UNITED STATES PATENT AND TRADEMARK OFFICE

BEFORE THE BOARD OF PATENT APPEALS
AND INTERFERENCES

Ex parte GHAZALA HASHMI and MICHAEL SEUL[1]

Appeal 2008-5415
Application 11/439,697
Technology Center 1600

Decided: December 22, 2008

Before JAMES T. MOORE, *Vice Chief Administrative Patent Judge*, and RICHARD TORCZON and SALLY G. LANE, *Administrative Patent Judges*.

MOORE, *Vice Chief Administrative Patent Judge*.

DECISION ON APPEAL

STATEMENT OF CASE

The Appellants appeal under 35 U.S.C. § 134 (2002) from a final rejection of claims 45-48.[2] We have jurisdiction under 35 U.S.C. § 6(b) (2002).

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[1] The real party in interest is BioArray Solutions Ltd. (App. Br. 2).
[2] Claims 1-44 and 49-61 have been canceled. (App. Br. 2).

The Appellants' claims are directed to methods of identifying genes associated with disease by differentiating homozygous, heterozygous and wild-type alleles[3] in a target sample.

The specification describes that "[a] number of mutations and polymorphisms are significant only if they are homozygous, and therefore, to be useful in such cases, the assay must be capable of discriminating heterozygotes from homozygotes." (Specification p. 2, ll. 11-14).

For example, cystic fibrosis ("CF") is a recessive disorder that develops when two copies of the abnormal gene, i.e., the CF transmembrane conductance regulator gene, are present. (*See id*. p. 2, ll. 18-21). Therefore, genomic analysis for a recessive disorder such as CF requires determining not merely whether a mutation or polymorphism exists in an allele of a "target sample," but whether the sample is "homozygous for mutant or polymorphic alleles," indicating the existence of the disease.

The specification further describes that "[i]n a multiplexed hybridization assay, cross-hybridization among mis-matched, but closely homologous, probes and amplicons can generate false positive signals." (*Id*. p. 2, ll. 9-11). Therefore, according to the specification, an effective assay of a target sample "should also be designed to minimize such effects." (*Id*. p. 2, l. 11).

Claim 45 is the only independent claim in the application. The Appellants do not argue any claims or rejections separately. Therefore, we select independent claim 45 to decide the appeal. See 37 C.F.R. §

[3] An allele is one of a group of genes that occur at a given location.

41.37(c)(1)(vii)(2006). Accordingly, the remaining claims stand or fall with claim 45.

Claim 45 reads as follows:

> 45. A method of differentiating homozygous, heterozygous and wild-type alleles in a target sample comprising mutant or polymorphic alleles or wild-type alleles using results obtained from a probe array, wherein individual probes in the array are designed to detect designated mutant or polymorphic alleles or wild-type alleles through hybridization of said probes and targets, where the results compensate for hybridization between probes and targets other than their designated targets, comprising:
>
> amplifying the genomic regions in the target sample that are predicted to include said mutant or polymorphic alleles or said wild-type alleles, thereby producing
>
> a first set of labeled amplicons having a region complementary to a subsequence of said mutant or polymorphic alleles (if said mutant or polymorphic alleles are present in said target sample) and
>
> a second set of labeled amplicons having a region complementary to a subsequence of said wild-type alleles (if said wild-type alleles are present in said target sample);
>
> providing an array of probe pairs, the first member of each pair including a complementary subsequence to a subsequence of a first-set amplicon and the second member of each pair including a complementary subsequence to a subsequence of a second-set amplicon;
>
> contacting under hybridizing conditions said array of probe pairs with said amplicons;
>
> detecting hybridization between probes and said amplicon sets based on the presence of signals from the labeled, hybridized amplicons, said signals being corrected to adjust for hybridization between probes and amplicons other than their designated amplicons as follows:

> (i) determining the intensity of signals from hybridization of first-set amplicons and from hybridization of second-set amplicons, as corrected for background signals, and
>
> (ii) determining the ratio of said signals, wherein the ratio of the intensity of first-set amplicon signals to the intensity of second-set amplicon signals is designated as a first ratio (a), and wherein the ratio of the intensity of second-set amplicon signals to the intensity of first-set amplicon signals is designated as second ratio (b); and defining three relative ranges of values for said ratios such that:
>
> > (i) the lowest range of ratio (a) indicates that said target sample is homozygous for wild-type alleles and the lowest range of ratio (b) indicates that said target sample is homozygous for mutant or polymorphic alleles,
> >
> > (ii) a middle range of either ratio (a) or (b) indicates that said target sample is heterozygous, and
> >
> > (iii) the highest range of ratio (a) indicates that said target sample is homozygous for mutant or polymorphic alleles and the highest range of ratio (b) indicates that said target sample is homozygous for wild-type alleles.

(Additional indentation added, see 37 C.F.R. §1.75(i))(Reference numerals added).

THE EVIDENCE

The Examiner relies upon the following as evidence in support of the rejections:

Carrico	US 5,200,313	Apr. 06, 1993
Chan	US 2002/0119455 A1	Aug. 29, 2002
Rowlen	US 2006/0286570 A1	Dec. 21, 2006
Wang	US 2007/0009954 A1	Jan. 11, 2007
Yasuno	US 2007/0031829 A1	Feb. 08, 2007
Barany	US 2007/0042419 A1	Feb. 22, 2007
Choi	US 2007/0042400 A1	Feb. 22, 2007

Zhang, et al., *Reconstruction of DNA Sequencing by Hybridization*. Bioinformatics 19:14-21(2003).

THE REJECTION

The following rejection is before us for review:

Claims 45-48 stand rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the enablement requirement.

We REVERSE.

ISSUE

Have the Appellants established that the Examiner erred in determining that that the specification does not provide sufficient guidelines to enable of one of ordinary skill in the art to practice the claimed invention without undue experimentation?

FINDINGS OF FACT

The record supports the following findings of fact by a preponderance of the evidence.

1. Zhang describes,with respect to DNA sequencing, "that the hybridization process is prone to errors and that the future of DNA sequencing by hybridization is predicated on the ability to successfully cope with such errors." (Zhang p. 14).

2. Chan describes that "[i]mperfect hybridization has led to difficulties in generating adequate sequence [and that e]rror in hybridization is amplified many times." (Chan [0018]).

3. Carrico describes that "the extent and specificity of hybridization is affected by" the purity of the nucleic acid preparation, the length of homologous base sequences, ionic strength of the incubation solution, incubation temperature, nucleic acid concentration and incubation time,

denaturing reagents, the presence of volume exclusion agents, and washing or rinsing hybridization product. (Carrico 12:5-46).

4. Barany describes, "For allele-specific oligonucleotide hybridization ("ASO"), the mutation must be known, hybridization and washing conditions must be known, cross-reactivity is difficult to prevent, closely-clustered sites due to interference of overlapping primers cannot undergo multiplex detection, and mutant DNA cannot be detected in less than 5% of background of normal DNA." (Barany [0036]).

5. Choi describes, "In conventional methods of preparing nucleic acid, polysaccharides such as starch often co-precipitate with nucleic acid... [making it] difficult to manipulate nucleic acids by amplification methods... or by ... hybridization detection." (Choi [0035]).

6. Choi also describes that "[p]olysaccharides may also inhibit digestion with restriction endonucleases and other enzymatic manipulations." (*Id*.).

7. Yasuno describes that while "…DNA hybridization is sequence-specific, it is difficult to completely exclude hybridizations towards very similar nucleotide sequences." (Yasuno [0037]).

8. Rowlen describes that "[d]etection of molecular recognition events such as DNA hybridization, antibody-antigen interactions, and protein-protein interactions becomes increasingly difficult as the number of recognition events to be detected decreases." (Rowlen [0004]).

9. The instant specification describes that "[i]n a multiplexed hybridization assay, cross-hybridization among mis-matched, but closely

homologous, probes and amplicons can generate false positive signals." (Specification, p. 2, ll. 9-11).

10. According to the specification, an effective assay of a target sample "should be designed to minimize such effects." (*Id*. p. 2, l. 11).

11. The specification describes that "[a] number of mutations and polymorphisms are significant only if they are homozygous, and therefore, to be useful in such cases, the assay must be capable of discriminating heterozygotes from homozygotes." (Specification p. 2, ll. 11-14).

12. The specification also describes that cystic fibrosis ("CF") is a recessive disorder associated with the CF transmembrane conductance regulator gene. (*Id*. p. 2, ll. 18-21).

13. The specification describes that a "method to enhance the reliability of hybridization-mediated multiplexed analysis of polymorphisms (hMAP) is to determine the ratio of the signals generated by the capture of the target matched and mismatched probes and to set relative ranges of values indicative of normal and heterozygous or homozygous variants." (Id. p. 6, ll. 13-17).

14. The specification also describes that "[p]robes used in the detection of mutations in a target sequence hybridize with high affinity to amplicons generated from designated target sites, when the entire amplicon, or a subsequence thereof, is fully complementary ("matched") to that of the probe, but hybridize with a lower affinity to amplicons which have no fully complementary portions ("mismatched")." (Id. p. 10, ll. 26-27 to p. 11, ll. 1-3).

15. The specification describes an example in which a number of patient samples were amplified then analyzed using conventional dot blot hybridization methods and the methods and reagents of the claimed invention. (Id. p. 35, ll. 13-20).

16. The specification describes that a comparison of the results for the conventional dot blot method for detecting mutations and wild type alleles and the results for the claimed method showed 100% "concordance." (Id. pp. 35-36 and Table V).

PRINCIPLES OF LAW

"That some experimentation is necessary does not constitute a lack of enablement; the amount of experimentation, however, must not be unduly extensive." *Amgen, Inc. v. Chugai Pharmaceutical Co., Ltd.*, 927 F.2d 1200, 1212 (Fed Cir. 1991). Factors considered in determining whether undue experimentation would have been required to practice the claimed invention include "(1) the quantity of experimentation necessary, (2) the amount of direction or guidance presented, (3) the presence or absence of working examples, (4) the nature of the invention, (5) the state of the prior art, (6) the relative skill of those in the art, (7) the predictability or unpredictability of the art, and (8) the breadth of the claims." *In re Wands*, 858 F.2d 731, 737 (Fed. Cir. 1988).

ANALYSIS

Claims 45-48 stand rejected under 35 U.S.C. § 112, first paragraph, as failing to comply with the enablement requirement. (Final Rejection, Oct. 26, 2007, p. 2). The representative claim, claim 45, recites in part,

> A method of differentiating homozygous, heterozygous and wild-type alleles in a target sample comprising mutant or polymorphic alleles or wild-type alleles using results obtained from a probe array, wherein individual probes in the array are designed to detect designated mutant or polymorphic alleles or wild-type alleles through hybridization of said probes and targets....

(App. Br. 5, Claims Appendix).

The Examiner's Rejection

Based on the factors set forth in *In re Wands*, 858 F.2d 731, 737 (Fed. Cir. 1988), the Examiner found that the claims contain subject matter which was not described in the specification in such a way as to enable one skilled in the art to which it pertains, or with which it is most nearly connected, to make and/or use the invention. (Final Rejection, Oct. 26, 2007, pp. 3-9).

In particular, the Examiner found the prior and post-filing art teach "numerous problems confronting those of ordinary skill in the art[]" relating to the claimed invention that "have not been addressed by the instant disclosure." (*Id*. 4).

Specifically, the Examiner found that Zhang teaches "that the hybridization process is prone to errors and that the future of DNA sequencing by hybridization is predicated on the ability to successfully cope with such errors." (*Id*.)(quoting Zhang p. 14). Similarly, the Examiner found that Chan teaches that "[i]mperfect hybridization has led to difficulties in generating adequate sequence [and that e]rror in hybridization is amplified many times." (Final Rejection, p. 4)(citing Chan [0018]).

More specifically, the Examiner found that Carrico describes that "the extent and specificity of hybridization is affected by ... principal conditions,"

including, the purity of the nucleic acid preparation, the length of homologous base sequences, ionic strength of the incubation solution, incubation temperature, nucleic acid concentration and incubation time, denaturing reagents, the presence of volume exclusion agents, and washing or rinsing hybridization product. (Final Rejection, pp. 4-6)(citing Carrico 12:5-46).

The Examiner further relied upon Barany for teaching that "[f]or allele-specific oligonucleotide hybridization ("ASO"), the mutation must be known, hybridization and washing conditions must be known, cross-reactivity is difficult to prevent, closely-clustered sites due to interference of overlapping primers cannot under multiplex detection, and mutant DNA cannot be detected in less than 5% of background of normal DNA." (Final Rejection, p. 6)(citing Barany [0036]).

The Examiner also referenced Choi for the teaching that "[i]n conventional methods of preparing nucleic acid, polysaccharides such as starch often co-precipitate with nucleic acid... [making it] difficult to manipulate nucleic acids by amplification methods... or by ... hybridization detection." (Final Rejection, p. 6)(citing Choi [0035]). The Examiner also found that Choi teaches that "[p]olysaccharides may also inhibit digestion with restriction endonucleases and other enzymatic manipulations." (*Id.*).

The Examiner found that Yasuno describes that while "DNA hybridization is sequence-specific, it is difficult to completely exclude hybridizations towards very similar nucleotide sequences." (Final Rejection, p. 6)(citing Yasuno [0037]).

Additionally, the Examiner found that Rowlen teaches that "[d]etection of molecular recognition events such as DNA hybridization, antibody-antigen interactions, and protein-protein interactions becomes increasingly difficult as the number of recognition events to be detected decreases." (Final Rejection, p. 7)(citing Rowlen [0004]). The Examiner particularly "noted that the claimed method places no lower limit on the ability to accurately and reproducibly detect any binding between polymer and unit specific markers." (Final Rejection, p. 7).

The Examiner found that the issues described in these references "directly impact the enablement of the claimed invention." (*Id*.). The Examiner further determined that "the instant disclosure fails to identify how these art-recognized issues are to be overcome such that the full scope of the invention can be practiced without the public having to resort to undue experimentation." (*Id*.).

The Examiner additionally found that the claimed method requires imposing a correction factor for signals that may result from probes hybridizing with an unintended target. (*Id*. 8). However, according to the Examiner, "[t]he specification is silent as to how one is to compensate for hybridization between probe and a complementary sequence when the binding energy for intended target/amplicon is the same as that for the unintended target/amplicon." (*Id*.).

Consequently, based on the scope of the claims, the limited guidance provided by the specification, the state of the prior art and the unpredictable nature of the art to which the claimed invention is directed, the Examiner determined that the claims are not enabled by the disclosure. (*Id*. 9).

The Appellants' Contentions

The Appellants challenge the Examiner's enablement rejection of claims 45-48 by asserting that some of the references cited by the Examiner " actually present their respective solutions to the 'problems' with sequencing by hybridization or genotyping (e.g., Zhang et al.'s algorithm for solving the problems in sequencing by hybridization) and this is itself evidence of enablement." (Reply Br. 1-2). The Appellants further assert that "[i]f there really is a problem [in sequencing or genotyping by hybridization], one skilled in the art could adopt the solutions proposed in one or more of the cited references." (*Id*. 2).

On balance, we find that the evidence of record supports the Appellants. In rejecting claims under 35 U.S.C. § 112, first paragraph, as failing to comply with the enablement requirement, the Examiner has the intial burden of establishing why the scope of protection provided by a claim is not adequately enabled by the disclosure. *In re Wright*, 999 F.2d 1557, 1562 (Fed. Cir. 1993).

Here, while the Examiner has described a number of references addressing "problems confronting those of ordinary skill in the art" relating to the subject of claimed invention, the Examiner has not established that these problems actually impacted the claimed invention in such a way that one skilled in the art could not resolve the problems without undue experimentation.

Indeed, as the Appellants have asserted, in some instances, the references themselves provide solutions to the problems which they address.

Moreover, the specification describes an example demonstrating that the claimed method, as taught by the specification, is effective for its intended purpose, that is, detecting homozygous wild type, heterozygous and homozygous mutant alleles.

Specifically, the specification describes an example in which a number of patient samples were amplified then analyzed using conventional dot blot hybridization methods and the methods and reagents of the claimed invention. (Id. p. 35, ll. 13-20). According to the specification, a comparison of the results of the conventional dot blot method for detecting mutations and wild type alleles with the results of the claimed method showed 100% concordance. (Id. pp. 35-36 and Table V).

The effectiveness of the claimed method, evidenced by these comparative results, reasonably supports a conclusion that the method described in the specification (by which the results were achieved) is enabled.

Without establishing that the disclosure fails to teach skilled artisans how to make and use the full scope of the claimed invention without undue experimentation, the Examiner has not satisfied the initial burden of proof required to reject the claims.

Consequently, we find that the Examiner erred in rejecting the claims for lack of enablement.

Accordingly, we reverse the Examiner's rejections.

CONCLUSION OF LAW

On the record before us, the Examiner has not established a prima facie case for rejecting the claims for lack of enablement.

DECISION

The Rejection of claims 45-48 under 35 U.S.C. § 112, first paragraph, as failing to comply with the enablement requirement is REVERSED.

REVERSED

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