

UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE PATENT TRIAL AND APPEAL BOARD

SAREPTA THERAPEUTICS, INC.,
Petitioner

v.

GENZYME CORPORATION,
Patent Owner.

Case: IPR2026-00150
Patent No. 12,031,894

PATENT OWNER'S RESPONSE

TABLE OF CONTENTS

Table of Authorities	iv
Table of Exhibits	vi
I. Introduction	1
II. State of the Prior Art for SV-AUC of rAAV Particles	5
A. Determining the Quantity of a Viral Particle Species by Absorbance Detection Typically Requires Determining the Size of the Encapsidated Genome	5
B. Particle Friction Matters for SV-AUC	7
C. The POSA Understood that Genome-Containing rAAV Particles Were Not of Identical Size and Shape	10
III. The '894 Patent	13
A. Disclosed Innovation of the '894 Patent	13
1. Overcoming the Challenge of Determining rAAV Species Concentration by AUC	14
2. One-Step rAAV Preparation Analysis by AUC	16
a. Example 5: Application of the Standard Curve	16
b. The Power of the Innovative AUC Method Compared to Southern Blot	18
B. Prosecution History	23
C. Person of Skill in the Art	23
D. Challenged Claims	24
IV. Claim Construction	25
V. Ground 1: Sarepta Fails to Establish That Le Bec and de la Maza, In View of	

Sommer Rendered Claims 1-7, 10-11, and 20-30 Obvious.....	26
A. Claim 1: Sarepta Fails to Establish that Claim 1 Would Have Been Obvious as of the Effective Date	26
1. Le Bec and de la Maza, in View of Sommer, Did Not Render Obvious “determining the genome size of one or more species of the individual variant viral particles” with SV-AUC.....	29
a. The POSA Would Not Have Been Motivated to “determin[e] the genome size of one or more species of the individual variant viral particles” with SV-AUC	34
i. The POSA Would Not Have Been Motivated to Use SV-AUC Instead of SE-AUC to Determine Genome Size.....	37
ii. Le Bec’s Data Belie a Linear Relationship Between Genome Size and Sedimentation Coefficient	38
iii. Carter Publications Teach Away From a Reliable Linear Relationship Between Genome Size and rAAV Particle Density	42
iv. Carter Laboratory Publications Belie a Linear Relationship Between Genome Size and Sedimentation Coefficient.....	44
v. The POSA Would Not Have Ignored the Effect of Friction on Sedimentation Rate.....	47
vi. As Dr. Berkowitz Testified, the Prior Art Showed that rAAV Particle Shape Was Unpredictable.....	53
b. The POSA Would Not Have Reasonably Expected that Sedimentation Coefficients Could Be Used to Determine Fragmented Genome Size in rAAV Particles	55
2. Le Bec and de la Maza, In View of Sommer Did Not Render Obvious “determining the quantity of one or more species of the variant viral particles in the heterogeneous mixture of viral particles”	58
B. Claim 2: The Cited Prior Art Fails to Teach or Suggest an SV-AUC Method “wherein each species of the individual variant viral particles ... is fully resolved”	60
C. Claims 3-7, 10-11, and 20-30 Depend from Claim 1 and Are Not Unpatentable for the Same Reasons as Claim 1	63
VI. Ground 2: The Petition Fails to Establish Le Bec and de la Maza, in view of	

Sommer and Cole, Renders Claims 12-19 Obvious	63
VII. Objective Indicia of Nonobviousness Support the Patentability of the Challenged Claims	63
A. The Claimed Method’s Capability to Determine Genome Size and Quantify Fragmented Particles Was Surprising and Unexpected.....	64
B. Industry Praise Is Further Evidence of Nonobviousness	66
C. The Claimed Method Satisfied a Long-Felt Need for Characterizing rAAV Impurities	69
VIII. Conclusion	73

TABLE OF AUTHORITIES

	Page(s)
Cases	
<i>Amneal Pharms. LLC v. Cubist Pharms. LLC</i> , IPR2020-00193, Paper 7 (PTAB May 29, 2020)	57
<i>Arendi S.A.R.L. v. Apple Inc.</i> , 832 F.3d 1355 (Fed. Cir. 2016)	4, 55
<i>ATD Corp. v. Lydall, Inc.</i> , 159 F.3d 534 (Fed. Cir. 1998)	4, 54
<i>Belden Inc. v. Berk-Tek LLC</i> , 805 F.3d 1064 (Fed. Cir. 2015)	36
<i>Coherus Biosciences Inc. v. AbbVie Biotechnology Ltd.</i> , IPR2016-01018, Paper 12 (PTAB Feb. 2, 2017).....	58
<i>Creo Prod., Inc. v. Presstek, Inc.</i> , 166 F. Supp. 2d 944 (D. Del. 2001).....	28, 47
<i>In re Cyclobenzaprine</i> , 676 F.3d 1063 (Fed. Cir. 2012)	64
<i>Depuy Spine, Inc. v. Medtronic Sofamor Danek, Inc.</i> , 567 F.3d 1314 (Fed. Cir. 2009)	58
<i>Forest Labs., LLC v. Sigmapharm Labs., LLC</i> , 918 F.3d 928 (Fed. Cir. 2019)	64, 66
<i>Henny Penny Corp. v. Frymaster LLC</i> , 938 F.3d 1324 (Fed. Cir. 2019)	4, 42
<i>Intelligent Bio-Sys., Inc. v. Illumina Cambridge Ltd.</i> , 821 F.3d 1359 (Fed. Cir. 2016)	55
<i>InTouch Techs., Inc. v. VGO Commc'ns</i> , 751 F.3d 1327 (Fed. Cir. 2014)	55
<i>Janssen Pharms., Inc. v. Teva Pharms. USA, Inc.</i> , 97 F.4th 915 (Fed. Cir. 2024)	64

<i>Jones v. Hardy</i> , 727 F.2d 1524 (Fed. Cir. 1984)	28, 47
<i>Leo Pharm. Prods., Ltd. v. Rea</i> , 726 F.3d 1346 (Fed. Cir. 2013)	37, 54, 64
<i>In re Magnum Oil Tools Int’l Ltd.</i> , 829 F.3d 1364 (Fed. Cir. 2016)	26
<i>Momenta Pharms., Inc. v. Bristol-Myers Squibb Co.</i> , IPR2015-01537, Paper 37 (PTAB Dec. 22, 2016)	58
<i>Ortho–McNeil Pharm., Inc. v. Mylan Labs., Inc.</i> , 520 F.3d 1358 (Fed. Cir. 2008)	64
<i>Quanergy Sys. v. Velodyne Lidar USA, Inc.</i> , 24 F.4th 1406 (Fed. Cir. 2022)	69
<i>Rockwell Int’l Corp. v. U.S.</i> , 147 F.3d 1358 (Fed. Cir. 1998)	28, 47
<i>Sandoz Inc. v. AbbVie Biotechnology Ltd.</i> , IPR2017-01823, Paper 16 (PTAB Feb. 9, 2018).....	58
<i>In re Soni</i> , 54 F.3d 746 (Fed. Cir. 1995)	64
<i>Transocean Offshore Deepwater Drilling, Inc. v. Maersk Drilling USA, Inc.</i> , 699 F.3d 1340 (Fed. Cir. 2012).....	72
<i>Trivascular, Inc. v. Samuels</i> , 812 F.3d 1056 (Fed. Cir. 2016)	55
<i>W.L. Gore & Assocs., Inc. v. Garlock, Inc.</i> , 721 F.2d 1540 (Fed. Cir. 1983)	28, 42, 47
<i>Application of Wesslau</i> , 353 F.2d 238 (C.C.P.A. 1965)	42

TABLE OF EXHIBITS

Exhibit No.	Description
2001	Declaration of Katherine A. Helm
2002	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 78 – Joint Stipulation to Amend Scheduling Order
2003	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 59 – Notice of Service of Plaintiff’s Initial Claim Charts on Infringement
2004	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 68 – Notice of Service of Defendant’s Initial Invalidity Contentions
2005	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 28 - Notice of Service of Plaintiff’s First Set of Requests for Production of Documents and Things
2006	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 35 - Notice of Service of Defendant’s First Set of Requests of Production of Documents and Things to Plaintiff and First Set of Interrogatories and Initial Disclosures
2007	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 37 - Notice of Service of Plaintiff’s Second Set of Interrogatories and Second Set of Requests for Production of Documents and Things
2008	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 45 – Notice of Service of Plaintiff’s First Set of Interrogatories and Third Set of Requests for Production of Documents and Things
2009	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 67 - Notice of Service of Defendant’s Second Set of Requests of Production of Documents and Things to Plaintiff and First Set of Interrogatories
2010	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 83 - Notice of Service of Defendant’s Amended Second Set of Requests of Production of Documents and Things to Plaintiff and First Set of Interrogatories
2011	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-

Exhibit No.	Description
	00882) D.I. 87 - Notice of Service of Defendant's Third Set of Requests of Production of Documents and Things to Plaintiff
2012	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 91 – Notice of Service of Plaintiff's Responses to Defendant's Third Set of Requests for Production of Documents and Things
2013	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 92 - Notice of Service of Plaintiff's Fourth Set of Requests for Documents and Things
2014	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 99 – Notice of Service of Plaintiff's Fifth Set of Requests for Documents and Things
2015	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 102 – Notice of Service of Defendant's Fourth Set of Requests of Production of Documents and Things to Plaintiff
2016	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 110 – Notice of Service of Plaintiff's Sixth Set of Requests for Production of Documents and Things
2017	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) D.I. 76 – Second Amended Complaint
2018	<i>Genzyme Corporation v. Sarepta Therapeutics Inc.</i> (1:24-cv-00882) - Defendant's Supplemental Initial Invalidity Contentions
2019	Burnham et al., Analytical Ultracentrifugation as an Approach to Characterize Recombinant AAV Vectors, 26 <i>Human Gene Therapy Methods</i> 228 (2015) ("Burnham 2015")
2020	F. Collaud et al., Preclinical Development of an AAV8-hUGT1A1 Vector for the Treatment of Crigler-Najjar Syndrome, 12 <i>Molecular Therapy: Methods & Clinical Development</i> 157, 166-67 (March 2019)
2021	E. Spies, Assessing the Quality of Adeno-Associated Virus Gene Therapy Vectors by Sedimentation Velocity Analysis, <i>AUC Insights</i> (March 2020)
2022	<i>Novartis Gene Therapies, Inc. v. Genzyme Corp.</i> , IPR2023-01044, Paper 12 (Jan. 23, 2024), Patent Owner's Request for Rehearing by the Director

Exhibit No.	Description
2023	US 2017/0362608 to McLaughlin et al. (“McLaughlin”)
2024	<i>Novartis Gene Therapies, Inc. v. Genzyme Corp.</i> , IPR2023-01044, Paper 1 (Jun. 1, 2023).
2025	Q. Xie et al., The Atomic Structure of Adeno-Associated Virus (AAV-2), a Vector for Human Gene Therapy, 99 <i>PNAS</i> 10405-410 (Aug. 6, 2002)
2026	T. Matsushita et al., Adeno-Associated Virus Vectors Can Be Efficiently Produced Without Helper Virus, 5 <i>Gene Therapy</i> 938-45 (February 10, 1998)
2027	J.F. Wright, Product-Related Impurities in Clinical-Grade Recombinant AAV Vectors: Characterization and Risk Assessment, 2 <i>Biomedicines</i> 80-97 (March 3, 2014)
2028	D. Curran-Everett, Explorations in Statistics: Hypothesis Tests and P-values, 33 <i>Advances in Physiology Education</i> 81-86 (June 2009)
2029	Oliver, Christopher, Diffusion Coefficient and Molecular Weight of Type 5 Adenovirus by Photon-Correlation Spectroscopy, <i>Biochimica et Biophysica Acta</i> , 437 (1976) 589-598 (October 21, 1975)
2030	Tanford, Charles, Physical Chemistry of Macromolecules (1961)
2031	Declaration of Karen G. Fleming
2032	Curriculum Vitae of Karen G. Fleming
2033	Declaration of Barrie J. Carter (“Carter”)
2034	Curriculum Vitae of Barrie J. Carter
2035	Google Scholar Results for Citations to Burnham et al., Analytical Ultracentrifugation as an Approach to Characterize Recombinant AAV Vectors, 26 <i>Human Gene Therapy Methods</i> 228 (2015) (July 27, 2026)
2036	BioAnalysis, “Empty/Full/Partial Capsid Assay Using Sedimentation Velocity Analytical Ultracentrifugation (SV-AUC),” available at https://bioanalysisllc.com/sv-auc-empty-partial-full-capsid-assay-gene-therapy/ (accessed July 27, 2026).
2037	Sigma-Aldrich, “Strategies for the Separation and Analysis of Empty and Filled Capsids During Viral Vector Production,” available at https://www.sigmaaldrich.com/US/en/technical-documents/technical-article/pharmaceutical-and-

Exhibit No.	Description
	biopharmaceutical-manufacturing/gene-therapy-manufacturing/strategies-separation-analysis-empty-filled-capsids?srsId=AfmBOor6raGiKOeAeg-obMoKHCf4pXXje_9H4YFJM1T8BSW0bX_GKNRe (accessed July 27, 2026).
2038	D.M. Lipinski et al., Clinical Applications of Retinal Gene Therapy, 32 <i>Progress in Retinal and Eye Research</i> 22-47 (2013).
2039	Deposition Transcript of Steven Berkowitz, Ph.D., IPR2026-00150 (July 29, 2026)
2040	A. Roy et al., “Antigenic Differences Among AAV Species of Different Densities” in <i>Replication of Mammalian Parvoviruses</i> (Ward, D., and Tattersall, P., eds.), Cold Spring Harbor Laboratory, Cold Spring Harbor, NY, 193-204 (1978)
2041	B. Kokona et al., Particle Size Distribution Resolution Limit of AAV High-Speed Sedimentation Velocity Protocol Relative to Low-Speed Sedimentation Velocity Protocol, Mass Photometry, and Charge-Detection Mass Spectrometry, 114 <i>J. Pharm. Sci.</i> 103878 (2025).
2042	S.A. Berkowitz et al., Rapid High-Resolution Size Distribution Protocol for Adeno-Associated Virus Using High Speed SV-AUC, 689 <i>Anal. Biochem.</i> 115482 (2024).
2043	S.A. Berkowitz & T. Laue, Boundary Convection During Sedimentation Velocity in the Optima Analytical Ultracentrifuge, 631 <i>Anal. Biochem.</i> 114306 (2021).
2044	Declaration of Karen G. Fleming, Ph.D. in Support of Patent Owner’s Response (“Fleming”).

I. Introduction

The claimed sedimentation velocity analytical ultracentrifugation (“SV-AUC” or “AUC”)¹ method of the ’894 patent was a breakthrough for quantifying recombinant adeno-associated viral (“rAAV”) particles in gene therapy. It was the first single assay capable of identifying and quantifying all species of rAAV particles in a heterogeneous mixture, including potentially dangerous empty capsids and fragmented genome-containing rAAV impurities.

Heterogeneous mixtures of rAAV particles containing impurities, such as genome fragments, pose clinical risks, including immunotoxicity, that compromise the safety and efficacy of gene therapy products. EX2027, 87; EX1026, 6; EX2039, 119:9-120:7; *see also* EX2033 (“Carter”), ¶¶25-32 (citing EX1029, 15, 18, 19-20); EX2044 (“Fleming”), ¶¶27-45. But there was no practical method for identifying and quantifying rAAV particles containing unknown genome fragments. Patent Owner (“Genzyme”) developed a single experiment method to conquer this challenge in a surprising and unexpected way.

The inventors identified an unexpected linear relationship between genome size and rAAV particle sedimentation coefficient. By applying this relationship according to the claimed method, Genzyme developed a single-step SV-AUC

¹ Unless otherwise specified, AUC without the “SV” prefix refers to SV-AUC.

experiment to quantify rAAV species in a heterogeneous mixture. The linear relationship between genome size and rAAV sedimentation coefficient defied expectations. The person of ordinary skill in the art (“POSA”) understood that a particle’s sedimentation coefficient (S) depends on both its density and friction between the particle and solution. Even if a particle’s density varied linearly with the size of the genome—a proposition contradicted by AAV particle data—the POSA would have expected rAAV particles to have different shapes, sizes, and chemical properties. Different rAAV particle species would experience varying amounts of friction causing sedimentation coefficients to vary unpredictably.

Sarepta fails to carry its burden to prove obviousness by a preponderance of the evidence for at least three reasons:

First, the primary reference for all Grounds, Le Bec (EX1006), indicates a nonlinear relationship between sedimentation coefficient and genome size. Sarepta’s contention that Le Bec’s data (EX1006) shows a linear relationship between sedimentation coefficient and genome size is undermined by Sarepta’s improper statistical analysis. Le Bec, in fact, would have motivated the POSA away from using SV-AUC to “determin[e] the genome size.” EX1001, claim 1.

Second, Sarepta’s argument that a prior art mention of a *qualitative* linear relationship between rAAV genome size and particle density is evidence of a *quantitative* linear relationship between rAAV genome size and sedimentation

coefficient is erroneous. Sarepta argues the POSA would have assumed that density is the only variable that impacts rAAV sedimentation coefficient, because “the size and shape of the particles are approximately the same.” Petition, 44. When confronted with contradictory evidence, Sarepta expert, Dr. Steven Berkowitz, conceded that it “appears to be the case” that rAAV particles can have different sizes and shapes. EX2039, 116:23-117:6.

Sarepta heavily relies on publications from Dr. Barrie Carter’s laboratory at NIH, which did “groundbreaking work” in rAAV. Petition, 2. Carter has been recognized as one of the “fathers of rAAV vectors.” Petition, 11-12 (citing EX1040, 491). But Sarepta misinterprets Dr. Carter’s publications. Dr. Carter submits a declaration (EX2033, “Carter”) refuting Sarepta’s characterization of his publications. He explains that his publications teach the opposite of what Sarepta claims: the POSA would have understood that rAAV particles do *not* have approximately the same size and shape.

Dr. Carter’s publications describe AAV particles that contain identical genomes, but have different sedimentation coefficients caused by “a change in structural conformation.” EX1013, 1136. In deposition, Sarepta’s expert Dr. Berkowitz agreed that the Carter publications did not indicate a quantitative linear relationship between particle density and genome size (EX2039, 99:9-100:3) and that rAAV particles can have different sizes and shapes (*id.*, 116:4-117:15).

Third, objective indicia support nonobviousness. Le Bec and the Carter publications would have led the POSA to conclude that a linear relationship between sedimentation coefficient and rAAV genome size was not merely improbable, but scientifically unsupported. As Dr. Berkowitz testified at deposition, the prior art indicated an “unpredictability of the structure” of rAAV particles. EX2039, 175:6-177:5. Further, industry has praised the claimed method. Beckman Coulter GmbH, the only AUC instrumentation manufacturer, extolled the importance of the inventors’ method. A publication coauthored by Le Bec, in fact, remarked on the reliability of the method. Dr. Berkowitz’s own publication cited the inventors’ publication describing claimed method. In sum, Genzyme’s inventors addressed a long-felt need for a single experiment rAAV impurity assay.

Sarepta’s Petition “selectively culled from the prior art to fit the parameters of the patented invention.” *ATD Corp. v. Lydall, Inc.*, 159 F.3d 534, 546 (Fed. Cir. 1998). It ignored disclosures contradicting its narrative. The Board “must avoid ‘hindsight bias and must be cautious of arguments reliant upon ex post reasoning,’” as Sarepta engages in here. *Arendi S.A.R.L. v. Apple Inc.*, 832 F.3d 1355, 1361 (Fed. Cir. 2016). Evaluating “the prior art ... for all its teachings, not selectively,” Sarepta fails to carry its burden to establish the claims are obvious by a preponderance of evidence. *Henny Penny Corp. v. Frymaster LLC*, 938 F.3d 1324,

1332 (Fed. Cir. 2019). A Final Written Decision finding claims 1-7 and 10-30 (the “Challenged Claims”) not unpatentable is warranted.

II. State of the Prior Art for SV-AUC of rAAV Particles

A. Determining the Quantity of a Viral Particle Species by Absorbance Detection Typically Requires Determining the Size of the Encapsidated Genome

Determining the quantity of a species of rAAV particles in a heterogeneous mixture by calculating the area under the peak in an SV-AUC distribution profile acquired with absorbance typically requires knowing the size of the encapsidated genome. Fleming, ¶42; Petition, 32 (citing EX1010, 127). This is because the capsid protein and the encapsidated DNA absorb light to different degrees. Thus, the correlation between the peak area and rAAV particle species concentration depends on the size of the genome. Fleming, ¶51; EX1001, 24:27-31.

Absorbance detection was the preferred method of detection for AUC instruments at the time of the '894 patent's effective date. EX1025, 164; Fleming, ¶¶39-42. The Petition's primary reference, Le Bec, uses absorbance detection in its examples at a wavelength of 276 nm. EX1006, 15:19-28. An rAAV particle absorbs UV light according to the Beer–Lambert law where ϵ is the weight extinction coefficient; c is the molar concentration; ℓ is the path length.

$$A = \epsilon c \ell$$

EX1009, 152; Fleming, ¶40; Petition, 49. According to Beer–Lambert, the absorbance signal is directly proportional to solute concentration. *Id.*

The capsid and genome absorb light to significantly different degrees. This is illustrated by Figure 1B of the '894 patent. Figure 1B shows an “AAV2 mixture contain[ing] empty capsids (‘Empty Cap’) and full genome capsids (‘Intact Vector’) A plot of concentration in units of detection, $C(S)$, versus sedimentation coefficient (Svedberg units, S) showing that AUC can be used to measure the concentration of empty capsids and full genome capsids from an 80%/20% mixture).” EX1001, 8:24-35.

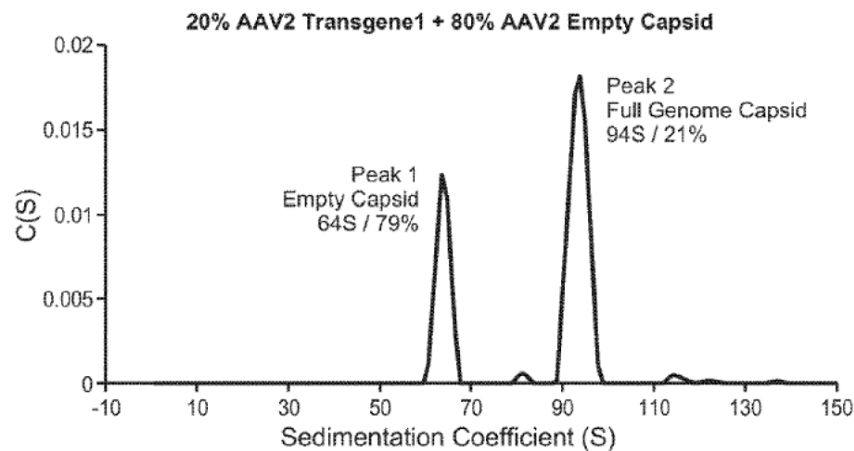


FIG. 1B

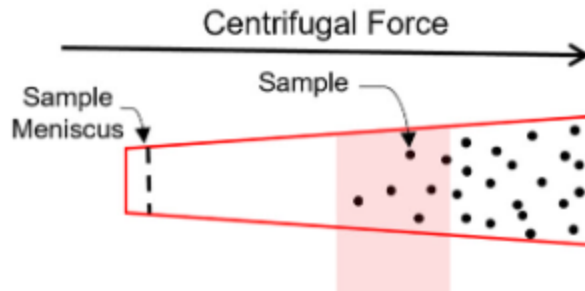
EX1001, Fig. 1B. The peak corresponding to empty capsids, present in four-fold excess to the full genome capsids, has a visibly smaller area than the full genome rAAV peak. If the peak area reflected actual concentration, the empty capsid peak at 64S would have been four-times the area as the full genome capsid peak at 94S. This discrepancy is attributable to the different absorbances of empty capsid and

full rAAV particles at the absorbance wavelength. The extinction coefficient can account for this difference. Fleming, ¶41; EX1001, 24:41-44.

For a genome-containing rAAV particle, which is a combination of protein (from the capsid) and DNA (genome), the extinction coefficient will depend on the amounts of protein and DNA in a particle. EX1010, 127. Extinction coefficients of empty and fully intact rAAV particles had been estimated. *Id.* EX1001, 24:44-47. To calculate extinction coefficients for rAAV particles containing fragmented genome, the POSA would need to know the molecular weights of both the capsid protein and encapsidated DNA. EX1010, 127; Fleming, ¶42. Before the '894 patent's invention there was no way to determine genome molecular weight from an SV-AUC experiment. This meant that the SV-AUC analysis was non-quantitative for a heterogeneous population that included fragmented genomes. Fleming, ¶¶46-47. As the 2014 Le Bec publication shows (EX1006), the POSA's use of SV-AUC as of the priority date was limited to qualitative analyses. EX2039, 77:1-14; *see also id.*, 144:23-145:2.

B. Particle Friction Matters for SV-AUC

SV-AUC measures the sedimentation rate of particles in solution when subjected to the centrifugal force generated by spinning the sample at high speed.



Fleming, ¶¶34-36 (citing EX2029, EX2030).² When centrifugal force is applied the particles begin to sediment towards the bottom of the cell. Buoyant and frictional forces oppose this sedimentation. Different particles experience different buoyant and/or frictional forces.

The sedimentation and diffusion of the particles over time and radius of the experiment is described by the Lamm equation, which can be used to generate sedimentation coefficients for particle species within the sample. Fleming, ¶50.

The sedimentation coefficient is defined by the Svedberg equation:

$$s = M (1 - \bar{v}\rho) / (N_a f)$$

where s is the sedimentation coefficient of the particle, M is the molar mass of the particle, $\bar{v}$ is the partial specific volume of the particle (the inverse of the density of the particle), ρ is the density of the solvent, N_a is Avogadro's number, and f is the

² See <https://www.beckman.com/resources/technologies/analytical-ultracentrifugation/experiments>

frictional coefficient of the particle. Petition 11 (citing EX1025, 165-66; EX1009, 146; EX1024, 216); Fleming, ¶¶35-36, 84.

By contrast, in sedimentation equilibrium AUC (“SE-AUC”) the distribution of a particle species “is a function of its buoyant mass” without the need to consider friction. Fleming, ¶87. The Berkowitz 2015 publication explains that SE-AUC “was this mode of sedimentation analysis that was most commonly used many years ago to assess the [molecular weight] MW of proteins and their complexes since it was totally independent of the frictional properties of the protein molecule, which made *MW assessment via SV-AUC much more difficult.*” EX1024, 241.

Berkowitz 2015 explains the complicated picture of forces impacting the sedimentation coefficient beyond size and shape:

From these simple equations it can be seen that these three forces acting on the sedimenting molecule depend on several factors that are unique properties of the protein being studied. Those factors include (1) the protein’s buoyant mass (which depends on its amino acid composition, glycosylation, and the density of the formulation buffer) (2) its frictional properties, which depend on its overall size and shape (conformation or three-dimensional structure), (3) its hydration (the amount of water bound on its surface) and (4) solvation or preferential solvation effects (exclusion of excipients, or binding of excipients at the protein surface).

EX1024, 216. In testimony, Dr. Berkowitz provided additional details regarding the complex interactions between particle and solvent, including an “adhesion” force from “dragging additional solvent with the particle as it migrate[s]” and “charge” effects. EX2039, 44:16-47:8.

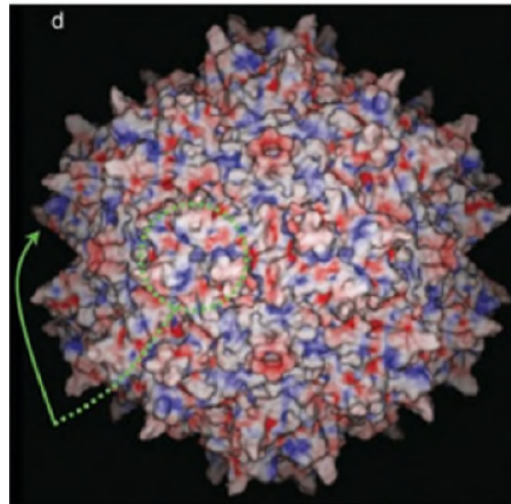
C. The POSA Understood that Genome-Containing rAAV Particles Were Not of Identical Size and Shape

Encapsidated genome size was understood to cause variations in rAAV particle size and shape. Carter, ¶¶40-47; Fleming, ¶158. Partial or fragmented genomes encapsidated in rAAV particles were observed to impact the conformation of the capsid and its infectability and immunotoxicity. EX1026, 6. Gao explains that “encapsidation of full or partial AAV genomes may lead to capsid conformational changes ... on capsid structure expos[ing] both phospholipase A2 domain and nuclear localization signal domains and facilitates endosome escape and viral infection.” EX1026, 6. Gao further explains that “partial viral genomes may also contribute to the side effects of the [partially empty] virions due to their different capsid conformations as compared with [completely empty] particles.” *Id.*

AAV vectors, including rAAV, are “composed of an icosahedral assembly of 60 capsid proteins (VP1, -2, -3) assembled together in a defined stoichiometry.” EX2027, 81. A detailed crystallographic analysis of AAV-2 describes the complexity of the outer surface of an rAAV particle. EX2025, 10409; Carter, ¶30;

Fleming, ¶30. This can be seen in Figure 3d, which shows the electrostatic surface potential of AAV-2 calculated with SPOCK running from -10 (red) to +10 (blue).

EX2025, 10408.



EX2025, Fig. 3d.

“AAV assembly” was understood to include a step “in which progeny DNA strands are packaged into preformed capsids.” EX1012, 71; *see also id.* 79-80. This does not mean that all rAAV particles are the same size and shape. Carter, ¶¶44-47; Fleming, ¶91. Publications by Myers and Carter (EX1012, 71) and de la Maza and Carter in March 1980 (EX1013, 1129, 1136) showed that even particles with the same DNA and capsid protein content can have different sizes and shapes. Carter, ¶¶39-45; Fleming, ¶93.

De la Maza (March 1980) observed two AAV particle species having different densities, “AAV (1.41 [g/cm³]) and (1.45 [g/cm³]),” for which “the major components had an identical DNA content and DNA/protein ratio but differed

slightly in their sedimentation rates and differed significantly in their relative infectivity.” EX1013, 1129. Myers theorized that the AAV (1.45) species may be a precursor to the AAV (1.41) species or an end product, indicating complexity in rAAV particle assembly that included structural changes. EX1012, 75, 81; Carter, ¶45.

Because sedimentation coefficient is proportionate to buoyant density, which increases with particle density in the Svedberg equation, if friction were identical for the two rAAV particles the POSA would have expected the denser particle, AAV (1.45), to have a higher *S* value. Fleming, ¶93; Carter, ¶¶41-42. But that is not what was observed:

We found that the 1.41 component has a slightly higher *S* value (111) than the 1.45 particle (109) Assuming that the masses of the two types of particles are equivalent, the data suggest that a change in structural conformation accounts for these differences between the two classes of particles. This may account for the antigenic differences between the 1.45 and 1.41 particles.

EX1013, 1136. Although the 1.45 and 1.41 particles contain identical genomes and the same protein/DNA ratio, they are structurally distinct. Critically, the denser particle (1.45) sediments more slowly (*S*=109) than the less dense particle (1.41, *S*=111). Carter, ¶42. This is the opposite of what Sarepta’s theory predicts and demonstrates that friction, not density, can dominate the sedimentation coefficient.

Myers (EX1012) and de la Maza (March 1980) (EX1013), along with other prior art, meant the POSA would have understood that rAAV particles could have significant variations in size and shape. Carter, ¶¶46-47.

III. The '894 Patent

The '894 patent represents an important advance in the manufacture and purification of rAAV preparations. It provides the gene therapy field with a method to reliably identify and quantify concentrations of each species (*e.g.*, empty capsids, rAAV with fragmented genomes, and fully intact rAAV particles) in an rAAV preparation. Fleming, ¶¶46-50. The claimed method remains the industry's "gold standard." *Infra* §VII.

The prior method, Southern blot, used a specific complementary DNA probe sequence to detect rAAV species. The probe, however, could miss fragmented genome species. EX1001, 1:44-58. In contrast, the claimed AUC method provides a single experiment to detect and quantify multiple rAAV species, including species containing fragmented genomes, that either evaded detection or required multiple experiments to detect. *Id.*

A. Disclosed Innovation of the '894 Patent

"[A] major problem in producing pure AAV vector preparations is the presence of capsids with ***partial or fragmented genomes***." EX1001, 48:38-40 (emphasis added). "[T]he variant genomes" include partial or fragmented genomes

that “are smaller than the intact recombinant viral genome (*e.g.*, truncated genomes).” *Id.*, 25:19-21. The ’894 patent provides a single method, SV-AUC, which can analyze a sample in a matter of hours and provide quantitative characterization of rAAV preparations. *Id.*, 43:49-59. During rAAV production, for example, AUC facilitates “monitor[ing] [rAAV preparation] at time intervals (*e.g.*, one or more times).” *Id.*, 11:45-51.

1. Overcoming the Challenge of Determining rAAV Species Concentration by AUC

Extinction coefficients (*supra* §II.A), had been determined for empty capsids and fully intact rAAV particles. EX1001, 24:44-47 (citing EX1010, 127). However, “it [was] not possible to determine empirically the extinction coefficient of fragmented genomes,” because they are “of unknown size and sequence” *Id.*, 24:56-59; 45:2-5; 49:57-61; Fleming, ¶¶51-55. To overcome this obstacle, the inventors determined that the “calculated S values can be plotted to generate a standard curve to which recombinant viral species of unknown molecular weight or genome size can be compared to determine the molecular weight of the unknown species.” EX1001, 24:59-67; *see also id.*, 45:2-8.

The application of this innovative method is illustrated in Example 5, which demonstrates the implementation of “AUC ... as a tool to quantify the aberrant packaging of fragmented genomes in rAAV vector preps.” EX1001, 49:39-50:67; Fleming, ¶¶53. EX1001, 50:1-19. Figure 8 “shows the linear relationship between

sedimentation coefficient and vector genome size,” provides the standard curve of Example 5, correlating genome size and S value. *Id.*, 9:36-40. Data plotted and fit in Figure 8 surprisingly “demonstrated a highly linear relationship ($R^2=0.9978$) between sedimentation coefficient and genome size.” *Id.*, 49:66-67.

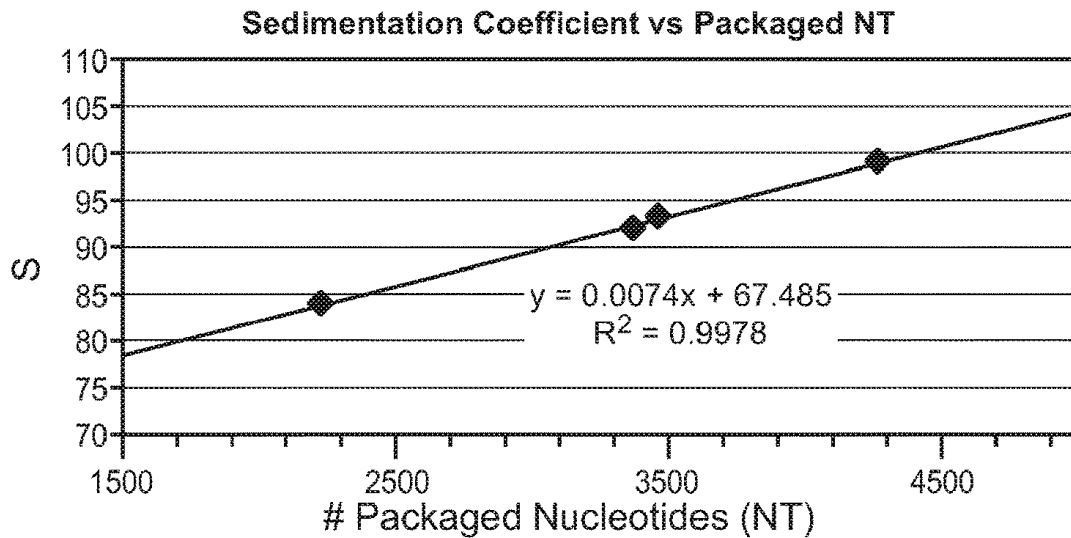


FIG. 8

Id., Fig. 8; Fleming, ¶54.

Generating the standard curve allowed the inventors to determine the molecular weight, genome size, and extinction coefficient at 260 nm of unknown rAAV species, for example, partial or fragmented species. EX1001, 49:42-50:67 (Example 5, Table 5); Figs. 8, 9A. Once the standard curve was generated, a study was conducted to show “that assessment of capsid genome size using AUC data correlates with assessment of genome size by Southern blot.” *Id.*, 9:41-43; 50:21-42; Fleming, ¶55.

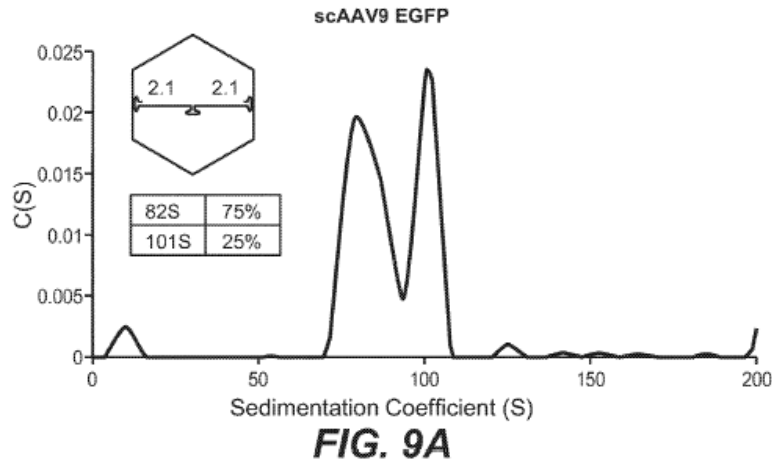
2. One-Step rAAV Preparation Analysis by AUC

The inventors applied their method to quantitatively analyze the compositions of several samples to demonstrate how the method identified peaks corresponding to rAAV species containing partial or fragmented genomes and determined their concentrations. Fleming, ¶56.

a. Example 5: Application of the Standard Curve

“To demonstrate the utility of AUC to detect genome fragments, a self-complementary vector comprising AAV2 ITRS, a minimal CBA promoter, and an EGFP transgene was packaged into an AAV9 capsid.” EX1001, 50:21-24. After purifying the preparation to eliminate empty capsids, the remaining vector particles were “analyzed by AUC” and “[t]he standard curve was then used to assign genome size to each of the resolved genome containing capsids.” *Id.*, 50:27-31; Fleming, ¶57.

In Figure 9A, a “plot of differential sedimentation coefficient distribution value, $c(s)$, vs sedimentation coefficient in Svedberg units (S), yields the distribution of sedimentation coefficients for a [self-complementary] scAAV9 EGFP vector preparation. Single stranded monomeric (82S) and double stranded dimeric (101S) species are labeled with the corresponding sedimentation coefficients and relative abundance values (%).”



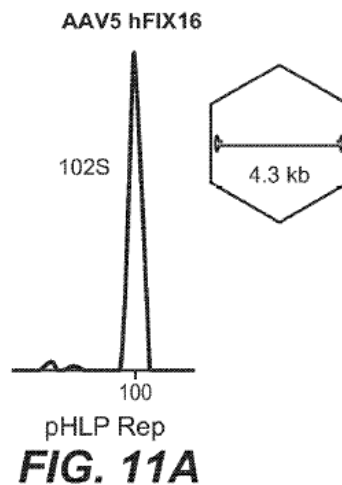
EX1001, 9:41-50; 50:21-31; FIG. 9A.³ Applying the standard curve, the inventors were able to determine the approximate genome sizes of the rAAV species corresponding to the 101S and 82S peaks. *Id.*, 50:28-36. By determining the approximate molecular weights of the two components it was possible to calculate their respective estimated extinction coefficients at 260 nm and, therefore, their relative concentrations of 25% for the 101S peak double-stranded species and 75% for the 82S peak single stranded species. *Id.*, 50:28-36; FIG. 9A. The AUC results showed good agreement with Southern blot results. Fleming, ¶¶58-63; EX1001, 50:43-46; FIG. 9B.

³ Peaks with lower *S* values than empty capsids are not rAAV species. Fleming ¶58; EX1023, 27.

b. The Power of the Innovative AUC Method Compared to Southern Blot

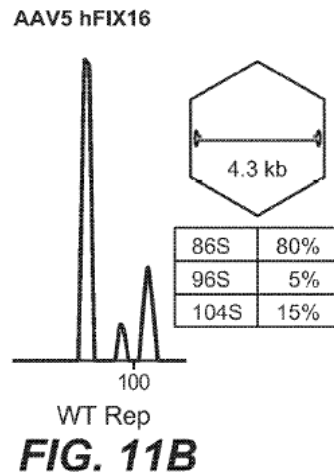
The power of the disclosed AUC method was further revealed through a comparison of AUC with Southern blot analysis. Fleming, ¶64. A “single stranded AAV5 Factor IX vector, AAV5FIX, (FIG. 11A-B) and a single stranded AAV5hSMN vector (FIGS. 11C-D) were generated using” the modified helpers, pHLP Rep, “as described above, but cap sequences of AAV2 were replaced by cap sequences of AAV5.” EX1001, 51:39-44.

The predicted sedimentation coefficient for the AAV5 FIX vector capsid was approximately 101S, and AUC analysis of AAV2/SFIX in the presence of the modified pHLP Rep helper “revealed a homogenous profile with the majority of the vector (90%) sedimenting with an S value of the expected size.”



Id., 51:44-50, FIG. 11A. “In contrast, AAV5 FIX vector generated using [the unmodified wild-type, WT Rep,] generated a strikingly different AUC profile Instead of a predominant peak at 101S, this profile revealed more capsid

heterogeneity, with the majority of the AAV5 FIX (80%) sedimenting at a lower S value of 86S, likely representing packaging of a fragmented genome,” whereas “only 15% of the AAV5 FIX vector capsids sedimented at the correct S value of ~104S.”



Id., 51:50-60, FIG. 11B; Fleming, ¶65.

AAV5SMN vectors made using the same wild-type and mutated helpers also had different profiles. Fleming, ¶66. “AAV5 SMN vectors generated in the presence of” the modified pHLP Rep helper “showed less heterogeneity by AUC analysis, with a single capsid species sedimenting at the S value of 101S consistent with packaging of a genome of the predicted size of ~4.4 kb.”

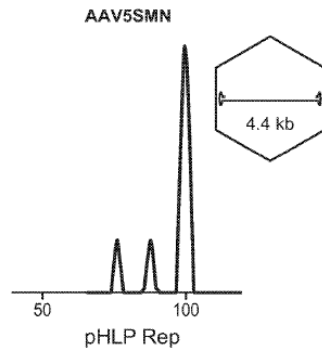


FIG. 11C

EX1001, 51:61-52:1, FIG. 11C. “In contrast, the AUC profile for the same vector genome packaged using ‘wild-type’” WT Rep helper “revealed three distinct AAV vector species, with sedimentation coefficients of 100S...92S (representing a fragmented genome of approximately 3300 [nucleotides]) and 80S (representing a fragmented genome of 2000 nucleotides).”

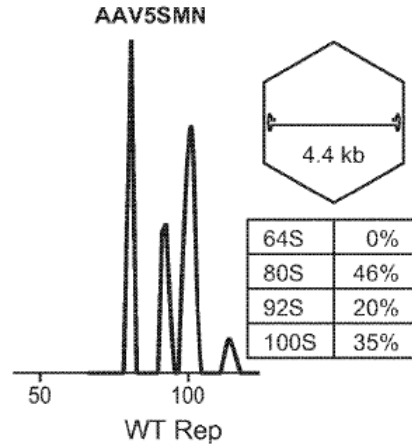


FIG. 11D

Id., 52:1-10, FIG. 11D; Fleming, ¶66.

Southern blot analysis of the AAV5SMN preparations was consistent with the AUC results for the pHLP helper (FIG. 11C) showing a homogenous profile of

the full-length vector (FIG. 12A, lane 2) compared with packaging of full-length and fragmented SMN genomes (FIG. 12A, lane 1). EX1001, 52:11-27.

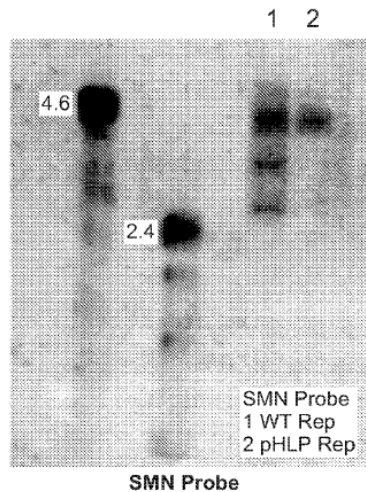
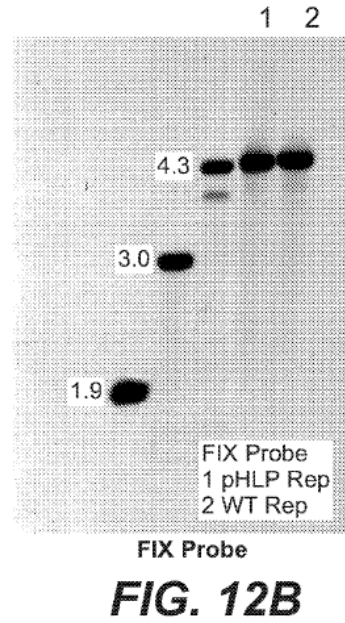
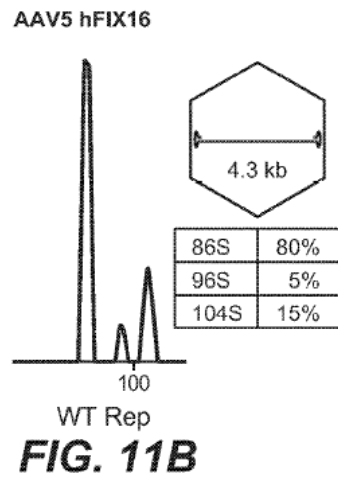
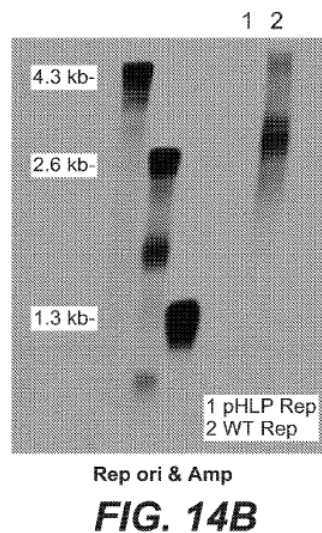


FIG. 12A

Southern blot analysis of the FIX vector preparation made with the pHLP modified helper agreed with the AUC result (FIG. 11A), showing predominantly full-length AAV (FIG. 12B, lane 1). In contrast, “AUC analysis of this AAV5FIX vector (FIG. 11B)” made with the WT-Rep helper “showed that 80% of the capsids contained a fragmented genome (~3000 nucleotides)” but these fragments “were **undetected** by the FIX probe” in a Southern blot experiment. *Id.*, 52:24-27 (emphasis added); Fleming, ¶67.



When an Amp^R specific probe was used in a Southern blot experiment, the vector genome species from the FIX vector preparation sedimenting at 86S containing a ~3 kb fragment (FIG. 11B) was detected.



EX1001, 52:36-45, FIG. 14B. But the Amp^R probe did **not detect** the fully intact genome in the rAAV5 FIX vector prep generated with the modified pHLP Rep

helper. *Id.* Thus, multiple experiments were required to identify the species in the preparation by Southern blot; Fleming, ¶¶68.

These comparative experiments illustrate the power of the inventors' AUC method. Unlike Southern blot, AUC is not sequence dependent, which allows for the detection and quantification of all rAAV species in a single experiment.

Fleming, ¶¶64-69.

B. Prosecution History

The Examiner allowed the '894 patent over references, including Le Bec (EX1002, 27-36) after Genzyme explained that the claimed innovative method, when applied under the specific conditions set forth in the claims, demonstrated that SV-AUC could in fact be used to resolve and characterize disparate rAAV species, including those with fragmented genomes, in a heterogeneous mixture. EX1002, 146; EX1001, 9:36-40. Prior to '894 patent's disclosure, the POSA would have been unable to use SV-AUC to determine encapsidated genome size.

Fleming, ¶¶70-72.

C. Person of Skill in the Art

Petitioner asserts that the POSA working in the field of the '894 patent in January 2015 would have had at least a Ph.D. in biology, chemistry, biochemistry, biophysics, pharmaceutical sciences, or a related discipline, with two or more years of industry, laboratory, and/or clinical experience in analyzing or characterizing

biomolecules, including viruses or viral vectors. Alternatively, according to Petitioner, the POSA would have had at least a Master's or Bachelor's Degree in biology, chemistry, biochemistry, biophysics, pharmaceutical sciences, or a related discipline, with a corresponding number of additional years of experience analyzing or characterizing biomolecules, including viruses or viral vectors. Petition, 32-33.

A POSA may work as part of a team with scientists familiar with the development, formulation, and/or administration of viruses or viral vectors for gene therapy and quality standards required for such products. Fleming, ¶¶19-20; Carter, ¶¶12-13. For purposes of this proceeding, Patent Owner applies this general framework, but reserves the right to apply a modified definition as the case proceeds.

D. Challenged Claims

Claim 1 of the '894 patent is the only independent claim. Therefore, all claims share the elements recited by this claim. Claim 1 recites:

Claim	Element
1[pre]	A method of quantifying one or more species of individual variant viral particles comprising <i>fragmented recombinant adeno-associated viral (rAAV) genomes</i> in a heterogeneous mixture of viral particles, said method comprising:
1[a]	(i) subjecting the heterogeneous mixture of viral particles to analytical ultracentrifugation under boundary sedimentation velocity conditions to generate sedimenting boundaries, wherein the boundary sedimentation velocity is from about 3,000 rpm to about 20,000 rpm;

1[b]	(ii) measuring the rate of movement or migration of the sedimenting boundaries, wherein movement or migration of each species of individual viral particles in the heterogeneous mixture of viral particles results in distinct sedimenting boundaries, each distinct sedimenting boundary corresponding to a resolvable species of viral particle, and wherein the individual viral particles comprise empty particles without genome, particles with full genomes and particles with fragmented genomes;
1[c]	(iii) determining the genome size of one or more species of the individual variant viral particles in the heterogeneous mixture of viral particles; and
1[d]	(iv) determining the quantity of one or more species of the variant viral particles in the heterogeneous mixture of viral particles.

EX1001, 55:1-26; Fleming, ¶¶73-76.

The remaining Challenged Claims depend from claim 1.

Dependent claim 2, recites “wherein each species of the individual variant viral particles in a heterogeneous mixture of viral particles is fully resolved.”

As explained below, Sarepta’s Grounds do not establish the unpatentability of claims 1 and 2. Because all claims depend either directly, or indirectly, from claim 1, all Challenged Claims are not unpatentable.

IV. Claim Construction

Genzyme does not dispute the Petition’s “plain and ordinary meaning” constructions for the purpose of this proceeding. Petition, 33; Fleming, ¶¶77.

V. Ground 1: Sarepta Fails to Establish That Le Bec and de la Maza, In View of Sommer Rendered Claims 1-7, 10-11, and 20-30 Obvious

Sarepta bears the burden of proving by a preponderance of the evidence that the Challenged Claims are unpatentable as anticipated and/or obvious based on the art and arguments set forth in its Petition. *In re Magnum Oil Tools Int’l Ltd.*, 829 F.3d 1364, 1381 (Fed. Cir. 2016) (“[T]he Board must base its decision on arguments that were advanced by a party ...”). The Petition fails to establish that it would have been obvious for the POSA to determine the size of the encapsidated genome in a variant viral particle from the particle’s sedimentation coefficient. The Petition also fails to establish it would have been obvious to determine the quantity of a variant viral particle species. For at least these reasons, the Petition has not established that claim 1 and its dependent claims are obvious.

A. Claim 1: Sarepta Fails to Establish that Claim 1 Would Have Been Obvious as of the Effective Date

Sarepta’s hindsight-driven theory is that the POSA would have found it obvious to determine the size of fragmented genomes as a function of the sedimentation coefficient of the viral particles in which they are contained. Petition, 38-50. No prior art reference—not Le Bec, not de la Maza (1978), not any other publication—ever taught, suggested, or even hypothesized that sedimentation coefficient could be used to determine the size of a fragmented genome in an intact rAAV particle.

Sarepta’s argument that the POSA would have understood that sedimentation coefficient depends solely on particle density is premised on the assumption that the “POSA would understand that the size and shape of intact, non-aggregated AAV particles are approximately the same, regardless of the size of the genome packaged in the capsid.” Petition, 45; EX1004, ¶170. The art as of the effective date, including de la Maza (1978) (EX1007), Berkowitz 2007 (EX1024), Myers and Carter (EX1012), de la Maza (March 1980) (EX1013), and Gao (EX1026), demonstrates otherwise. Fleming, ¶¶78-108; Carter, ¶¶35-47.

The POSA would have understood that viral particle shape and/or size varies unpredictably. Carter, ¶¶54-59; Fleming, ¶107; EX2039, 175:6-177:5. Differences in size and shape impact friction and thereby cause unpredictable variations in sedimentation coefficient. This variability would have been understood to exclude the possibility of any reliable linear relationship—and potentially any predictive correlation—between genome size and sedimentation coefficient. *Supra* §§II.B–C. The lack of such a linear relationship in Le Bec was consistent with, and reinforced, the POSA’s understanding of non-linearity between genome size and sedimentation coefficient. *Supra* §§I, II.B–C. This would have further discouraged the POSA from seeking to identify and exploit such a relationship to determine genome size. Fleming, ¶107.

Sarepta ignores numerous teachings of the art and falls prey to “hindsight syndrome by reasoning backward from the teaching of the patent itself.” *Creo Prod., Inc. v. Presstek, Inc.*, 166 F. Supp. 2d 944, 966 (D. Del. 2001) (internal quotation marks omitted), *aff’d*, 305 F.3d 1337 (Fed. Cir. 2002); *see also Rockwell Int’l Corp. v. U.S.*, 147 F.3d 1358, 1364 (Fed. Cir. 1998); *Jones v. Hardy*, 727 F.2d 1524, 1529 (Fed. Cir. 1984); *W.L. Gore & Assocs., Inc. v. Garlock, Inc.*, 721 F.2d 1540, 1553 (Fed. Cir. 1983). The hallmarks of hindsight are present here. Thirty-seven years passed between de la Maza (1978) and the ’894 patent without anyone discovering the linear relationship or its claimed application.

Sarepta also omits discussion of de la Maza (March 1980)—a publication that directly contradicts Sarepta’s theory, and is even cited in Le Bec. EX1006, 18 n.15, 45 (citing EX1013); Petition, 26. Sarepta constructs its obviousness theory by ignoring the failure of Le Bec’s data to show linearity. Sarepta omits the p-value, the one statistical metric that would have revealed the absence of linearity, from its regression analysis of Le Bec’s data. Sarepta also disregards knowledge regarding the unpredictable variability in rAAV particle size and shape (and viral particles more generally). The Petition omits teaching in references it cites including de la Maza (1978), de la Maza (March 1980), and Gao that contradict its obviousness narrative by describing conformational changes to AAV capsids. The Petition,

therefore, fails to establish the POSA's motivation or reasonable expectation of success in arriving at claim 1's method.

1. Le Bec and de la Maza, in View of Sommer, Did Not Render Obvious “determining the genome size of one or more species of the individual variant viral particles” with SV-AUC

Neither Le Bec nor de la Maza (1978) teach or suggest determining genome size of partial or fragmented rAAV particles from sedimentation coefficients generated by SV-AUC. Le Bec groups rAAV species together without differentiating rAAV particles containing fragmented genomes. Fleming, ¶¶109-110. The Petition does not address that Le Bec's AUC studies fail to resolve peaks associated with rAAV particles containing fragmented genomes. Petition, 38-47. Accordingly, the POSA would not, and could not, have relied on Le Bec to develop claim 1's method of determining the size of fragmented genomes in a heterogeneous mixture of viral particles.

Le Bec does not attempt to identify or quantify rAAV particles containing fragmented genomes. EX2039, 76:7-77:14; 143:23-146:15. Le Bec's analysis was qualitative, did not account for the different extinction coefficients of each particle and did not result in quantitative concentration measurements. EX2039, 77:1-14; *see also id.*, 143:23-146:15; *supra* §II.A. As Dr. Berkowitz explained, Le Bec's use of SV-AUC was about “try[ing] to get optimum performance in terms of reducing the amount of empty capsids” versus rAAVs containing “full” genomes

in rAAV vector production—not developing a quantitative assay to identify and quantify rAAV species. EX2039, 64:23-66:6. Le Bec’s category of “full AAV viral particles” includes multiple peaks as shown in Le Bec’s Figure 2 having genomes of varying sizes, such as fragmented genomes that Le Bec does not attempt to analyze. Fleming, ¶¶111-112.

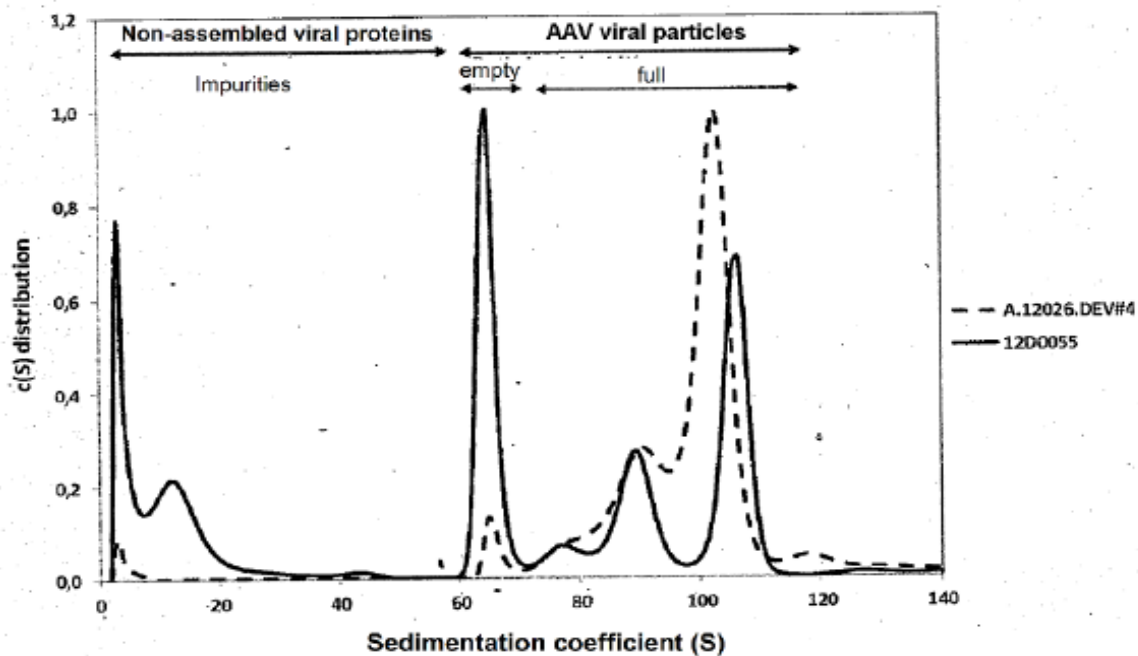


Figure 2

EX1006, 6:3-7; 18:8-11; Fig. 2; Fleming, ¶112. Le Bec explains that “[t]he distribution profile of the species allows the identification of **two categories of population**: a) <60S corresponding to non-assembled viral 15 proteins and/or contaminants” and “b) 60S-110S corresponding to intact AAV viral particles” and “**two subpopulations** with empty AAV viral particles at 65S and full AAV viral

particles between 80S and 110S, with the viral genome in single-stranded form at 90S and double-stranded form at 105S.” EX1006, 18:11-25 (emphasis added).

The Petition concedes that “Le Bec reports that the peak at 65S corresponds to empty rAAV particles and that the peak at 105S corresponds to an rAAV particle with a complete double-stranded, self-complementary genome,” without identifying specific *S* values for peaks having partial genomes. Petition, 43 (citing EX1006, 18:21-25). Dr. Berkowitz conceded that beneath the broad peaks of Figure 2, may lie peaks corresponding to unidentified genome species. EX2039, 75:12-76:5. Dr. Berkowitz testified that because Le Bec was not concerned with characterizing the different species in the composition, its authors made no effort to sharpen the peaks to identify different species of genome-containing rAAV particles. EX2039, 64:23-65:12. Dr. Berkowitz also agreed that the POSA would have considered the usefulness of SV-AUC to depend on its “ability to resolve [] distinct peaks for each species,” because “if for some reason they overlap, you are missing information.” EX2039, 54:16-56:8. Thus, the POSA would have assessed Le Bec’s distribution as missing significant amounts of information regarding rAAV species in the mixture. Fleming, ¶111. Because of this deficiency, the Petition guesses at approximate *S* values for other rAAV particle species from Le Bec’s poorly resolved distribution profiles. Petition, 43 (citing EX1006, 18:23-25, Figures 2 and 3). Given the poor resolution of peaks between 60S and 110S, the

POSA would not have considered Le Bec useful for characterizing, much less quantifying, fragmented genomes. Fleming, ¶113.

Le Bec's Figure 3 is even worse. The Petition and Berkowitz Declaration never address the agglomeration of what appears to be multiple peaks ranging from about 75S to about 110S in Figure 3 of Le Bec. Fleming, ¶115.

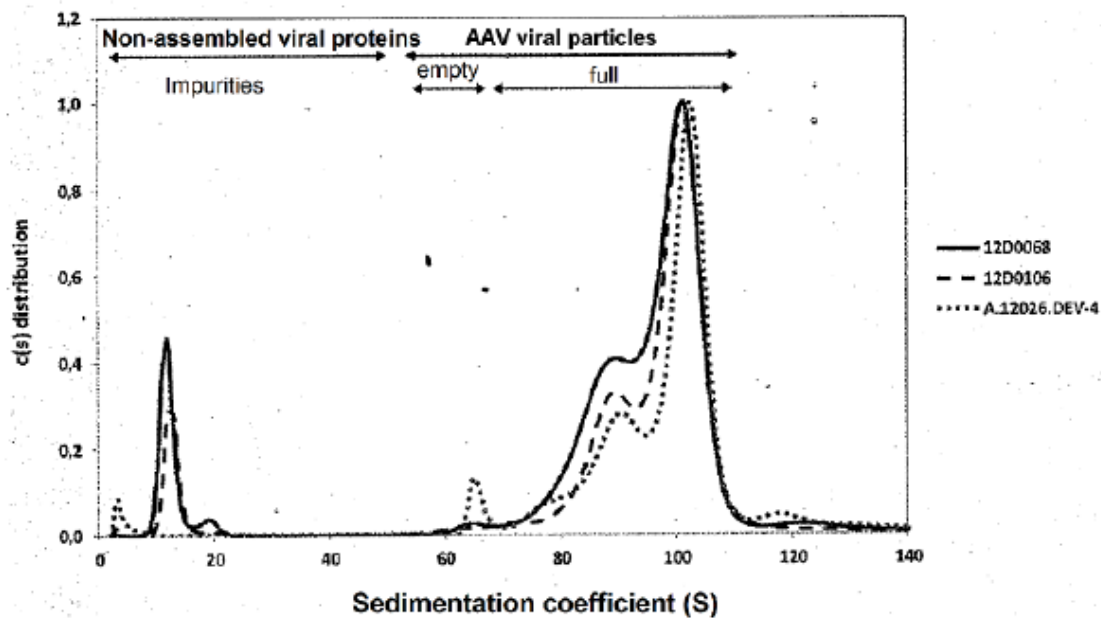


Figure 3

EX1006, Fig. 3. This commingling of species masks the presence of multiple peaks, whereas the POSA would have understood that for heterogeneous mixtures of particles, the use of AUC under boundary sedimentation conditions depends on resolving peaks of different species. EX2039, 62:18-24; Fleming, ¶115. Dr. Berkowitz admitted that “an issue like the peaks overlapping that you see in figure

3” would “make it really very difficult to quantitate different species” under the peaks. EX2039, 69:1-24; Fleming, ¶¶114, 116.

The ’894 patent explains that “if multiple species of solute are present, this may lead to the formation of multiple sedimenting boundaries, each corresponding to a resolvable species.” EX1001, 20:10-13. “[T]he presence of multiple species with similar sedimentation coefficients,” as in Le Bec’s Figures 2 and 3, will cause the boundary to be broader than expected on the basis of diffusion alone.” *Id.*, 17:41-43; *see also* EX2039, 75:4-76:5. The POSA would have considered the broad peaks in Figures 2 and 3 to conceal peaks corresponding to partial or fragmented species. Fleming, ¶117; EX2039, 75:12-21 (Figure 2); 69:1-24 (Figure 3). Because these peaks are not resolved, the POSA would not have been motivated to apply Le Bec’s method to determine the size of fragmented genomes in rAAV particles. Fleming, ¶117.

De la Maza (1978) does not even use SV-AUC, or any sedimentation velocity method, on intact viral particles. Fleming, ¶118; Carter, ¶37. Its sedimentation velocity studies are conducted on non-encapsidated DNA. EX1007, 195. Despite the glaring deficiencies of the teachings of Le Bec and de la Maza (1978), the Petition argues that the “POSA would have been motivated to combine Le Bec with de la Maza” to arrive at the claimed method. Petition, 36. This argument fails. Fleming, ¶¶118-122.

**a. The POSA Would Not Have Been Motivated to
“determin[e] the genome size of one or more species of
the individual variant viral particles” with SV-AUC**

The Petition’s obviousness challenge to claim 1 fails at the threshold motivation requirement. Although the POSA may have been familiar with Le Bec’s AUC methodology and de la Maza’s density-genome size observations, the record does not support that the POSA would have been motivated to combine their teachings to arrive at the claimed method. Fleming, ¶123. Sarepta’s motivation argument suffers from at least five significant failures.

First, Sarepta’s argument ignores its own expert’s publication and testimony stating that the POSA seeking to determine molecular weight would not use SV-AUC; the POSA would use SE-AUC because it is not impacted by friction. EX2039, 52:16-53:6; EX1024, 241; Fleming, ¶123.

Second, the POSA analyzing Le Bec’s data would have concluded that no cognizable relationship—linear or otherwise—exists between sedimentation coefficient and genome size. Le Bec did not give the POSA any indication that its AUC method would be suitable to identify the genome size of one or more species of the individual variant viral particles in a heterogeneous mixture of viral particles, as Sarepta alleges. Petition, 33-37. As explained (*supra* §V.A.1), Le Bec focused on determining the approximate amount of empty versus full capsids in an rAAV mixture. EX1006, 15:17-20:3. But Le Bec did not differentiate rAAV

particles containing full genomes from rAAV particles containing genome fragments (*id.*, 17:29-20:3), nor did it attempt to determine the molecular weights of genomes, or quantify rAAV species. EX2039, 144:3-15. Le Bec's method also failed to achieve the peak resolution necessary for the POSA to determine the size of the one or more fragmented genomes. *Supra* §V.A.1; Fleming, ¶¶124-125.

Third, de la Maza (1978) demonstrated, and Dr. Berkowitz conceded, that AAV particle density is not linearly related to genome size. EX2039, 101:3-11; 105:19-106:9. The POSA would not have understood the qualitative observation that Sarepta relies on, stating "the particle density has an approximately direct linear relationship to the strand length" (Petition, 2, 21, 25), to support the claimed quantitative method. Fleming, ¶126.

Fourth, de la Maza (March 1980) refutes Sarepta's reliance on de la Maza's 1978 publication to argue that the linear relationship between particle density and genome size also applies to sedimentation coefficient. *Supra* §II.C. De la Maza (March 1980) discloses rAAV particles having identical DNA content but different densities and sedimentation coefficients. Fleming, ¶127; Carter, ¶¶37-39, 41 (citing EX1013, 1136; EX2040). The authors attribute this to conformational differences in the capsids. *Id.* Further, the POSA would not have understood de la Maza's disclosure (Petition, 34-37) as applicable to the sedimentation coefficient of intact rAAV particles. Fleming, ¶127.

Fifth, the POSA understood that rAAV particles vary in shape and size, which impacts their frictional coefficients. EX1023, 18, 31. Moreover, chemical interactions, including hydration and adhesion, affect friction. Dr. Berkowitz conceded that he failed to address myriad issues that affect rAAV particle friction in SV-AUC, including hydration and other chemical properties. EX2039, 82:14-83:7; 78:18-21; 130:12-131:1; 134:16-135:11. The POSA would not have had any reason to expect a reliable relationship between genome size and sedimentation coefficient. *Supra*, §II.B; Fleming, ¶128.

Finally, Sommer merely supplies extinction coefficient equations, which require the molecular weight of the encapsidated genome as an input. EX1010, 127. The Petition relies on Le Bec and de la Maza to render obvious determining the size of fragmented genomes. Petition, 34-37, 51. But because the combination of Le Bec and de la Maza would not have motivated the POSA to determine genome size by generating the sedimentation coefficients of fragmented genomes (*supra* §V.A.1), the POSA would not have been motivated to develop the method of claim 1. Fleming, ¶129.

“[O]bviousness concerns whether a skilled artisan not only *could have made* but *would have been motivated to make* the combinations or modifications of prior art to arrive at the claimed invention.” *Belden Inc. v. Berk-Tek LLC*, 805 F.3d 1064, 1073 (Fed. Cir. 2015)) (emphasis original). The combination of Le Bec and

de la Maza did not give the POSA any reason to develop an SV-AUC method to determine the genome size of fragmented genomes or accurately determine their quantity in a heterogeneous mixture of viral particles. Fleming, ¶¶129-130. The nearly four decades that elapsed between de la Maza's publication and the '894 patent's effective filing date reflects the nonobviousness of the claimed method. *Leo Pharm. Prods., Ltd. v. Rea*, 726 F.3d 1346, 1354 (Fed. Cir. 2013).

i. The POSA Would Not Have Been Motivated to Use SV-AUC Instead of SE-AUC to Determine Genome Size

According to Sarepta's expert, Dr. Berkowitz, the POSA would have considered SE-AUC—not SV-AUC—the preferred method to determine rAAV particle molecular weight. Fleming, ¶131. Indeed, de la Maza (1978) undermines Sarepta's motivation argument to use SV-AUC to determine molecular weight. The AAV density measurements in de la Maza (1978) were made using sedimentation equilibrium, which separates particles based solely on buoyant mass and is independent of friction. EX1007, 195; EX2039, 52:16-53:6.

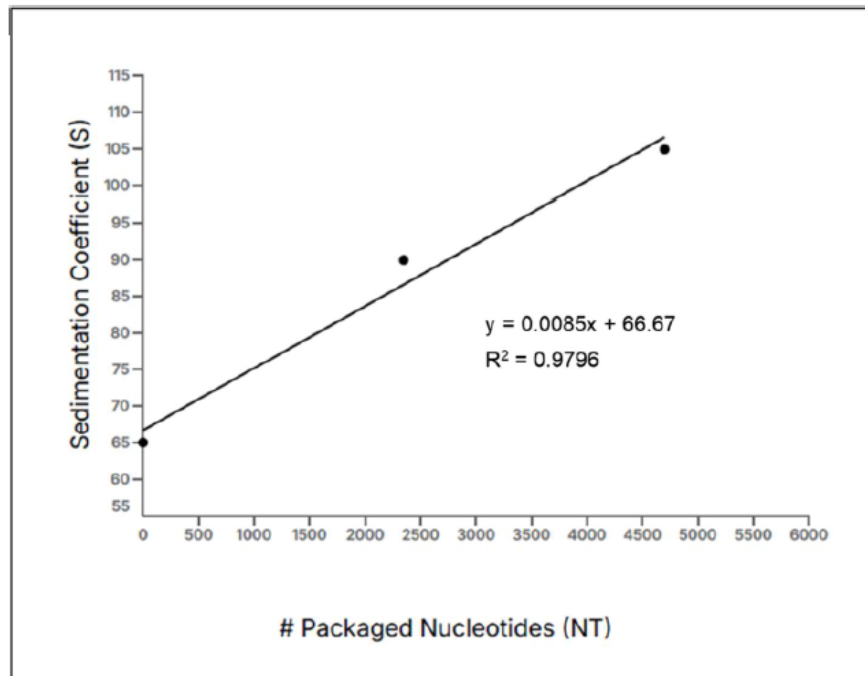
As Berkowitz 2015 explains, SE-AUC “was totally independent of the frictional properties of the protein molecule, which made MW assessment via SV-AUC much more difficult.” EX1024, 241; *see also* EX2039, 52:16-24 (“[e]quilibrium tends to be the best” AUC technique to determine a particle's molecular weight.); 146:3-10. Therefore, SE-AUC “was this mode of

sedimentation analysis that was most commonly used many years ago to assess the MW of proteins and their complexes” EX1024, 241. Thus, the POSA would not have been motivated to use SV-AUC to determine genome size. Fleming, ¶132.

ii. Le Bec’s Data Belie a Linear Relationship Between Genome Size and Sedimentation Coefficient

Sarepta’s argument that plotting sedimentation coefficient and packaged nucleotide data in Le Bec’s Figure 2 would have shown a “linear relationship between the sedimentation coefficient and the length” is based on an erroneous linear regression analysis. Petition, 53-54 (citing EX1004, ¶194). The plot contains only three data points, and none of them fall on the drawn line. Petition, 54 (citing EX1004, ¶194); *supra* §V.A. Drawing a “best fit” line is different from conducting a meaningful linear regression. Fleming, ¶133.

Both cursory visual inspection and a mathematical analysis of Sarepta’s plot refute that Le Bec’s data show a linear relationship between sedimentation coefficient and genome size. Fleming, ¶¶79-83, 134. With only three data points spanning a limited range, the deviation of each point from the line represents a large percentage of the total variation. The POSA would recognize this as a poor fit, not a confirmed linear relationship. Fleming, ¶¶80, 134-135.



Petition, 54 (citing EX1004, ¶194). A best-fit line can always be drawn through any set of points—that is a mathematical operation, not a scientific conclusion. A linear relationship requires statistical evidence. Petitioner drew a line; it did not establish statistical significance or linearity. Fleming, ¶81.

A statistical analysis of Le Bec’s Figure 2 data confirms a lack of linearity. The p-value, which indicates the probability of observing the results if no true linear relationship exists between the variables, does not support the linearity of the plotted Figure 2 data. Fleming, ¶82. A low p-value is strong evidence of a linear relationship, while a high p-value indicates that likely no linear relationship exists. EX2028, 82. The standard significance threshold is $p \leq 0.05$, meaning there is less than a 5% probability the observed relationship is due to chance, which corresponds to a 95% confidence level. *Id.* More stringent thresholds include $p \leq$

0.01 (99% confidence) and $p \leq 0.001$ (99.9% confidence) for situations requiring stronger evidence. *Id.*

Sarepta fails to apply the accepted linear regression analysis. Petition, 53-54. The slope in Sarepta's linear regression is not statistically significant at the 95% confidence level. The p-value is 0.091258, which substantially exceeds the standard significance threshold of 0.05. Fleming, ¶135. Thus, the POSA would not have considered Le Bec's data to establish a linear relationship between genome size and sedimentation coefficient. Fleming, ¶¶133-139.

In contrast, the data in Figure 8 of the '894 patent (*see also* EX2019, Figure 5A) confirmed a linear relationship between sedimentation coefficient and genome size. The nucleotide values, taken from page 16 of the Petition, are reproduced below. Fleming, ¶136.

Nucleotides	Sedimentation Coefficients (S)
2200	84
3400	92
3450	93
4300	99

The linear regression outputs are summarized in the table below:

SUMMARY OUTPUT	
<i>Regression Statistics</i>	
Multiple R	0.99882
R Square	0.997642

Adjusted R Square	0.996463
Standard Error	0.366599
Observations	4

	<i>Coefficients</i>	<i>Standard Error</i>	<i>t Stat</i>	<i>p-value</i>
Intercept	68.20201174	0.838356933	81.352	0.000151
X Variable 1	0.007130483	0.000245116	29.0903	0.00118

This summary agrees closely with the R^2 value in the patent (.9976 vs .9978). EX1001, 49:64-67. In this linear regression, the p-value for the slope of the linear regression is 0.00118, which is highly significant ($p \ll 0.05$) and passes even the more stringent threshold of $p \leq 0.01$ (99% confidence). This confirms that the linear relationship between sedimentation coefficient and genome size in the '894 patent data is statistically robust. Fleming, ¶137. The p-value for the intercept (0.000151) is also highly significant, passing the $p \leq 0.001$ threshold (99.9% confidence). This analysis confirms the inventors' representation that the '894 patent provides the first documentation of the linear relationship between sedimentation coefficient and genome size for rAAV particles. Fleming, ¶137.

Sarepta's failure to conduct a proper regression analysis of Le Bec—which the POSA would have done—is a glaring error in its obviousness analysis. Fleming, ¶139. "It is impermissible within the framework of section 103 to pick

and choose from any one reference only so much of it as will support a given position, to the exclusion of other parts necessary to the full appreciation of what such reference fairly suggests to one of ordinary skill in the art.” *Application of Wesslau*, 353 F.2d 238, 241 (C.C.P.A. 1965); *see also Henny Penny*, 938 F.3d at 1332; *W.L. Gore*, 721 F.2d at 1551. Based on a proper regression analysis of Le Bec’s data, the POSA would not have concluded that there was a linear relationship, or a predictive relationship at all, between sedimentation coefficient and genome size. Fleming, ¶139. This validates the claimed method’s nonobviousness.

iii. Carter Publications Teach Away From a Reliable Linear Relationship Between Genome Size and rAAV Particle Density

Sarepta’s Petition relies on an assumption central to its obviousness argument: the existence of “a linear relationship between the density of an AAV particle and the size of the nucleic acid packaged inside the capsid.” Petition, 7, *see also id.*, 12, 21, 25, 33, 36, 39, 43, 46, 51, 62. At deposition, however, Sarepta’s expert Dr. Berkowitz disavowed this assumption, agreeing that the data in de la Maza (1978) contradict it:

Q. So the data in the exhibit that we were just discussing [EX1007] doesn’t seem to indicate a direct linear relationship between density and DNA strand length? Right?

A. I guess you would have to say that basically. Yes.

EX2039, 101:3-11; *see also id.* 105:2-107:20.

Indeed, table 1 of de la Maza (1978) shows three isolated AAV particles having identical densities, 1.41 g/cm³, but genomes of different sizes. EX1007, 195. The identification of three particles with the same densities but different amounts of encapsidated DNA refutes Sarepta's linear relationship argument:

Q. So from this, would this data tend to indicate that the density of an AAV particle is not solely determined by the size of the genome inside?

[Objection]

A. It would look like that's the case. Yes.

EX2039, 99:9-100:3.

Sarepta's argument relies on an out-of-context quote from another Carter laboratory publication, de la Maza (April 1980), which states "the particle density has an approximately direct linear relationship to the strand length." Petition, 2, 21, 25 (quoting EX1008, 3202). At deposition, Dr. Berkowitz admitted that this quote is based on "unpublished observations" (EX1008, 3202 n.3), which he has never seen, EX2039, 105:2-6, and is inconsistent with the data in de la Maza (1978) discussed above. EX2039, 107:10-20. Further, another paper, de la Maza (March 1980) disclosed two particles with the same capsid protein and genome content but different densities. EX1013, 1129. From the Carter laboratory's publications, the POSA would have understood that the discussion of the Carter

laboratory's "unpublished observations" was meant to support a qualitative observation that AAV particles were assembled by inserting DNA into preformed capsids. EX2039, 106:24-107:20; 151:3-6; Carter, ¶49. Dr. Carter explains that this qualitative statement regarding unpublished observations did not provide the POSA with a foundation to build an analytical method for quantifying rAAV species, particularly when there is contradictory quantitative data. Carter, ¶¶48-53.

iv. Carter Laboratory Publications Belie a Linear Relationship Between Genome Size and Sedimentation Coefficient

The Petition's other central assumption, that rAAV particles "have the same size and shape" (Petition, 25, 44-46), omits consideration of other Carter laboratory publications, including de la Maza (March 1980) and Myers, which Sarepta cites. EX1012; EX1013.

De la Maza (March 1980) indicated no predictable relationship between sedimentation coefficient and genome size. De la Maza (March 1980) observed two AAV particle species, "AAV (1.41) and (1.45)," for which "the major components had an identical DNA content and DNA/protein ratio but differed slightly in their sedimentation rates and differed significantly in their relative infectivity." EX1013, 1129. De la Maza (March 1980) explains that the different densities (1.41 g/cm³ versus 1.45 g/cm³), sizes (the denser particles were "10 to 20% smaller in diameter"), and sedimentation coefficients (111 and 109) observed

for two AAV particles with the same DNA and protein content indicate differences in AAV sizes and conformations. EX1013, 1129, 1136; Carter, ¶¶38-42.

Particularly noteworthy, de la Maza (March 1980) shows that the lower density particle AAV (1.41) sediments faster ($S=111$) than the higher density particle (1.45, $S=109$). EX1013, 1129, 1136. This is opposite of what would be expected if they experienced identical friction. Fleming, ¶95; Carter, ¶42. If density were the only variable affecting sedimentation coefficient—as Sarepta argues (Petition, 45)—the denser particle would sediment faster. It does not. This means that the higher density of the AAV (1.45) particle is more than offset by the additional friction it experiences compared to the lower density AAV (1.41) particle. Fleming, ¶95; Carter, ¶42; EX2039, 92:3-10; 110:18-112:16.

Based on de la Maza (March 1980)—as Dr. Berkowitz admits—the POSA would **not** have concluded that rAAV particles have “approximately the same size and shape” (Petition, 25, 45, 46). EX2039, 113:6-115:15. Referring to EX1004, ¶168, Dr. Berkowitz testified:

Q. Now we just discussed this Exhibit 1013 of de la Maza and Carter, showing that particles appear like they can have different sizes and shape? Right?

[Objection]

A. That appears to be the case. Yes.

EX2039, 116:4-117:15.

This example disproves the Petition's central argument: that the POSA would have assumed that sedimentation coefficient was solely a function of rAAV particle density. Fleming, ¶¶93-95; EX2039, 117:16-118:8. The de la Maza (March 1980) study of intact rAAV particles by sedimentation velocity confirms the potential for conformational changes that dramatically impact the sedimentation rate for particles containing identical genomes. Fleming, ¶¶93-95; Carter, ¶¶40-47. From these disclosures, the POSA would have understood that it was not possible to determine genome size from the sedimentation coefficient of an intact rAAV particle.

Another Carter laboratory publication, Myers (EX1012), further disproves that the mechanism of assembly “in which progeny DNA strands are packaged into preformed capsids” supports Sarepta's argument that the POSA would have assumed that rAAV particles are “of approximately the same size, regardless of the length of the DNA.” Petition, 12 (citing EX1033, 983-84; EX1034, 495; EX1012, 71-72, 79-82). Myers discusses the same viral particles from the de la Maza publications, AAV(1.41) and AAV(1.45), having the same DNA content and DNA:protein ratio, but different densities, sizes, and sedimentation coefficients. EX1012, 81; Carter, ¶¶44-45. Myers theorizes that AAV(1.45) may have been a precursor to AAV(1.41), or, alternatively, itself an end product. EX1012, 81. Under either theory, the POSA would have understood that the rAAV assembly

process can result in differently shaped particles. Carter, ¶46; EX2039, 155:5-158:21. The Petition's obviousness theory fails to address this prior art teaching.

Sarepta clearly falls prey to "hindsight syndrome by reasoning backward from the teaching of the patent itself" and disregarding the art as a whole. *Creo*, 166 F. Supp. 2d at 966, *aff'd*, 305 F.3d 1337 (Fed. Cir. 2002); *see also Rockwell* 147 F.3d at 1364; *Jones*, 727 F.2d at 1529; *W.L. Gore*, 721 F.2d at 1553. This includes a failure to address publications from the same laboratory as de la Maza that contradict the obviousness grounds.

v. The POSA Would Not Have Ignored the Effect of Friction on Sedimentation Rate

De la Maza (March 1980) demonstrates that sedimentation coefficients could not be reliably used to calculate genome size. Moreover, the literature, including Dr. Berkowitz's own publications, indicated more generally that changes in viral particle shape were known to be an issue impacting sedimentation coefficient that could not be disregarded. *Supra* §§II.B–C. Sarepta attempts to bridge its failure of proof regarding rAAV particles having identical size and shape with a conclusory characterization of publications from 1978 and 1980 relating to the mechanism of genome packaging. Petition, 43-44 (citing EX1007, 203; EX1008, 3202; EX1004, ¶165). The de la Maza (March 1980) account of conformational changes in capsids having identical DNA content causing particles to sediment at different rates refutes Sarepta's argument. *Supra* §V.A.1.a.iv;

Carter, ¶¶38-42; Fleming, ¶¶93-95. Several other references, including one by Sarepta's expert, Dr. Berkowitz, confirm that viral particles can undergo changes in size and shape that unpredictably alter their sedimentation coefficients. Fleming, ¶99; Carter, ¶¶44-47, 54-59.

Berkowitz 2007 explained that viral particles can exhibit structural changes that impact their sedimentation coefficients in unforeseen ways. EX1023, 31-32. Neither the Petition nor the Berkowitz declaration explains why the POSA would have considered decades-old publications on AAV packaging as confirming that “the size and shape of intact, non-aggregated AAV particles are approximately the same, regardless of the size of the genome packaged in the capsid” (Petition, 45), when publications like Berkowitz 2007 indicated the opposite—nonuniformity in particle size and shape. Carter, ¶¶33-34; Fleming, ¶99.

Sarepta contends that certain references “show[] transmission electron microscopy images of full and empty AAV particles with the same size and shape.” Petition, 44; EX1026, 2, Figure 1; EX1041, 59, Figure 2; Ex 1004, ¶¶166-168. The POSA would not have considered the electron microscopy (“EM”) images to indicate that viral particles would have the same size and shape regardless of genome content. Visually reviewing images of bulk particles is not suited to detecting the types of surface changes that would impact a particle's

frictional coefficient, like changes to fibrous spikes on viral particle surfaces.

Fleming, ¶102.

Moreover, EM is limited to analyzing the physical topology of a particle. Dr. Berkowitz concedes that EM would not detect “chemical differences between particles that could impact the[] hydrodynamic forces” that affect friction in an SV-AUC experiment. EX2039, 47:19-49:15; 50:7-20.

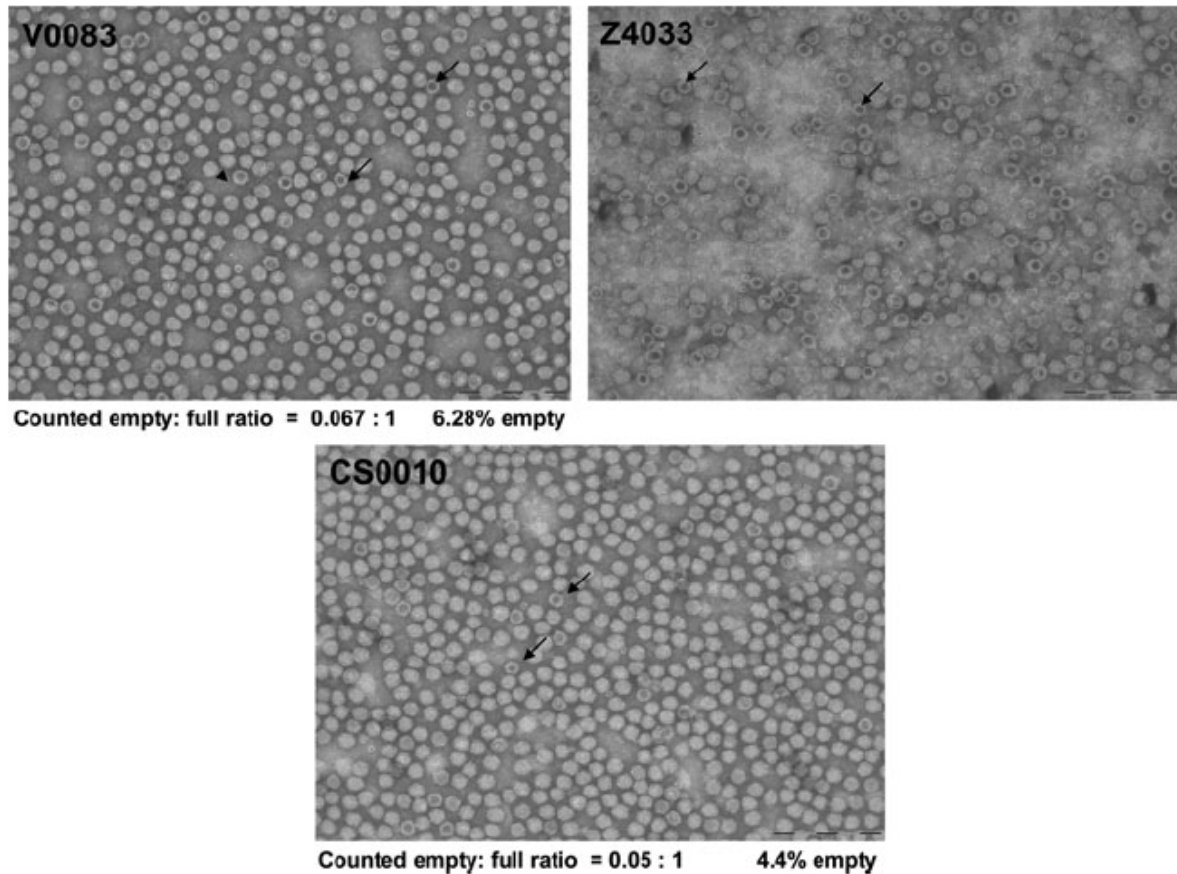
Contrary to the Petition’s argument, Gao (EX1026) and Lock (EX1041) do not “show[] transmission electron microscopy images of full and empty AAV particles with the same size and shape.” Petition, 44; Fleming, ¶102; Carter, ¶55.

Gao explains that electron micrographs are acceptable for “qualitatively studying the structures of virions.” EX1026, 6. The POSA would understand Gao, however, to refute relying on EM to evaluate quantitative issues, including whether differences in particle size and shape could impact friction in SV-AUC experiments. Fleming, ¶102; Carter, ¶56. Dr. Berkowitz acknowledged that neither his declaration nor Gao quantified rAAV particle size and shape based on Gao’s EM images. EX2039, 122:2-123:1. He further acknowledged that frictional coefficient, in contrast, is quantitative. EX2039, 123:13-124:7. Without quantitative analysis, the POSA could not have possibly used EM to conclude that all particle species had approximately the same size and shape. Fleming, ¶¶102, 146-149; Carter, ¶54.

Moreover, as explained (*supra* §II.C), Gao states that “encapsidation of full or partial AAV genomes may lead to capsid conformational changes” EX1026, 6. In fact, Gao (EX1026) indicates that partially empty particles (*i.e.*, particles containing partial genomes) do have different conformations than empty particles. EX1026, 6 (partially empty virions exhibit “different capsid conformations as compared with [completely empty] particles”). The POSA would have understood that conformational changes to the capsid are relevant to AUC analysis because they affect the sedimentation coefficient by impacting the frictional coefficient of a particle. *Supra* §II.B; Fleming, ¶102; Carter, ¶¶41, 57. Indeed, Dr. Berkowitz testified that conformational changes could affect the chemical interactions (*e.g.*, hydration and solvation) between rAAV particles and the solution in SV-AUC experiments, which could cause changes to frictional coefficients. EX2039, 128:16-130:10. Dr. Berkowitz admitted, however, that his analysis was limited to particle size and shape, and that he did not consider chemical interactions known to impact friction. EX2039, 130:12-131:1; 134:16-135:4; Fleming, ¶150.

Lock (EX1041) confirms Gao’s characterization of EM, stating that “[e]ven in the sharpest images, some particles appear only partially stained, and this creates ambiguity in characterizing them as empty or full.” EX1041, 57. The POSA would disagree with Sarepta’s contention that Lock “show[s] transmission electron microscopy images of full and empty AAV particles with the same size and

shape.” Petition, 44. Lock’s images indicate a qualitative difference in particle shapes and sizes. Fleming, ¶¶103, 151; Carter, ¶58.



EX1041, 59. Moreover, the POSA would have considered the frictional coefficient to require a quantitative assessment of particle shape and chemical properties.

Neither Lock nor Gao supports Sarepta’s reliance on their qualitative images for assessing quantifiable differences in particle friction. Carter, ¶59; Fleming, ¶151. Despite relying on qualitative EM images to conclude that rAAV particles have the same size and shape regardless of genome content, Dr. Berkowitz testified that it was difficult to “quantitate partials” via visual inspection of EM images.

EX2039, 133:9-134:21. Dr. Berkowitz also explained that negative staining used in EM experiments dehydrates the particles so that they are not the same size and shape as in their native environment:

Q. And the Lock and Gao electron microscopy images that you looked at, those were done through the negative staining technique that you are referring to?

A. Yes.

Q. So it is possible that the particles are not the same size and shape that they would be in a sedimentation velocity AUC experiment?

[Objection]

A. It is possible, yes, because they are not in their native environment.

EX2039, 139:20-140:20; Fleming, ¶152.

Dr. Berkowitz lists other techniques, including X-ray crystallography, cryo-EM, and size exclusion chromatography, which may provide more high-resolution information about particle size, shape, and chemical properties than the bulk EM images in the Petition. EX2039, 137:15-138:15; 138:21-139:8; 140:19-141:17. But Dr. Berkowitz concedes that he did not consider any of these techniques, in part, because “they were more recent” because access to this type of information was “much more limited” before 2015. EX2039, 142:5-22.

For all of the above reasons, the POSA would not have considered bulk EM images as useful for assessing differences in particle size or shape that could

impact the sedimentation rate of different rAAV particle species. Carter, ¶¶54-59; Fleming, ¶¶153-156. The information about rAAV particle structure that was available before 2015, would have led the POSA to conclude the rAAV particles were not the same size and shape.

vi. As Dr. Berkowitz Testified, the Prior Art Showed that rAAV Particle Shape Was Unpredictable

Sarepta does not cite any prior art that describes the use of sedimentation coefficients to determine the size of a fragmented genome for viral particles of any type. Indeed, despite evidence of rAAV particles containing partial genomes, Le Bec did not attempt to use sedimentation coefficients to calculate genome size. Fleming, ¶154. According to Dr. Berkowitz's testimony, one of the reasons for this may have been the POSA's perception based on the literature, such as the SV-AUC study in de la Maza (March 1980) of the AAV (1.45) and AAV (1.41) particles, that rAAV particle structures were unpredictable:

Q. So I asked whether the fact that the denser particle sediments at a lower rate than the less dense particle ... is an indicator that there is unpredictable friction in AAV particles?

[Objection]

A. It is unpredictable structure of the particle, how it is behaving. It plays out in terms of whatever the particle structure is there, then that reflects the physical properties basically. So I am sort of maybe getting

when you say unpredictability about friction, you are talking about the unpredictability of the structure.

EX2039, 175:24-176:17; *see also* 174:16-175:17; 176:19-177:5.

There is no dispute that nearly forty years elapsed from de la Maza's 1978 publication to the priority date. Sarepta failed to identify a reference that even *hypothesized* that such a linear relationship existed, much less applied it, to determine the size of fragmented genomes in viral particles. "[T]his considerable time lapse" is an indicator that Sarepta "only traverses the obstacles to this inventive enterprise with a resort to hindsight." *Leo*, 726 F.3d at 1356; Fleming, ¶155. If the claimed method were obvious, someone would have done it, or at least proposed it, in the 37 years between de la Maza (1978) and the '894 patent's priority date.

The Petition uses the Challenged Claims as a roadmap to "selectively cull[] from the prior art to fit the parameters of the patented invention." *ATD Corp.*, 159 F.3d at 546. Sarepta ignores that the teachings of de la Maza (March 1980), Gao, Berkowitz 2007, and Berkowitz 2015, among others, fail to support Sarepta's conclusions of obviousness. An obviousness analysis requires examining what the POSA would have done without the inventors' guidance. *Arendi*, 832 F.3d at 1361 (stating that "the court must avoid 'hindsight bias and must be cautious of arguments reliant upon ex post reasoning.'"); *Trivascular, Inc. v. Samuels*, 812

F.3d 1056, 1066 (Fed. Cir. 2016) (the Board must “be careful not to allow hindsight reconstruction of references . . . without any explanation as to how or why the references would be combined to produce the claimed invention”); *InTouch Techs., Inc. v. VGO Commc’ns*, 751 F.3d 1327, 1352 (Fed. Cir. 2014) (finding that the “expert also succumbed to hindsight bias in her obviousness analysis.”). These references make clear that the POSA would not have expected a linear relationship between *S*-values and genome size, and would have not been motivated to use sedimentation coefficients to determine genome size. Fleming, ¶156.

b. The POSA Would Not Have Reasonably Expected that Sedimentation Coefficients Could Be Used to Determine Fragmented Genome Size in rAAV Particles

Sarepta fails to carry its burden to show that the POSA “would have had a reasonable expectation of success in” combining teachings “to achieve the claimed invention.” *Intelligent Bio-Sys., Inc. v. Illumina Cambridge Ltd.*, 821 F.3d 1359, 1367–68 (Fed. Cir. 2016) (internal quotation marks and citations omitted). The POSA’s lack of a reasonable expectation of success is highlighted by the proper statistical analysis of Le Bec’s data. *Supra* §V.A.1.a.ii; Fleming, ¶157. Sarepta’s reasonable expectation of success argument fails for much the same reasons as its motivation argument. *Supra* §§V.A.1.a-V.A.1.a.vi; Fleming, ¶157.

The POSA would not have reasonably expected to be able to determine the genome size of an rAAV particle species using SV-AUC. Le Bec's data show a decidedly nonlinear relationship between genome size and sedimentation coefficient. Fleming, ¶158. The POSA would not have considered de la Maza's qualitative mention of a linear relationship between particle density and genome size to suggest that a linear relationship exists between sedimentation coefficient and genome size, because it failed to account for variability in rAAV particle size and shape. *Supra* §V.A.1.a.v. De la Maza (1978) showed that AAV particle density is not necessarily determined by the genome size. Carter, ¶37; *Supra* §V.A.1.a.iii. De la Maza (March 1980) showed that even rAAV particles having identical DNA content can have different sedimentation coefficients. Carter, ¶¶38-42; *Supra* §V.A.1.a.iv. Dr. Berkowitz testified that these results indicated "unpredictability of the structure" of rAAV particles. *Supra* §V.A.1.a.vi; EX2039, 175:24-176:17; *see also id.*, 174:16-175:17; 176:19-177:5.

The POSA would never have assumed that "AAV particles are approximately the same, regardless of the size of the genome packaged in the capsid" (Petition, 45). Carter, ¶¶35-47. The POSA would not have ignored the effects of friction. *Supra* §§II.B, V.A.1.a.v; Fleming, ¶158. Thus, the POSA would not have predicted a linear relationship between encapsidated genome size and sedimentation coefficient. Fleming, ¶158.

In his own publications, Sarepta's expert, Dr. Berkowitz, described viral particles having differing shapes and sizes affecting the relative friction of those particles. Fleming, ¶159. Berkowitz 2007 explained that some particles were observed to lose surface spikes, which impacted their sedimentation coefficient. EX1023, 31-32. Berkowitz 2007 observed that some "adenovirus monomers [may] have lost their fiber spikes" resulting in "significantly reduced hydrodynamic drag" decreasing friction therefore leading to "a higher sedimentation coefficient than the intact adenovirus particle." *Id.*, 31. This was a phenomenon observed for other viral particles as well. *Id.* ("Similar effects have been observed for the bacterial virus T2 with regard to an extended versus compact geometric arrangement of its tail spikes or fibers "). The POSA would have expected that a similar issue would arise for rAAV particles, which also exhibit "spikes" projecting from their capsids. Fleming, ¶159. Thus, the POSA would not have expected a predictive correlation between sedimentation coefficient and the size of the encapsidated genome as Sarepta alleges. Petition, 52-53; Fleming, ¶159.

A reasonable expectation of success argument that fails to account for, and rebut, this unpredictability cannot prove obviousness. *Amneal Pharms. LLC v. Cubist Pharms. LLC*, IPR2020-00193, Paper 7 at 22-32 (PTAB May 29, 2020); *Sandoz Inc. v. AbbVie Biotechnology Ltd.*, IPR2017-01823, Paper 16 at 14 (PTAB Feb. 9, 2018); *Coherus Biosciences Inc. v. AbbVie Biotechnology Ltd.*, IPR2016-

01018, Paper 12 at 4 (PTAB Feb. 2, 2017); *Momenta Pharms., Inc. v. Bristol-Myers Squibb Co.*, IPR2015-01537, Paper 37 at 12-13 (PTAB Dec. 22, 2016). As a “touchstone of obviousness,” a reasonable expectation of success requires not only an “expectation that prior art elements are capable of being physically combined, but also that the combination would have *worked for its intended purpose*.” *Depuy Spine, Inc. v. Medtronic Sofamor Danek, Inc.*, 567 F.3d 1314, 1326 (Fed. Cir. 2009) (emphasis added). The POSA would not have reasonably expected to be able to reliably determine the size of a fragmented encapsidated genome from a particle’s sedimentation coefficient based on Le Bec and de la Maza, in view of Sommer. Fleming, ¶¶160-161.

2. Le Bec and de la Maza, In View of Sommer Did Not Render Obvious “determining the quantity of one or more species of the variant viral particles in the heterogeneous mixture of viral particles”

Sarepta’s arguments with respect to element (iv) of claim 1, which requires “determining the quantity of one or more species of the variant viral particles in the heterogeneous mixture of viral particles” (Petition, 47), fail for the same reasons described above for claim element (iii). *Supra* §V.A.1. Le Bec’s concentration analyses were qualitative and did not account for the different extinction coefficients of each particle or for quantitative concentration measurements. EX2039, 77:1-14; *see also id.*, 144:23-145:2; *supra* §II.A. To quantify viral particles from a C(s) distribution obtained with absorbance spectroscopy detection,

the detection method applied by Le Bec, the POSA requires additional information about the viral particles, for example the extinction coefficient of the particles at the measured wavelength. Petition, 47-50; Fleming, ¶162; EX2039, 145:4-146:15 (Dr. Berkowitz testifying that Le Bec does not attempt to obtain the molecular weights or extinction coefficient of its rAAV particles).

Calculating an extinction coefficient requires determining the molecular weight of DNA packaged in the rAAV particle, *i.e.*, determining the size of the genome. Petition, 51-52; *see also id.*, 32; Fleming, ¶163. Sarepta's obviousness argument with respect to element (iv) of claim 1, therefore, relies on its Ground 1 argument for claim element 1(iii). Petition, 51 (citing Section IX.A.1.d).

When absorbance spectroscopy at given wavelengths (*e.g.*, Le Bec follows absorbance at 276 nm) is applied in AUC methods to monitor sedimentation of viral particles, knowing the proportion of peak area to concentration requires knowing additional information about the particles. For example, the extinction coefficient (ϵ) of each species is typically used to calculate molar concentration, because different rAAV species will absorb certain wavelengths of light to different degrees. *Supra* §II.A; Fleming, ¶¶164-165; EX1001, 24:41-44 (“[E]xtinction coefficients are used to calculate molar concentration and the actual percent value of the intact vector peak from absorbance data.”).

Consistent with Berkowitz 2007, the inventors explained that at the time of the invention, “it [was] not possible to determine empirically the extinction coefficient of particular species of recombinant viral particles (*e.g.*, viral particles with fragmented genomes of unknown size and sequence).” EX1001, 24:56-59. By finding a way to determine the genome size, the inventors were able to calculate extinction coefficients of variant rAAV species and determine the relative concentrations of these species. Fleming, ¶¶166-168. This innovation helped lead to the invention of claim 1.

B. Claim 2: The Cited Prior Art Fails to Teach or Suggest an SV-AUC Method “wherein each species of the individual variant viral particles ... is fully resolved”

The POSA would not have considered either of Le Bec’s SV-AUC plots to support Sarepta’s contention that “each species of the individual variant viral particles is fully resolved” in Le Bec’s Figures 2 and 3. Petition, 50 (citing EX1006, 18:8-32). It is clear that species appearing in the sedimentation plots of both Figures 2 and 3 of Le Bec are not fully resolved. *Supra* §V.A.1. This makes sense because the POSA would have understood that Le Bec was not attempting to quantify peaks corresponding to different rAAV species containing fragmented genomes, as Dr. Berkowitz testified. EX2039, 76:7-77:7.

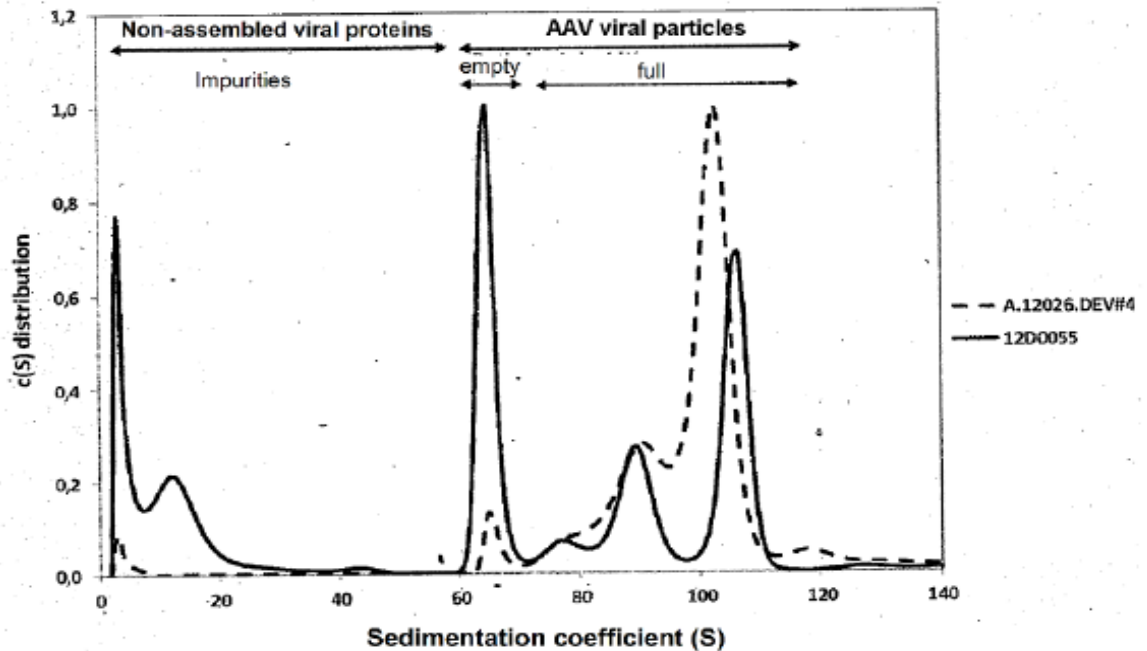


Figure 2

EX1006, Figure 2. The peak corresponding to empty capsids, as well as AAV particles at approximately 75S and 90S, according to Sarepta (Petition, 50) are not fully resolved from each other. Fleming, ¶169.

Le Bec's Figure 3 is even worse. The Petition and Berkowitz Declaration never address the agglomeration of what appears to be multiple peaks ranging from about 75S to about 110S in Figure 3 of Le Bec. Fleming, ¶170.

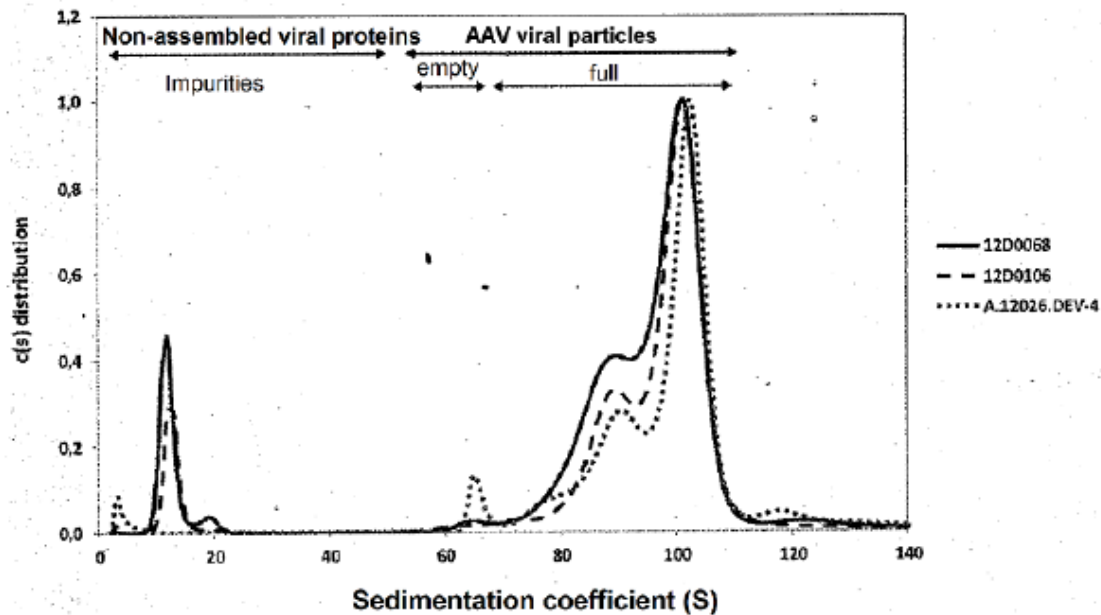


Figure 3

EX1006, Fig. 3. This commingling of species masks the presence of multiple peaks, whereas the POSA would have understood that for heterogeneous mixtures of particles, the use of AUC under boundary sedimentation conditions depends on resolving peaks of different species. Because Le Bec's peaks overlap and commingle, it is impossible to assign specific sedimentation coefficients to individual species or to integrate their areas for quantification. Fleming, ¶170; EX2039, 75:12-21 (Figure 2); 69:1-24 (Figure 3). The POSA would not understand which species is assigned to or contributes to each peak.

Le Bec's Figures 2 and 3 sharply contrast with the fully resolved peaks of the figures of the '894 patent. Fleming, ¶171. Sarepta fails to point to any reference cited in the Petition presenting fully resolved peaks, as recited in claim 2. Full

resolution of peaks is not a cosmetic requirement—it is a functional prerequisite for the claimed method. Fleming, ¶171. Without fully resolved peaks, it is impossible to accurately integrate the area under each peak, which is the basis for quantifying the concentration of each species. Le Bec’s broad, overlapping peaks in Figures 2 and 3 would have made accurate quantification impossible, which is precisely why Le Bec never attempted to quantify individual rAAV species. Fleming, ¶171. Accordingly, Sarepta fails to prove claim 2 was obvious.

C. Claims 3-7, 10-11, and 20-30 Depend from Claim 1 and Are Not Unpatentable for the Same Reasons as Claim 1

Because claims 3-7, 10-11, and 20-30 depend from claim 1, and Sarepta’s only challenge to claim 1 is in Ground 1, Sarepta’s failure to prove claim 1 obvious also results in a failure of proof for claims 3-7, 10-11, and 20-30 of the ’894 patent. Fleming, ¶172.

VI. Ground 2: The Petition Fails to Establish Le Bec and de la Maza, in view of Sommer and Cole, Renders Claims 12-19 Obvious

Because claims 12-19 depend from claim 1, and Sarepta’s only challenge to claim 1 is in Ground 1, Sarepta’s failure to prove claim 1 obvious also results in a failure of proof for claims 12-19 of the ’894 patent.

VII. Objective Indicia of Nonobviousness Support the Patentability of the Challenged Claims

Genzyme’s evidence of objective indicia of nonobviousness further “guard[s] as a check against hindsight bias” that Sarepta applied here. *In re*

Cyclobenzaprine, 676 F.3d 1063, 1079 (Fed. Cir. 2012). The objective indicia evidence, particularly when taken together, is “not just a cumulative or confirmatory part of the obviousness calculus but constitutes independent evidence of nonobviousness.” *Leo*, 726 F.3d at 1358 (Fed. Cir. 2013) (quoting *Ortho–McNeil Pharm., Inc. v. Mylan Labs., Inc.*, 520 F.3d 1358, 1365 (Fed. Cir. 2008) (internal quotation marks omitted).

A. The Claimed Method’s Capability to Determine Genome Size and Quantify Fragmented Particles Was Surprising and Unexpected

The claimed method surprisingly and unexpectedly provides the POSA with a single experiment that determines the size and quantity of impurity rAAV particles containing fragmented genomes. *See Forest Labs., LLC v. Sigmapharm Labs., LLC*, 918 F.3d 928, 937 (Fed. Cir. 2019) (holding that “in considering unexpected results, courts ask whether ‘the claimed invention exhibits some superior property or advantage that a person of ordinary skill in the relevant art would have found surprising or unexpected.’”) (quoting *In re Soni*, 54 F.3d 746, 750 (Fed. Cir. 1995)).

Le Bec and de la Maza (1978), the references Sarepta alleges were the closest prior art to the claimed invention, indicate to the POSA that it was not possible to determine the size of a fragmented genome from a sedimentation coefficient. *Janssen Pharms., Inc. v. Teva Pharms. USA, Inc.*, 97 F.4th 915, 933 (Fed. Cir. 2024) (“The starting reference point for evaluating unexpectedness is the

closest prior art.”) (cited references omitted). Le Bec never attempts to determine genome size from sedimentation coefficients of viral particles containing fragmented genomes. *Supra* §V.A.1. Le Bec focuses on the much less ambitious goal analyzing “empty and full AAV viral particles” by grouping rAAV particles containing partial genomes together with those having intact genomes as “full AAV viral particles.” *Supra* §V.A.1; EX1006, 18:21-25; Figure 2; Fleming, ¶¶173-174. Subjecting Le Bec’s data to statistical analysis fails to indicate a linear relationship between genome size and sedimentation coefficient. *Supra* §V.A.1.a.ii; Fleming, ¶174.

De la Maza (1978) does not attempt to subject intact particles to SV-AUC. *Supra* §V.A.1; Fleming, ¶175; Carter, ¶37. De la Maza (March 1980), however, shows that rAAV particles containing the same DNA content can nonetheless have different sedimentation coefficients, which is attributable to conformational change. EX1013, 1136; *Supra* §V.A.1.a.iv; Carter, ¶¶38-42; Fleming, ¶175. Several other references indicated that rAAV particles adopted different structural conformations depending on their genome content. *Supra* §II.C; Carter, ¶¶44-47; Fleming, ¶175.

The inventors did the unexpected. Despite the art indicating no particular relationship between sedimentation coefficient and genome size, they found that a statistically significant linear relationship does exist. Fleming, ¶176. By applying

this relationship in the claimed method, it was possible in a single experiment to determine genome size and quantify rAAV particles containing fragmented genome. This unexpected advantage led to AUC becoming the “gold standard” in rAAV analysis, as explained below. *Forest Labs.*, 918 F.3d at 937.

B. Industry Praise Is Further Evidence of Nonobviousness

A 2015 peer-reviewed publication co-authored in part by the inventors of the '894 patent, Burnham and O’Riordan, and colleagues (EX2019), publicized experiments disclosed in the '894 patent, including Examples 1-6. *See, e.g.*, EX2019, 232, Figure 1 (EX1001, Example 1 (FIGS. 1A-B, 2A-B)), 233, Figure 2 (EX1001, Example 2, Table 4, FIGS. 3A-3B), 234, Figures 3A-B (EX1001, Example 3, FIGS. 6B-C), 235, Figure 4 (EX1001, Example 4, FIG. 7A-7C), 236, Figure 5 (EX1001, Example 5, FIGS. 8, 9A-9C), 237, Figure 6D (EX1001, Example 6, FIGS. 11A); Fleming, ¶177. Between its publication in 2015 and July 2026, the Burnham publication has been cited over 200 times according to Google Scholar. EX2035. Citations in peer-reviewed references are a reflection that the work is of importance in the field in which it was published. Fleming, ¶178. Dr. Berkowitz is among the authors to have cited the Burnham publication multiple times. EX2041, 3, 5 (citing EX2019); EX2042, 3 (citing EX2019); EX2043, 7, 8 (citing EX2019).

Additionally, several industry sources have made comments that reflect the value of the claimed AUC method. A 2020 white paper by Beckman Coulter GmbH, the manufacturer of instruments for AUC analysis, highlighted the importance of the inventors' contribution, stating:

Burnham and colleagues showed that sedimentation velocity ... [AUC] experiments are able to characterize the quality of viral vector productions. *A single AUC experiment provides insights into the composition of the encapsulated DNA, the success of the purification, the presence of aggregates and the ratio between empty, partially filled and filled viral particles....* Taken together, the AUC is a valuable tool to analyze rAAV vectors notwithstanding the composition and length of the transgene or the viral serotype.

EX2021 (citing EX2019) (emphasis added). Thus, Genzyme's work helped make SV-AUC the "gold standard in the detection and characterization of AAV particles." *Id.*; Fleming, ¶179.

BioAnalysis LLC, a contract research organization whose business is built around the AUC method for gene therapy applications, also recognizes that SV-AUC "has emerged as the gold standard technique for the empty/partial/full capsid assay in gene therapy product development," offering "unmatched resolution, accurate quantitation, and regulatory compatibility" and representing "the preferred method for characterizing viral vectors such as AAV, lentivirus, and other delivery systems." EX2036. Compared to alternative methods, including transmission EM,

UV/Vis spectroscopy, charge detection mass spectrometry, mass photometry, and SEC-MALS, SV-AUC is superior for quantifying rAAV species. *Id.*; Fleming, ¶180.

Sigma-Aldrich, a major scientific reagent and instrumentation provider, describes AUC as “widely recognized as a gold standard method for determining the empty/full AAV capsid ratio.” It extolls the method for “high resolution separation, allowing for quantitation of empty partial and full capsids where partial capsids contain partial or truncated fragments of the gene or capsid with contaminant from the host cell,” and that it is “serotype independent.” EX2037; Fleming, ¶181.

Citing the inventors’ work, a 2018 publication co-authored by an inventor of the Petitions’ primary prior art reference (EX1006), Christine Le Bec, and others, stated that “for the determination of empty-to-full capsid ratio, here we used AUC, a method that has been demonstrated to provide a reliable readout.” EX2020, 166-67 (citing EX2019). It is not surprising the rAAV field, including Le Bec, would rely on the AUC inventors’ method. Fleming, ¶182. As explained herein, the Le Bec prior art reference relied on by the Petition (EX1006) did not provide a method that could quantify the ratio of different variant rAAV species within an rAAV preparation.

The method disclosed and claimed in the '894 patent was the innovation that made AUC a valuable tool to identify the species present in rAAV compositions and measure the “the ratio between empty, partially filled and filled viral particles.” EX2021; Fleming, ¶183. Sarepta neither demonstrates that AUC was used in the prior art to identify the size of each species present in a rAAV composition, nor that anyone recognized that it could quantify the concentrations of those rAAV species, particularly partially filled rAAV species.

The praise is directed to the utility of the claimed method for “determining the genome size of one or more species of the individual variant viral particles in the heterogeneous mixture of viral particles; and [] determining the quantity of one or more species of the variant viral particles” in a single experiment. Fleming, ¶184. Because the praise is tied to the elements expressly recited in the claims, it supports nonobviousness. *Quanergy Sys. v. Velodyne Lidar USA, Inc.*, 24 F.4th 1406, 1417 (Fed. Cir. 2022) (“We presume a nexus when the patentee shows that the asserted objective evidence is tied to a specific product and that product embodies the claimed features, and is coextensive with them.”) (internal quotations omitted).

C. The Claimed Method Satisfied a Long-Felt Need for Characterizing rAAV Impurities

Prior to the '894 patent's effective date, there was a long-felt and unsolved need for a method that accurately quantified the concentration of the rAAV

particles containing fragmented genomes. EX2019, 228; Carter, ¶¶25-29; Fleming, ¶185. As impurities, rAAV particles containing partial genomes could be immunotoxic. Carter, ¶28; Ex. 1026, 6. The inventors' contemporaneous peer-reviewed publication, submitted in April 2015 and published in September 2015, explained:

[N]o established approaches exist for assessing the homogeneity of rAAV vector preparations that include measuring both the presence and fractional content of empty viral particles, full genome-containing viral vectors, and vectors with fragmented genomes or non-transgene-related DNA contaminants. Thus, identifying and developing sensitive and accurate methods to assess vector quality are needed.

EX2019, 228. Ensuring the safety and efficacy of rAAV-based gene therapies requires a method to analyze the manufacture of rAAV compositions, and ensure the quality of final compositions. EX2019, 229.

For example, “[d]epending on the origin of the cells used for AAV generation, *i.e.*, human or non-human origin, genotoxicity and immunotoxicity are two theoretical risks.” Ex. 2027, 87; Carter, ¶27; Fleming, ¶185. The European Medicine Agency's licensure assessment of the gene therapy Glybera[®] raised quality objections over encapsidated residual baculovirus genetic sequences co-purified with the product that were below the detection level of the impurity assay. Ex. 1029, 15, 18, 19-20; Carter, ¶27; Fleming, ¶185. Dr. Berkowitz also testified

to the importance of characterizing all rAAV species in a gene therapy composition, because of risks associated with safety and efficacy.

Q. In characterizing a gene therapy composition, if there is an error in the analysis of the amounts of different species present, that could impact the infectivity of that composition? Right?

[Objection]

A. Right.

Q. And infectivity would be a concern for the efficacy of that composition? Right?

A. Yes.

Q. And infectivity could also be a concern for the safety of that composition? Right?

A. Yes.

Q. So would you agree that the stakes of analyzing an AAV composition accurately are high for its efficacy and safety?

[Objection]

A. Absolutely. Yes.

EX2039, 119:9-120:7; Carter, ¶29; Fleming, ¶186.

Prior to the inventions claimed in the '894 Patent, determining the DNA content of recombinant viral vectors was typically performed by Southern blot

analysis. Fleming, ¶187. Southern blot requires a sequence-specific probe and cannot quantify different species of rAAV in a heterogeneous rAAV mixture. Fragmented genomes of unknown sequence could evade detection. *Supra* §III.A.2.b. Failure to detect and quantify a fragmented genome-containing rAAV impurity could compromise the safety of a gene therapy composition by, for example, causing immunotoxicity effects. EX2027, 87; EX1026, 6; Fleming, ¶187.

Le Bec demonstrates that even researchers who used AUC as part of their characterization work in the rAAV field did not develop, and could not derive, the quantitative, comprehensive methodology claimed in the '894 patent. Fleming, ¶188. Le Bec used AUC qualitatively to track empty and genome-containing rAAV particles, and never attempted to characterize rAAV particles containing fragmented genomes. *Supra* §V.A.1.

The claimed invention was the first single assay that could distinguish full, properly-formed rAAV particles from the empty capsids and fragmented genome-containing rAAV particles, and quantify each component. The claimed invention met the previous unsolved need to identify impurities that could compromise the safety of gene therapy compositions. *See Transocean Offshore Deepwater Drilling, Inc. v. Maersk Drilling USA, Inc.*, 699 F.3d 1340, 1354 (Fed. Cir. 2012) (Holding that inventor and expert testimony can establish an invention met a long-felt need.). The failures of others and the long-felt need to develop a quantitative assay

to characterize rAAV species in a gene therapy composition confirms the nonobviousness of the claimed invention.

VIII. Conclusion

For the foregoing reasons, the Board should find the Challenged Claims not unpatentable. Sarepta's obviousness theory depends entirely on hindsight. No prior art reference ever taught, suggested, or hypothesized that sedimentation coefficient could be used to determine the size of a fragmented genome in an intact rAAV particle. The primary reference, Le Bec, fails statistical significance testing for linearity—a fact Sarepta conceals by omitting the p-value. Moreover, the high p-value would have been concerning to the POSA and taught away from reliance on Le Bec in accordance with the invention of the '894 patent.

The other primary reference, de la Maza (1978), used SE-AUC on released DNA, not intact particles, and de la Maza (March 1980) directly contradicts Sarepta's core assumption by showing that the denser of two identical-genome particles sediments more slowly. Sarepta's expert's publications and deposition testimony warn against assuming uniform frictional properties for viral particles. And the co-author of de la Maza (1978), and leader of the laboratory from which it emerged, Dr. Barrie Carter, has submitted a declaration confirming that the POSA would not have made the assumptions Sarepta attributes to his laboratory's

publication. Finally, objective indicia, including unexpected results, industry
praise, and long-felt need, independently confirm nonobviousness.

Respectfully submitted,

Dated: August 5, 2026

/ Blaine M. Hackman /
Blaine M. Hackman
Reg. No. 67,479
Counsel for Patent Owner

WORD COUNT CERTIFICATION

Pursuant to 37 C.F.R. §42.24(d), I certify that this Patent Owner Response contains 13,980 words (excluding the title page, table of contents, table of exhibits, claim listing, this certificate, and the certificate of service), as determined by Microsoft Word.

Dated: August 5, 2026

/ Blaine M. Hackman /
Blaine M. Hackman
Reg. No. 67,479
Counsel for Patent Owner

CERTIFICATE OF SERVICE

I certify that today in:

Sarepta Therapeutics, Inc., v. Genzyme Corp., IPR2026-00150
(U.S. Patent 12,031,894)

I caused to be served a copy of:

PATENT OWNER'S RESPONSE
EXHIBITS 2025-2032

upon:

Sarepta Therapeutics, Inc. c/o:

robertwilson@quinnemanuel.com
annetoker@quinnemanuel.com
jimglass@quinnemanuel.com

via:

electronic service to the email addresses above.

Dated: August 5, 2026

By: / Blaine Hackman /
Blaine Hackman, Reg. No. 67,479
Dechert LLP
Three Bryant Park
1095 Avenue of the Americas
New York, NY 10036-6797
Tel: 212-698-3500
Fax: 212-698-3599
blaine.hackman@dechert.com