UNITED STATES DISTRICT COURT
FOR THE DISTRICT OF MASSACHUSETTS

CHR. HANSEN HMO GMBH, Plaintiff, v. GLYCOSYN LLC, Defendant.	C.A. No. 1:22-cv-11090

COMPLAINT

1. This is an action for a declaratory judgment of invalidity and non-infringement of United States Patent No. 9,970,018 (the "'018 Patent," attached as Exhibit A) pursuant to the Declaratory Judgment Act, 28 U.S.C. §§ 2201-2202, and the Patent Laws of the United States, 35 U.S.C. §§ 1 *et seq.*

THE PARTIES

2. Plaintiff Chr. Hansen HMO GmbH ("Chr. Hansen HMO") is a private limited liability company organized under the laws of Germany, with its principal place of business at Maarweg 32, D-53619 Rheinbreitbach, Germany.

3. Chr. Hansen HMO was formerly known as Jennewein Biotechnologie GmbH. Upon acquisition of all shares of Jennewein Biotechnologie GmbH through Chr. Hansen Holding A/S (DK) on September 22, 2020, Jennewein Biotechnologie GmbH was renamed to Chr. Hansen HMO GmbH effective May 04, 2021. For purposes of this Complaint, "Chr. Hansen HMO" refers to both Chr. Hansen HMO and where appropriate its predecessor, Jennewein.

4. Jennewein was a pioneer in the development of 2'-FL, a human milk oligosaccharide ("HMO"). After years of research and development, it was the first company to

develop a commercially successful process for making 2'-FL and was the first company to obtain regulatory approval for 2'-FL made by fermentative production using a genetically engineered *E. coli* from the U.S. Food and Drug Administration ("FDA").

5. Defendant Glycosyn LLC, on information and belief, is a Massachusetts limited liability company having an address of Sawyer Road, Suite 120, Waltham, MA 025453.

6. Glycosyn alleges that it is the owner of the entire right, title, and interest in the '018 Patent.[1]

NATURE OF THE ACTION

7. In this action, Chr. Hansen HMO seeks a judicial declaration that it does not infringe any of the claims of the '018 Patent, and that those claims are invalid.

8. Chr. Hansen HMO seeks this relief because, *inter alia*, Glycosyn previously sued Jennewein, Chr. Hansen HMO's corporate predecessor, for infringement of the '018 Patent, and because on June 14, 2022, Glycosyn sued Abbott Laboratories ("Abbott") in the U.S. District Court for the Western District of Texas, *Glycosyn LLC v. Abbott Laboratories*, C.A. No. 6:22-cv-619 (the "Waco Action"), based on the manufacture, use, offer for sale, and/or sale by Abbott of infant formula containing 2'-FL manufactured by Jennewein.

9. As will be explained in this Complaint, Glycosyn's previous actions against Jennewein and its pending lawsuit against Abbott create an actual and justiciable controversy between Chr. Hansen HMO and Glycosyn.

[1] Glycosyn's counsel, Mintz Levin, has a security interest giving it "all proceeds derived" from the '018 Patent to secure $3.8 million in unpaid bills. In addition, in May 2022, Glycosyn recorded another security interest with Gingko Bioworks.

JURISDICTION AND VENUE

10. This Court has subject matter jurisdiction under 28 U.S.C. §§ 2201, 2202, and 1338, as a declaratory judgment action arising under the Patent Laws, Title 35 of the United States Code.

11. Venue is proper in this district under 28 U.S.C. §§ 1391(b) and (c), and the Court has personal jurisdiction under Glycosyn, because Glycosyn is, on information and belief, a Massachusetts limited liability company with its principal place of business in Massachusetts. Moreover, Glycosyn has previously availed itself of this forum based on the lawsuit it previously filed against Jennewein, styled *Glycosyn LLC v. Jennewein Biotechnologies GmbH*, Case No. 1:18-cv-10423-PBS, which Glycosyn recently voluntarily dismissed (after it was pending for more than 4 years). In that action, Glycosyn alleged infringement by Jennewein of U.S. Patent No. 9,453,230 (the "'230 Patent"), the great grandparent of the '018 Patent, based on Jennewein's manufacture and sale of 2'-FL, the same accused product as in the Waco Action.

BACKGROUND – THE '018 PATENT

12. The '018 Patent bears the title "BIOSYNTHESIS OF HUMAN MILK OLIGOSACCHARIDES IN ENGINEERED BACTERIA" (Ex. A ('018 patent)) and states that it issued on May 15, 2018.

13. The '018 Patent identifies Massimo Merighi, John M. McCoy, and Matthew Ian Heidtman as the inventors and lists Glycosyn LLC as the assignee.

14. The '018 Patent issued with 28 claims. Claim 1 is the only independent claim, all other claims are dependent on claim 1.

BACKGROUND – GLYCOSYN'S DISTRICT COURT ACTION

15. On March 5, 2018, Glycosyn filed a complaint with the U.S. District Court for the District of Massachusetts against Jennewein, alleging infringement of U.S. Patent No. 9,453,230.

Glycosyn LLC v. Jennewein Biotechnologies GmbH, Case No. 1:18-cv-10423-PBS (the "District Court Action").

16. On June 26, 2018, the District Court Action was stayed pursuant to 28 U.S.C. § 1659(a) pending a final determination in the ITC Investigation, described below. The ITC determination became final after the Federal Circuit issued its decision on September 17, 2021. Instead of moving to lift the stay, nearly 9 months later, on June 14, 2022, Glycosyn filed a Notice of Voluntary Dismissal of the District Court Action on June 14, 2022, the same day it filed suit against Chr. Hansen HMO's customer, Abbott, in the Western District of Texas, Waco Division.

BACKGROUND – THE ITC INVESTIGATION

17. On April 2, 2018, Glycosyn filed a Complaint with the U.S. International Trade Commission under Section 337, 19 U.S.C. 1337, alleging infringement by Jennewein of the '230 Patent, the great grandparent of the '018 Patent. The Complaint was amended on May 16, 2018, to include the '018 Patent. The Commission instituted an investigation, *Certain Human Milk Oligosaccharides and Methods of Producing the Same*, Inv. No. 337-TA-1120 (the "ITC Investigation"), by publication of a notice in the Federal Register on June 21, 2018. During the course of the investigation, Glycosyn withdrew its claims of infringement of the '230 Patent.

18. On September 9, 2019, the ALJ issued an Initial Determination finding a violation by Jennewein of Section 337 based on two strains used in its production process, the #1540 strain and the #1540 derivative strain (also referred to as the #2410 strain). As used herein, the "#1540 strain" refers to both of these strains. The ALJ declined to adjudicate 2'-FL made by Jennewein using an alternative strain, the TTFL12 strain.

19. After review, on May 19, 2020, the Commission affirmed the finding of infringement as to the #1540 strain but reversed the ALJ's determination as to the TTFL12

strain, finding that the ALJ erred by not considering the strain and ruling that 2'-FL made using the TTFL12 strain was non-infringing based on the fact that it lacked a "an exogenous functional β-galactosidase gene comprising a detectable level of β-galactosidase activity," either literally or under the doctrine of equivalents, as required by claim 1 of the '018 Patent. The Commission found that "[u]nlike the accused #1540 strain and its derivative, there is no evidence that a *lacZ*Ω fragment was inserted into the TTFL12 strain or any of its precursors." Comm'n Op. at 18. An exclusion order issued by the Commission on May 19, 2020 specifically exempted 2'-FL made using the TTFL12 strain.

20. On September 17, 2021, the Federal Circuit affirmed the ITC's determination of infringement under the doctrine of equivalents as to the #1540 strain. It is well-established, however, that ITC determinations of infringement and validity do not have preclusive effect. *Texas Instruments Inc. v. Cypress Semiconductor Corp.*, 90 F.3d 1558, 1569 (Fed. Cir. 1996); *see also Tandon Corp. v. U.S. Int'l Trade Comm'n*, 831 F.2d 1017, 1019 (Fed. Cir. 1987) ("[O]ur appellate treatment of decisions of the Commission does not estop fresh consideration by other tribunals."). Glycosyn did not appeal the ITC's finding that 2'-FL made using the TTFL12 strain did not infringe.

BACKGROUND – THE RULE 177 CUSTOMS PROCEEDING

21. On June 3, 2020, Jennewein submitted a letter to the IPR Branch of U.S. Customs and Border Protection ("Customs"), requesting a ruling pursuant to 19 C.F.R. part 177 that 2'-FL made using the #1242 strain did not infringe the claims of the '018 Patent and therefore was outside the scope of the ITC's exclusion order (the "Rule 177 Request").

22. Glycosyn opposed Jennewein's Rule 177 Request. After written submissions and a hearing in which both parties participated, Customs issued a ruling on August 19, 2020, finding that Jennewein met its burden of showing that 2'-FL made using the #1242 strain did not infringe

because, like the TTFL12 strain, the #1242 strain has no *lacZ* gene and no *lacZ*Ω gene fragment, and therefore lacked a "an exogenous functional B-galactosidase gene comprising a detectable level of β-galactosidase activity" as required by claim 1 of the '018 Patent.

BACKGROUND – THE WACO ACTION AGAINST ABBOTT

23. On June 14, 2022, the same day as it dismissed its district court case against Jennewein in the District of Massachusetts, Glycosyn filed a complaint for patent infringement against Chr. Hansen HMO's customer, Abbott Laboratories, in the U.S. District Court for the Western District of Texas, Waco Division, *Glycosyn LLC v. Abbott Laboratories*, C.A. No. 6:22-cv-619 (the "Waco Action"). A copy of the complaint in the Waco Action (the "Waco Complaint") is attached as Exhibit B.

24. In the Waco Complaint, Glycosyn alleges that "Abbott has made, used, offered for sale, and/or sold 2'-FL made via Jennewein's infringing processes, in violation of Glycosyn's patent rights and 35 U.S.C. § 271(g)." Waco Compl., ¶16. Glycosyn refers to Jennewein as "Abbott's manufacturing partner." *Id*. at ¶68. It further alleges that "Abbott's complicity with Jennewein has caused Glycosyn extraordinary harm." *Id*. at ¶76. In total, Glycosyn's Waco Complaint references Jennewein fifty times.

25. Chr. Hansen HMO's 2'-FL products do not infringe and have not infringed, either directly or indirectly, literally or under the doctrine of equivalents, any valid claim of the '018 Patent.

26. In view of Glycosyn's allegations that 2'-FL made and sold by Chr. Hansen HMO infringes the '018 Patent, and its patent infringement claims against Chr. Hansen HMO's downstream customer, Abbott, based on purchase of that 2'-FL product, a substantial controversy exists between the parties that is of sufficient immediacy and reality to warrant declaratory relief.

FIRST CAUSE OF ACTION – DECLARATORY JUDGMENT OF INVALIDITY

27. Chr. Hansen HMO incorporates by reference all preceding paragraphs as if fully set forth herein.

28. As set forth below, Chr. Hansen HMO seeks a declaration that the claims of the '018 Patent are invalid based on obviousness under 35 U.S.C. § 103 (or anticipation under 35 U.S.C. § 102), and based on indefiniteness, and lack of enablement and written description under 35 U.S.C. § 112, and the requirements of 35 U.S.C. § 101, or any other judicially-created bases for invalidity or unenforceability.

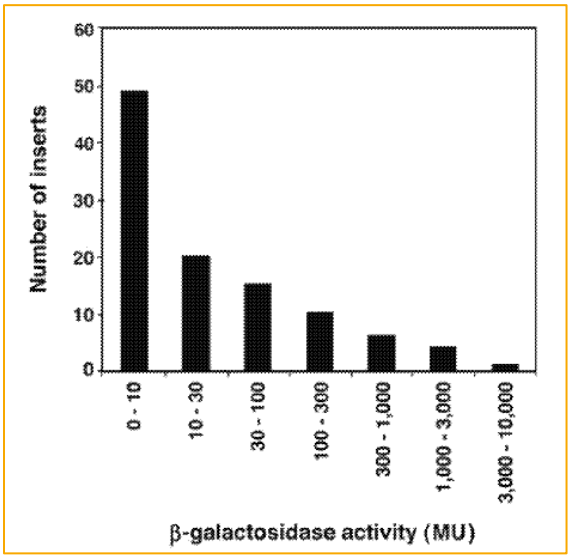
A. Invalidity Based on Prior Art

29. U.S. Patent No. 7,521,212 ("Samain") issued on April 21, 2009, from an application filed on May 24, 2002. Through a Patent Cooperation Treaty application and a French application, Samain claims priority to July 7, 1999. Thus, Samain is prior art to the '018 patent under at least pre-AIA 35 U.S.C. § 102(a), (b), and (e).

30. Samain is titled "Method for Producing Oligopolysaccharides." It details Dr. Samain and his team's efforts to develop a process for making 2'-FL using *E. coli* bacteria dating back to 1999. Specifically, the Samain team used genetic engineering to harness enzymes in bacteria to make 2'-FL, resulting in a less resource-intensive process than the traditional methods. Accordingly, Samain is unquestionably in the same field of endeavor (production of oligosaccharides) as and is analogous art to the '018 patent.

31. Kawano, et al., "Detection of low-level promoter activity within open reading frame sequences of *Escherichia coli*," Nucleic Acids Research, 2005, 33:19 ("Kawano") published in 2005. Thus, Kawano is prior art to the '018 patent under at least 35 U.S.C. § 102(a), (b), and (e).

32. Kawano teaches inserting a β-galactosidase gene into a gene construct in *E. coli*, producing low-level β-galactosidase activity, including 130-220 Miller Units. Kawano also teaches obtaining engineered *E. coli* strains showing low-level β-galactosidase activity when a wild-type *E. coli* β-galactosidase gene (*lacZ*) is located near specific promoter-like sequences, such as in the *E. coli* chromosome. Specifically, Kawano in Figure 1 shows β-galactosidase activity of about 0 to about 10,000 Miller Units, with the largest number of inserts producing below 10 Miller units:



33. Like Samain, Kawano is also directed to the same field of endeavor as the '018 patent. Moreover, Kawano is reasonably pertinent because it would have logically lent itself to solving the problem faced by the inventors of the '018 patent, for example, balancing the need for β-galactosidase activity low enough so as not to destroy the lactose feedstock used to make 2'-FL, but high enough to consume the residual lactose after 2'-FL production. For instance, Kawano set out to find additional promoters for the *lacZ* gene in *E. coli* producing Miller units within the ranges claimed by the '018 Patent. Thus, Kawano is analogous art.

34. The pertinent prior art also includes Drouillard, *et al.*, "Large Scale Synthesis of H-Antigen oligosaccharides by Expressing *Helicobater pylori* α1,2-Fucosyltranferase in Metabolically Engineered *Escherichia coli* Cells," *Angew. Chem.* 2006, 118, 1810-12 ("Drouillard"), Geisser, *et al.*, "Separation of lactose from human milk oligosaccharides with simulated moving bed chromatography," *Journal of Chromatography A*, 1092 (2005), 17-23 ("Geisser"), and World Intellectual Property Organization Publication No. WO 2010/115935 ("Dekany"). Drouillard, Geisser, and Dekany all demonstrate the known difficulty of purifying 2'-FL away from lactose. Lactose, being a feedstock, would necessarily be present in significant amounts in the fermentation medium, and would be difficult and impractical to separate from 2'-FL, particularly at a large scale, due to these known difficulties in separating 2'-FL from lactose.

35. Each of Drouillard, Geisser, and Dekany is prior art to the '018 patent. Drouillard published in 2006 in Volume 118 of the *Angewandte Chemie* (German for "Applied Chemistry") journal. Geisser published in 2005 in the *Journal of Chromatography A*. Both of these references are prior art to the '018 Patent under at least 35 U.S.C. § 102(a) and (b).

36. Likewise, Dekany published on October 11, 2010, as a World Intellectual Property Organization Publication. Dekany was filed on April 7, 2010, and claims priority to a Danish application filed on April 7, 2009. Thus, Dekany is prior art to the '018 patent under at least 35 U.S.C. § 102(a).

37. Like Samain and Kawano, each of Drouillard, Geisser, and Dekany is within the same field of endeavor as the '018 patent. Moreover, these three references are reasonably pertinent because they each would have logically lent themselves to solving the problem faced by the inventors of the '018 patent, for example, balancing the need for β-galactosidase activity low enough so as not to destroy the lactose feedstock but high enough to consume the residual

lactose after 2'-FL production. Accordingly, each of Drouillard, Geisser, and Dekany is analogous art.

38. These prior art publications show that the difficulty of purifying fucosylated oligosaccharides like 2'-FL, particularly from lactose, was known. According to Drouillard, for example, "Human milk is unique in containing large quantities of fucosylated oligosaccharides, and these molecules cannot readily be purified from alternative natural sources." Geisser similarly explains that "[t]he predominance of lactose in the carbohydrate fraction of milk makes it difficult to separate and analyze the complex oligosaccharides," including 2'-FL.

39. Dekany states that "*2'-O*-fucosyllactose [*i.e.* 2'-FL] has been synthesised by both chemical and enzymatic methodologies but commercially attractive production processes have not been developed due to lack of efficient purification and synthetic approaches." These publications are therefore consistent with the knowledge in the art that purification of HMOs like 2'-FL from lactose was difficult at the time of the invention, particularly so at a commercial scale.

40. Because all of the claim elements were known, and would have been combined by a person having ordinary skill in the art in known ways without undue experimentation, the combination of Samain, Kawano, and any of Drouillard, Geisser, and Dekany render the claims of the '018 patent obvious. It would have been obvious to use a low level of β-galactosidase activity in Samain's *E. coli* strain, less than wild-type activity, to solve the known problem of purifying oligosaccharides from lactose, as taught by Drouillard, Geisser or Dekany, but keeping the level low enough to avoid preventing 2'-FL production as taught by Samain. β-galactosidase was known to destroy lactose. In other words, it would be obvious to one of skill in the art that by reducing the amount of β-galactosidase made, they would get proportionally more 2'-FL.

Thus, in view of the teachings in Drouillard, Geisser, and Dekany, a person of ordinary skill in the art would have been motivated to incorporate a low level of β-galactosidase activity into Samain's fucosylated oligosaccharide-producing strains.

41. For example, a person of ordinary skill in the art would have genetically engineered the *E. coli* bacterium in Samain to express a low level of β-galactosidase activity with a reasonable expectation of success from the teachings of Kawano. As discussed above, Kawano teaches how to engineer *E. coli* to have a level of β-galactosidase activity falling within the claimed range of "between 0.05 and 200 units."

42. Indeed, during prosecution of one of the '018 Patent's great grandparent (the '230 Patent), Glycosyn admitted that a person of ordinary skill could engineer a low level of β-galactosidase activity. A low level of β-galactosidase activity, including in the broadly claimed range of 0.05 to 200 units, would have been desirable to avoid limiting 2'-FL production while still allowing excess lactose to be eliminated. A low level of activity would also have been expected to allow some amount of 2'-FL to be produced. Moreover, there is no evidence that it was difficult for the inventors, who wanted to avoid the Samain patent, to come up with a low level of β-galactosidase activity as a solution to this known issue.

43. The inventors of the '018 Patent deleted the native *E. coli* β-galactosidase gene, *lacZ* (as Samain did), then inserted an entire copy of the open reading frame of *lacZ* (*i.e.* encoding the LacZ enzyme), which had no promoter, into a different gene (*lon*) in a way that purportedly yielded reduced expression of the β-galactosidase enzyme. The inserted gene purportedly harnessed a weak promoter near the *lon* gene into which it was inserted to drive a low-level expression of β-galactosidase. The '018 Patent claims that the resultant low level of β-

galactosidase activity does not eliminate the intracellular lactose pool, which allows production of 2'-FL. '018 Patent at 4:65-5:18, 18:23-25.

44. Based on Kawano's teachings, one of ordinary skill in the art would have been able and motivated to insert a functional *lacZ* gene into an endogenous gene such that the resultant bacterium comprises a low level of β-galactosidase activity, wherein the β-galactosidase activity is between 0.05 and 200 Miller units.

45. The combination of Samain, Kawano, and any one of Drouillard, Geisser, and Dekany teaches the limitation "(ii) an exogenous functional β-galactosidase gene comprising a detectable level of β-galactosidase activity that is reduced compared to that of a wild-type *E. coli* bacterium, wherein the level of β-galactosidase activity comprises between 0.05 and 200 units." For example, it would have been obvious to one of ordinary skill in the art to modify Samain to insert, into an endogenous gene, an exogenous wild-type β-galactosidase gene having low activity (between 0.05 and 200 Miller Units). Samain itself teaches that the "genetically modified cells" of the claims are bacteria whose genome has been altered by the introduction of one or more phenotype-producing genes.

46. As Dr. Prather (Glycosyn's retained technical expert in the ITC proceeding) admitted, it was known how to insert genes into bacterial gene constructs, including inserting functional *lacZ* genes that produced β-galactosidase activity between 0.05 and 200 units. For example, Kawano teaches inserting a β-galactosidase gene into a gene construct in *E. coli*, producing low-level β-galactosidase activity, including 130-220 Miller units.

47. Even though Samain taught $LacZ^-$ strains, one of skill in the art in 2011 was well-aware that Samain's strains in fact possessed Miller units of β-galactosidase activity yet still produced fucosylated oligosaccharides. For example, Giacomini at page 88 shows 23 Miller

units for strain JM 109, the same host strain used in Samain. Giacomini, *et al*. *FEMS Microbiol.. Letters*, 100 (1992) 87-90.

48. One of skill in the art would have arrived at the lowest possible β-galactosidase level to minimize destroying the lactose substrate. Furthermore, one of ordinary skill would have known they could avoid the explicit disclosure of the Samain patent simply by having a miniscule, non-zero amount of β-galactosidase activity, such as the lower 0.05 Miller unit limit recited by the '018 Patent's claims.

49. Kawano teaches obtaining engineered *E. coli* strains showing low-level β-galactosidase activity when a wild-type *E. coli* β-galactosidase gene (*lacZ*) is located near promoter-like sequences, such as in the *E. coli* chromosome. Kawano accomplished this by placing short chromosomal fragments randomly into an upstream region of a *lacZ* gene.

50. One of ordinary skill in the art would have inserted a functional *lacZ* gene into an endogenous gene such that the resultant bacterium comprises a low level of β-galactosidase activity, wherein said β-galactosidase activity comprises between 0.05 and 200 Miller units based on the teachings of Kawano. One of ordinary skill in the art would have been motivated to modify Samain based on Kawano to insert a β-galactosidase gene into an endogenous gene in the *E. coli* genome with Kawano's promoter-like sequences to obtain low-level β-galactosidase activity between 0.05 and 200 Miller units. For example, the ordinarily skilled artisan would have been motivated to have made this combination to address the known problem of purification of fucosylated oligosaccharides like 2'-FL from lactose while not destroying too much lactose substrate during production.

51. Further, a person of ordinary skill in the art would have been motivated to modify Samain with a reasonable expectation of success by introducing a low-level of β-galactosidase

activity according to Kawano to aid in the purification of the fucosylated oligosaccharides produced by eliminating other oligosaccharides present, like lactose.

52. It was further known to measure units of β-galactosidase activity according to the Miller assay.

53. Alternatively, the JM109 strain in Samain already possessed a low-level of β-galactosidase activity, and Samain did not alter this property in producing fucosylated oligosaccharides. A strain of JM109 (DE3) was later confirmed to be able to produce 2'-FL despite having detectable β-galactosidase activity. Thus, if, as Glycosyn has erroneously asserted, the measured Miller units do not have to result from β-galactosidase activity found in the exogenous functional β-galactosidase gene, but could be attributable to some other source, then Samain would anticipate claim 1 of the '018 Patent as well as several of the dependent claims of the patent.

54. Accordingly, it would have been obvious to one of ordinary skill in the art to select a low level of β-galactosidase activity to avoid destroying the lactose feedstock necessary for an engineered strain to produce a lactose-containing oligosaccharide, such as a fucosylated lactose.

55. The limitations of the dependent claims of the '018 Patent are likewise disclosed in the prior art, and these claims would have been obvious at the time of the alleged invention of the '018 Patent in view of the prior art and knowledge of one of skill in the art. For example, claim 2 of the '018 patent recites the following: "The method of claim 1, wherein said colanic acid synthesis gene comprises an *E. coli wcaJ*, *wzxC*, *wcaD*, *wza*, *wzb*, or *wzc* gene." As admitted by Glycosyn's expert in the ITC Investigation, Dr. Prather, Samain teaches the additional limitation of claim 2.

56. Claim 3 recites the following: "The method of claim 2, wherein said colanic acid synthesis gene comprises a *wcaJ* gene." As admitted by Dr. Prather, Samain teaches the additional limitation of this claim. Specifically, Samain discloses an inactivating mutation in a colanic acid synthesis gene.

57. Claim 4 recites "The method of claim 1, wherein the bacterium comprises an increased intracellular guanosine diphosphate (GDP)-fucose level, wherein the increased intracellular GDP-fucose level is at least 10% more than the level of GDP-fucose in a wild-type bacterium." It would have been obvious to increase the GDP-fucose level in a bacterium producing 2'-FL at time of the alleged invention of the '018 Patent in view of the prior art and knowledge of one of skill in the art. For example, GDP-fucose was a known substrate of the enzyme that creates 2'-FL, and thus it would have been obvious to one of ordinary skill in the art to increase its level in any 2'-FL-producing bacterium. Furthermore, it was known that an inactivating mutation in a colanic acid synthesis gene would have increased GDP-fucose levels over that in a wild-type bacterium.

58. Claim 5 of the '018 patent recites the following: "The method of claim 1, wherein said exogenous lactose-accepting fucosyltransferase gene encodes α(1,2) fucosyltransferase and/or α(1,3) fucosyltransferase." Dr. Prather admitted that Samain teaches the added limitation of this claim as well. For example, Samain discloses an exogenous lactose accepting fucosyltransferase gene comprising an α(1,2)-fucosyltransferase gene, an α(1,3) fucosyltransferase gene, or an α(1,4)-fucosyltransferase gene.

59. Claim 6 recites "The method of claim 5, wherein said α(1,2)-fucosyltransferase gene comprises a *Bacteroides fragilis wcfW* gene." This was obvious to use at time of the alleged invention of the '018 Patent in view of the prior art and knowledge of one of skill in the

art. For example, it was known in the art that the *Bacteroides fragilis wcfW* gene was an α(1,2)-fucosyltransferase gene that produces 2'-FL.

60. Claim 7 is limited to making 3-FL and is thus irrelevant to producing 2'-FL. That said, the production of 3-FL was known to those of skill in the art at the time of the alleged invention of the '018 Patent.

61. Claim 8 of the '018 patent recites the following: "The method of claim 1, wherein said exogenous functional β-galactosidase gene comprises an *E. coli lacZ* gene." The prior art renders this claim obvious. It was known that *E. coli lacZ* was a functional β-galactosidase gene, and it was also known how to insert the *lacZ* gene into a gene construct. It was therefore obvious to use an *exogenous E. coli lacZ* gene to produce β-galactosidase activity. It was also obvious to one of ordinary skill in the art to use an exogenous *lacZ* gene to express β-galactosidase having β-galactosidase activity in an *E. coli* strain engineered to produce a fucosylated oligosaccharide (such as JM109, as taught by Samain). The genotype of strain JM109 is ΔlacZ. *lacZ* is the name of the wild-type *E. coli* β-galactosidase gene.

62. Claim 9 involves insertion of a lacZ gene into an endogenous *lon* gene in the 2'-FL producing bacterium. Chr. Hansen's 2'-FL producing bacterium does not contain a *lon* gene. Chr. Hansen HMO's 2'-FL producing bacteria are descendants of the *E. coli* B strain. The *E. coli* B strain does not bear a *lon* gene, and neither do Chr. Hansen's 2'-FL production strains. Hence, Chr. Hansen HMO's 2'-FL producing bacterium does not have a *lacZ* gene or a *lacZ* gene fragment such as *lacZα* or *lacZΩ* inserted into a *lon* gene. Thus this claim is irrelevant here.

63. Claim 10 of the '018 Patent recites the following: "The method of claim 1, wherein said bacterium further comprises a functional lactose permease gene." As Dr. Prather

admitted, Samain teaches the additional limitation of this claim. In particular, Samain discloses a functional lactose permease gene, *E. coli lacY*.

64. Claim 11 recites "The method of claim 10, wherein said lactose permease gene is an endogenous lactose permease gene." It was obvious to use the endogenous lactose permease gene of the bacterium according to its known function at time of the alleged invention of the '018 Patent in view of the prior art and knowledge of one of skill in the art.

65. Claim 12 of the '018 Patent states: "The method of claim 10, wherein said lactose permease gene comprises an *E. coli lacY* gene." As Dr. Prather admitted, Samain teaches the additional limitation of claim 12. Specifically, Samain discloses a functional lactose permease gene, *E. coli lacY*.

66. Claim 13 recites "The method of claim 1, wherein said bacterium further comprises an exogenous *E. coli rcsA* or *E. coli rcsB* gene." It was obvious to use an exogenous *E. coli rcsA* or *E. coli rcsB* gene in a 2'-FL producing bacterium at time of the alleged invention of the '018 Patent in view of the prior art and knowledge of one of skill in the art. The *E. coli rcsA* and *E. coli rcsB* gene were known prior to the invention to be positive regulators of the bacterial polysaccharide and colanic acid synthesis pathway, and particularly for increasing production of the 2'-FL feedstock, GDP-fucose, as taught by Samain *e.g.* in Examples 1 and 8; Dumon, C. et al (2001) *Glycoconjugate J.* 18: 465-474, and Drouillard et al; where it was shown that overexpression of rcsA has beneficial effect on fucosylated lactose biosynthesis. It was thus obvious to one of ordinary skill in the art to use exogenous *rcsA* and *rcsB* genes according to their known functions to increase GDP-fucose and thus increase production of 2'-FL in a bacterium at the time of the invention.

67. Claim 14 recites "The method of claim 1, wherein said bacterium further comprises an inactivating mutation in a *lacA* gene." LacA was known at the time of the invention as a lactose transacetylase, which acetylates lactose, thereby reducing the amount of lactose available for 2'-FL biosynthesis in the cell. It was obvious to one of ordinary skill in the art at the time of the invention to inactivate the *lacA* gene to increase the intracellular lactose pool and increase 2'-FL production in the 2'-FL producing bacterium.

68. Claim 15 recites "The method of claim 1, wherein said bacterium further comprises an exogenous sialyltransferase gene" and is therefore irrelevant to Chr. Hansen's methods for making 2'-FL as they do not employ a sialyltransferase.

69. Claim 16 recites "The method of claim 15, wherein said exogenous sialyltransferase gene encodes an α(2.3)sialyl transferase" and is therefore irrelevant to Chr. Hansen's methods for making 2'-FL as they do not employ a sialyltransferase.

70. Claim 17 recites "The method of claim 1, wherein said bacterium further comprises a deficient sialic acid catabolic pathway comprising a null mutation in an endogenous N-acetylneuraminate lyase gene or a null mutation in an endogenous N-acetylmannosamine kinase gene." This pathway is irrelevant to making 2'-FL.

71. Claim 18 of the '018 Patent recites the following: "The method of claim 1, wherein the level of β-galactosidase activity comprises between 0.05 and 5 units." The prior art renders this claim obvious. For example, it would have been obvious to one of skill in the art to select as low a level of β-galactosidase activity as possible to avoid destroying the lactose feedstock necessary for an engineered *E. coli* to make a fucosylated lactose oligosaccharide, as taught by the prior art including Samain.

72. Claim 19 is directed to an inactivating mutation in a *lon* gene in the 2'-FL producing bacterium. The *lon* gene is not present in Chr. Hansen's 2'-FL producing bacterium, because Chr. Hansen's 2'-FL producing bacteria are descendants of the *E. coli* B strain. The *E. coli* B strain does not bear a *lon* gene, and neither do Chr. Hansen's 2'-FL production strains. Thus this claim is irrelevant here.

73. Claim 20 recites "The method of claim 1, wherein said bacterium comprises an increased intracellular lactose level, wherein the increased intracellular lactose level is at least 10% more than the level in a wild-type bacterium." It was obvious to create a bacterium for producing 2'-FL having at least 10% more intracellular lactose than the level in a wild-type bacterium at time of the alleged invention of the '018 Patent in view of the prior art and knowledge of one of skill in the art. For example, it was obvious that, to produce 2'-FL in a bacterium, it was advantageous to have an increased intracellular lactose level versus the wild-type bacterium, ideally by 10% or more. Furthermore, it was obvious at the time of the invention that the claimed genetic mutations, inactivations, and deletions in the bacterium recited in claim 1 would have increased the amount of intracellular lactose by 10% or more.

74. Claim 21 recites, "The method of claim 1, wherein said exogenous functional β-galactosidase gene is an *E. coli* lacZ gene lacking an operably linked promoter, and said colanic acid synthesis gene comprises an *E. coli wcaJ*, *wzxC*, *wcaD*, *wza*, *wzb*, or *wzc* gene." As admitted by Glycosyn's expert in the ITC Investigation, Dr. Prather, the prior art including Samain teaches limitations of claim 21. The prior art also renders this claim obvious. It was known that *E. coli lacZ* was a functional β-galactosidase gene, and it was also known how to insert the *lacZ* gene into a bacterium. *lacZ* is the name of the wild-type *E. coli* β-galactosidase gene. It was therefore obvious at the time of the invention to use an exogenous *E. coli lacZ* gene

to produce β-galactosidase activity in a bacterium. It was also obvious to one of ordinary skill in the art to use an exogenous *lacZ* gene to express β-galactosidase producing β-galactosidase activity in an *E. coli* strain engineered to produce a fucosylated oligosaccharide, as taught by Samain and Giacomini. It was also known that gene expression uses a promoter operably linked to the gene, and a *lacZ* gene without a promoter would yield reduced β-galactosidase activity. One of ordinary skill in the art at the time of the invention knew, e.g. from Samain, Kawano, and Giacomini, that eliminating or reducing lacZ gene expression, by gene or promoter elimination, would reduce β-galactosidase activity. Furthermore, Kawano teaches obtaining engineered *E. coli* strains with low-level β-galactosidase activity when a wild-type *E. coli* β-galactosidase gene (*lacZ*) is located near non-optimal, promoter-like sequences. One of ordinary skill in the art at the time of the invention would thus have found claim 21 obvious.

75. Claim 22 is directed to a bacterium with a genotype of a) ampC::(PtrpBλcI+), PlacI q (ΔlacI-lacZ)lacY+, ΔwcaJ, thyA::Tn10, Δlon::(kan, lacZ+); or (b) ampC::(PtrpBλcI+), PlacI q (ΔlacI-lacZ)lacY+, ΔwcaJ, thyA::Tn10, Δlon::(kan, lacZ+), ΔlacA; which are not relevant to Chr. Hansen HMO's production of 2'-FL.

76. Claim 23 of the '018 Patent recites the following: "The method of claim 1, wherein said exogenous functional β-galactosidase gene is inserted into an endogenous gene." The prior art renders this claim obvious. It was obvious to one of ordinary skill in the art to insert an exogenous functional β-galactosidase gene into an endogenous gene to express a β-galactosidase enzyme having β-galactosidase activity in an *E. coli* strain that was engineered to produce a fucosylated oligosaccharide. Methods of inserting genes, including *lacZ* genes into bacterial gene constructs and endogenous genes, were known. *E. coli* strain JM109 (discussed in Samain) also was measured to have units of β-galactosidase activity from 23 to 356 Miller Units.

77. Moreover, it would have been obvious to one of ordinary skill in the art to select expression of a β-galactosidase enzyme with low-level β-galactosidase activity in order to avoid destroying the lactose feedstock necessary for an engineered strain, such as ones using JM109 as a host, to produce a lactose-containing oligosaccharide, such as a fucosylated lactose. It would also have been obvious to select a range of β-galactosidase activity substantially less than the activity of wild-type cells (1000 Miller Units) for this purpose. It was also widely known how to measure units of β-galactosidase activity according to the Miller assay. Methods for obtaining low-level expression of genes, including enzymes, were also known, for example as shown in Kawano.

78. Claim 24 recites the following: "The method of claim 1, wherein said exogenous functional β-galactosidase gene comprises a recombinant β-galactosidase gene engineered to produce a detectable level of β-galactosidase activity that is reduced compared to the level of β-galactosidase activity in a wild-type *E. coli* bacterium." In this context, *recombinant* means genetically engineered DNA prepared by transplanting or splicing genes from one species into the cells of a host organism of a different species.

79. The prior art renders claim 24 obvious. For example, it would have been obvious to one of ordinary skill in the art to use a recombinant exogenous functional β-galactosidase gene to express in *E. coli* a β-galactosidase enzyme having reduced β-galactosidase activity, as discussed for claim 21. This would have been the case at least because Kawano describes use of a recombinant exogenous functional β-galactosidase gene to express in *E. coli* a β-galactosidase enzyme having reduced β-galactosidase activity.

80. Claim 25 recites the following: "The method of claim 24, wherein the level of β-galactosidase activity comprises between 0.05 and 5 units." The prior art renders claim 25

obvious. For example, it would have been obvious to one of ordinary skill in the art to select as low a level of β-galactosidase activity as possible to avoid destroying the lactose feedstock necessary for an engineered *E. coli* to make a fucosylated lactose oligosaccharide. As explained previously, this low level would have included activity as low as 0.05 to 5 units.

81. Claim 26 states: "The method of claim 1, wherein the level of β-galactosidase activity comprises between 0.05 and 4 units." The prior art renders claim 26 obvious. For example, it would have been obvious to one of ordinary skill in the art to select as low a level of β-galactosidase activity as possible to avoid destroying the lactose feedstock necessary for an engineered *E. coli* to make a fucosylated lactose oligosaccharide. As explained previously, this low level would have included activity as low as 0.05 to 4 units.

82. Claim 27 recites: "The method of claim 1, wherein the level of β-galactosidase activity comprises between 0.05 and 3 units." The prior art renders claim 27 obvious. For example, it would have been obvious to one of ordinary skill in the art to select as low a level of β-galactosidase activity as possible to avoid destroying the lactose feedstock necessary for an engineered *E. coli* to make a fucosylated lactose oligosaccharide. As explained previously, this low level would have included activity as low as 0.05 to 3 units.

83. Claim 28 recites the following: "The method of claim 1, wherein the level of β-galactosidase activity comprises between 0.05 and 2 units." The prior art renders claim 28 obvious as well. For example, it would have been obvious to one of ordinary skill in the art to select as low a level of β-galactosidase activity as possible to avoid destroying the lactose feedstock necessary for an engineered *E. coli* to make a fucosylated lactose oligosaccharide. As explained previously, this low level would have included activity as low as 0.05 to 2 units.

84. Accordingly, the claims of the '018 Patent that are potentially relevant to Chr. Hansen HMO's production of 2'-FL are invalid as obvious. The prior art recited above is illustrative and not limiting. Further, there are no secondary considerations that outweigh the strong showing of obviousness here.

B. Invalidity Under 35 U.S.C. §§ 101 and 112

85. The claims of the '018 Patent are also invalid based on indefiniteness, and lack of enablement and written description, 35 U.S.C. § 112, and failure to meet the requirements of 35 U.S.C. § 101.

1. Indefiniteness

86. The claims of the '018 Patent are invalid because the claim term "β galactosidase activity comprises between 0.05 and [200/5/4/3/2] units" is indefinite under 35 U.S.C. § 112, ¶ 2. "β galactosidase activity comprises between 0.05 and [200/5/4/3/2] units" is indefinite for at least three reasons: 1) recitation of a specific range in which β-galactosidase activity must fall but simultaneously failing to limit that range by describing it with the term "comprises;" 2) failing to define at what point the β-galactosidase activity must be present during the production of 2'-fucosyllactose ("2'-FL"); and 3) defining "units" of activity by reference to a publication by J.H. Miller[1], "Experiment 48 Assay of β-Galactosidase," Experiments in Molecular Genetics, Cold Spring Harbor, NY (1972) 352-55 ("Miller"), where the measurements of units can yield different results. Indeed, the Miller assay as conceived by Miller was a general assay to teach students how to run an enzymatic assay, not one that aimed for sensitivity and accuracy/reliability of the results.

87. First, the phrase "β-galactosidase activity comprises between 0.05 and 200 units" defines an explicit range using the open-ended term *comprises*, thus giving no reasonable certainty as to whether or not the claimed activity must fall within this range. Defining a specific

range normally means just that – the value must fall within the limited range defined. The claim language presents an inescapable conflict that prevents a person of ordinary skill in the art from understanding the scope of the claim. This cannot be considered a mere drafting error; the specification of the patent goes so far as to expressly define the term "comprising": "[t]he transitional term 'comprising,' which is synonymous with 'including,' 'containing,' or 'characterized by,' is inclusive or open-ended and does not exclude additional, unrecited elements or method steps." '018 Patent at 11:49-52. As this phrase appears in the sole independent claim of the '018 Patent, and dependent claims reciting narrower ranges use "comprises" identically, all of the patent's claims are invalid under 35 U.S.C. § 112 for being indefinite.

88. Even if one of ordinary skill in the art could perform the assay in Miller to calculate units of β-galactosidase activity, the term is still indefinite. Miller does nothing to address the fact that, by using *comprises*, the claim encompasses any amount of activity, rendering this term meaningless. Even when following Miller's assay procedures, persons of ordinary skill in the art cannot know whether an activity outside of 0.05 to 200 Miller units infringes these claims because, by using the term *comprises*, the claim does not exclude values outside of the range. Additionally, the variability inherent in the Miller assay (explained below) greatly compounds this lack of clarity. In short, the phrase "β-Galactosidase comprises between 0.05 and [200/5/4/3/2] units" has no reasonably-certain bounds and is therefore indefinite.

89. Second, the claim term "β-galactosidase activity comprises between 0.05 and [200/5/4/3/2] units" is indefinite because the term "unit" as derived from Miller is fatally ambiguous due to its inherent variability. Different applications of the test set forth in Miller can render radically different results, and the '018 Patent never elaborates on Miller or resolves any

of these inconsistencies. In fact, Miller itself describes multiple competing methods that may lead to inconsistent results, rendering the Miller definition of "units" indefinite. Even more problematic is that regardless of what Miller methodology is used, the assay yields wildly variable, unpredictable results that may or may not provide units inside or outside of the claimed range as described, for example, in Giacomini, et al., "Experimental conditions may affect reproducibility of the β-galactosidase assay," *FEMS Microbiology Letters*, 100 (1992) 87-90 ("Giacomini").

90. Giacomini shows that following the methods taught in Miller, wildly different units of β-galactosidase activity were obtained. For example, in Table 1, the authors show quite different Miller units for a lacZ$^-$ strain when β-galactosidase activity tested using one of two alternative assay procedures explicitly authorized by Miller to correct for cellular debris in the samples: a correction factor of 1.75xOD_{550}, or centrifugation. The JM109 strain showed 23 or 356 units using one or the other, while the DH1 strain showed 1878 or 2580 units. Giacomini at Table 1, p. 88.

91. Giacomini also shows Miller units differing about 20% when using the two permeabilization methods taught in Miller (toluene vs. SDS-chloroform), and that even values below zero were possible. Giacomini at Tables 1, 2, pp. 88-89. Extending the reaction time of the Miller assay further halved the measured units of activity. *Id.* at Table 4, p. 89. Giacomini concludes at page 90 that the Miller assay "is not particularly reproducible, at least in *E. coli*." The authors further note that "it appears to be essential to standardize the different parameters," and that "data appearing in the literature cannot be accurately compared unless the exact experimental conditions are stated." *Id.* Therefore, the Miller assay required by every claim will

provide fundamentally different values depending on the conditions used. The ’018 Patent nowhere explains how to resolve this known ambiguity.

92. Other factors, including differing amounts of SDS-chloroform, culture volume and stage, and ONPG concentration also dramatically altered the measured units. *Id.* at Tables 3, 5-7, p. 89-90. The procedure of the Miller assay uses one *drop* of SDS and two *drops* of chloroform as cell permeabilizing agents. The ambiguous unit of a “drop” can cause variations in results, especially if the assay is done by different individuals, in laboratories, or using different measuring tools—despite the permeability of cell being a factor that greatly influences β-galactosidase activity. As shown above, the variability in measured units when following explicitly allowed methods in Miller can simultaneously provide infringing and non-infringing results. One of ordinary skill in the art therefore could not determine the boundaries of the claims with reasonable certainty.

93. Furthermore, natural and man-made variations in the culturing step can also alter the data measured and thus give different results for the units calculated, particularly at the lower end of the claimed range, as this is very close to zero or undetectable activity.

94. Additionally, the Miller assay uses an arbitrary number (*e.g.*, A420/min/mL of cells/OD600) as unit, rather than a standard enzyme unit (e.g. EU or U) which is defined as the amount of enzyme capable of catalyzing the reaction of 1 micromole of substrate per minute. Using the standard U, it is possible to compare the enzyme activities from multiple laboratories using different spectrophotometers, cuvettes, and reagents. However, in the case of the Miller unit, it is impossible to reliably and quantitatively compare β-galactosidase activities between different studies.

95. Because the Miller units of β-galactosidase activity cannot reliably be determined using the Miller assay, and both infringing and non-infringing results could be obtained, the '018 Patent's claims are indefinite. Miller unit measurement is imprecise, particularly at the low levels of β-galactosidase activity claimed in the '018 Patent, and therefore the required range of Miller units renders the claims indefinite. The '018 Patent does not clearly teach one of ordinary skill which competing methodology in Miller to use to determine infringement, leaving doubts as to whether a product falling within the claimed range for one of the Miller methodologies but not the other would fall within the claim scope.

96. Third, the asserted claims are also indefinite because they do not state at what point β-galactosidase activity needs to be present to determine infringement. Regarding activity, the '018 Patent purports to teach maintaining β-galactosidase activity to avoid interfering with the cell's construction of 2'-FL, yet high enough to remove lactose after construction of 2'-FL to allow for purification of the 2'-FL away from residual lactose. Yet neither the claims nor the specification make reasonably clear when this β-galactosidase activity should be present and even if it must be present during production of 2'-FL. It is even unclear whether the engineered *E. coli* bacterium can infringe the claims while it is not actually producing 2'-FL. Again, such uncertain claims prevent a person of ordinary skill in the art from understanding the claim scope.

97. For the above reasons, the '018 Patent's claims are invalid under 35 U.S.C. § 112, ¶ 2 for being indefinite.

2. Lack of Enablement, Written Description and Utility

98. The claims of the '018 Patent are also invalid for failing to satisfy the enablement requirement of 35 U.S.C. § 112, ¶ 1.

99. The ’018 Patent’s specification does not enable the full scope of the claims of the ’018 Patent. Specifically, the specification fails to teach how to make 2’-FL for the full range of the claimed β-galactosidase activity.

100. The claims of the ’018 Patent are also invalid for failing to satisfy the written description requirement of 35 U.S.C. § 112, ¶ 1. Glycosyn argues that its claims cover Chr. Hansen HMO’s α-complementation. Alpha-complementation is a genetic engineering technique that uses non-functional fragments of the *E. coli* β-galactosidase gene (*lacZ*) to create controllable expression of an active β-galactosidase enzyme in a genetically engineered cell. The *lacZ* gene fragments are called *lacZα* and *lacZΩ*. When expressed separately, each fragment produces a non-functional portion of the LacZ protein, and neither portion has β-galactosidase activity alone. When *lacZα* and *lacZΩ* are simultaneously expressed in the same cell, their portions of the LacZ protein assemble into one of the four identical subunits of the LacZ β-galactosidase enzyme, which then assemble with three additional subunits into the homotetrameric form of the β-galactosidase enzyme, which then possesses β-galactosidase activity. However, the ’018 Patent’s specification never mentions α-complementation, *lacZα*, or *lacZΩ*. Rather, the patent recites a “functional β-galactosidase gene” or “*lacZ*” only. *See, e.g.*,’018 Patent at 2:62-3, claim 1. Because α-complementation and the *lacZ*α and *lacZ*Ω are nowhere mentioned (or even suggested) in the ’018 Patent, the claims of the ’018 Patent, if they are interpreted broadly (as Glycosyn does) to cover α-complementation, fail to show possession of the full scope of the term “functional … β-galactosidase gene,” and therefore fail to meet the written description requirement of 35 U.S.C. § 112, ¶ 1.

101. The claims of the ’018 Patent are also invalid because they fail to enable the production of 2’-FL under 35 U.S.C. § 112, ¶ 1. In particular, there is no benefit to having a

lower limit of β-galactosidase activity of 0.05 Miller units, which does not produce enough activity to reduce lactose or enable purification of 2'-FL. Furthermore, activity in the upper portion of the cited range (up to 200 Miller units) would effectively prevent the formation of 2'FL by consuming the lactose necessary to produce 2'-FL. Therefore, the claims of the '018 Patent do not enable the production of 2'-FL in the activity ranges specified in the claims and lack a full scope of enablement under 35 U.S.C. § 112, ¶ 1.

102. In addition, the claims of the'018 Patent lack enablement under 35 U.S.C. § 112 because the Miller unit ranges claimed are not enabled. For example, Giacomini concludes at page 90 that the Miller assay "is not particularly reproducible, at least in *E. coli*." The authors further note that "it appears to be essential to standardize the different parameters," and that "data appearing in the literature cannot be accurately compared unless the exact experimental conditions are stated." *Id.* Therefore, the Miller assay required by every claim will provide fundamentally different values depending on the conditions used. The '018 Patent nowhere explains how to resolve this known ambiguity, and has not enabled producing 2'-FL using the claimed bacteria and Miller units.

103. In addition, the '018 patent lacks enablement for failing to teach how the claimed *E. coli* bacterium, or the β-galactosidase gene in it, has to be altered such that the bacterium exhibits a β-galactosidase activity between 0.05 and 200 Miller units. The patent purports to describe how a functional wild-type β-galactosidase gene is inserted into the *lon* locus of an *E. coli* bacterium, and that the resulting *E. coli* strain exhibits between 1 and 2 Miller units. To this end, a functional wild-type, but promoter-less *E. coli lacZ*$^{+}$ gene was inserted into the *lon* locus of an *E. coli* strain which had its endogenous β-galactosidase gene deleted (see Example 1 in combination with Fig. 14). As illustrated in Fig.12, a large portion of the *lon* gene was replaced

by said functional wild-type, but promoter-less *E. coli lacZ*$^{+}$ gene, which was inserted into the *lon* locus in reverse orientation (as compared to the *lon* gene) (see e.g. Fig. 12 in combination with SEQ ID NO:7). However, this genetic alteration does not lead to an *E. coli* bacterium that possesses β-galactosidase activity. First, the *lacZ* gene being inserted into the *lon* locus lacks a promoter, which is an essential element of a gene for it to function. Since the *lacZ* gene has been inserted in reverse orientation into the *lon* locus (Fig. 12 in combination with Fig. 13 and SEQ ID NO: 7), the endogenous promoter of the original *lon* gene cannot provide a substitute promoter and mediate expression of the inserted *lacZ* gene. For these reasons, the *lacZ* gene inserted into the *lon* locus is most likely not expressed at all. The patent discloses no evidence about expression of the protein coding region of the *lacZ* gene within this *lon* locus to show expression, e.g. Northern-blots or Rt-PCR data).

104. Although it is disclosed that the *E. coli* strain bearing the protein-coding region of the *lacZ* gene inserted into its *lon* locus shall exhibit a β-galactosidase activity of 1 to 2 Miller units, the 1 to 2 units described in the patent merely represent background noise that was inadvertently considered and incorrectly assigned as β-galactosidase activity. It was well known that that Miller units can be observed in β-galactosidase deficient *E. coli* strains, i.e. in *E. coli* cells that do not express a β-galactosidase. *See* Giacomini, *et al*., *supra*. Because the application does not contain any information that the promoter-less *lacZ* gene being inserted into the *lon* locus is expressed at all, and the Miller values with 1 to 2 units are in the range of the unspecific background of the Miller assay, one of ordinary skill in the art would not have found that the 1 to 2 Miller units described by the patent as being possessed by the one *E. coli* example represent β-galactosidase activity.

105. No other ways to obtain the claimed *E. coli* strain that contains a functional β-galactosidase gene and exhibits low β-galactosidase activity between 0.05 and 200 Miller units when cultivated in the presence of lactose than example 1 are disclosed in the '018 Patent. Hence, a skilled artisan would find the subject matter of the claims lack enablement.

106. The claims of the '018 Patent are also invalid for failure to meet the requirements of 35 U.S.C. § 101, including but not limited to lack of utility. In particular, there is no benefit to having a lower limit of β-galactosidase activity of 0.05 Miller units, which was inserted into the claims for the purpose of avoiding the prior art of Samain and not for any utility, and does not produce enough activity to be useful, *e.g.* to reduce lactose or otherwise enable purification of 2'-FL.

107. Accordingly, Chr. Hansen HMO seeks and is entitled to a declaratory judgment that each of the claims of the '018 patent is invalid for at least the reasons set forth above. A judicial determination of the respective rights of the parties regarding invalidity of these patent claims is necessary and appropriate under 28 U.S.C. § 2201 to resolve the parties' dispute regarding invalidity of the '018 Patent.

SECOND CAUSE OF ACTION – DECLARATORY JUDGMENT OF NON-INFRINGEMENT

108. Chr. Hansen HMO incorporates by reference all preceding paragraphs as if fully set forth herein.

109. An actual controversy exists concerning the '018 Patent due at least to Glycosyn's assertions in the ITC Investigation and the Waco Action that Chr. Hansen HMO has infringed the '018 Patent through its manufacture and sale of 2'-FL.

110. Glycosyn's wrongful assertion of the '018 Patent against 2'-FL sold by Chr. Hansen HMO and products containing such 2'-FL has caused and will continue to cause Chr. Hansen HMO irreparable injury and damage.

111. The '018 Patent claims a manufacturing process, specifically methods of producing complex sugars in a genetically modified *E. coli* bacterium. The only allegedly inventive aspect of the patent is an exogenous gene that produces low levels of β-galactosidase activity (0.05 to 200 units, measured using the Miller protocol). β-galactosidase is an enzyme that naturally occurs in *E. coli* and that interferes with 2'-FL production. The prior art includes disclosures of producing 2'-FL by eliminating β-galactosidase activity altogether and discloses all other aspects of claim 1 of the '018 Patent, as well as the dependent claims of the patent, as discussed above.

112. Chr. Hansen HMO's processes for producing 2'-FL, including those using the #1540 strain, do not include at least the use of "an exogenous functional β-galactosidase gene comprising a detectable level of β-galactosidase activity that is reduced compared to that of a wild-type *E. coli* bacterium, wherein the level of β-galactosidase activity comprises between 0.05 and 200 units" as required by claim 1 of the '018 Patent, the only independent claim of the patent, and claims 2-28, all of which are dependent on claim 1. In particular, the inserted β-galactosidase gene in the #1540 strain accused of infringement in the Waco Action has a level of β-galactosidase activity, if any, that is less than the claimed range.

113. Further, the strains used in Chr. Hansen HMO's processes to make 2'-FL do not infringe the '018 Patent because they do not contain "an exogenous functional β-galactosidase gene" or its equivalent. For example, the #1540 strain contains two gene fragments, the *lacZ-α* and *lacZ-Ω* fragments, neither of which can produce a β-galactosidase enzyme (*i.e.*, neither of

which is a "functional" β-galactosidase gene) on its own, and therefore does not literally infringe. Nor are these two fragments equivalent to an exogenous functional β-galactosidase gene.

114. Moreover, the claims at issue, if properly interpreted, must exhibit a level of β-galactosidase activity resulting from the inserted β-galactosidase gene between 0.05 and 200 units substantially throughout 2'-FL biosynthesis, *i.e.*, fermentation. Chr. Hansen HMO's processes for making 2'-FL, including those using the #1540 strain, fail to satisfy this requirement as well.

115. Chr. Hansen HMO's 2'-FL and its processes for making 2'-FL also do not infringe any of the above-referenced limitations under the doctrine of equivalents because, among other things, there are substantial differences between Chr. Hansen HMO's 2'-FL and its processes for making 2'-FL and the referenced limitations, and Chr. Hansen HMO's 2'-FL and its processes for making 2'-FL do not perform substantially the same function, in substantially the same way, to achieve substantially the same result, as the referenced limitations.

116. Accordingly, for at least the above reasons, Chr. Hansen HMO's 2'-FL and its processes for making 2'-FL do not infringe claim 1 of the '018 Patent or claims 2-28 of the '018 Patent, all of which depend from claim 1.

117. Further, Chr. Hansen HMO has not actively encouraged or induced others to infringe the '018 Patent with knowledge that such acts will lead to infringement. Moreover, as shown in this Complaint, Chr. Hansen HMO has a good-faith belief of non-infringement. Therefore, Chr. Hansen HMO has not induced infringement of any of the claims of the '018 Patent.

118. Because Chr. Hansen HMO's 2'-FL and its processes for making 2'-FL do not meet each limitation of the claims of the '018 Patent, Chr. Hansen's HMO's 2'-FL and its

processes for making 2'-FL do not infringe, directly or indirectly, those claims, either literally or under the doctrine of equivalents.

119. Accordingly, Chr. Hansen HMO seeks and is entitled to a declaratory judgment that its 2'-FL and processes for making 2'-FL do not infringe the claims of the '018 Patent. A judicial determination of the respective rights of the parties regarding non-infringement of these patent claims is necessary and appropriate under 28 U.S.C. § 2201 to resolve the parties' dispute regarding alleged infringement of the '018 Patent.

120. Moreover, given the positions taken by Glycosyn, for example in the Waco Complaint and in Glycosyn's letter to Abbott (Exhibit 15 to the Waco Complaint), Chr. Hansen HMO seeks a declaratory judgment that any *E. coli* strain having its endogenous *lacZ* gene deleted or functionally inactivated, but containing a *lacZα* gene (regardless of whether this is the truncated *lacZα* version encoded by the M13 prophage and/or another *lacZα* gene that has been inserted into the *E. coli* bacterium), does not infringe the '018 Patent in the absence of a *lacZΩ* gene.

PRAYER FOR RELIEF

WHEREFORE, Chr. Hansen HMO prays for the following relief:

(A) a declaration that the claims of the '018 Patent are invalid and/or unenforceable;

(B) a declaratory judgment that its 2'-FL and processes for making 2'-FL do not infringe, directly or indirectly, literally or under the doctrine of equivalents, the claims of the '018 Patent;

(C) a declaratory judgment that any Chr. Hansen HMO *E. coli* strain having its endogenous *lacZ* gene deleted or functionally inactivated, but containing a *lacZα* gene (regardless of whether this is the truncated *lacZα* version encoded by the M13

prophage and/or another *lacZα* gene that has been inserted into the *E. coli* bacterium), does not infringe the '018 Patent in the absence of a *lacZΩ* gene;

(D) an injunction barring Glycosyn from asserting or threatening Chr. Hansen HMO, its customers or its potential customers, or any other third parties purchasing or using 2'-FL made or sold by Chr. Hansen HMO or products containing 2'-FL made or sold by Chr. Hansen HMO, with infringement of the '018 Patent;

(E) a declaration that this action is an exceptional case under 35 U.S.C. § 285;

(F) an award to Chr. Hansen HMO of its attorneys' fees and costs incurred in this Action; and

(G) a grant of any other relief that this Court deems just and proper.

Dated: July 7, 2022

CHR. HANSEN HMO GMBH

By its Attorneys,

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