Filed on behalf of: **Junior Party, Broad** Paper No. ____

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UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE PATENT TRIAL AND APPEAL BOARD

THE BROAD INSTITUTE, INC., MASSACHUSETTS INSTITUTE OF TECHNOLOGY, and PRESIDENT AND FELLOWS OF HARVARD COLLEGE,

Patents 8,697,359; 8,771,945; 8,795,965; 8,865,406; 8,871,445; 8,889,356; 8,889,418; 8,895,308; 8,906,616; 8,932,814; 8,945,839; 8,993,233; 8,999,641; and 9,840,713; and Applications 14/704,551 and 15/330,876,

Junior Party,

v.

SIGMA-ALDRICH CO., LLC,

Application 15/456,204,

Senior Party.

Patent Interference No. 106,133 (DK)
(Technology Center 1600)

BROAD MOTION 3

(to designate claims as not corresponding to Count 1)

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I. PRECISE RELIEF REQUESTED

Junior Party, The Broad Institute, Inc., Massachusetts Institute of Technology, and President and Fellows of Harvard College (collectively "Broad"), moves pursuant to 37 C.F.R. §§ 41.121(a)(1)(i) and 41.207(b)(2) that the PTAB designate five categories of Broad's claims (Categories A-E) as not corresponding to Count 1:

Category A: *Staphylococcus aureus* Cas9 protein ("SaCas9");

Category B: Cas9 chimeric CRISPR enzyme;

Category C: Cas9 with two or more nuclear localization signals ("NLSs");

Category D: Cas9 fused to specified protein domains; and

Category E: Claims that are generic as to RNA and also do not specify integration of a donor polynucleotide sequence ("Donor Template Integration" claims), i.e., the only claims that should remain designated as corresponding to Count 1 are those that are either (i) limited to single molecule RNA ("sgRNA"), or (ii) require Donor Template Integration and are not otherwise separately patentable.

If this motion is granted and Broad Motion 1 is denied, the following claims would remain designated as corresponding to Count 1: claims 4, 11 and 18 of U.S. Patent No. 8,697,359 ("359 patent") (Ex. 2011), claim 5 of U.S. Patent No. 8,771,945 ("945 patent") (Ex. 2015), all claims (1-30) of U.S. Patent No. 8,795,965 ("965 patent") (Ex. 2012), claims 2, 5 and 30 of U.S. Patent No. 8,906,616 ("616 patent") (Ex. 2014), claims 8-9, 14, 16 and 27 of U.S. Patent No. 9,840,713 ("713 patent") (Ex. 2043) and claims 14-16 of Application No. 14/704,551 ("551 application") (Ex. 2051).

II. DESCRIPTION OF APPENDICES

Appendix A is an Exhibit List, Appendix B is the Statement of Material Facts, Appendix C is a Summary Chart of Grounds and Claims, and Appendix D is a Claim Differentiation Chart.

III. BACKGROUND AND SUMMARY

A. Count 1

Count 1 is an "or" count drawn to (Broad half, Broad US Patent 8,697,359, claim 18) a eukaryotic CRISPR-Cas9 system comprising Cas9 and sgRNA that comprises a guide sequence fused to a tracr sequence that targets and hybridizes to a DNA target sequence in a eukaryotic cell (Ex. 2011), or (Sigma half, Sigma application 15/456,204 ("the 204 application"), claim 31) a method for using a CRISPR-Cas9 system in a eukaryotic cell to create a double-stranded break in target DNA and integrate a donor template. Paper 1 at 11-13. The Sigma half of Count 1 requires Donor Template Integration but is generic with regard to the RNA configuration encompassing both sgRNA and dual-molecule RNA ("dualRNA") configurations; the Broad half of Count 1 requires sgRNA but does not require Donor Template Integration. *Id*.; MF 1-2.

B. Broad's Currently Designated Claims Cover Many Different Inventions That Are Separately Patentable From Count 1

Broad's claims currently designated as corresponding to Count 1 include many drawn to inventions that are patentably distinct from the subject matter of Count 1. These separately patentable inventions include four categories: use of (a) SaCas9, (b) chimeric Cas9 formed of two fragments, each from a different Cas9 ortholog, (c) Cas9 having two or more NLSs, and (d) Cas9 with fused and heterologous protein domains. None of the claims directed to those improvements and selections are anticipated by, or obvious in view of, Count 1 if considered as prior art alone or in proper combination with the prior art. Notably, in the 106,132 Interference ("132 Interference"), Sigma filed a declaration from Dr. Paula Cannon specifically opining that claims to a "chimeric Cas9 protein," "a Cas9 protein that includes a Protein Transduction Domain ('PTD')" such as an NLS, and "one or more mutation(s) in the Cas9 RuvC/HNH domain(s)" would not have been obvious to a POSA as of early December 2012 in view of 132 Interference Proposed Count 2. Ex.

2463 (132 Cannon Decl.) ¶ 35; MF 3. These categories correspond to categories (b)-(d) above, and Proposed Count 2 in the 132 Interference and Count 1 (as well as Proposed Count 3) here are materially the same with respect to the relevant Cas9 and NLS related limitations. Nothing in the Broad half of Count 1 affects Sigma's admissions regarding lack of claim correspondence of these subject matters, as confirmed by Broad's expert, Dr. Seeger. Ex. 2464 (Seeger Decl.) ¶ 174.

C. Broad's Non-Limited/Generic RNA Claims Also Not Limited To Donor Template Integration Should Be Designated As Not Corresponding

In addition to those four categories, Broad's claims that are both generic as to RNA and not limited to Donor Template Integration should also be designated as not corresponding to Count 1. The only claims that should correspond to Count 1 are those that either are limited to use of sgRNA (and so correspond to the Broad half of Count 1) or recite Donor Template Integration (and so correspond to the Sigma half of Count 1). MF 27-48.

First, Broad's half of Count 1 requires that the CRISPR-Cas9 system use sgRNA. But Broad's generic claims that do not limit the RNA configuration to sgRNA are currently designated as corresponding to Count 1, even claims that do not use the term "guide RNA." These claims are indisputably not limited to sgRNA, and thus do not correspond to the Broad half of Count 1.

In addition, Broad submits that its claims that do use the term "guide RNA," without specifying a fused or chimeric RNA limitation, are also not limited to the sgRNA configuration. Broad recognizes that in Interference No. 106,115 ("115 Interference"), the PTAB construed "guide RNA" to be limited to a "single-molecule RNA configuration." Ex. 2121, Paper No. 877, at 44. Broad respectfully disagrees with that construction, especially given the accompanying evidence that, at the relevant time, persons of ordinary skill in the art ("POSAs") would have understood the plain and ordinary meaning of the term "guide RNA," in particular after publication of Jinek 2012, to be generic and not limited to sgRNA. Ex. 2202, Jinek 2012.

For instance, Sigma's 204 application, at issue here, uses the term "guide RNA" consistent with its plain and ordinary meaning and exemplifies that POSAs understood the term "guide RNA" as a generic term not limited to either the sgRNA or the dualRNA configuration. Sigma's 204 specification explains that "[i]n other embodiments, the guide RNA comprises a single molecule…" and "[i]n other embodiments, the guide RNA can comprise two separate molecules." Ex. 2074 (204 application) at [0077]. Moreover, ToolGen's contemporaneous 14/685,510 application ("510 application"), at issue in Interference 106,126 ("the 126 Interference"), similarly states that "guide RNA" can refer to either "single-chain RNA" or "two RNA." Ex. 2068 ¶¶ [168]-[170] ("In the present invention, the ***guide RNA*** may consist of ***two RNA***, i.e., CRISPR RNA (crRNA) and transactivating crRNA (tracrRNA) ***or be a single-chain RNA*** (sgRNA) produced by fusion of an essential portion of crRNA and tracrRNA.").

Neither Sigma nor ToolGen independently coined the term "guide RNA" or defined it in their patent applications in a manner inconsistent with its plain and ordinary meaning. To the contrary, both Sigma and ToolGen used the term "guide RNA" in accord with its plain and ordinary meaning, consistent with Jinek 2012's use of the term, to include both sgRNA and dualRNA. For example, Jinek 2012 states: "In this ternary complex, the ***dual tracrRNA:crRNA*** structure acts as ***guide RNA*** that directs the endonuclease Cas9 to the cognate target DNA."[1] Ex. 2202, Jinek 2012, at Figure S1 description. Certainly, a POSA's understanding of the term "guide RNA" in the context of dualRNA and sgRNA CRISPR-Cas9 systems after June 2012 would be guided by the disclosure of Jinek 2012, which proposed the use of sgRNA in CRISPR-Cas9 systems. Thus, the disclosures from Sigma and ToolGen, along with references available at the time (including as detailed by Dr. Seeger (Ex. 2464 (Seeger Decl.) ¶¶ 146-83), show that a POSA

[1] All emphases in this brief are supplied unless otherwise noted.

in 2012 understood "guide RNA" had a plain and ordinary meaning that was generic to both sgRNA and dualRNA CRISPR-Cas9 systems.

Broad's specification did not redefine "guide RNA" in a manner contrary to this plain and ordinary meaning. Indeed, the Broad half of Count 1 uses 359 patent dependent claim 18 that specifically requires "a guide sequence fused to a tracr sequence," rather than independent claim 15, which recites "guide RNA." If "guide RNA" really meant sgRNA, there would be no need to use the dependent claim as the Broad half of Count 1. The presumption raised by claim differentiation of dependent claims directed to sgRNA shows that Broad did not provide a clear and unambiguous redefinition of "guide RNA" different than the plain and ordinary meaning of the term "guide RNA." The broadest reasonable interpretation ("BRI") of "guide RNA" is not limited to sgRNA.

Second, Broad's claims that do not require Donor Template Integration do not correspond to Sigma's half of Count 1. Sigma has argued persistently throughout the prosecution of its applications that Donor Template Integration claims are patentably distinct from claims that are to targeting and cleavage of DNA in a eukaryotic cell by a CRISPR-Cas9 system. *See, e.g.*, Ex. 2074 (April 9, 2018 Petition) at 7-9. Sigma has argued in particular that claims that do not recite integration of a donor template do not correspond to a count that includes such a requirement, even if they recite other forms of cleavage and editing/repair. Ex. 2074 (October 13, 2020 Suggestion of Interference) at 7. Sigma has repeatedly argued that Donor Template Integration and other forms of cleavage and repair were separately patentable inventions, and that a POSA would not have reasonably expected success in integrating a donor template even if the art showed recognition of successful cleavage in a eukaryotic cell. Ex. 2074 (April 9, 2018 Petition) at 9, 24-25; Ex. 2074 (April 29, 2019 Applicant Remarks) at 19 (citing Ex. 2465 April 29, 2019 Cannon Decl. ¶¶ 97-

98). Accordingly, Broad's claims that require neither sgRNA nor Donor Template Integration should be designated as not corresponding to Count 1.

IV. ARGUMENT

Broad now takes up each of the categories A-E set forth above in turn and shows for each category why the claims in the category are separately patentable from, and should be designated as not corresponding to, Count 1.

A. Broad's Claims Limited To SaCas9 Should Be Designated As Not Corresponding To Count 1

All of the Involved Claims of U.S. Patent No. 8,865,406 ("406 patent") (Ex. 2017), U.S. Patent No. 8,895,308 ("308 patent") (Ex. 2013) and Application No. 15/330,876 ("876 application") (Ex. 2064) are limited to using the *Staphylococcus aureus* Cas9 protein ("SaCas9") or a nucleotide sequence encoding SaCas9. As one example, claim 1 of the 406 patent recites: "a second regulatory element operable in a eukaryotic cell operably linked to a nucleotide sequence encoding **a *Staphylococcus aureus* Cas9 protein**." Ex. 2017, 406 patent, claim 1. Thus, these claims are directed to the selection of SaCas9 as the Cas9 component of the CRISPR-Cas9 system.

With respect to anticipation, Count 1 does not recite that the Cas9 be SaCas9 (or any particular Cas9 protein). MF 4; Paper 1 at 12-13. Thus, Count 1 does not anticipate claims that require SaCas9. Ex. 2464 (Seeger Decl.) ¶¶ 175-77.

With respect to obviousness, there is no teaching or suggestion in Count 1 or the prior art to use SaCas9 in CRISPR-Cas9 systems in eukaryotic cells. MF 5; Ex. 2464 (Seeger Decl.) ¶¶ 177-96. Moreover, CRISPR-Cas9 systems using SaCas9 possess a surprising combination of small size and high efficacy in eukaryotes. MF 6; Ex. 2017, 406 patent at 83:1-25-84:1-23. These features render CRISPR-SaCas9 systems advantageous for use in eukaryotic cell-based applications, especially where vector delivery using the highly versatile adeno-associated virus

(AAV) is favored because AAV vectors are notoriously space-constrained. MF 7; Ex. 2464 (Seeger Decl.) ¶ 193; Ex. 2017, 406 patent at 83:1-25-84:1-23.

As of 2012, there were more than 600 bacterial Cas9 orthologs that had been identified. MF 8-9; Ex. 2226, Ran 2015; Ex. 2464 (Seeger Decl.) ¶ 181. Although SaCas9 was known as one of those orthologs, knowledge of SaCas9's existence would not have suggested the use of, or provided a POSA any reasons for the selection of, SaCas9 for use in eukaryotes over any of the other Cas9 orthologs. Ex. 2464 (Seeger Decl.) ¶ 180-93. Where, as here, the prior art choices are vast, the prior art must provide some reason to make the selection in order to find obviousness. *Ortho-McNeil Pharm., Inc. v. Mylan Labs., Inc*., 520 F.3d 1358, 1364 (Fed. Cir. 2008). Here, the prior art failed to provide, or even suggest, a reason to select SaCas9 out of the vast choices of Cas9 orthologs. Ex. 2464 (Seeger Decl.) ¶¶ 180-93. Even if, as found in the 115 Interference, the prior art does not teach away from SaCas9, a lack of a teaching away does not equate to obviousness. *Rembrandt Wireless Tech., LP v. Samsung Elecs. Co., Ltd.*, 853 F.3d 1370, 1379-80 (Fed. Cir. 2017) ("[T]he absence of a formal teaching away in one reference does not automatically establish a motivation to combine it with another reference in the same field."). Rather, even without a teaching away, an affirmative showing of motivation to select SaCas9 is required to show that the selection would have been obvious. There is no evidence of such motivation here.

The small size of SaCas9 alone did not provide motivation to select that particular ortholog. Ex. 2464 (Seeger Decl.) ¶¶ 185-93; MF 10. At the relevant time, the most commonly studied Cas9 ortholog was from *Streptococcus pyogenes* ("SpCas9") (MF 8), which achieved high efficiencies in eukaryotic cells, as shown by later disclosed indel formation tests. *Id*.; Ex. 2201, Cong 2013; *see also* Ex. 2202, Jinek 2012, at 816. At 1368 amino acids in length, SpCas9 is substantially larger than SaCas9 (1053 amino acids in length). MF 11; *see* Ex. 2464 (Seeger Decl.) ¶¶ 178-92. Later

study of Cas9 proteins smaller than SpCas9 has not shown any improvement in efficiency as a result of smaller size. At least one group of researchers studied *S. thermophilus* (StCas9), a smaller ortholog of 1121 amino acids. Ex. 2464 (Seeger Decl.) ¶¶ 185-92; *see also* Ex. 2239, Gasiunas 2012, at E2579. No benefit was demonstrated with the smaller StCas9 ortholog, and so that provided no motivation for a POSA to look at even smaller orthologs or then to choose SaCas9 out of all those smaller orthologs. Even if a POSA would have been focused on reduced size (and there is no contemporaneous evidence suggesting that was the case), there was nothing pointing the POSA to SaCas9. MF 10.

A POSA also would not have been motivated to investigate SaCas9 because its amino acid sequence was very different than previously characterized Cas9s. Ex. 2464 (Seeger Decl.) ¶ 189. As noted above, SpCas9 was the most commonly studied Cas9 ortholog in the prior art. *Id*. The sequence identity between SaCas9 and SpCas9 is at 17%, particularly low for orthologous proteins. *Id*.; Ex. 2227, Nishimasu 2015, at 1114. If a POSA considered sequence identity, it would have steered the POSA away from selecting SaCas9 based on its lack of sequence homology with the most efficient and most frequently used Cas9 ortholog, SpCas9. Ex. 2464 (Seeger Decl.) ¶ 189.

In the 115 Interference, the PTAB noted that a SaCas9 system was one of a number of "model systems" established to study CRISPR-Cas9 functionality, citing Ex. 2215, Sapranauskas 2011. Ex. 2121, 115 Interference, Paper 877 at 33. However, with regard to SaCas9 Sapranauskas cites to other scientists who never reported any success in using SaCas9 systems in eukaryotes. Additionally, research from Sapranauskas and others establishes that there was little to no predictability as to which Cas9 orthologs could be used in systems that would be effective in eukaryotes. Thus, Sapranauskas identifying SaCas9 as one of the "model systems" would not have provided a POSA motivation to use the SaCas9 ortholog in eukaryotic cells. Under such

circumstances, the development of a eukaryotic CRISPR-SaCas9 system would not have been obvious. *See* Ex. 2464 (Seeger Decl.) ¶ 178. Sigma's arguments made during prosecution of the 204 application about the unpredictability of nearly every aspect of the eukaryotic CRISPR-Cas9 system, including those associated with the Cas9 protein, confirm this lack of predictability and lack of motivation to work with any different Cas9. *See, e.g.*, Ex. 2074 (April 29, 2019 Applicant Remarks) at 19, 27-28 (citing Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 76-77); Ex. 2466 (October 17, 2017 Urnov Decl.) ¶¶ 15-17; MF 12.

In addition, the use of a CRISPR-SaCas9 system in eukaryotic cells has unexpected attributes demonstrating non-obviousness. Ex. 2464 (Seeger Decl.) ¶ 193; MF 13. For example, Broad scientists demonstrated that CRISPR-SaCas9 systems produced indels *in vivo* with efficiencies comparable to those achieved using SpCas9 ortholog, in stark contrast to the poorer performance of other Cas9 orthologs. *Id.* The robust *in vivo* activity specific to SaCas9 was neither expected nor predictable based on the state of the art. Ex. 2464 (Seeger Decl.) ¶ 193. These surprising advantages would not have been expected from Count , alone or in combination with the prior art, thus, rendering the involved SaCas9 claims non-obvious. *Id.*; *In re Chupp*, 816 F. 2d 643, 646 (Fed. Cir. 1987) (superior, unexpected properties in one of a spectrum of common properties can be sufficient to show nonobviousness); *Application of Albrecht*, 514 F. 2d 1389, 1396 (C.C.P.A. 1975).

Finally, the commercial success of systems using SaCas9 is powerful objective evidence. SaCas9 has become an in-demand research tool for CRISPR-Cas9 systems and is used for more therapeutic applications than any other Cas9 ortholog. MF 14; Ex. 2464 (Seeger Decl.) ¶ 194. Commercial success is a secondary indicia of non-obviousness that must be considered in an obviousness inquiry. *Bristol-Myers Squibb Co. v. Teva Pharm. USA, Inc.,* 752 F.3d 967, 977 (Fed.

Cir. 2014) ("Secondary considerations of nonobviousness 'must always when present be considered,' and can serve as an important check against hindsight bias."); *Power Integrations, Inc. v. Fairchild Semiconductor Intern., Inc.*, 711 F.3d 1348, 1368 (Fed. Cir. 2013) ("[O]bjective considerations, which [are] sometimes refer[red] to as 'secondary considerations,' [are] essential components of [an] obviousness analysis. Objective evidence of nonobviousness can include [] commercial success.") (internal citations omitted).

For these reasons, and as set forth in detail in the Seeger Declaration, there is nothing in Count 1 alone or in combination with the prior art that would have taught, suggested, or motivated a POSA to use SaCas9, and certainly not in a eukaryotic cell. Ex. 2464 (Seeger Decl.) ¶¶ 175-96.

B. Broad's Claims That Require A Chimeric Cas9 Should Be Designated As Not Corresponding to Count 1

All of the involved claims of Broad's U.S. Patent No. 8,889,418 ("418 patent") (Ex. 2060) are limited to a Cas9 protein that is a chimeric enzyme comprising a first fragment and a second fragment, wherein each of the first and second fragments are from a different Cas9 protein from an enumerated list. MF 15. As one example, claim 1 of the 418 patent recites: "A modified Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR) enzyme wherein the enzyme is a **Cas9 protein that is a chimeric CRISPR enzyme** in that it **comprises a first fragment and a second fragment,** wherein **each of the first and second fragments is from a different Cas9 protein**…." Ex. 2060, 418 patent, claim 1. Thus, these claims are directed to the improvement in which the Cas9 is not a wild type Cas9 from a single organism, but rather is a chimeric Cas9 that includes fragments from different Cas9s. Claims 1-22 of the 418 patent are to the Cas9, a nucleic acid encoding the Cas9, or a vector comprising such a nucleic acid; claims 23-28 are to eukaryotic CRISPR-Cas9 systems/methods using such a chimeric Cas9 with RNA. Such

chimeric Cas9s can lead to enhanced or new functions. *See* Ex. 2060, col. 82; Ex. 2464 (Seeger Decl.) ¶¶ 197-202.

With respect to anticipation, Count 1 does not recite that the Cas9 has any chimeric aspect. Paper 1 at 12-13. Thus, Count 1 does not anticipate claims that require chimeric Cas9. Ex. 2464 (Seeger Decl.) ¶ 200.

With respect to obviousness, nothing in Count 1 or in the prior art teaches, suggests, or provides motivation to a POSA to design a chimeric Cas9 that is comprised of two fragments from Cas9 proteins from different organisms. MF 16; Ex. 2464 (Seeger Decl.) ¶¶ 200-08. As set forth in the 418 patent, 4:21-25, "[t]hese chimeric Cas9 proteins may have a higher specificity or a higher efficiency than the original specificity or efficiency of either of the individual Cas9 enzymes from which the chimeric protein was generated." MF 17; Ex. 2060. The 418 patent also outlines unexpected benefits of a chimeric Cas9:

> The benefits of making chimeric Cas9 include:
> a. reduce toxicity;
> b. improve expression in eukaryotic cells;
> c. enhance specificity;
> d. reduce molecular weight of protein, make protein smaller by combining the smallest domains from different Cas9 homologs; and
> e. Altering the PAM sequence requirement.

MF 18; Ex. 2060, 418 patent at 83:45-52; *see also* Ex. 2464 (Seeger Decl.) ¶¶ 201-07.

Notably, in the 132 Interference Sigma has taken the position that claims to chimeric Cas9 are nonobvious over its Proposed Count 2 (Ex. 2124 (132 Sigma Mot. 1) at 27:11-28:2), which is limited to Donor Template Integration, and which does not materially differ from Count 1 as to the Cas9 limitation. Sigma submitted expert testimony from Dr. Paula Cannon who specifically opined that claims to a "chimeric Cas9 protein" "would not have been obvious to a POSITA in view of proposed Count 2 in early December 2012." Ex. 2463 (132 Cannon Decl.) ¶ 35; MF 3.

Consistent with this position, during prosecution of the involved 204 application, Sigma argued, again via its expert Dr. Cannon, that modifications to the Cas9 protein such as adding an NLS could "affect Cas9 protein folding and the final protein structure" such that it "could interfere with Cas9 function, for instance, by interfering with a binding site or catalytic domain." Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 59-60 (one could not "predict with any reasonable certainty whether ***any*** functional domain… will be properly exposed when expressed as a fusion with Cas9"); *id*. ¶¶ 59-60 ("Indeed, there was no reasonable expectation that the use and arrangement of any functional domain…. in the CRISPR-Cas system, *e.g.* as a fusion to a Cas9 would be operative…" because modifications to the protein could affect protein folding and structure.). Sigma specifically argued that "[b]ecause a protein's function is inextricably linked to its folded structure, proper folding after (or during) translation is crucial" and that misfolded proteins would be subject to degradation in the eukaryotic environment. Ex. 2074 (April 29, 2019 Applicant Remarks) at 30 (citing Ex. 2465 (April 29, 2019 Cannon Decl.) ¶ 88). While Sigma offered these arguments in support of the novelty of adding an NLS, a POSA would have expected a more significant modification—such as use of a chimeric Cas9 with two fragments each from a different Cas9 ortholog—to be even more likely to affect protein folding and structure, and so a POSA would have understood that such a suggestion was less predictable as to what would happen to the Cas9 than adding an NLS. There would have been no expectation of success with a chimeric Cas9 system for use for use in eukaryotic cells. Ex. 2464 (Seeger Decl.) ¶¶ 180-93.

For these reasons, and as set forth in the Seeger Declaration, there is nothing in Count 1 alone or in combination with the prior art that would have taught, suggested, or motivated a POSA to use a chimeric Cas9, and certainly not in a eukaryotic cell. Ex. 2464 (Seeger Decl.) ¶¶ 197-208.

C. Broad's Claims To The Use Of A Cas9 With Two Or More NLSs Should Be Designated As Not Corresponding To Count 1

The next set of the Broad involved claims that should be designated as not corresponding to Count 1 are those claims requiring the use of "two or more" nuclear localization signals ("NLSs"). Ex. 2464 (Seeger Decl.) ¶¶ 209-32. These claims include all of the claims of the U.S. Patent No. 8,871,445 ("445 patent") (Ex. 2029) and U.S. Patent No. 8,932,814 ("814 patent") (Ex. 2037) as well as claim 7 of the 233 patent (Ex. 2024), claims 9-11 of the 551 application (Ex. 2051), and claim 34 of the 876 application (Ex. 2064). One example of these claims is claim 1 of the 445 patent, which recites "wherein the CRISPR-Cas system **comprises two or more nuclear localization signals (NLSs)**." Ex. 2029, 445 patent, claim 1. These claims are directed to the improvement in which the Cas9 has two or more NLSs. This invention provides for significantly greater nuclear localization, unexpectedly without the potential negative consequences that can arise from the engineering of a protein that must fold and undergo multiple configuration changes in order to perform its function.

With respect to anticipation, the Sigma half of Count 1 specifically recites that the Cas9 has "only one NLS," not two or more. MF 19; Paper 1 at 12-13. The Broad half of Count 1 does not indicate anything about the Cas9 with regard to NLSs. Thus, Count 1 does not anticipate these claims that require two or more NLSs.

With respect to obviousness, neither Count 1 nor the prior art provides any teaching or suggestion to use two or more NLSs in a CRISPR-Cas system in a eukaryotic cell, nor was there a reasonable expectation of success in using two or more NLSs. MF 20; Ex. 2464 (Seeger Decl.) ¶¶ 209-32. As an initial matter, the Sigma half of Count 1 expressly teaches away from more than one NLS, stating that the Cas9 has "only one NLS."

Additionally, a POSA would have understood that adding amino acids to a protein such as Cas9 could alter its folding affecting its structure and function in ways that were not predictable. MF 21-23, 25; Ex. 2464 (Seeger Decl.) ¶¶ 216-17, 222-28. Indeed, throughout the prosecution of its 204 application, Sigma argued that a POSA would not expect adding a single NLS to work, and that a POSA would not expect the protein to be operative if one NLS was added, resulting in no reasonable expectation of success. As Sigma stated, "when a protein is expressed as a fusion, such as with an NLS or epitope or chimeric tag, there are unexpected results, further confirming that there was no reasonable expectation of success as to a eukaryotic CRISPR-Cas9 system wherein the Cas9 includes one or more NLSs." Ex. 2074 (April 29, 2019 Applicant Remarks) at 27 (citing Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 76-77); MF 21-23.

The POSA thus had no foreknowledge of whether fusing two or more NLSs to the Cas9 protein of Count 1 would disrupt its structure or function in a eukaryotic cell. This was so even though the prior art contained publications that showed the use of multiple NLSs with other proteins. There is no evidence, nor was there a reasonable expectation, that the use and arrangement of any functional domain (*e.g.*, two or more NLSs), in the CRISPR-Cas system, *e.g.*, as a fusion to a Cas9, would be operative because functional domains may be buried or shielded during Cas9 protein folding, affecting the activity of the functional domains and the Cas9 protein itself. *Id.*; Ex. 2264, Turner 1996 (abstract evidences that significant effects as to function, including loss of function, can result from converting to fusion protein); *see also* Ex. 2263, Brothers 2003 (abstract: "tags do have a significant effect on protein localization and function"). Also, tagging a protein with two NLSs could interfere with function; for instance, by interfering with a binding site or catalytic domain. Ex. 2464 (Seeger Decl.) ¶¶ 222-28; Ex. 2258, Fieck 1992, at 1785.

Notably, Sigma made this exact argument during prosecution of the 204 application as to addition of a single NLS, eventually securing claims reciting "only one NLS." Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 59-60 (one could not "predict with any reasonable certainty whether ***any*** functional domain… will be properly exposed when expressed as a fusion with Cas9"); *id*. ¶¶ 59-60 ("Indeed, there was no reasonable expectation that the use and arrangement of any functional domain…. in the CRISPR-Cas system, *e.g.*, as a fusion to a Cas9, would be operative…" because modifications to the protein could affect protein folding and structure.). Ex. 2074 (April 29, 2019 Applicant Remarks) at 30 (citing Ex. 2465 (April 29, 2019 Cannon Decl.) ¶ 88 (arguing that "[b]ecause a protein's function is inextricably linked to its folded structure, proper folding after (or during) translation is crucial" and that misfolded proteins would be subject to degradation in the eukaryotic environment). Ex. 2074 (October 17, 2017 Applicant Remarks); MF 21-23.

Additionally, in the 132 Interference, Sigma has taken the position that claims to a Cas9 protein that includes a Protein Transduction Domain (PTD), such as an NLS, are nonobvious over its Proposed Count 2. Ex. 2124 (132 Sigma Mot. 1) at 27:11-28:2; MF 3. Proposed Count 2 in the 132 Interference is limited to Donor Template Integration but does not materially differ from Count 1 here as to the NLS limitation. Sigma also submitted expert testimony from Dr. Cannon who specifically opined that claims to a "a Cas9 protein that includes a Protein Transduction Domain ("PTD")" "would not have been obvious to a POSITA in view of proposed Count 2 in early December 2012." Ex. 2463 (132 Cannon Declaration) ¶ 35.

Moreover, even if Count 1's recitation of "only one NLS" is not considered as a teaching away, Sigma's arguments during prosecution of the 204 application make clear that just because one NLS might function in the system, a POSA would not have had a reasonable expectation that two or more NLSs would function based on that information about one NLS. Sigma filed a

declaration from Dr. Fyodor Urnov arguing that "protein functional domains are not mix-and-match interchangeable…." Ex. 2466 (October 17, 2017 Urnov Decl.) ¶¶ 15-17; MF 21-23. He explicitly testified that "you cannot, from a statement 'domain A functions when fused to protein B, which also retains its function,' derive the statement 'domain C will also function when fused to that protein, which will function in that fusion as well." *Id*. Thus, just because a single NLS might function when fused to Cas9 does not mean that another domain "will also function when fused to that protein, which will function in that fusion as well."

Additionally, the Broad inventors demonstrated unexpected results, showing that using two NLSs fused to Cas9 resulted in significantly greater co-localization of CRISPR-Cas9 components in the nucleus of a eukaryotic cell than could be expected. MF 24. In a eukaryotic cell, expression of a protein such as Cas9 is completed in the cytoplasm—outside of the nucleus. Ex. 2464 (Seeger Decl.) ¶ 212. The Cas9 protein must then be localized to the nucleus in order to form a complex with its RNA counterparts if the desire is to act on a genomic DNA target, also contained within the nucleus. *Id.* Two or more NLSs unexpectedly aided in the localization of the CRISPR-Cas9 system to the nucleus of a eukaryotic cell ***without*** inhibiting functionality of the Cas9. Ex. 2464 (Seeger Decl.) ¶¶ 202-21; Ex. 2001, Zhang B1, Figure 1B; Ex. 2201, Cong 2013, Figure 1A); Ex. 2793.

In the work detailed in their first provisional patent application, U.S. Application No. 61/736,527 ("Zhang B1") (Ex. 2001), filed December 12, 2012, the Broad inventors tested systems with a single NLS and systems with two NLSs for nuclear localization of both SpRNase III and SpCas9. Ex. 2464 (Seeger Decl.) ¶¶ 218-21. While a single NLS on the C-terminal end of SpRNase III (but not the N-terminus) was sufficient to target that protein to the nucleus (*Id*.; Ex. 2001, Zhang Bl, ¶ 170), a version of SpCas9 with a NLS fused to each end of the protein exhibited a significantly

higher degree of nuclear localization, and was unexpectedly more efficient over and above what a POSA may have expected from the prior art disclosures that in other systems use of more than one NLS could increase nuclear localization. MF 24. For example, in an August 2012 update of key data (Ex. 2793, Email from Le Cong RE: CRISPR Update - Mammalian System 20120829, dated Aug. 29, 2012), one experiment showed a synergistic effect by using two NLSs resulting in nearly four times the cleavage vs the best achieved with one NLS. Ex. 2464 (Seeger Decl.) ¶ 221.

There is no teaching, suggestion, motivation, or reason in Count 1 or in other prior art that would have caused a POSA to use two or more NLSs for eukaryotic CRISPR-Cas9 systems, nor is there a showing that a POSA would have had a reasonable expectation that such a Cas9 would have worked, let alone significantly enhanced nuclear localization. *See also* Ex. 2464 (Seeger Decl.) ¶¶ 209-32.

D. Broad's Claims That Are Limited To A Cas9 Fused To Specified Protein Domains Or Including Heterologous Domains Should Be Designated As Not Corresponding To Count 1

These claims require that the Cas9 be fused to specified protein domains or include heterologous domains and are thus patentably distinct. All of the claims (1-43) of Broad's 233 patent (Ex. 2024), all claims (1-28) of U.S. Patent No. 8,999,641 ("641 patent") (Ex. 2047), claims 18-19, 25, 29-30 and 36 of the 713 patent (Ex. 2043), and claim 21 of the 876 application (Ex. 2064) are in this category. As one example, claim 1 of the 641 patent recites that the Cas9 part of the system comprises:

> b) a second regulatory element operable in a eukaryotic cell operably linked to a nucleotide sequence encoding **a fusion of a Type-II Cas9 protein and one or more protein domains,**
>
> wherein:
>
> components (a) and (b) are located on same or different vectors of the system, **the Cas9 protein comprises one or more mutations in a catalytic domain,** the

guide RNA comprises a tracr sequence which is 30 or more nucleotides in length, the Cas9 protein and the guide RNA do not naturally occur together,

…, and wherein

the **one or more protein domains comprises** an epitope tag, a reporter, or a domain having transcription activation activity, transcription repression activity, transcription release factor activity, histone modification activity, RNA cleavage activity, nucleic acid or cellular molecule binding activity, activity as a light-responsive cytochrome heterodimer, transposase activity, integrase activity, recombinase activity, resolvase activity, invertase activity, protease activity, nuclease activity, transcription-protein recruiting activity, cellular uptake activity or antibody presentation activity.

Ex. 2047, 641 patent, claim 1. These claims are directed to the improvement in which the binding function of the Cas9 is used and the Cas9 is fused to one or more of a set of enumerated functional protein domains. This allows for a number of different mechanisms of action by the Cas9 and not just the DSBs that occur with the use of wild type Cas9.

With respect to anticipation, neither half of Count 1 recites that the Cas9 is fused to specified protein domains. MF 26; Paper 1 at 12-13. Thus, Count 1 does not anticipate these claims. With respect to obviousness, there is no teaching or suggestion in Count 1 or the prior art directing a POSA to modify the naturally occurring Cas9 protein sequences as set forth in these claims. MF 27; Ex. 2464 (Seeger Decl.) ¶¶ 233-40. Both halves of Count 1 cover the use of Cas9 as part of the CRISPR-Cas9 system and do not teach, suggest, or motivate a POSA to make these modifications to Cas9 for use in eukaryotic cells. And, as noted above with regard to chimeric Cas9 and the use of two or more NLSs, a POSA would have been hesitant to both blindly modify the Cas9 and fuse it to protein domains, as it would have been expected that such engineering of the Cas9 could cause a lack of function in a eukaryotic cell. *Id*.

There are unexpected benefits to fusing functional domains to Cas9 as doing so can "turn the Cas9/gRNA CRISPR system into a generalized DNA binding system [which] can execute functions beyond DNA cleavage." MF 28; Ex. 2464 (Seeger Decl.) ¶ 239; Ex. 2024, 233 patent at

73:22-29. The 233 patent notes, for example, that "fusing functional domains onto a catalytically inactive Cas9, dCas9, … can impart novel functions, such as transcriptional activation/repression, methylation/demethylation, or chromatin modifications." Ex. 2024, 233 patent at 73:22-29. These unexpected benefits demonstrate the non-obviousness of Broad's claims directed to fusing functional domains to Cas9. *See generally*, Ex. 2464 (Seeger Decl.) ¶¶ 233-40.

Additionally, in the 132 Interference Sigma has taken the position that claims to "chimeric and mutated Cas9 protein" are nonobvious over its Proposed Count 2 (Ex. 2124 (132 Sigma Mot. 1) at 27:11-28:2), which is limited to Donor Template Integration, but which does not materially differ from Count 1 as to the Cas9 limitation. Sigma submitted expert testimony from Dr. Cannon who specifically opined that claims to a "one or more mutation(s) in the Cas9 RuvC/HNH domain(s)" "would not have been obvious to a POSITA in view of proposed Count 2 in early December 2012." Ex. 2463 (132 Cannon Decl.) ¶ 35. And, as noted above in Sections B and C, during prosecution of the involved 204 application, Sigma argued and submitted expert testimony that similar modifications of the Cas9 protein would produce unexpected results and that there was no such expectation of success for modifications to a Cas9 protein. Ex. 2074 (April 29, 2019 Applicant Remarks) at 30 (citing Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 59-60, 88)). Indeed, Sigma argued that "when a protein is expressed as a fusion, such as with an NLS or epitope or chimeric tag, there are unexpected results, further confirming that there was no reasonable expectation of success as to a eukaryotic CRISPR-Cas9 system wherein the Cas9 includes one or more NLSs." Ex. 2074 (April 29, 2019 Applicant Remarks) at 29.

While Sigma directed these arguments to use of an NLS generally, they apply here as well to the more significant modifications contemplated by these claims. Ex. 2464 (Seeger Decl.) ¶¶ 237-38. Indeed, Sigma submitted expert testimony that one could not "predict with any reasonable

certainty whether ***any*** functional domain… will be properly exposed when expressed as a fusion with Cas9," (Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 59-60), and further that it was a "key principle in protein biochemistry" that knowledge regarding the function of one domain fused to a protein was not informative of whether domains will function. *Id*. ¶ 88; Ex. 2466 (October 17, 2017 Urnov Decl.) ¶¶ 15-17 (("[Y]ou cannot, from a statement 'domain A functions when fused to protein B, which also retains its function,' derives the statement 'domain C will also function when fused to that protein, which will function in that fusion as well."); *see also* Ex. 2464 (Seeger Decl.) ¶ 229.

For these reasons, and as set forth in the Seeger Declaration, there is nothing in Count 1 alone or in combination with the prior art that would have taught, suggested, or motivated a POSA to use a modified Cas9 including one being fused to one or more protein domains, and certainly not in a eukaryotic cell. *See* Ex. 2464 (Seeger Decl.) ¶¶ 233-40.

E. Broad's Non-Limited RNA Claims That Are Not Donor Template Integration Claims Should Be Designated As Not Corresponding To Count 1

As formulated, the Broad half of Count 1 is directed to eukaryotic CRISPR-Cas9 systems that require sgRNA (also called "chimeric RNA"). And the Sigma half of Count 1 requires Donor Template Integration.

Many of Broad's involved claims are not limited to sgRNA systems and thus do not correspond to the Broad half of Count 1. Ex. 2464 (Seeger Decl.) ¶¶ 112-42. These claims fall into one of three groups: 1) claims that do not require an RNA component at all; 2) claims that are generic as to the RNA component and do not use the term "guide RNA"; and 3) claims that use the term "guide RNA" but are still generic as to the RNA component under the proper construction of "guide RNA." Broad concedes that the term "guide RNA" was construed in the 115 Interference as limited to the single-molecule RNA configuration. Ex. 2121, 115 Interference, Paper 877 at 33.

However, in light of the evidence presented herewith, it is submitted that the BRI of "guide RNA" does not limit that term to single molecule RNA. *See also* Ex. 2464 (Seeger Decl.) ¶¶ 112-42.

Additionally, of the generic claims that do not correspond to the Broad half of the Count, only three recite Donor Template Integration and so correspond to the Sigma half of Count 1—which is generic as to the RNA. These claims are claim 13 of the 445 patent; claim 30 of the 616 patent; and claim 14 of the 713 patent. As to the remainder of the generic claims not reciting Donor Template Integration, as set out below, they should not be designated to Count 1 as they do not correspond to either half of the Count. *See also* Ex. 2464 (Seeger Decl.) ¶¶ 248-51. Notably, in the 132 Interference, Sigma argued that claims to "cleavage alone" are "patentably distinct" from claims directed to Donor Template Integration. Ex. 2124 (132 Sigma Mot. 1) at 1:12-13, 2:18-30, 3:1-7, 4:14-23, 5:3-7 ("[U]sing a CRISPR-Cas9 system in a eukaryotic cell to cleave DNA and *thereafter to integrate a donor polynucleotide into that cleaved DNA via HDR* is patentably distinct from (not obvious in view of) simply using a CRISPR-Cas9 system in a eukaryotic cell to cleave DNA.") (emphasis in original).

Broad first identifies its claims that are generic as to RNA configuration, then those generic claims that recite Donor Template Integration. Broad then addresses why the generic claims that are not Donor Template Integration claims should be designated as not corresponding to Count 1.

1. Broad's Claims Not Limited To Single-Molecule RNA Designated To The Current Count

a. Claims 15, 17-26, 28-41 Of The 713 Patent And Claims 1-24 Of The 418 Patent Do Not Use The Term "Guide RNA" And Therefore Are Indisputably Not Limited To Single-Molecule RNA

In Broad's 713 patent, independent Claims 15 and 26 (as well as most of the claims that depend from 15 or 26) are directed to methods of genome editing using a CRISPR-Cas system and those claims do not specify any particular configuration of the RNA components of the system;

moreover, these claims do not use the term "guide RNA." MF 29; Ex. 2043, 713 patent; *see* App'x C. In addition, independent claim 1 of the 418 patent does not recite (and neither do dependent claims 2-24) an RNA component at all, let alone sgRNA. *See* Ex. 2464 (Seeger Decl.) ¶¶ 114-18.

Claim 15 of the 713 patent is illustrative. In that claim, the method comprises delivering to target sequences in the cell of a plant or animal a Cas9 protein and RNA that is simply described as "a guide sequence linked to a tracr mate sequence; and a tracr sequence." Ex. 2043, 713 patent, claim 15; Paper No. 13 at 75-76. Thus, claim 15 (and likewise claim 26 with similar language) does not specify whether the RNA is dualRNA or sgRNA; thus, these claims are generic with respect to the RNA configuration. Additionally, these claims do not use the term "guide RNA," but merely set forth the three RNA sequences that are present. Dependent claims 17-25 and 28-41 of the 713 patent are similarly RNA construct-agnostic. *See* Ex. 2464 (Seeger Decl.) ¶¶ 114-15. Dependent claims 16 and 27, in contrast, specify that the RNA sequences are "within a chimeric RNA."

Claims 1-24 of the 418 patent (Ex. 2060) are directed to Cas9 and do not require RNA at all, let alone indicating the configuration of any RNA that may be used with the claimed Cas9s. MF 30. Thus, these claims too are not limited to the use of sgRNA as in the Broad half of Count 1. *See* Ex. 2464 (Seeger Decl.) ¶¶ 116-17.

Claims 15, 17-26, and 28-41 of the 713 patent (Ex. 2043) and claims 1-24 of the 418 patent (Ex. 2060) are non-limited RNA claims that do not correspond to the Broad half of Count 1 because they are indisputably not limited to sgRNA.

b. Broad's Claims That Use The Term "Guide RNA" Also Cover Both Single- And Dual-Molecule RNA Systems

In addition to the claims in the 713 and 418 patents that do not specify the use of sgRNA/chimeric RNA, a number of Broad's other claims are also generic as to the RNA but use

the term "guide RNA." In the 115 Interference Decision on Motions, the PTAB construed the term "guide RNA" used in Broad's claims as limited to sgRNA. Ex. 2121, 115 Interference, Paper 877 at 33. Broad respectfully disagrees with that prior decision and asserts that the evidence provided herewith shows that the BRI of "guide RNA" in Broad's claims is not so limited. *See also* Ex. 2464 (Seeger Decl.) ¶¶ 119-42.

The BRI standard applies in determining the meaning of "guide RNA" as set forth in Broad's involved claims for this Interference. *See Bamberger v. Cheruvu*, 55 U.S.P.Q.2d 1523, at *2 (B.P.A.I. 1998) (BRI applies in interference proceedings). Respectfully, the BRI of "guide RNA" is dictated by how the term was used generally in the art in 2012, including as it was used in Jinek 2012, by Sigma in the involved 204 application and other related applications, by ToolGen in its 510 application that is in the 126 Interference, and how it is used in Broad's patents and applications (including the doctrine of claim differentiation, if Broad is not using "guide RNA" generically, several dependent claims are superfluous)—rather than being limited based on the conclusion that there was no ordinary meaning but there was a purported "definition" of "guide RNA" in a single sentence in Broad's specification.

(1) *The Intrinsic Evidence Supports A Generic Understanding Of "Guide RNA"*

The intrinsic record for Broad's involved patents and applications indicates that "guide RNA" includes both dualRNA and sgRNA. Ex. 2464 (Seeger Decl.) ¶¶ 129-42. In reviewing Broad's Involved Claims, a clear pattern is established. An independent claim uses the term "guide RNA." A dependent claim then narrows guide RNA to an sgRNA guide RNA (*i.e.*, by describing the guide RNA as a "fused" RNA or a "chimeric" RNA construct). *See* Ex. 2011, 359 patent at Claims 1, 4, 8, 11, 15, and 18; Ex. 2015, 945 patent at Claims 1 and 5; Ex. 2013, 308 patent at

Claims 1, 6 and 10 and 25, 29, and 30; Ex. 2014, 616 patent at Claims 1, 2, and 5; and Ex. 2043, 713 patent at Claims 1, 8, and 9.

The doctrine of claim differentiation applies in this case: "The presence of a dependent claim that adds a particular limitation gives rise to a presumption that the limitation in question is not present in the independent claim." *Ex Parte William M. Cresse*, Appeal No. 2008-4746, 2008 WL 5264251, at *4 (B.P.A.I. Dec. 16, 2008) (citing *Phillips v. AWH Corp*., 415 F.3d 1303, 1315 (Fed. Cir. 2005)). While claim differentiation is a presumption, it is a strong presumption that is not to be cast aside lightly. *InterDigital Communs., LLC v. ITC*, 690 F.3d 1318, 1324 (Fed. Cir. 2012).

And that is especially so here where there are many dependent claims that will be rendered superfluous if "guide RNA" is construed as limited to sgRNA. In the hierarchy of claim construction tenets, "[i]t is highly disfavored to construe terms in a way that renders them void, meaningless, or superfluous." *Wasica Fin. GmbH v. Cont'l Auto. Sys., Inc*., 853 F.3d 1272, 1288 n.10 (Fed. Cir. 2017). Accordingly, "claim differentiation . . . is clearly applicable" when the only meaningful difference between the two claims is eliminated by reading in a limitation of the dependent claim into the independent claim. *Wenger Mfg., Inc. v. Coating Mach. Sys., Inc.*, 239 F.3d 1225, 1233 (Fed. Cir. 2001).

The below claim series indicate instances in addition to the 359 patent as already noted, where claim differentiation is required so that a dependent claim does not become superfluous:[2]

- U.S. Patent No. 8,895,308: Claim 1 describes a method using CRISPR-Cas system having "guide RNA" that hybridizes to the target sequence. Claim 6 covers the "method of claim 1, wherein the guide RNA comprises a guide sequence and a tracr sequence." Claim 10 requires the "method of claim 6, wherein the guide sequence and tracr sequence are chimeric." If the "guide RNA" of Claim 1 is already defined

[2] The full text of each claim series is set forth in Appendix 4.

to be chimeric RNA (*i.e.*, a single-molecule RNA), Claim 10 is superfluous over Claim 6. Ex. 2013; MF 31.

- U.S. Patent No. 8,906,616: Claim 1 describes a CRISPR-Cas system having a "guide RNA" polynucleotide sequence comprising a guide sequence, a tracr mate sequence, and a tracr sequence. Claim 2 covers the "composition of claim 1, wherein the modified guide RNA comprises a chimeric guide sequence and a tracr sequence." Claim 5 covers the "composition of claim 1, wherein the modification comprises fusing the tracr mate sequence and the tracr sequence through an artificial loop." If the "guide RNA" of Claim 1, which includes a guide sequence and a tracr sequence, is already defined to be chimeric or fused, Claims 2 and 5 are superfluous. Ex. 2014; MF 32.

Other Involved Claims also show that the term "guide RNA" should be interpreted generically. Whenever Broad intended a chimeric guide RNA, the nature of the RNA is expressly stated in the claim. For example, in the 965 patent, all of the claims explicitly require that the guide RNA be "composed of a chimeric RNA." Ex. 2012, 965 patent at independent claims 1, 10, 17, and 26. Notably, in the 965 patent (Ex. 2012), none of the dependent claims include limitations that the components of the guide RNA be fused or bound to one another (MF 33)—that characteristic is already taught by the explicit use of the term "chimeric" in the independent claims.

That "guide RNA" is a generic term is also supported by the examples in the specifications. Broad's patents disclose preferred embodiments that are dual-molecule RNA systems. MF 34; Ex. 2011, 359 patent at 43:49-53, 44:5-8; Ex. 2464 (Seeger Decl.) ¶¶ 138-41. If "guide RNA" were limited to single-molecule RNA systems, however, such preferred embodiments would not be covered by the patent claims. Such a result "is rarely, if ever, correct." *Ex Parte Andrew Graham, Ando Feyh, & Bernhard Gehl*, Appeal No. 2017-009616, 2018 WL 4356999, at *3 (P.T.A.B. Aug. 23, 2018); *MBO Labs., Inc. v. Becton, Dickinson & Co*., 474 F.3d 1323, 1333 (Fed. Cir. 2007) ("[A] claim interpretation that excludes a preferred embodiment from the scope of the claim is rarely, if ever, correct.") (citation omitted).

(2) The Extrinsic Evidence Shows The Plain Meaning Of "Guide RNA"

A proper BRI claim construction analysis should evaluate evidence of how a POSA would have understood the plain and ordinary meaning of the term "guide RNA" at the relevant time. *See* Ex. 2464 (Seeger Decl.) ¶¶ 120-27. Broad submits that even prior to publication of Jinek 2012, a POSA would have understood the plain meaning of "guide RNA" to be a generic term. However, certainly after publication of Jinek 2012, a POSA would so conclude. Jinek 2012 proposed in June 2012 the use of sgRNA systems for CRISPR-Cas9 and also showed dualRNA systems. Thus, after June 2012, a POSA would be guided by Jinek's generic use of the term "guide RNA."

Jinek 2012 very clearly states: "In this ternary complex, the ***dual tracrRNA:crRNA*** structure acts as ***guide RNA*** that directs the endonuclease Cas9 to the cognate target DNA." MF 41; Ex. 2202, Jinek 2012, at Figure S1, description. Thus, Jinek 2012 taught a POSA that "guide RNA" was a generic term covering systems with either sgRNA or dualRNA. *See, e.g.,* Ex. 2202, Jinek 2012, at Fig. S1, caption; Ex. 2464 (Seeger Decl.) ¶¶ 121-23.

Consistent with Jinek 2012, Sigma's 204 application shows that Sigma's inventors understood that "guide RNA" was a generic term. The 204 specification explains that "[i]n some embodiments, ***the guide RNA comprises a single molecule***…" and "[i]n other embodiments, ***the guide RNA can comprise two separate molecules***." Ex. 2074 (204 application) at [0077]; MF 35. Claim 31 uses the generic term "guide RNA," while dependent claims 33 and 34 specify that the "***guide RNA is a single molecule***" and "***the guide RNA is two molecules***" respectively. Paper 12 at 2; MF 36.

Similarly, in ToolGen's 510 application at issue in the 126 Interference, the ToolGen inventors state that "guide RNA" can refer to either "single-chain RNA" or "two RNA." Ex. 2068, ¶¶ [168]-[170] ("In the present invention, the guide RNA may consist of two RNA, i.e., CRISPR

RNA (crRNA) and transactivating crRNA (tracrRNA) or be a single-chain RNA (sgRNA) produced by fusion of an essential portion of crRNA and tracrRNA."). And in ToolGen's 510 application original claim 1 used the generic term "guide RNA." MF 37. Original dependent claims 3 and 4 narrowed the RNA to either the dual-molecule (claim 3) or single-molecule (claim 4) RNA species within the generic "guide RNA" genus. MF 37-40; Ex. 2067, 2062, Claims 3 and 4; Ex. 2068, WO 2014/065596 ("WO 596"), Claims 3 and 4.

Sigma's and ToolGen's applications, when considered in view of Jinek 2012 and other multiple references available at the time, provide substantial evidence that POSAs at the time of the invention understood "guide RNA" to include systems with either sgRNA or dualRNA. Ex. 2464 (Seeger Decl.) ¶¶ 120-27.

(3) Broad Did Not Act As Its Own Lexicographer Nor Redefine "Guide RNA"

In the 115 Interference, the PTAB limited the term "guide RNA" to single molecule RNA primarily because of one sentence within the voluminous specification for Broad's patents. While a patentee may act as their own "lexicographer" and redefine a term contrary to its plain and ordinary meaning, "a clear and unambiguous definition" limiting the term "contrary to its plain and ordinary meaning" is required. *Bradium Techs. LLC v. Iancu*, 923 F.3d 1032, 1044 (Fed. Cir. 2019). Here, the sentence at issue is neither clear nor unambiguous.

In the detailed description of the invention, the Broad inventors state the following:

> In aspects of the invention the terms "chimeric RNA", "chimeric guide RNA", "guide RNA", "single guide RNA" and "synthetic guide RNA" are used interchangeably and refer to the polynucleotide sequence comprising the guide sequence, the tracr sequence and the tracr mate sequence.

Ex. 2011, 359 patent at 12:6-16. This statement does not "clearly express an intent" to redefine the plain and ordinary meaning of the generic term "guide RNA" to be single molecule RNA. *See Ex Parte Charles John Berg, Jr. & John David Norcom*, Appeal No. 2010-004063, 2012 WL

1744485, at *2 (B.P.A.I. Apr. 25, 2012). Instead, it only relates to the use of the term "guide RNA" "in aspects of the invention," not in the overall context of the invention or in the specification as a whole. As noted previously, multiple claims and the disclosures of preferred embodiments in the specification indicate that guide RNA may also include a dual molecule RNA system. Thus, any interpretation of this sentence to limit "guide RNA" conflicts with other aspects of the specification, the claims and examples. Because there certainly is not a "clear and unambiguous" definition provided as to "guide RNA," the plain and ordinary meaning of the term should prevail.

Additionally, when definitions are presented in Broad's patents, those definitions do not use the "in aspects of the invention" language. Instead, as shown by one example in the very next paragraph of the specification (and in multiple other paragraphs), when Broad's patents define terms they used express and non-limiting definitional language, stating that "as used herein the term [term] means [definition]." Ex. 2011, 359 patent at 12:17-20; *see also id.* at 12:21-23. This is a clear and unambiguous definition, completely unlike the qualified "aspects" language regarding "guide RNA." *See also* Ex. 2464 (Seeger Decl.) ¶¶ 132-41.

Lastly, even if the cited specification language were definitional, the language does ***not*** limit "guide RNA" to any specific RNA configuration. Rather, it recites that the term guide RNA "***refers to***" "the polynucleotide sequence comprising the guide sequence, the tracr sequence and the tracr mate sequence." Ex. 2011, 359 patent at 12:6-10. That is, to the extent the sentence is seen as definitional, it indicates "guide RNA" refers to three RNA sequences (the guide sequence, the tracr sequence, and the tracr mate sequence). But the definition (assuming *arguendo* it is a definition) indicates "guide RNA" refers to the sequences but is silent on and does not require that the guide sequence and tracr mate sequence be fused to the tracr sequence or otherwise limit the configuration of these three sequences within the "guide RNA."

When the proper BRI of "guide RNA" is determined based on the intrinsic and extrinsic evidence showing a plain and ordinary meaning and with no clear and unambiguous redefinition, "guide RNA" as used in Broad's Involved Claims includes both sgRNA and dualRNA systems.

2. Of The Large Set of Claims Not Limited To sgRNA, Only Three Require Donor Template Integration

The Sigma half of Count 1, unlike the Broad half, is generic as to the RNA configuration, but requires Donor Template Integration. Of the claims identified above as not requiring sgRNA—and thus not corresponding to the Broad half of Count 1—only three claims require Donor Template Integration as in the Sigma half of Count 1 and are not otherwise separately patentable: 445 patent, claim 13; 616 patent, claim 30; and 713 patent claim 14. *See also* Ex. 2464 (Seeger Decl.) ¶ 250. These claims should remain designated as corresponding to Count 1.

The remainder of Broad's generic claims—those that do not recite Donor Template Integration—should be designated as not corresponding to Count 1.

3. Broad's Generic/Non-Limited RNA Claims That Do Not Recite Integration Of A Donor Template Do Not Correspond To Either Half Of The Count

If this Interference is continued with Count 1, Broad's "generic" non-limited RNA claims that do not recite integration of a donor template should be designated as not corresponding to Count 1 in the interests of fairness and efficiency.

It is presumed that a "claim corresponds to a count if the subject matter of the count, treated as prior art to the claim, would have anticipated or rendered obvious the subject matter of the claim." 37 C.F.R. § 41.207(b)(2). This is not, however, a per se rule that must rigidly be applied, even if doing so would work an inequitable outcome. Ex. 2305, Rules of Practice Before the Board of Patent Appeals and Interferences, 69 FR 49960-01, at *49994 (Comment 186/Answer).

Should the Interference proceed with Count 1, failing to de-designate generic/non-limited RNA claims that do not correspond with Sigma's half of Count 1 would work an unfairness to Broad and unfairly reward Sigma for limiting its claims in prosecution.

First, Sigma has made it clear that the invention captured by its claims, including the claim representing its half of Count 1, is limited to Donor Template Integration.

In fact, in the 132 Interference, Sigma argued that claims to "cleavage alone" are "patentably distinct" from its claims directed to Donor Template Integration: Ex. 2124 (132 Sigma Mot. 1) at 1:12-13. Sigma specifically noted that there are three interferences currently pending before the Board directed to "cleavage only in a eukaryotic cell," and that

> Sigma is ***properly*** not a party to those pending 'cleavage only' interferences because all of Sigma's involved claims are directed solely to the ***patentably distinct*** 'cleavage plus integration' technological advance in the art.

Id. at 4:14-23; *id*. at 4:21-23 ("[A]ll of Sigma's involved claims are directed solely to the patentably distinct 'cleavage plus integration' technological advance in the art"). By stating Sigma is not properly a party to interferences involving the generic eukaryotic invention, Sigma acknowledges that it did not invent that subject matter. Sigma further argued that:

> using a CRISPR-Cas9 system in a eukaryotic cell to cleave DNA and *thereafter to integrate a donor polynucleotide into that cleaved DNA via HDR* is patentably distinct from (not obvious in view of) simply using a CRISPR-Cas9 system in a eukaryotic cell to cleave DNA.

Id. at 5:3-7 (emphasis in original); *see also id.* at 2:18-30, 3:1-7, 4:14-23, 5:4-7. Sigma is arguing that the involved claims of both parties in the 132 Interference be limited only to those expressly reciting Donor Template Integration. *Id.* at 27:1-8; MF 42-47.

Consistent with this position, throughout prosecution of Sigma's 204 application, Sigma argued that integration of a donor template via a CRISPR-Cas9 system in a eukaryotic cell is a distinct invention from cleavage and repair of DNA by other means. *See, e.g.*, Ex. 2074 (October

13, 2020 Applicant Remarks) at 8-9. MF 48-50. Accordingly, Sigma's claims involved in this Interference are entirely limited to donor template integration claims.

For instance, during prosecution of the involved 204 application, Sigma's claims requiring integration of a donor sequence were indicated as allowable over CVC's P1 and P2 during an Interview. During the Interview, the Examiner also suggested that Sigma remove the limitation requiring integration of a donor sequence to include cleavage only claims. Sigma responded it would not because "the Applicant has concluded that CRISPR cleavage + donor sequence integration claims, as more specifically recited in the new claims presented herein, ***are patentably distinct from CRISPR cleavage-only claims.***" Ex. 2074 (October 13, 2020 Applicant Remarks) at 8-9. Sigma thus recognized that it could not seek, in the same patent, claims to integration of a donor sequence and claims directed only to cleavage and repair, as they were separately patentable.

Similarly, in suggesting an interference with CVC during the course of prosecuting the 204 application, Sigma made clear that only claims that explicitly recite Donor Template Integration correspond to a count with such a requirement—and that claims to other forms of cleavage and repair or editing did not correspond. Specifically, in October 2020, Sigma filed a suggestion of Interference during prosecution of the 204 application, proposing a count that required Donor Template Integration. Sigma argued that only claims from CVC's applications that expressly recite Donor Template Integration would correspond to its proposed count—not claims directed to more generally "cleaving or editing" or "modulating transcription" of a gene product. Ex. 2074 (October 13, 2020 Suggestion of an Interference).

Having limited its claims to only those requiring Donor Template Integration in order to secure allowance, Sigma should not be permitted to challenge Broad's entitlement to generic RNA claims that are not so limited.

Second, because Broad's best proofs include *dual-molecule* RNA systems *without* a donor template, they are excluded by Count 1. Ex. 2464 (Seeger Decl.) ¶¶ 151-66; MF 51. Specifically, if the PTAB proceeds with Count 1, Sigma will likely argue that Broad cannot rely on its early dual-molecule RNA proofs to establish a reduction to practice for Broad's half of the Count because that half is limited to sgRNA—but that result would make no sense unless the genus and species were separately patentable inventions. Sigma will also likely argue that Broad cannot rely on those same early proofs to establish reduction to practice for Sigma's half of the Count because they do not involve Donor Template Integration. *Id*. Accordingly, Broad's earliest proofs are not covered by the first half of Count 1 because they are not single-molecule, and they are not covered by the second half of Count 1 because they do not use a donor template. Thus, while prohibited from relying on dual molecule work because of the awkward construction of Count 1, Broad's generic/non-limited and dual-molecule RNA claims still will be at risk. That should not happen.

To be clear, Broad believes that if this case proceeds to priority on Count 1, the evidence will establish that the Broad invented the sgRNA eukaryotic CRISPR invention prior to Sigma, and that it was the first to use a CRISPR-Cas9 system for Donor Template Integration into the DNA in a eukaryotic cell as well. Proceeding with Count 1 in this case with all of the currently designated claims, however, raises the specter of the potentially improper and inequitable result that Broad could be denied generic claims solely because the two halves of Count 1 import separate limitations (sgRNA; Donor Template Integration) that are not part of the fundamental breakthrough of getting any CRISPR-Cas9 system to work in eukaryotic cells.

Designating Broad's generic RNA claims as not corresponding to Count 1 is consistent with the Federal Circuit's decision in *Eli Lilly & Co. v. Bd. of Regents of Univ. of Wash.*, 334 F. 3d 1264, 1268 (Fed. Cir. 2003). There, the Federal Circuit upheld the PTAB's decision to not apply

a one-way test for determining interference-in-fact, as that would be over-inclusive in determining whether potentially interfering claims were to the "same patentable invention." *Id.* Specifically, the Federal Circuit explained, affirming the PTAB's decision, that a "genus [that] was invented before" a species is "separately patentable from, the species" for the purposes of determining an interference-in-fact. *Id.* at 1270. The Federal Circuit found that the PTAB had the discretion to adopt a two-way test to prevent "the proliferation of unnecessary, wasteful interference proceedings concluding that both parties are entitled to patents in situations in which the claimed inventions do not define the same patentable invention, but merely overlap in scope." *Id.* at 1268.

The current case is critically similar to *Eli Lilly* in that, given the complexity and unpredictability of the CRISPR-Cas9 art, the development of working species (the single molecule RNA system; the integration of donor templates) should not grant rights to the genus where the another working species (dual-molecule without integration of a donor template) was invented first. The development of eukaryotic CRISPR technology was a breakthrough technological advance for which there was no reasonable expectation of success regardless of whether the RNA components were in one or two molecules, and regardless of the use of a donor template or any other repair mechanism. In unpredictable arts, multiple species can be separately patentable.

For the foregoing reasons, if the Interference proceeds on Count 1, Broad requests that the PTAB designate its generic/non-limited RNA eukaryotic CRISPR claims that do not recite Donor Template Integration as not corresponding to Count 1.

V. CONCLUSION

For the foregoing reasons, Broad respectfully requests that the PTAB grant this motion.

Dated: December 3, 2021

Respectfully submitted,

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APPENDIX A: LIST OF EXHIBITS CITED

Ex.	Description
2001	U.S. Application 61/736,527, Zhang et al., December 12, 2012.
2011	U.S. Patent No. 8,697,359, issued on April 15, 2014, to Feng Zhang ("the 359 Patent").
2012	U.S. Patent No. 8,795,965, issued on August 5, 2014, to Feng Zhang ("the 965 Patent").
2013	U.S. Patent No. 8,895,308, issued on November 25, 2014, to Feng Zhang and Fei Ran ("the 308 Patent")
2014	U.S. Patent No. 8,906,616, issued on December 9, 2014, to Feng Zhang et al. ("the 616 Patent")
2015	U.S. Patent No. 8,771,945, issued on July 8, 2014, to Feng Zhang ("the 945 Patent")
2017	U.S. Patent No. 8,865,406, issued on October 21, 2014, to Feng Zhang and Fei Ran ("the 406 Patent")
2024	U.S. Patent No. 8,993,233, issued on March 31, 2015 to Feng Zhang et al. ("the 233 Patent")
2029	U.S. Patent No. 8,871,445, issued on October 28, 2014, to Le Cong and Feng Zhang ("the 445 Patent")
2037	U.S. Patent No. 8,932,814, issued on January 13, 2015, to Le Cong and Feng Zhang ("the 814 Patent")
2043	U.S. Patent No. 9,840,713, issued on December 12, 2017 to Feng Zhang ("the 713 Patent")
2047	U.S. Patent No. 8,999,641, issued on April 7, 2015 to Feng Zhang et al. ("the '641 Patent")
2051	U.S. Patent Application No. 14/704,551, Ex. 22 to Sanjana Declaration ("NIH application") (Ex. 2411)
2060	U.S. Patent No. 8,889,418 issued on November 18, 2014, to Feng Zhang, et al. ("the 418 Patent")
2064	U.S. Application 15/330,876, Zhang, et al., November 7, 2016 (the '876 Application)
2067	International PCT Application PCT/KR2013/009488, filed on October 23, 2013
2068	International PCT Publication WO 2014/065596 A1, filed on October 23, 2013
2074	Excerpts of U.S. Patent Application 15/456,204, Chen et al., dated March 10, 2017
2121	Paper 877, Declaration of Interference, Interference 106,015, September 10, 2020.
2124	Paper 482, Sigma Motion 1 (to Substitute Proposed Count 2 for Count 1), Interference 106,132, November 19, 2021.

Ex.	Description
2201	Cong et al., *Multiplex Genome Engineering Using CRISPR/Cas Systems*, 339(6121) SCIENCE 819-823 (2013) with Supplemental Material.
2202	Jinek et al., *A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity*, 337(6096) SCIENCE 816-821 (2012) with Supplemental Material.
2215	Sapranauskas et al., *The* Streptococcus thermophilus *CRISPR/Cas system provides immunity in* Escherichia coli, 39(21) NUCL. ACIDS RES. 9275-9282 (2011).
2226	Ran et al., *In vivo genome editing using* Staphylococcus aureus *Cas9*, 520 Nature 186-191 (2015).
2227	Nishimasu et al., *Crystal Structure of* Staphylococcus aureus *Cas9*, 162 Cell 1113-1126 (2015).
2239	Gasiunas et al., Cas9-crRNA ribonucleoprotein complex mediates specific DNA cleavage for adaptive immunity in bacteria, 109(39) PNAS e2579-86 (2012).
2258	Fieck, A., et al., Modifications of the E. coli Lac repressor for expression in eukaryotic cells: effects of nuclear signal sequences on protein activity and nuclear accumulation, Nucleic Acids Res. 20(7) 1785-1791 (1992) (Ex. 1235)
2263	Brothers, S.H., et al., Unexpected Effects of Epitope and Chimeric Tags on Gonadotropin-Releasing Hormone Receptors: Implications for Understanding the Molecular Etiology of Hypogonadotropic Hypogonadism, The Journal of Clinical Endocrinology & Metabolism 88(12):6107–6112 (2003) (Ex. 2203)
2264	Turner, J.R., et al., Carboxyl-terminal Vesicular Stomatitis Virus G Protein-tagged Intestinal Na^+-dependent Glucose Cotransporter (SGLT1), J. Biol. Chem., 271(13): 7738–7744 (1996) (Ex. 2239)
2305	Rules of Practice Before the Board of Patent Appeals and Interferences, 69 Fed. Reg. 49960-01.
2454	Declaration of Christoph Seeger, Interference 106,126, executed May 28, 2021.
2463	Declaration of Paula M. Cannon, Ph.D., Interference 106,132, dated November 19, 2021.
2464	Declaration of Christoph Seeger, executed December 3, 2021.
2465	Declaration of Paula M. Cannon, Ph.D., Application No. 15/456,204 Prosecution History, dated April 29, 2019.
2466	Declaration of Fyodor Urnov, Application No. 15/456,204 Prosecution History, dated October 17, 2017.
2793	Email from Le Cong to Feng Zhang, Shuailiang Lin, David Cox, and Michael Yim, dated August 29, 2012, with 20 page attachment, 21 pages total

APPENDIX B: STATEMENT OF MATERIAL FACTS

1. Count 1 is an "or" count drawn to (Broad half, Broad US Patent 8,697,359, claim 18) a eukaryotic CRISPR-Cas9 system comprising Cas9 and RNA that comprises a guide sequence fused to a tracr sequence that targets and hybridizes to a DNA target sequence in a eukaryotic cell (Ex. 2011, 359 patent, claim 18), or (Sigma half, Sigma application 15/456,204 ("the 204 application"), claim 31) a method for using a CRISPR-Cas9 system in a eukaryotic cell to create a double-stranded break in target DNA and integrate a donor template. Paper 12 at 13.

2. The CRISPR-Cas9 system of the Sigma half of Count 1 requires integration of a donor template; the Broad half of Count 1 requires sgRNA but does not require integration of a donor template. *Id*.

3. In the 132 Interference, Sigma submitted expert testimony that the following would not have been obvious to a POSITA as of early December 2012 in view of Sigma's proposed count in the 132 Interference: "a Cas9 protein that includes a Protein Transduction Domain ('PTD')"; "one or more mutation(s) in the Cas9 RuvC/HNH domain(s)"; and "chimeric Cas9 protein." Ex. 2124 (132 Sigma Mot. 1) at 27:11-28:2; Ex. 2465 (132 Cannon Decl.) ¶ 35.

4. Count 1 does not recite any particular ortholog of Cas9 protein, including SaCas9. Paper 1 at 12-13; Broad Motion 1 at 4.

5. There is no teaching or suggestion in Count 1 or the prior art to use SaCas9 in CRISPR-Cas9 systems in eukaryotic cells. *See* Ex. 2464 (Seeger Decl.) ¶¶ 177-96.

6. CRISPR-Cas9 systems using SaCas9 possess a combination of small size and high efficacy in eukaryotes. Ex. 2017, 406 patent, 83:1-25-84:1-23; Ex. 2464 (Seeger Decl.) ¶ 193.

7. Small size and high efficacy in eukaryotes render CRISPR-SaCas9 systems advantageous for use in eukaryotic cell-based applications where vector delivery using the highly

versatile adeno-associated virus (AAV) is favored because AAV vectors are space-constrained. Ex. 2464 (Seeger Decl.) ¶¶ 193, 245; Ex. 2017, 406 patent, 83:1-25-84:1-23.

8. As of 2012, SpCas9 was the most commonly studied Cas9 ortholog. Ex. 2464 (Seeger Decl.) ¶ 177; Ex. 2215.

9. As of 2012, more than 600 bacterial Cas9 orthologs that had been identified. Ex. 2226, Ran 2015; Ex. 2464 (Seeger Decl.) ¶ 181.

10. As of 2012, there was nothing in the art pointing the POSA to use SaCas9 in eukaryotic cells including nothing pointing to SaCas9 out of the many known "small" Cas9 orthologs that are similar in size to, or smaller than, SaCas9. Ex. 2464 (Seeger Decl.) ¶¶ 185-93.

11. SpCas9 is larger than SaCas9. *Id.* ¶¶ 178-92.

12. During prosecution of the 204 application, Sigma argued that many aspects of the eukaryotic CRISPR-Cas9 system, including those associated with the Cas9 protein, were unpredictable. *See, e.g.*, Ex. 2074 (April 29, 2019 Applicant Remarks) at 19, 27-28; Ex. 2466 (October 17, 2017 Urnov Decl.) ¶¶ 15-17.

13. Broad determined that using an CRISPR-SaCas9 system in a eukaryotic cell provides a surprising combination of benefits not taught or suggested by the prior art, namely high efficiency and small size. *See* Ex. 2464 (Seeger Decl.) ¶¶ 193-94.

14. SaCas9 is used for more therapeutic applications than any other Cas9 ortholog. *Id.* ¶ 194; *see* Ex. 2017, 406 Patent at 83:25-84:23; Exs. 2687, 2517, 2686.

15. All claims of Broad's 418 patent (Ex. 2060) require that the Cas9 is not taken from a single organism but rather is a chimeric Cas9 that includes two fragments from different Cas9. Ex. 2060 at 83:45-52; Ex. 2464 (Seeger Decl.) ¶¶ 197-202.

16. Count 1 and the prior art do not teach, suggest, or provide motivation to a POSA to

design a chimeric Cas9 that is comprised of two fragments from different organisms. Ex. 2464 (Seeger Decl.) ¶¶ 197-202; Paper 1 at 12-13; Broad Motion 1 at 4.

17. Ex. 2060, 418 patent at 4:21-25 recites "[t]hese chimeric Cas9 proteins may have a higher specificity or a higher efficiency than the original specificity or efficiency of either of the individual Cas9 enzymes from which the chimeric protein was generated."

18. The 418 patent (Ex. 2060 at 83:45-52) recites benefits of a chimeric Cas9. *See also* Ex. 2464 (Seeger Decl.) ¶¶ 201-07.

19. The Sigma half of Count 1 specifically recites that the Cas9 has "only one NLS," while the Broad half of Count 1 does not indicate anything about the Cas9 with regard to NLSs. Paper 1 at 12-13.

20. Neither Count 1 nor the prior art provides any teaching or suggestion to use two or more NLSs in a CRISPR-Cas system in a eukaryotic cell, nor was there a reasonable expectation of success in using two or more NLSs. Ex. 2464 (Seeger Decl.) ¶¶ 209-32.

21. During prosecution of the 204 application, Sigma submitted expert testimony from Dr. Cannon that modifications to the Cas9 protein such as adding an NLS could "affect Cas9 protein folding and the final protein structure" such that it "could interfere with Cas9 function; for instance, by interfering with a binding site or catalytic domain." Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 59-60.

22. During prosecution of the 204 application, Sigma argued that "[b]ecause a protein's function is inextricably linked to its folded structure, proper folding after (or during) translation is crucial" and that misfolded proteins would be subject to degradation in the eukaryotic environment. Ex. 2074 (April 29, 2019 Applicant Remarks) at 30.

23. During prosecution of the 204 application, Sigma argued that "when a protein is

expressed as a fusion, such as with an NLS or epitope or chimeric tag, there are unexpected results, further confirming that there was no reasonable expectation of success as to a eukaryotic CRISPR-Cas9 system wherein the Cas9 includes one or more NLSs." Ex. 2074 (April 29, 2019 Applicant Remarks) at 27 (citing Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 76-77).

24. Broad determined that, unexpectedly, the use of two or more NLSs resulted in CRISPR-Cas9 systems with significantly improved localization to the nucleus, as compared to systems lacking two or more NLSs. *See generally* Ex. 2464 (Seeger Decl.) ¶¶ 202-21; Ex. 2001, Zhang B1, Figure 1B; Ex. 2201, Cong 2013, Figure 1A; Ex. 2793.

25. A POSA would have understood that adding amino acids to a protein such as Cas9 could alter its folding affecting its structure and function in ways that were not predictable. Ex. 2464 (Seeger Decl.) ¶¶ 216-17, 222-28; Ex. 2258 at 1785-1790; Exs. 2263-2265; Ex. 2465 (April 29, 2019 Cannon Decl.) ¶¶ 59-60 (one could not "predict with any reasonable certainty whether ***any*** functional domain… will be properly exposed when expressed as a fusion with Cas9"); *id*. ¶¶ 59-60.

26. Count 1 does not recite that the Cas9 is fused to specified protein domains. Paper 1 at 12-13; Broad Motion 1 at 4.

27. There is no teaching or suggestion in Count 1 or the prior art to modify the naturally occurring Cas9 to include protein domains. Ex. 2464 (Seeger Decl.) ¶¶ 233-40.

28. The 233 patent notes that there is a benefit to fusing functional domains to Cas9, as doing so can "to turn the Cas9/gRNA CRISPR system into a generalized DNA binding system [which] can execute functions beyond DNA cleavage." Ex. 2024 at 73:22-37; Ex. 2464, ¶ 239.

29. Broad's 713 patent (Ex. 2043), includes claims, including claims 15-26 and 28-41, that are not limited to single-molecule RNA and do not contain the term "guide RNA." *See* Ex.

2464 (Seeger Decl.) ¶¶ 114-18.

30. Independent claim 1 and dependent claims 2-24 of Broad's 418 patent (Ex. 2060) do not recite any RNA component. *See* Ex. 2464 (Seeger Decl.) ¶¶ 116-17.

31. The 308 patent, claim 1 describes a method using CRISPR-Cas system having "guide RNA" that hybridizes to the target sequence and claim 6 covers the "method of claim 1, wherein the guide RNA comprises a guide sequence and a tracr sequence." Ex. 2013.

32. The 616 patent, claim 1 describes a CRISPR-Cas system having a "guide RNA" polynucleotide sequence comprising a guide sequence, a tracr mate sequence, and a tracr sequence, claim 2 covers the "composition of claim 1, wherein the modified guide RNA comprises a chimeric guide sequence and a tracr sequence," and claim 5 covers the "composition of claim 1, wherein the modification comprises fusing the tracr mate sequence and the tracr sequence through an artificial loop." Ex. 2014.

33. In the 965 patent (Ex. 2012), none of the dependent claims include limitations that the components of the guide RNA be fused or bound to one another in a chimeric manner.

34. The Broad patents disclose preferred embodiments that are dual-molecule RNA systems. Ex. 2011, 359 patent at 43:49-53, 44:5-8; Ex. 2464 (Seeger Decl.) ¶¶ 138-41.

35. Sigma's 204 specification explains that "[i]n some embodiments, the guide RNA comprises a single molecule…" and "[i]n other embodiments, the guide RNA can comprise two separate molecules." Ex. 2074 (204 specification) at [0077].

36. Sigma's 204 application, claim 31 uses the generic term "guide RNA," while dependent claims 33 and 34 specify that the "guide RNA is a single molecule" and "the guide RNA is two molecules" respectively. Paper 12 at 2.

37. ToolGen's patent applications explicitly define "guide RNA" to encompass both

dual- and single-molecule RNA configurations:

> In the present invention, ***the guide RNA may consist of two RNA***, i.e., CRISPR RNA (crRNA) and transactivating crRNA (tracrRNA) ***or be a single-chain RNA*** (sgRNA) produced by fusion of an essential portion of crRNA and tracrRNA.

Ex. 2068, ¶¶ [168]-[170]; Ex. 2067, ToolGen PCT, ¶¶ [0168]-[0169]; Ex. 2062, 510 application, ¶¶ [0094]-[0095].

38. In the original claims of the ToolGen PCT application and the 510 application, the inventors included claims reciting "guide RNA," without any restriction as to RNA configuration. Ex. 2067, ToolGen PCT; Ex. 2062, 510 application original claims.

39. In the original claims of the ToolGen PCT application and the 510 application, the inventors included claims 3 and 4, which respectively limited that "guide RNA" to a dualRNA (a dual molecule RNA) and a "single-chain" guide RNA (a single molecule RNA). *Id*.

40. In the disclosures from Ex. 2067 and 2062 referenced in MFs 36 and 37, ToolGen used the term "guide RNA" consistent with Jinek 2012's use of the term to include both sgRNA and dualRNA.

41. Jinek 2012 states: "In this ternary complex, the dual tracrRNA:crRNA structure acts as guide RNA that directs the endonuclease Cas9 to the cognate target DNA." Ex. 2202, Jinek 2012, at Figure S1 description.

42. Sigma moved to change the count in the 132 Interference. Ex. 2124.

43. In the 132 Interference, the current count is an "or" count directed on one hand (the Sigma half) to Donor Template Integration, and on the other hand (the CVC half), to Non-Template activity. *Id*.

44. Sigma stated in Motion 1 in the 132 Interference that "using a CRISPR-Cas9 system in a eukaryotic cell to cleave DNA and thereafter to integrate a donor polynucleotide into that

cleaved DNA via HDR is patentably distinct from (not obvious in view of) simply using a CRISPR-Cas9 system in a eukaryotic cell to cleave DNA." *Id.* at 5:3-7.

45. Sigma stated in Motion 1 in the 132 Interference that "in early December 2012 a POSITA would not have had a reasonable expectation that such a process in a CRISPR-Cas9 system would be successful in eukaryotic cells." *Id*. at 6:23-29.

46. In Motion 1 in the 132 Interference, Sigma identified Donor Template Integration claims as the only involved claims corresponding to 132 Interference Proposed Count 2. *Id.* at 27:1-8.

47. In Motion 1 in the 132 Interference, Sigma stated that there are three interferences pending before the PTAB directed to Non-Template CRISPR-Cas9 activity in a eukaryotic cell, but that "Sigma is ***properly*** not a party to those pending 'cleavage only' interferences because all of Sigma's involved claims are directed solely to the patentably distinct 'cleavage plus integration' technological advance in the art." *Id.* at 4:14-23.

48. Sigma has argued that integration of a donor template via a CRISPR-Cas9 system in a eukaryotic cell is a distinct invention from cleavage and repair of DNA by other means. *See, e.g.*, Ex. 2074 (October 13, 2020 Applicant Remarks) at 8-9.

49. In October 2020, Sigma filed a suggestion of Interference during prosecution of the 204 application for CVC's 680 application, proposing a Count that required integration of a donor template. Ex. 2074 (October 13, 2020 Suggestion of an Interference).

50. Sigma argued that only claims from CVC's applications that expressly recite donor template integration would correspond to its proposed Count. *Id*.

51. Broad's best proofs include dual-molecule RNA systems without a donor polynucleotide. Ex. 2464 (Seeger Decl.) ¶¶ 20, 151-66.

APPENDIX C: CHART OF GROUNDS AND CLAIMS

Patent	Claims Currently Designated	SaCas9	Chimeric Cas9	Two or more NLSs	Cas9 Domains	No RNA at all	Generic RNA	sgRNA or donor template remaining designated	Donor template
8,867,359	1-20						1-3, 5-10, 12-17, 19-20	4, 11, 18	
8,771,945	1-29						1-4, 6-29	5	
8,795,965	1-30							ALL 1-30	
8,865,406	1-30	ALL					ALL		
8,871,445	1-30			ALL			1-12, 14-30		13
8,889,356	1-30						ALL		2, 4
8,889,418	1-28		ALL			1-24	25-28		24, 26-28
8,895,308	1-30	ALL					1-9, 11-28		15, 26
8,906,616	1-30						1, 3-4, 6-29	2, 5, 30	30
8,932,814	1-30			ALL			ALL		2, 14
8,945,839	1-28						ALL		
8,993,233	1-43			7	ALL		ALL		
8,999,641	1-28				ALL		ALL		
9,840,713	1-41				18-19, 25, 29-30, 36		1-7, 10-13, 15, 17-26, 28-41	8-9, 14, 16, 27	14
14/704,551	2, 4-18			9-11			2, 4-13, 17-18	14-16	14-16
15/330,876	1, 16-21, 30-40	ALL		34	21		1, 16-21, 30-39		

APPENDIX D: CLAIM DIFFERENTIATION EXAMPLES

U.S. Patent 8,895,308	
Claim 1: Describes method using CRISPR-Cas system having "a CRISPR-Cas system guide RNA that hybridizes with the target sequence" 1. A method of altering expression of at least one gene product in a eukaryotic cell containing and expressing a DNA molecule having a target sequence and encoding said gene product comprising introducing into said eukaryotic cell an engineered, non-naturally occurring Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-CRISPR associated (Cas) system comprising one or more vectors comprising: a) a first regulatory element operable in a eukaryotic cell operably linked to at least one nucleotide sequence encoding a CRISPR-Cas system guide RNA that hybridizes with the target sequence, and b) a second regulatory element operable in a eukaryotic cell operably linked to a nucleotide sequence encoding a Staphylococcus aureus Cas9 protein, wherein the CRISPR-Cas system further comprises one or more nuclear localization signal(s) (NLS(s)), and components (a) and (b) are located on same or different vectors of the system, whereby the guide RNA targets the target sequence and the Cas9 protein cleaves the DNA molecule; the method further comprising inserting DNA into a cleaved strand of the DNA molecule; whereby expression of the at least one gene product is altered; and, wherein the Cas9 protein and the guide RNA do not naturally occur together.	**Claim 6**: The method of claim 1, wherein the guide RNA comprises a guide sequence and a tracr sequence. **Claim 10**. The method of claim 6, wherein the guide sequence and tracr sequence are chimeric.

U.S. Patent 8,906,616	
Claim 1: Claim 1 describes a CRISPR-Cas system having a "guide RNA" polynucleotide sequence comprising a guide sequence, a tracr mate sequence, and a tracr sequence. 1. An engineered, non-naturally occurring composition comprising a Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)-CRISPR associated (Cas) (CRISPR-Cas) system having a guide RNA polynucleotide sequence, wherein the polynucleotide sequence comprises (a) a guide sequence capable of hybridizing to a target sequence in a eukaryotic cell, (b) a tracr mate sequence, and (c) a tracr sequence wherein (a), (b) and (c) are arranged in a 5' to 3' orientation, wherein when transcribed, the tracr mate sequence hybridizes to the tracr sequence and the guide sequence directs sequence-specific binding of a CRISPR complex to the target sequence, wherein the to CRISPR complex comprises a Type II Cas9 protein complexed with (1) the guide sequence that is hybridized to the target sequence, and (2) the tracr mate sequence that is hybridized to the tracr sequence, wherein in the polynucleotide sequence, one or more of the guide, tracr and tracr mate sequences are modified.	**Claim 2**: The composition of claim 1, wherein the modified guide RNA comprises a chimeric guide sequence and a tracr sequence. **Claim 5:** The composition of claim 1, wherein the modification comprises fusing the tracr mate sequence and the tracr sequence through an artificial loop.

U.S. Patent 8,697,359	
Claim 1: Claim a method using CRISPR-Cas system having a "guide RNA RNA that hybridizes with the target sequence." A method of altering expression of at least one gene product comprising introducing into a eukaryotic cell containing and expressing a DNA molecule having a target sequence and encoding the gene product an engineered, non-naturally occurring Clustered Regularly Interspaced Short Palindromic Repeats (CRISPR)--CRISPR associated (Cas) (CRISPR-Cas) system comprising one or more vectors comprising: a) a first regulatory element operable in a eukaryotic cell operably linked to at least one nucleotide sequence encoding a CRISPR-Cas system guide RNA that hybridizes with the target sequence, and b) a second regulatory element operable in a eukaryotic cell operably linked to a nucleotide sequence encoding a Type-II Cas9 protein, wherein components (a) and (b) are located on same or different vectors of the system, whereby the guide RNA targets the target sequence and the Cas9 protein cleaves the DNA molecule, whereby expression of the at least one gene product is altered; and, wherein the Cas9 protein and the guide RNA do not naturally occur together.	**Claim 4**: The method of claim 1, wherein the guide RNAs comprise a guide sequence fused to a trans-activating cr (tracr) sequence.

CERTIFICATE OF FILING AND SERVICE

I hereby certify that on December 3, 2021, a true and complete copy of the foregoing **BROAD MOTION 3** (to designate claims as not corresponding to Count 1) is being filed and served by 8:00 pm ET via the Interference Web Portal (SO ¶ 105.3; Paper 27 at 11). Service copies are being sent by email by 11:00 pm ET to counsel for Senior Party as follows:

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