolated from the supernatant.

44. The purification of the rEPO from the undesirable molecules of the host cell’s supernatant is necessary to provide a rEPO that is a pharmaceutically acceptable product. If the rEPO is not properly purified from the supernatant, it cannot be used for its intended purpose.

45. The amino acid sequence shown in FIG. 6 of the patents-in-suit is the translation product of the erythropoietin gene used in the mammalian cell culture of ‘016 claim 10. I understand that a final rEPO product may not have all 166 amino acids shown in FIG. 6.

46. I am told that courts specifically created the doctrine of obviousness-type double patenting to preclude applicants from extending the duration of their patent term to a single invention by claiming obvious variants of that invention in later patents. To protect the public from an unwarranted extension of the patent monopoly, a terminal disclaimer limiting the patent terms of these patents to the full life of the earlier '016 patent should have been required. Since the '016 patent has now expired, the patents-in-suit should likewise be expired or invalidated.

47. All of the limitations of the five Amgen patents-in-suit would have been considered routine and obvious to those of skill in the art once they were in possession of the process for recovering purified recombinant EPO from a mammalian cell culture claimed in the '016 patent.

THE CLAIMS-IN-SUIT PROVIDE NO PATENTABLE DISTINCTION BEYOND '016 PATENT CLAIM 10

48. The limitations of the claims-in-suit not explicitly set forth in claim 10 of the '016 patent fall into several categories, none of which can provide a patentable distinction:

(i) Well known routine recombinant engineering techniques (e.g., the use of viral promoter DNA, amplified DNA, exogenous DNA, CHO cells, DHFR, suitable nutrient conditions, etc.).

(ii) the utility of the product, which included well known properties of EPO, and which otherwise would have been obvious and reasonably expected in light of the well known properties of EPO (e.g., the *in vivo* biological activity of causing bone cells to increase production of reticulocytes and red blood cells, use as a pharmaceutical composition, use for treating kidney dialysis patients, and use for humans),

(iii) well known routine pharmaceutical techniques (e.g., use of a pharmaceutically

acceptable diluent, adjuvant or carrier).

'933 Patent

Claim 3

49. Claim 3 of the '933 patent reads as follows:

> 3. A non-naturally occurring glycoprotein product of the expression in a mammalian host cell of an exogenous DNA sequence comprising a DNA sequence encoding human erythropoietin said product possessing the in vivo biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells.

50. All of the elements of claim 3 are either found within claim 10 of the '016 patent or would be obvious to a person of ordinary skill in the art with knowledge of claim 10 of the '016 patent. Without limitation, I note the product claimed by claim 3 of the '933 patent would have been obvious over the product used in or formed by the process set forth in claim 10 of the '016 patent.

51. Claim 3 of the '933 patent covers the recombinant EPO product used in claim 10 of the '016 patent. Although I have not considered how one could distinguish a "naturally occurring" protein from a "non-naturally occurring" protein, it is my opinion that *non-naturally occurring erythropoietin*, if it means anything, would have been obvious in view of the "recombinant erythropoietin." The erythropoietin of in claim 10 of the '016 patent is *a glycoprotein product of the expression in a mammalian host cell.* Mammalian derived cells, such as CHO cells, were known at the time of the '016 patent submission to glycosylate secreted proteins. Such cells were specifically known to glycosylate other recombinant secreted proteins. Since EPO is naturally glycosylated, there is every expectation that the rEPO recovered in claim 10 of the '016 patent would be glycosylated as well. The cell culture of the '016 patent uses a DNA sequence encoding erythropoietin. *Human* EPO was and remains the EPO of greatest

interest to scientists and pharmaceutical companies. Claim 3 also calls for the "in vivo biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells." Recombinant EPO from the cell culture of claim 10 of the '016 patent would have every expectation to inherently exhibit this property. I base this belief on several facts. First, as I discussed above, it was well known in 1983 that other active recombinant proteins could be isolated from the supernatant of mammalian cells manipulated to express the recombinant proteins. Second, based on what was known in 1983, someone of ordinary skill in the field would have had a reasonable expectation that recombinant erythropoietin would be appropriately glycosylated through expression in a mammalian host cell and would be erythropoietic, i.e., that it would have the *in vivo* biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells. Third, to one skilled in the art in 1983, it would be obvious to test purified or partially purified preparations of rEPO for biological activity. In fact, if this were not done the ultimate goals of this purification could not be achieved. The purified rEPO was only useful if it had biological activity. Therefore, although one of skill in the art had a reasonable expectation for success in making a biologically active rEPO, it is inconceivable to me that one would purify rEPO without confirming biological activity. Indeed, I note that throughout the '016 patent, it is claimed that the isolated rEPO is biologically active. For example, see Abstract, Background, and Brief Summary. Finally, I note that Amgen has conceded in discourse with the Patent and Trademark Office that once a clone for the human EPO gene was isolated, all of the steps to synthesize, purify, and prepare active rEPO for clinical trials followed in a straightforward manner. (See discussion below.) Therefore, there would have been a reasonable expectation that the rEPO used in the '016 patent possessed the *in vivo biological property (or activity) of causing bone marrow cells to increase production of*

reticulocytes and red blood cells. Thus, the glycoprotein product of claim 3 would have been obvious in view of claim 10 of the '016 patent.

Claim 7

52. Claim 7 of the '933 patent reads as follows:

> 7. The glycoprotein product according to claim 3, 4, 5 or 6 wherein the host cell is a non-human mammalian cell.

53. The use of a mammalian cell culture as set forth in claim 10 of the '016 patent would have suggested to one of ordinary skill in the art in 1983 that *non-human mammalian cells* could have been used. See paragraphs 9-14 and 15-22 above for a description of the state of the art as to the routine use of non-human mammalian cells for protein expression. Thus, claim 7 depending from claim 3 recites a glycoprotein that would have been obvious over the EPO used in the process of claim 10 of the '016 patent.

Claim 8

54. Claim 8 of the '933 patent reads as follows:

> 8. The glycoprotein product according to claim 7 wherein the non-human mammalian cell is a CHO cell.

55. As explained above at paragraphs 9-14, *CHO cells* would have been a routine choice for those of ordinary skill in the art in 1983 for use as the host cell. Therefore, claim 8 would have been obvious over claim 10 of the '016 patent.

Claims 9 and 12

56. Claims 9 and 12 read as follows:

> 9. A pharmaceutical composition comprising an effective amount a glycoprotein product effective for erythropoietin therapy according to claim 1, 2, 3, 4, 5 or 6 and a pharmaceutically acceptable diluent, adjuvant or carrier.

> 12. A pharmaceutical composition comprising an effective amount of a glycoprotein product effective for erythropoietin therapy according to claim 7 and a pharmaceutically acceptable diluent, adjuvant or carrier.

57. One of ordinary skill in the art in 1983 would have understood the purified recombinant EPO of the ‘016 patent was intended for use in *pharmaceuticals*. As in any pharmaceutical, one of ordinary skill in the art would have found it routine to combine an effective amount of the recombinant EPO purified according to claim 10 of the ‘016 patent with a *diluent, adjuvant or carrier*. Therefore, claim 9 depending from claim 3 and claim 12 would have been obvious in view of the purification of recombinant EPO as claimed in the ‘016 patent.

Claims 11 and 14

58. Claims 11 and 14 read as follows:

> 11. A method for treating a kidney dialysis patient which comprises administering a pharmaceutical composition of claim 9 in an amount effective to increase the hematocrit level of said patient.
>
> 14. A method for treating a kidney dialysis patient which comprises administering a pharmaceutical composition of claim 12 in an amount effective to increase the hematocrit level of said product.

59. As discussed above at paragraphs 25-28, it was well known in the art in 1983 that EPO could be used for *treating kidney dialysis patients to increase a patient’s hematocrit level*. The growing demands for these treatments led Amgen and others to seek a method to produce recombinant EPO. Based on the state of the art in recombinant DNA technology at that time, there was a reasonable expectation of success in producing rEPO useful for such treatment. Therefore, it would have been obvious in 1983 to one of ordinary skill in the art to use the purified recombinant EPO produced in claim 10 of the ‘016 patent to make a pharmaceutical composition for treating a kidney dialysis patient and inherently increase the hematocrit levels.

‘422 Patent Claim 1

60. Claim 1 of the ‘422 patent reads as follows:

> 1. A pharmaceutical composition comprising a therapeutically effective amount of human erythropoietin and a pharmaceutically acceptable diluent, adjuvant or carrier, wherein said erythropoietin is purified from mammalian cells grown in culture.

61. One of ordinary skill in the art in 1983 would have understood the purified recombinant EPO of the ‘016 patent was intended for use in *pharmaceuticals*. As in any pharmaceutical, one of ordinary skill in the art would have found it routine to combine a therapeutically effective amount of the recombinant EPO purified according to claim 10 of the ‘016 patent with a *pharmaceutically acceptable diluent, adjuvant or* carrier. As explicitly covered by the ‘016 patent, *erythropoietin is purified from mammalian cells grown in culture*. Given that it would have been obvious to use the process of claim 10 of the ‘016 patent to produce human rEPO, claim 1 of the ‘422 patent would have been obvious over claim 10 of the ‘016 patent.

‘698 Patent

Claim 4

62. Claim 4 of the ‘698 patent reads as follows:

> 4. A process for the production of a glycosylated erythropoietin polypeptide having the *in vivo* biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells comprising the steps of:
>
> (a) growing, under suitable nutrient conditions, vertebrate cells comprising promoter DNA, other than human erythropoietin promoter DNA, operatively linked to DNA encoding the mature erythropoietin amino acid sequence of FIG. 6; and
>
> (b) isolating said glycosylated erythropoietin polypeptide expressed by said cells.

63. I understand that Amgen's position is that claim 4 of the '698 patent covers example 10 of the patents-in-suit, and this is the example is used in the '016 patent. The process of claim 4 of the '698 patent would have been obvious over the "recombinant erythropoietin from a mammalian cell culture supernatant fluid" of claim 10 of the '016 patent.

64. The starting material used in the process of '016 claim 10 corresponds to a product obtainable from "isolating said glycosylated erythropoietin polypeptide expressed by said cells", that is, a mammalian cell culture supernatant fluid containing among other things, recombinant erythropoietin. As explained above in paragraphs 18-21 and 43 by expressing EPO in a mammalian cell culture as covered in claim 10 of the '016 patent, the resultant EPO will have been *glycosylated*. The EPO produced by claim 10 of the '016 patent is a *polypeptide*. Recombinant EPO from a mammalian cell culture as claimed in claim 10 of the '016 patent would have exhibited the *in vivo biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells.* Given that claim 10 of the '016 patent produces recombinant EPO, it necessarily follows that the mammalian cell culture was provided *suitable nutrient conditions.* Mammalian cells are *vertebrate* cells because mammals have backbones. As explained above in paragraphs 17-19, *promoter DNA other than human erythropoietin promoter DNA* was routinely used in recombinant protein synthesis in 1983. *FIG. 6* depicts the amino acid sequence of the expression product of human erythropoietin. This is the original complete 166-amino-acid sequence that would have been formed in the mammalian cells of claim 10 of the '016 patent when used to produce human EPO. Given that it would have been obvious to use the process of claim 10 of the '016 patent to produce human EPO, such use would necessarily involve *promoter DNA operatively linked to DNA encoding the mature*

erythropoietin amino acid sequence of FIG. 6. The elements of claim 4 are all thus covered by or obvious from the claimed process of claim 10 of the '016 patent.

Claim 5

65. Claim 5 of the '698 patent reads as follows:

> 5. The process of claim 4 wherein said promoter DNA is viral promoter DNA.

66. As discussed above at paragraph 18, it was routine in the art in 1983 to make use of *viral promoter DNA* during synthesis of recombinant proteins in a method such as that claimed in claim 10 of the '016 patent. In view of the discussion with respect to claim 4 and for this reason, claim 5 would have been obvious over claim 10 of the '016 patent.

Claim 6

67. Claim 6 of the '698 patent reads as follows:

> 6. A process for the production of a glycosylated erythropoietin polypeptide having the *in vivo* biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells comprising the steps of:
>
> (a) growing, under suitable nutrient conditions, vertebrate cells comprising amplified DNA encoding the mature erythropoietin amino acid sequence of FIG. 6; and
>
> (b) isolating said glycosylated erythropoietin polypeptide expressed by said cells.

68. I understand that Amgen's position is that claim 6 of the '698 patent covers example 10 of the patents-in-suit, and this is the example is used in the '016 patent. The process claimed by claim 6 of the '698 patent would have been obvious over the "recombinant erythropoietin from a mammalian cell culture" of claim 10 of the '016 patent.

69. The starting material used in the process of '016 claim 10 corresponds to a product obtainable from "isolating said glycosylated erythropoietin polypeptide expressed by said cells", that is, a mammalian cell culture supernatant fluid containing among other things,

recombinant erythropoietin. As explained above in paragraphs 32-35, the rEPO in a mammalian cell culture of claim 10 of the '016 patent would have been *glycosylated.* The rEPO of claim 10 of the '016 patent is a *polypeptide.* Recombinant EPO from a mammalian cell culture of claim 10 of the '016 patent would have exhibited the *in vivo biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells.* Given that claim 10 of the '016 patent produces rEPO, it necessarily follows that the mammalian cell culture was provided with *suitable nutrient conditions.* Mammalian cells are *vertebrate* cells because mammals have backbones. As explained above in paragraphs 19-22, *amplified DNA* was routinely used in recombinant protein synthesis in 1983. *FIG. 6* depicts the amino acid sequence of the expression product of human erythropoietin. This is the original complete 166-amino-acid sequence that would have been formed in the mammalian cells of claim 10 of the '016 patent when used to produce human EPO. Given that it would have been obvious to use the process of claim 10 of the '016 patent to produce human EPO, such use would necessarily involve *amplified DNA encoding the mature erythropoietin amino acid sequence of FIG. 6.* The elements of claim 6 are all thus covered by or obvious from the claimed process of claim 10 of the '016 patent.

Claim 7

70. Claim 7 of the '698 patent reads as follows:

> 7. The process of claim 6 wherein said vertebrate cells further comprise amplified marker gene DNA.

71. As discussed above in paragraphs 19-22, *amplified marker gene DNA* was routinely used in 1983 during synthesis of recombinant proteins in a method such as that claimed

in claim 10 of the '016 patent. In view of the discussion with respect to claim 6 and for this reason, claim 7 would have been obvious over claim 10 of the '016 patent.

Claim 8

72. Claim 8 of the '698 patent reads as follows:

> 8. The process of claim 7 wherein said amplified marker gene DNA is Dihydrofolate reductase (DHFR) gene DNA.

73. As discussed above in paragraphs 19-22, *Dihydrofolate reductase (DHFR) gene DNA* was routinely used as an amplified marker gene DNA in 1983 during synthesis of recombinant proteins in a method such as that claimed in claim 10 of the '016 patent. In view of the discussions with respect to claims 6 and 7 and for this reason, claim 8 would have been obvious over claim 10 of the '016 patent.

Claim 9

74. Claim 9 of the '698 patent reads as follows:

> 9. The process according to claims 2, 4 and 6 wherein said cells are mammalian cells.

75. The use of mammalian cells is explicitly covered by claim 10 of the '016 patent. In view of the discussions with respect to claims 4 and 6, claim 9 would have been obvious over claim 10 of the '016 patent.

'868 Patent

Claim 1

76. Claim 1 of the '868 patent reads as follows:

> 1. A process for the production of a glycosylated erythropoietin polypeptide having the *in vivo* biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells comprising the steps of:

> (a) growing, under suitable nutrient conditions, mammalian host cells transformed or transfected with an isolated DNA sequence encoding human erythropoietin; and
>
> (b) isolating said glycosylated erythropoietin polypeptide therefrom.

77. I understand that Amgen's position is that claim 1 of the '868 patent covers example 10 of the patents-in-suit, and this is the example used in the '016 patent. The process claimed by claim 1 of the '868 patent would have been obvious over the "recombinant erythropoietin from a mammalian cell culture" of claim 10 of the '016 patent.

78. The starting material used in the process of '016 claim 10 corresponds to a product obtainable from "isolating said glycosylated erythropoietin polypeptide expressed by said cells", that is, a mammalian cell culture supernatant fluid containing among other things, recombinant erythropoietin.. As explained above in paragraphs 32-35, by expressing EPO in a mammalian cell culture as covered in claim 10 of the '016 patent, the resultant EPO will have been *glycosylated.* The EPO produced by claim 10 of the '016 patent is a *polypeptide.* Recombinant EPO from a mammalian cell culture as claimed in claim 10 of the '016 patent would have exhibited the *in vivo biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells.* Given that claim 10 of the '016 patent produces recombinant EPO, it necessarily follows that the mammalian cell culture was provided *suitable nutrient conditions.* As explained above in paragraphs 15-19, it was routine in the art in 1983 when synthesizing recombinant proteins in mammalian cells *to transform or transfect the cells with the isolated DNA sequence* encoding the desired protein. It would have been a mere obvious choice to use the process of the '016 patent to produce *human erythropoietin.* The elements of claim 1 are all thus covered by or obvious from the claimed process of claim 10 of

the ‘016 patent. The elements of claim 1 are all thus covered by or obvious from the claimed process of claim 10 of the ‘016 patent.

Claim 2

79. Claim 2 of the ‘868 patent reads as follows:

> 2. The process according to claim 1 wherein said host cells are CHO cells.

80. Because glycosylation of EPO was necessary for the desired biological activity, and because *CHO cells* were readily available and well characterized, one of ordinary skill in the art in 1983 would have naturally chosen *CHO cells* as a host. In view of the discussion with respect to claim 1 and for this reason, claim 2 would have been obvious over claim 10 of the ‘016 patent.

‘349 Patent Claim 7

81. Claim 7 depends from any of claims 1-6. In order to present all elements of the claim, claim 7 can be read as follows:[19]

> A process for producing erythropoietin comprising the step of culturing, under suitable nutrient conditions, vertebrate cells [which can be propagated in vitro and which are capable upon growth in culture of producing erythropoietin in the medium of their growth in excess of 100, 500, or 1000 U of erythropoietin per 10^6 cells in 48 hours as determined by radioimmunoassay, said cells comprising non-human DNA sequences which control transcription of DNA encoding human erythropoietin].

82. The process of claim 7 of the ‘349 patent would have been obvious over the “recombinant erythropoietin from a mammalian cell culture supernatant fluid” of claim 10 of the ‘016 patent.

83. Although the claim specifies that the cells are capable of being propagated in vitro and are capable of producing human erythropoietin in excess of 100, 500 or 1000 U per 10^6 cells

[19] The bracketed text corresponds to what is claimed in claims 1-6.

in 48 hours, Dr. Lin did not provide any inventive contribution to the selection of the appropriate cells or to the well known amplification techniques used to increase the rates of production.

84. An RIA assay measures any material that reacts with the antibody used in the assay. In addition, the standard used in the radioimmunoassay (RIA) described in the '349 patent, as well as all the other parameters necessary to ensure a repeatable and consistent RIA, are not clearly set forth in the '349 patent. Thus, the claims of the '349 patent appear indefinite.

85. Given that claim 10 of the '016 patent produces recombinant EPO, it necessarily follows that the mammalian cell culture was provided *suitable nutrient conditions*. Mammalian cells are *vertebrate* cells because mammals have backbones.

86. Whether the mammalian cells of claim 10 of the '016 patent would have been capable of producing EPO at a rate on the order of what is enabled by the '349 patent was well within the ordinary skill in the art as of 1983. The techniques for amplifying production, as described in paragraphs 27-29 and 46, were readily available. The '349 patent does not disclose or claim any unknown method for making its rate of production possible. Since the scope of claim 7 of the '349 patent—which appears indefinite—must be limited to what was enabled in the '349 patent, and since such a scope would have been obvious over the claim 10 of the '016 patent, claim 7 must have been obvious over claim 10 of the '016 patent, if it were somehow capable of being construed to have a definite scope.

AMGEN'S ADMISSIONS AND OTHER EVIDENCE SUPPORTING MY OPINION THAT THE CLAIMS-IN-SUIT WOULD HAVE BEEN OBVIOUS OVER '016 PATENT CLAIM

Opinions of Amgen Expert Dr. Lodish in Prior Litigation Regarding Double-Patenting

87. In *In re Columbia University Litigation*, No. 09-MD-01592, D.Mass.—during which Amgen attacked the validity of a patent owned by Columbia University—Amgen's expert, Dr. Harvey Lodish provided an expert report (the "2004 Report") in support of an obviousness-type double patenting attack.[20] Dr. Lodish's 2004 Report showed that a later-issued Columbia University patent was obvious in light of an earlier-issued patent also owned by Columbia University. Like the patents-in-suit in the present case, the Columbia University patent being attacked by Amgen related to recombinant DNA engineering.

88. In his 2004 Report, Dr. Lodish admitted that, as of 1980 (which was the critical date in that prior litigation, and which is several years prior to the critical date in the present case), many of the techniques used in this field were obvious and well known.

89. In his 2004 Report, Dr. Lodish admitted that the glycosylation of proteins was obvious and well known in 1980.[21]

90. In his 2004 Report, Dr. Lodish also admitted that, as of 1980, the transformation of mammalian cells with exogenous DNA was obvious and well known.[22]

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[20] Apparently, this litigation involved many biotech companies accused of infringing Columbia University patents. Amgen was one of these companies and was a party to this litigation. It is not clear to me whether Dr. Lodish—who is an expert of Amgen in the present litigation—was retained by Amgen in the *Columbia University* litigation or was retained only by another parties in that litigation.

[21] *See* Para. 123 of INITIAL EXPERT REPORT OF HARVEY F. LODISH, Ph.D., August 27, 2004, *In re Columbia University Litigation*, No. 09-MD-01592, D.Mass.: "… **In my opinion, the requirement that a protein have an attached carbohydrate chain does not make it patentably distinct from the simple requirement that it be a protein.**" (Emphasis added.) *See also* paras. 141, 142, 143 and 145 of this report.

[22] See, e.g., paras. 55-64 of INITIAL EXPERT REPORT OF HARVEY F. LODISH, Ph.D., August 27, 2004, In re Columbia University Litigation, No. 09-MD-01592, D.Mass.

91. In his 2004 Report, Dr. Lodish also admitted that, as of 1980, the use of CHO cells for producing recombinant proteins was obvious and well known.[23]

92. In his 2004 Report, Dr. Lodish also admitted that, as of 1980, amplification of genes in mammalian cell cultures was obvious and well known.[24]

93. In his 2004 Report, Dr. Lodish also admitted that, as of 1980, the use of dihydroflate reductase (DHFR) was obvious and well known.[25]

94. In his 2004 Report, Dr. Lodish also admitted that, as of 1980, the use of viral promoters was obvious and well known.[26]

95. Dr. Lodish's side prevailed in the Colombia litigation. I understand that Columbia University withdrew its allegations of infringement of the later-issued patent. I agree with Dr. Lodish's opinions that as of 1980 many of the techniques used in this field were already obvious and well known, including the glycosylation of proteins, the transformation of mammalian cells with exogenous DNA, the use of CHO cells for producing recombinant proteins, amplification of genes in mammalian cell cultures, the use of dihydroflate reductase (DHFR), and the use of viral promoters.

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[23] Para. 64 of INITIAL EXPERT REPORT OF HARVEY F. LODISH, Ph.D., August 27, 2004, *In re Columbia University Litigation*, No. 09-MD-01592, D.Mass. See also paras. 144-148 of this report.

[24] See, e.g., paras. 65-67, 103, 133, 137, 162-168 of INITIAL EXPERT REPORT OF HARVEY F. LODISH, Ph.D., August 27, 2004, *In re Columbia University Litigation*, No. 09-MD-01592, D.Mass.

[25] See, e.g., paras. 65-67, 103, 133, 137, 162-168 of INITIAL EXPERT REPORT OF HARVEY F. LODISH, Ph.D., August 27, 2004, *In re Columbia University Litigation*, No. 09-MD-01592, D.Mass.

[26] See, e.g., para. 72 of INITIAL EXPERT REPORT OF HARVEY F. LODISH, Ph.D., August 27, 2004, *In re Columbia University Litigation*, No. 09-MD-01592, D.Mass.

More Opinions by Other Amgen Experts in Prior Litigations regarding the Selection, Transformation, and Culturing of CHO Cells

96. Dr. Davies, who I understand was an Amgen expert, admitted that the use of CHO cells to express human erythropoietin was merely one of a few obvious, identified and predictable solutions to the problem of reliably expressing large quantities of recombinant human protein, a point with which I agree:

> Judge Harris: Dr. Davies, could you tell us why the CHO cell was chosen as a host?
>
> The Witness: Why it was chosen? It's a cell which has been well studied for a number of years. It grows well in cell cultures. There are a number of cell lines which have been developed over many many years which grow well in cell culture. You can just simply take the cell, add nutrients and you can get high density cell cultures. The CHO cell is also the cell that has been used for many transfection and transformation studies and so it's probably one of the mammalian cell lines that is best understood in terms of its properties with respect to cell culture, and its properties with respect to introduction of DNA. And I suspect that it's largely for this reason, I think it's the most used standard mammalian cell line, so that's why it would be used. (AM-ITC 00178100-01)

97. Amgen's expert Dr. Wall also admitted:

> All that was necessary was to isolate the DNA sequence (either gDNA as discussed above, or cDNA as discussed below) and insert this into any suitable, well-known vector (not necessarily the one used in the patent) and then insert this into a suitable host cell. There were a variety of expression vectors available in 1983 and one would know that such a vector could be used to express protein once the gene sequence was known. [November 9, 2000 Expert Report of Professor Randolph Wall submitted in the High Court of Justice Chancery Division Patents Court. (AM-ITC 01049079-125) at 36-37.]
>
> Again by 1983 there were a number of host cells available which could be used to express proteins. With the EPO sequence disclosed by the patent, there was nothing at all which was not routine in constructing a host cell which could express the EPO gene. [*Id.* at 37.]
>
> As regards the conditions of transformation, culture and selection of the cells and the manner in which the plasmid integrates into the genome of the cells, this is, once more, just a matter of routine basic cloning. [*Id.* at 42.]

The foregoing admissions by Drs. Lodish, Davies and Wall support my opinion that the claims-in-suit would have been obvious over the claims of the '016 and '008 patents.

Amgen's Admissions Regarding to the Lack of Patentable Distinctiveness

98. I note that during the prosecution of the '868 patent, Amgen argued to the PTO that as of the time of the then asserted filing date of December 13, 1983, it was well known by those skilled in the art to obtain glycosylated proteins expressed in mammalian host cells. The Patent and Trademark Office rejected the pending claims as non-enabled and lacking adequate written description under Section 112. In particular, the PTO stated that:

> Applicant claims a method of preparing EPO, in part, by growing a host "capable of effecting post-translational glycosylation of polypeptides expressed therein." Applicant has provided no guidance for, and no working examples of, any test or procedure for determining which host cells have such capability and which do not. Without such a procedure, one of ordinary skill in the art would have no way to determine operable from inoperable embodiments of the claimed invention...Accordingly, it would require undue experimentation by one of ordinary skill in the art to practice the invention as claimed. (Office Action, dated 9/1/93, '868 patent prosecution)

Amgen traversed this rejection in part by arguing that it would have been obvious to the skilled worker to be able to make glycosylated proteins from available host cells. In particular, Amgen argued that "<u>numerous other mammalian cells [in addition to CHO and COS] capable of effecting glycosylation of expressed polypeptides were known to those skilled in the art at the time of the present invention</u>." Applicant Amendment and Response, dated January 3, 1994, '868 patent prosecution AM-ITC 00953641 (emphasis supplied). Thus, Amgen conceded during the prosecution of the '868 patent that the process for using host cells capable of effecting post-translational glycosylation was obvious at the time of the invention.

99. Amgen has admitted and has successfully argued in a prior proceeding that the patents-in-suit are to the same invention. Indeed, Amgen successfully argued to the Board of

Patent Appeals that the composition claims and the process claims "<u>are only different manifestations of the same invention</u>" and that "<u>the whole purpose and intent of the purified and isolated DNA sequence encoding human EPO (and host cells transfected therewith)...was to express *in vivo* biologically active human EPO</u>." Brief for the Senior Party Lin, Interference No. 102,097, dated 7/29/91 at 26, AM-ITC 00337678. I agree with these admissions by Amgen. In particular, I agree that the *in vivo* biological activity of EPO does not constitute a separate invention from EPO—and indeed it was the "<u>whole purpose</u>" behind Lin's activity in the EPO project.

100. Amgen succeeded in its argument that it should win the interference proceedings involving the '178 and '179 patent applications because it won the court case involving the '008 patent. In ruling in favor of Dr. Lin (Amgen) on the issue of whether Lin (Amgen) had priority over Fritsch (GI) on these two pending applications, the Board of Patent Appeals and Interferences (BPAI) ruled that the issues "are essentially identical to the issues considered in related Interference No. 102,096," which dealt with the '008 patent. *Fritsch v. Lin*, 21 U.S.P.Q.2d 1737, 1738 (BPAI 1992); *Fritsch v. Lin*, 21 U.S.P.Q.2d 1739, 1741 (BPAI 1992). The subject matter of the '008 patent was defined by the BPAI, for purposes of the interference proceeding, as

> A purified and isolated DNA sequence consisting essentially of a DNA sequence consisting essentially of a DNA sequence encoding human erythropoietin. [*Fritsch v. Lin*, 21 U.S.P.Q. 2d 1731 at 1733 (BPAI 1992).]

The subject matter of the '178 application was defined by the BPAI as:

> A non-naturally occurring glycoprotein product of the expression in a non-human eukaryotic host cell of an exogenous DNA sequence consisting essentially of a DNA sequence encoding human erythropoietin said product possessing the in vivo biological property of causing human bone marrow cells to increase production of reticulocytes and red blood cells and having an average

> carbohydrate composition which differs from that of naturally occurring human erythropoietin. [21 U.S.P.Q.2d at 1740.]

The subject matter of the '179 application was defined by the BPAI as:

> A process for the preparation of an *in vivo* biologically active glycosylated polypeptide comprising the steps of:
> (a) growing a mammalian host cell which is capable of effecting post-translational glycosylation of polypeptides expressed therein and which is transformed or transfected with an isolated DNA sequence encoding a polypeptide having a primary structural conformation sufficiently duplicative of that of naturally occurring human erythropoietin to allow possession of the *in vivo* biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells, or the progeny thereof, under nutrient conditions suitable to allow in sequence,
> (i) transcription within the host cell of said DNA to mRNA in the sequence of transcription reactions directed by the nucleotide sequence of said DNA;
> (ii) translation within said host cell of said mRNA to a polypeptide in the sequence of translation reactions directed by the nucleotide sequence of said transcribed mRNA;
> (iii) glycosylation within said host cell of said polypeptide in a pattern directed by the amino acid sequence of said translated polypeptide and sufficiently duplicative of the pattern of glycosylation of naturally occurring human erythropoietin to allow possession by the translated glycosylated polypeptide product of the *in vivo* biological property of causing bone marrow cells to increase production of reticulocytes and red blood cells; and
> (b) isolating the glycosylated polypeptide so produced.

In my view, the position taken by Amgen and by the BPAI that these are different manifestations of the same invention support my position that the claims-in-suit are not patentably distinct over '016 patent claim 10.

The Inseparability of the Utility of rEPO from rEPO itself.

101. As I noted above, I agree with Amgen's prior admission that "the whole purpose and intent of the purified and isolated DNA sequence encoding human EPO (and host cells transfected therewith) ... was to express *in vivo* biologically active human EPO." Brief for the Senior Party Lin, Interference No. 102,097, dated 7/29/91 at 26, AM-ITC 00337678. See also *Id.* at 57-58. The *in vivo* biological activity of the recombinant EPO that was the subject of the

claims in the Lin patent applications was the precise purpose and utility of the recombinant EPO being developed by Amgen. If the recombinant EPO was not biologically active it would be non-functional. I am not aware that at the time the application for the '016 patent was filed—or at the time the applications for the patents-in-suit were filed, or at any other time—that there was any other purpose or utility of the recombinant EPO other than it be "*in vivo* biologically active."

102. Furthermore, as noted in the Background Sections of the patents-in-suit, cols. 5-7, the *in vivo* biological properties of EPO were very well known in the prior art prior to 1983-1984. I am not aware that Lin discovered any additional new *in vivo* biological properties of EPO.

103. It appears, from the reports of Amgen's experts that I have reviewed, that Amgen is arguing that the subject matter of the '016 patent claim 10 could be practiced in the U.S. without fear of being accused of infringing the patents-in-suit, because one could practice the subject matter of the '016 patent claim 10 with biologically inactive recombinant erythropoietin. In other words, Amgen is in effect arguing that patents-in-suit have not effectively extended the term of the '016 patent claim 10 beyond twenty years from its filing date because one can now practice the '016 patent claim 10 process with inactive recombinant erythropoietin. It seems silly to me to practice a method such as this for recovering rEPO, when the rEPO composition has no *in vivo* biological activity, and it seems disingenuous to suggest that the public can now practice the '016 patent claim 10 in this way. Thus, in my view, a claim limitation requiring the recombinant erythropoietin to have *in vivo* biological activity cannot provide a patentable distinction over the recombinant erythropoietin of the '016 patent claim 10.

Dr. Fritsch's Ability to Make Biologically Active EPO

104. I have reviewed paras. 224-243 of the Lowe Report and paras. 207-226 of the Kellems Report, which paragraphs relate to the expression of rEPO at Genetics Institute ("GI") after the EPO gene was cloned. It appears that, once GI had isolated the correct DNA sequence and had transfected a mammalian host cell with the correct isolated DNA sequence, GI obtained *in vivo* biologically active EPO without any undue experimentation. Presumably, GI and Fritsch did not have access to the disclosure in the patents-in-suit at this time, but nevertheless GI and Fritsch were easily able to get *in vivo* biologically active erythropoietin from "recombinant erythropoietin from a mammalian cell culture." GI's experience supports my view that recombinant erythropoietin's *in vivo* biologically activity would have been a reasonable expectation of someone of ordinary skill in the field in 1983 in light of the "recombinant erythropoietin from a mammalian cell culture" of the '016 patent claim 10.

Amgen's Admissions Made in Defeating a Best Mode Attack

105. The lack of inventiveness in choosing mammalian host cells, and in particular in choosing mammalian host cells that grow EPO at higher rates, was shown in litigation on the '008 patent. In particular, in *Amgen Inc. v. Chugai Pharmaceutical Co. Ltd.*, 927 F.2d 1200, 18 U.S.P.Q.2d 1016 (Fed. Cir. 1991), the defendant argued that Amgen's failure to deposit or fully disclose the "specific genetically-heterogeneous strain of Chinese hamster ovary (CHO) cells at a rate greater than that of other cells"[27] rendered the '008 patent invalid under 35 U.S.C. Sec. 112 for failing to set forth the best mode—in particular, the best mammalian host cells known to Lin as of November 30, 1984 (the date Lin filed his fourth patent application, from which the patents-in-suit claim priority) for producing rEPO. The Federal Circuit found no violation of the

[27] *Chugai*, 927 F.2d at 1209, 18 U.S.P.Q.2d at 1023.

best mode requirement, quoting prior case law that "No problem exists when the microorganisms used are known and readily available to the public." *Chugai*, 927 F.2d at 1211, 18 U.S.P.Q.2d at 1025. This court decision confirms my view that the choice of host cells (including the choice of host cells that could grow EPO at higher rates) did not involve any inventive activity on Lin's part and is not a patentable distinction over the "recombinant erythropoietin from a mammalian cell culture" of '016 patent claim 10.

Other Statements Made in the USPTO

106. I have also reviewed statements made by Amgen and by the examiner in the prosecution histories of the patents-in-suit that support my opinion that each of the claims-in-suit would have been obvious in light of the subject matter of the '016 patent claim 10. The claims at issue during the prosecution when these statements were made are not identical to all the claims-in-suit, but the claims are similar enough that I feel these statements support my opinion.

107. During the prosecution of the '178 application—from which '933 patents claim priority—and while traversing a double patenting rejection, Amgen admitted the subject matter claimed in the '016 patent (which Amgen called the "Lai patent," claiming the "Lai process") was covered by the claims of the '178 application. Amgen argued

> The fact that both the starting material and final product of the Lai Process (neither of which are claimed in Lai patent) are *included within (dominated by)* the recombinant product claims of the present application is <u>not</u> a basis for a double patenting rejection.

(Italics added. Underlining in original.) Page 3 of January 11, 1990 Amendment in the 07/113,178 File Wrapper. (In a footnote, Amgen explained

> The Examiner must not confuse double patenting with "domination" which by itself does not give rise to double patenting. Domination occurs when one patent has claims which "read on" the invention defined by the claims of another patent, thus one patent dominates the other when

> practicing the invention of the other infringes the dominating patent (see In re Kaplan, supra).

I do not understand why Amgen made this admission, but "[t]he fact that … the … final product of the ['016 patent] … [is] *included within (dominated by)* the recombinant product claims of the present application" means that these recombinant product claims must be obvious in light of the '016 patent claim 10.

108. The claims of the '178 application that Amgen admitted dominated the final product of the '016 claims read as follows (emphasis added):

> 76. A non-naturally occurring glycoprotein product of the expression in a non-human eukaryotic host of an exogenous DNA sequence consisting essentially of a DNA sequence encoding human erythropoietin said product possessing the **in vivo biological property of causing human bone marrow cells to increase production of reticulocytes and red blood cells** and having an average carbohydrate composition which differs from that of naturally occurring human erythropoietin.
>
> 77. A glycoprotein product according to Claim 76 wherein the exogenous DNA sequence is a cDNA sequence.
>
> 78. A glycoprotein product according to Claim 76 wherein the exogenous DNA sequence is a genomic DNA sequence.
>
> 79. A glycoprotein product according to Claim 76, 77, or 78 wherein the host cell is a mammalian cell.
>
> 80. A glycoprotein product according to Claim 79 wherein the non-human eukaryotic host cell is a CHO cell.
>
> 81. A pharmaceutical composition comprising an effective amount of a glycoprotein product according to Claim 76 and a pharmaceutically acceptable diluent, adjuvant or carrier.
>
> 82. A method for providing erythropoietin therapy to a mammal comprising administering an effective amount of glycoprotein product of Claim 76.
>
> 83. A method according to Claim 76 wherein the therapy comprises enhancing hematocrit levels.

Thus, the pending claims in the '178 application required that the "glycoprotein product … possess[] the in vivo biological property of causing human bone marrow cells to increase production of reticulocytes and red blood cells …." Nevertheless, Amgen admitted that the '016 claims were "dominated by" and were "included within" these '178 claims. Thus, in 1990, Amgen's position was that the process of the '016 claims could not be practiced without being dominated by the '178 claims (regardless of whether the '016 patent claim 10 explicitly set forth the biological activity expected from rEPO). This admission by Amgen supports my opinion that "the in vivo biological property of causing human bone marrow cells to increase production of reticulocytes and red blood cells" is not a patentably distinct feature over rEPO itself.

109. During the prosecution of the '179 application—from which the '868, '349, '698 and '422 patents claim priority—the examiner made an obviousness-type double patenting rejection based on the '016 patent (Lai et al.). Amgen argued that a "two-way test" should be used. In withdrawing this obviousness-type double patenting rejection, the examiner agreed to apply the two-way test, but also noted that the pending claims of the '179 application were indeed obvious over the claims of the '016 patent:

> … And while the instantly claimed method is **an obvious variation of the process of Lai et al.** it is considered that applicant is not responsible for the delay in the prosecution of the instant application which resulted in the prior patenting of a later filed application to an invention derived from the instant invention. …

(Emphasis added.) '179 File History, Paper 34, 2/15/94 Office Action at 2. The claims pending at the time the examiner made this statement read as follows:

> 70. A process for the preparation of an in vivo biologically active glycosylated erythropoietin polypeptide comprising the steps of:
> (a) growing under suitable nutrient conditions, mammalian host cells transformed or transfected with an isolated DNA sequence encoding human erythropoietin; and

> (b) isolating said glycosylated erythropoietin polypeptide therefrom.
>
> 71. A process for the preparation of an in vivo biologically active glycosylated erythropoietin polypeptide comprising the steps of:
> (a) growing under suitable nutrient conditions, mammalian host cells transformed or transfected with an isolated DNA sequence selected from the group consisting of (1) the DNA sequences set out in FIGS. 5 and 6 or their complementary strands, (2) the protein coding sequences set out in FIGS. 5 and 6 or their complementary strands, and (3) DNA sequences which hybridize under stringent conditions to the DNA sequences defined in (1) and (2) and;
> (b) isolating said glycosylated polypeptide erythropoietin therefrom.
>
> 72. The process according to Claim 1 wherein said host cells are CHO cells.
>
> 73. The process according to Claim 1 wherein said host cells are COS cells.
>
> 74. The process according to Claim 1 wherein said DNA is cDNA.
>
> 75. The process according to Claim 1 wherein said DNA is genomic DNA.

I agree with the examiner's opinion that this claimed subject matter was *an obvious variation of the process* of the process claimed in the '016 patent. (Whether the examiner was correct in applying the two-way test or the one-way test is beyond the scope of what I have been asked to opine on.)

110. In dealing with an inventorship issued raised during one of the interference proceedings, the Patent and Trademark Office (PTO) has recognized that just given the isolation of the EPO gene needed for use in the mammalian cell culture, the process steps for making glycosylated biologically active EPO "<u>d[id] not require the exercise of inventive skill</u>." *Fritsch v. Lin*, 21 U.S.P.Q.2d 1737, 1739 (BPAI 1992) (emphasis supplied).

Amgen's Assertion that Lin is the Sole Inventor of All the Claims in the Patents-in-Suit and in the '008 Patent Supports my Position that the Claims-in-Suit Provide No Patentably Distinct Features over the Subject Matter of the '016 Patent

111. Amgen has listed Lin as the sole inventor of the patents-in-suit, because Amgen asserts that Lin was the individual who identified the DNA sequence claimed in the '008 patent and he asked other people in the Amgen EPO Project to perform additional non-inventive tasks, such as choosing host cells, expressing proteins from host cells, isolating rEPO from the host cell material, and preparing pharmaceutical compositions from purified rEPO.

112. The Board of Patent Appeals and Interferences stated in their Final Judgment of December 3, 1991, in an Interference proceeding in which Amgen was a party-in-interest and which involved the subject matter of the claims-in-suit, that:

> The record indicates that all the work at Amgen relating to expression of the EPO gene in mammalian host cells was directed and supervised by Dr. Browne, assisted by Ralph Smalling. Dr. Lin does not recall giving any instructions or suggestions as to how such expression should be carried out (PF V-3, 4). The effort to isolate the EPO glycoprotein expression product was carried out by Dr. Strickland, and Dr. Lin gave no specific instructions for accomplishing that task (PF V-6). [*Fritsch v. Lin,* 21 USPQ 2d 1739, 1991 WL 332571 *2 (BPAI 1991)]

113. From reading the foregoing, I conclude that Dr. Lin only provided Dr. Browne and Ralph Smalling with the DNA sequence and provided them with no further instructions. Thus, in my opinion, the expression of recombinant human EPO would have been obvious to one skilled in the art at the time, once the gene sequence for EPO was known. Otherwise, Dr. Lin should not have been listed by Amgen as the sole inventor on each of the patents-in-suit, since he provided no instructions for expressing EPO within a mammalian cell line to the people working on this aspect of the project. Similarly, Dr. Lin provided no instructions for isolating the EPO glycoprotein expression product. Therefore, the other people working with Dr. Lin on the EPO

project must have relied simply on the techniques and operating conditions known to those of ordinary skill in the art for expressing recombinant proteins in mammalian cells and isolating EPO therefrom.

114. Dr. Lin testified about his involvement (or rather his lack thereof) in various aspects of Amgen's EPO Project. Dr. Lin testified as follows on April 9, 1991 during the Interference proceedings (AM-ITC 00410949 - 00410952):

> Q. Now, the district court decision indicates that someone other than Dr. Lin did the work with the mammalian expression system. Do you see that in the third quoted paragraph?
> A. That's correct, yes.
> Q. And that is a correct statement; right?
> A. That is correct, yes.
>
> * * *
>
> Q. Now, you indicate in your declaration in paragraph 5 that the expression of EPO in the 293 cells, the COS cells, and the CHO cells was carried out by Dr. Jeff Browne?
> A. By Jeff Browne and his associates.
> Q. And his associates, one of whom was Mr. Smalling?
> A. That's correct, yes.
>
> * * *
>
> Q. And in that connection, you provided Dr. Browne with a DNA sequence which included the sequence coding human EPO; is that right?
> A. That's correct, yes.
> Q. And you also provided Dr. Browne with a monkey cDNA sequence which encoded monkey EPO; is that right?
> A. That's correct, yes.
> Q. And you then requested that Dr. Browne and his colleagues do whatever work was necessary to obtain expression in mammalian cells?
> A. That's correct, yes.
>
> * * *
>
> Q. Did you leave up to the judgment of Dr. Browne and his associates how to accomplish that result?
> A. Could you repeat the question again?

Q. Yeah. Did you rely on Dr. Browne and his colleagues for selecting the way to achieve that result?
A. 'That result' means expression?
Q. Expression in the mammalian host cell system.
A. Mostly, yes.

On the same date Dr. Lin also testified as follows (AM-ITC 00410953):

Q. So it was Dr. Browne and his colleagues who selected the vector for transfecting or transforming the cells; is that correct?
MR. SCOTT: Calling for hearsay of our witness.
THE WITNESS: Could you repeat the question.
BY MR. RICHTER:
Q. Let me rephrase the question in light of your counsel's objection. It was not you who selected the vector for transforming or transfecting the cells; is that correct?
A. That's correct, yes.
Q. You didn't design that vector?
A. No.
Q. And you didn't make it?
A. No.

On the same date Dr. Lin also testified as follows (AM-ITC 00410953 - 00410954):

Q. Did you give Dr. Browne any specific instructions as to the ***culture medium that should be used to grow the cells?***
A. No.

(Emphasis added.) Thus, Dr. Lin only provided Dr. Browne and Ralph Smalling with the DNA sequence and provided them with no further instructions. Similarly, Dr. Lin provided no instruction regarding the vectors and culture medium. Thus, he was not involved in transfecting host cells or expressing rEPO therefrom.

115. On April 9, 1991 Dr. Lin also testified as follows (AM-ITC 00410960 – 00410961):

THE WITNESS: ***I did not personally involve in the isolation of glycosolated polypeptide***.

BY MR. RICHTER: Who was personally involved in that effort?
A. I believe Tom Strickland and his associates.

Q. And it was Dr. Strickland who worked on the purification of erythropoietin at Amgen and his colleagues?
A. Yes. Tom Strickland and his colleagues.
Q. Did you assign that task to Dr. Strickland?
A. Yes.
Q. Did you give Dr. Strickland any specific instructions how to obtain purified erythropoietin?
A. I think I gave him some of the literature regarding the purification of the erythropoietin.
Q. I'm sorry, I didn't understand your answer.
A. Some of the literature regarding the purification of urinary erythropoietin, if I remember correctly.
Q. You gave him some literature?
A. Yeah, I believe so.

(Emphasis added.) Thus, Dr. Lin was not involved in isolating rEPO.

116. On this same date, Dr. Lin also testified as follows (AM-ITC 00410963 – 00410964):

Q. I'd like to refer you first to page 64 of the application, which is part of example 10, and you find the paragraph which begins "mammalian cell expression" at line 16?
A. Yes, I do.
Q. And it reads, "Mammalian cell expression products may be readily recovered in substantially purified form from culture media using HPLC (C4) employing an ethanol grading, preferably at [pH7]." Did I read that correctly?
A. That's correct, yes.
Q. Now, the HPLC procedure, which is referred to there, is a reference, or is one attempt, to isolate recombinant EPO from a mammalian expression system; is that correct?
A. That's correct, yes.
Q. Is it true that the suggestion or the idea of using HPLC (C4) in an effort to isolate erythropoietin from a mammalian expression system culture medium was not your suggestion or idea?
A. No.
Q. That is correct?
A. That's correct, yes.
Q. And do you know whose suggestion or idea it was?
A. Now I remember. This probably was Por Lai?[[28]]
Q. Por Lai?

[28] Dr. Lai is an inventor of the '016 patent.

A. Yeah. He was doing the protein sequencing at the time.

Thus, Dr. Lin was not involved in the isolation process of EPO at Amgen and that he provided this task to Dr. Thomas Strickland. Dr. Lin provided no other information to Dr. Strickland other than the known literature regarding isolation of human EPO.

117. Dr. Lin also admitted that the entire approach of using CHO cells for expressing a human glycoprotein, including use of amplification for expressing the recombinant protein at high levels was routine and predictable:

Q. And in 1983 as a matter of fact, the Amgen scientists had the technology to insert the human genomic clone for erythropoietin into a plasmid and transform or transfect it into a COS cell or a CHO cell. Is that right?

A. That's correct, yes.

Q. And other scientists in the industry also had that technology at this time?

A. Yes. That's correct. I think so. (AM-ITC 00177955)

Q. Tell me where I would have gone and said, give me CHO B11-3, .1.

A. Oh, there's so many people in the field who have used CHO B11, that -- apparently, many knew how to get it before we used it, so it's no problem. You get it from Dr. Chasin at Columbia University.

Q. Well, Dr. Chasin gives you CHO B11, correct?

A. Yes.

Q. CHO B11-3, .1, is something that has gone through some processes, hasn't it?

A. Don't try to use the term to confuse it. As I explained many times, B11-3, .1, means that the CHO B11 has been amplified with 30 nanomolar MTX and 100 nanomolar MTX. And it's a heterogeneous population. Anyone in the field can do it. (AM-ITC 00113336-37)

118. Dr. Lin admitted that his choice of CHO cells was driven by the need for a reliable and very stable cell line, in other words, by essentially the same concerns that would have led anyone in the art to use CHO cells to express a recombinant human glycoprotein:

Q. That's fine. And who suggested using CHO cells for

the expression of the EPO gene?

A. I may have suggested -- besides actually CHO cells, I also have actually look into other cells. I think there's another cell.

I talk to the people at ATCC at the time. There's only two stable cells that can be used -- I mean the stable cell line they

could use. One is the other cells. Now I cannot remember. There's another cell. It's also very stable for the purpose of production. So CHO cells and the other one, yes, which I -- I talked to one of the guys at ATCC.

Q. So your testimony is that you picked CHO cells because you called the ATCC, and they told you that's what you should use?

MR. MADRID: Objection.

Misstates the testimony.

THE WITNESS: No, no, no. I not call the ATCC.

I was looking for cells of which would be suitable for commercial production. Therefore, the cells -- cell line to be used had to be stable so that's why I would -- try to decide which cell line is to be used. So I call my friend at ATCC and talked to people to see what the -- what the cell line would be good to use. And the CHO cell come up as one. And the other one, I just cannot remember. There's another one that is also very stable. (March 28, 2007 Deposition of Dr. Lin at 63-65).

Q So before you called your friend at the ATCC -- strike that. You learned of CHO cells from your friend at the ATCC; right?

A Uh, yes. The CHO cell and the other cell, yeah.

Q You learned that -- you called your friend at the ATCC and said, "What cells should I use for stable mammalian expression?"; right?

A No, no.

MR. MADRID: Objection. Misstates the testimony.

THE WITNESS: No, that's not my question -- my question that I have for him is could you find a stable cell line that -- that -- you know, a stable cell line that -- for me, in your -- in your ATCC collection, what other cell line which are stable because I want stable cell line to be used for our research purpose, yeah -- for the EPO project purpose, yes.

BY MS. BEN-AMI:

Q And he directed you to a professor at Columbia?

A No, no, no, no, no, no, no. No, he did not direct me to professor at Columbia. He told me that CHO cell line and the

other cell line, that it -- stable cell line, and the other one maybe, even more stable than CHO cell. Yeah. So the cell -- the Columbia

University cell line is a different cell line. The CHO cell line was DHFR minus.

Q So it is a CHO cell line?

A That's correct. But it's DHFR gene minus. It's a different one. It's a variant of CHO cell. That's correct.

119. These admissions by Dr. Lin support my opinion that the choice of CHO cells and the use of other recombinant DNA engineering techniques would have been obvious to use in 1983 for making recombinant human EPO as a commercial product. As Dr. Lin acknowledged, it was certainly reasonable to expect that when expressed in a mammalian cell, the human EPO gene would yield an *in vivo* biologically active product:

Q. My question was, whether you had the expectation when you had the genomic EPO gene that when put into a mammalian cell and expressed, that the resulting EPO would be biologically active?

A I---

THE WITNESS: Of course, we would expect that it – to be -- to have that activity -- in the biological activity.

(March 29, 2007 Deposition of Dr. Lin at 368).

120. Reviewing this testimony reinforces my opinion that the expression of *in vivo* biologically active recombinant human EPO would have been obvious to one skilled in the art at the time, once the gene sequence for EPO was known. Otherwise, Dr. Lin should not have been listed by Amgen as the sole inventor on each of the patents-in-suit, since he provided no instructions for expressing EPO within a mammalian cell line to the people working on this aspect of the project. Similarly, Dr. Lin provided no instructions for isolating the EPO glycoprotein expression product, other than requesting that it be done. Therefore, the other people working with Dr. Lin on the EPO project must have relied simply on the techniques and operating conditions known to those of ordinary skill in the art for expressing recombinant proteins in mammalian cells and isolating EPO therefrom. This testimony also supports my

opinion of the level of skill in the field and my opinion of what someone of ordinary skill in the field would have found to be obvious. Thus, this supports my opinion that the claims-in-suit would have been obvious over the claims of the '016 and '008 patents.

121. Dr. Lin's lack of involvement in expression and isolation of rEPO—other than isolating the EPO gene—also supports my opinion of the level of skill in the field and my opinion of what someone of ordinary skill in the field would have found to be obvious, as well as my opinion that the claims-in-suit would have been obvious over the claims of the '016 patent.

XII. CONCLUSION

122. Amgen has already admitted that once the EPO gene has been cloned the remaining steps for producing *in vivo* biologically active rEPO are not inventive steps. This admission is confirmed by Genetics Institute near-contemporaneous work in this area. This admission is also confirmed by Amgen's listing of Dr. Lin as the sole inventor of the patents-in-suit, even though he does not appear to have done any inventive work beyond the isolation of the EPO gene. Also, Dr. Lodish has already admitted that—as early as 1980, several years before the filing of the applications that led to the patents-in-suit—many of the techniques used in this field were already obvious and well known, including the glycosylation of proteins, the transformation of mammalian cells with exogenous DNA, the use of CHO cells for producing recombinant proteins, amplification of genes in mammalian cell cultures, the use of dihydroflate reductase (DHFR), and the use of viral promoters. These admissions and the other facts related above support my opinion that each of the claims-in-suit would have been obvious over the subject matter of claim 10 of the '016 patent.

123. For all the reasons set forth above, it would have been obvious to one of skill in the art in 1983 to use the mammalian cell culture from which to recover recombinant EPO (as

claimed in the '016 patent) thereby producing the glycosylated, biologically active rEPO protein of the claims-in-suit. Accordingly, the processes, the proteins, the formulations and the treatments using same, all described in the claims-in-suit are just rewordings or obvious variations of the subject matter the '016 patent claim 10.

124. Claim charts setting forth my claim-by-claim analysis is included herewith as Exhibit 33.

125. I declare that the foregoing is true and correct to the best of my knowledge and belief.

Executed this 12th day of June 2007 at Boston, Massachusetts.

/s/ Edward Everett Harlow, Jr.
Edward Everett Harlow, Jr.

CERTIFICATE OF SERVICE

I hereby certify that this document filed through the ECF system will be sent electronically to the registered participants as identified on the Notice of Electronic Filing (NEF) and paper copies will be sent to those indicated as non-registered participants on the above date.

/s/ Nicole A. Rizzo
Nicole A. Rizzo

List of Exhibits

Exhibit 1 Curriculum Vitae

Exhibit 2 Li et al., "Biosynthesis of lipid-linked oligosaccharides," J. BIOL. CHEM., 254: 1600-1605 (1979).

Exhibit 3 Scahill, et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983).

Exhibit 4 Singer et al., "Characterization of a porcine genomic clone encoding a major histocompatibility antigen: Expression in mouse L cells," PROC. NAT'L ACAD. SCI. 79:1403-1407 (1982).

Exhibit 5 Haynes and Weissman, "Constitutive, long-term production of human interferons by hamster cells containing multiple copies of a cloned interferon gene," NUCL. ACIDS RES., 11: 687-706 (1983).

Exhibit 6 Sherwood and Goldwasser, "A radioimmunoassay for erythropoietin," BLOOD, 54: 885-893 (1979).

Exhibit 7 EPO 0 093 619 (Goeddel et al, 1983); Ex. 3, Scahill et al., "Expression and characterization of the product of a human immune interferon cDNA gene in Chinese hamster ovary cells," PROC. NAT'L ACAD. SCI. 80: 4654-4658 (1983).

Exhibit 8 EPO 0 117 059, 0 117 060 (Levinson et al, 1984).

Exhibit 9 McCormick et al., "Inducible expression of amplified human beta interferon genes in CHO cells," MOL. & CELL. BIOL., 4: 166-172 (1984).

Exhibit 10 Devos et al., "Purification of recombinant glycosylated human gamma interferon expressed in transformed Chinese hamster ovary cells," J. INTERFERON RES., 4: 461-468 (1984).

Exhibit 11 U.S. Pat. No. 4,766,075 (to Goeddel)

Exhibit 12 U.S. Pat. No. 4,356,270 (to Itakura)

Exhibit 13 U.S. Pat. No. 4,264,731 (to Shine)

Exhibit 14 U.S. Pat. No. 4,273,875 (to Manis)

Exhibit 15 U.S. Pat. No. 4,293,652 (to Cohen)

Exhibit 16 Srinivasan and Lewis, "Transfer of the Dihydrofolate Reductase Gene into Mammalian Cells Using Metaphase Chromosomes or Purified DNA,"

INTRODUCTION OF MACROMOLECULES INTO VIABLE MAMMALIAN CELLS, 27-45 (1980).

Exhibit 17 Milbrandt et al., "Organization of a Chinese Hamster Ovary Dihydrofolate Reductase Gene Identified by Phenotypic Rescue," MOL. CELL BIOL. 3: 1266-1273 (1983).

Exhibit 18 Nunberg et al., "Amplified dihydrofolate reductase genes are localized to a homogeneously staining region of a single chromosome in a methotrexate-resistant Chinese hamster ovary cell line," PROC. NAT'L ACAD. SCI., 75**:** 5553-5556 (1978).

Exhibit 19 U.S. Pat. No. 4399216 (Axel et al., filed 1980).

Exhibit 20 Urlaub and Chasin, "Isolation of Chinese hamster cell mutants deficient in dihydrofolate reductase activity," PROC. NAT'L ACAD. SCI., 77: 4216-4220 (1980).

Exhibit 21 Schimke et al. "Gene amplification and drug resistance in cultured murine cells," SCIENCE, 202: 1051-1055 (1978).

Exhibit 22 Miyake, Goldwasser et al, "Purification of Human Erythropoietin," J. BIOL. CHEM., 252: 5558-5564 (1977).

Exhibit 23 Goldwasser et al., "An assay for erythropoietin *in vitro* at the milliunit level," ENDOCRINOLOGY, 97: 315-323 (1975).

Exhibit 24 Cotes and Bangham, "Bio-assay of erythropoietin in mice made polycythæmic by exposure to air at a reduced pressure," NATURE, 191: 1065-1067 (1961).

Exhibit 25 Hammond et al., "Production, utilization and excretion of erythropoietin," ANN. N.Y. ACAD. SCI., 169: 516-527 (1968).

Exhibit 26 Weiss, et al., "Characterization of a monoclonal antibody to human erythropoietin," BIOCHEMISTRY, 79: 5465-5469 (1982).

Exhibit 27 Gray and Goeddel, "Cloning and Expression of Murine Immune Interferon cDNA," PROC. NAT'L. ACAD. SCI., 80:5842-5846 (1983).

Exhibit 28 Taniguchi et al., "Construction and Identification of a Bacterial Plasmid Containing the Human Fibroblast Interferon Gene Sequence," PROC. JAPAN ACAD., 55: 464-469 (1979).

Exhibit 29 Oi et al., "Immunoglobulin gene expression in transformed lymphoid cells," PROC. NAT'L. ACAD. SCI. 80:825-829 (1983).

Exhibit 30 Neuberger, "Expression and regulation of immunoglobulin heavy chain gene transfected into lymphoid cells," EMBO J. 2:1373-1378 (1983).

Exhibit 31	Sidman, "Differing requirements for glycosylation in the secretion of related glycoproteins is determined neither by the producing cell nor by the relative number of oligosaccharide units," J. BIOL. CHEM., 256: 9374-9376 (1981).
Exhibit 32	Seidman et al., "Antibody Diversity," SCIENCE, 202: 11 (1978).
Exhibit 33	Claim Charts