PATENT ASSIGNMENT COVER SHEET

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Stylesheet Version v1.2

EPAS ID: PAT3763683

SUBMISSION TYPE:	NEW ASSIGNMENT
NATURE OF CONVEYANCE:	ASSIGNMENT

CONVEYING PARTY DATA

Name	Execution Date
UAB RESEARCH FOUNDATION	01/14/2016

RECEIVING PARTY DATA

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PROPERTY NUMBERS Total: 1

Property Type	Number
Application Number:	14116902

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DATE SIGNED:	03/02/2016

Total Attachments: 15
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ASSIGNMENT
(Patent Application)

Patent Applications	Entitled: Method, Systems and Computer Program Products for Medical Brain Imaging Analysis filed with the U.S. Patent and Trademark Office: May 11, 2012; January 23, 2014 assigned serial no: PCT/US2012/37591; 14/116,902
Assignees	Frank Skidmore

The UAB Research Foundation, a corporation of the State of Alabama, having a principal place of business at 701 20th Street South, AB Suite 770, Birmingham, AL 35233 ("UABRF"), owns all right, title and interest in certain inventions and improvements disclosed in the above referenced patent applications.

For valuable consideration, UABRF:

1. agrees to assign, hereby assigns, and has assigned to the Assignee(s), the entire right, title, and interest in and to:

   (a) all intellectual property (including, without limitation, any innovation, information, invention, discovery, product, process, work or design) disclosed, embodied, shown, or claimed in the above-referenced patent applications, implicitly or explicitly;

   (b) the above-referenced patent applications, the right to claim priority to the above-referenced patent applications, all applications based in whole or in part upon the above-referenced patent applications, including, without limitation, all applications that are a provisional, non-provisional, design, divisional, continuation, continuation-in-part, registration, utility model, industrial design, reissue, renewal, substitute, extension, reexamination, post-grant review, inter partes review, supplemental examination or non-U.S. patent application or application for other rights based in whole or in part on the above-referenced patent applications;

   (c) all patents (including, without limitation, all U.S. and non-U.S. patents, registrations, utility models, industrial designs, design patents, counterparts, continuations, continuations-in-part, divisionals, reissues, renewals, substitutes, extensions, reexaminations, post-grant reviews, inter partes reviews and supplemental examinations) that are granted or issued upon, or that claim priority to, any and all applications described in (b) of this paragraph or that disclose or claim intellectual property described in (a) of this paragraph, in whole or in part; and

   (d) all claims for damages by reason of past infringement of any rights under the applications or patents described in (a), (b) or (c) of this paragraph (including provisional rights to reasonable royalties pursuant to 35 U.S.C. §154(d)) and the right to sue for and collect such damages and royalties for Assignee's own use.

2. authorizes and requests the U.S. Patent and Trademark Office or any other U.S. or non-U.S. agency to issue to the Assignee any and all patents, or other rights or documents, resulting from the intellectual property, patent applications and patents described in Section 1 of this Assignment;

3. agrees to sign all papers and documents, including without limitation, applications, declarations, oaths and petitions, and, at the Assignee's expense, perform any other acts that are necessary in connection with

prosecution of patent applications or intellectual property described in Section 1 of this Assignment and the enforcement of patents or other rights resulting from such patent applications or intellectual property;

4. agrees that the terms, covenants, and conditions of this Assignment shall inure to the benefit of the Assignee, his successors, assigns and other legal representatives, and shall be binding upon UABRF, as well as its successors and assigns; and

5. promises and affirms that it has not entered, and will not enter, into any assignment, contract, or understanding that conflicts with this Assignment.

Signature: Kathy Nugent Date: 1/14/16

Kathy Nugent, Ph.D.
Managing Director, UAB Research Foundation

STATE OF ALABAMA
JEFFERSON COUNTY

I, Karen Songer, a Notary Public in and for said County in said State, hereby certify that Kathy Nugent, Ph.D., whose name as Managing Director of the UAB Research Foundation, a corporation, is signed to the foregoing Assignment, and who is known to me, acknowledged before me on this day that, being informed of the contents of the Assignment, she, as such officer and with full authority, executed the same voluntarily for and as the act of said corporation.

Given under my hand this the 14th day of January, 2016.

(Seal)

NOTARY PUBLIC
My Commission Expires:

KAREN O. SONGER
My Commission Expires
March 20, 2018

Assignee hereby acknowledges receipt of the entire right, title, and interest in and to the intellectual property, patent applications and rights referenced in Section 1 above and which is owned by UABRF.

Signature: _____ Date: 01/14/2015

Name: Frank Skidmore

Witness: _____ Date: 1/14/15

Name:

ASSIGNMENT OF UABRF INTELLECTUAL PROPERTY DISCLOURES

UABRF IPD #	U2012-0045
UABRF IPD Entitled	Iterative imprinting of three dimensional brain maps
Assignees	Frank Skidmore

The UAB Research Foundation, a corporation of the State of Alabama, having a principal place of business at 701 20th Street South, AB Suite 770, Birmingham, AL 35233 ("UABRF"), owns all right, title and interest in certain inventions disclosed in the above referenced intellectual property disclosure ("IPD"), a copy of which is/are attached as Schedule 1 to this Assignment.

For valuable consideration, UABRF:

1. agrees to assign, hereby assigns, and has assigned to the Assignees, the entire right, title, and interest in and to all intellectual property (including, without limitation, any innovation, information, invention, discovery, product, process, work or design) disclosed, embodied, or shown in the above-referenced IPD, implicitly or explicitly, but excluding any intellectual property that has previously been disclosed to UABRF, other than in the IPD referenced above;

2. at the Assignee's expense, agrees to sign all papers and documents and perform any other acts that are necessary to perfect Assignee's title in the above referenced intellectual property;

3. agrees that the terms, covenants, and conditions of this Assignment shall inure to the benefit of the Assignee, his successors, assigns and other legal representatives, and shall be binding upon UABRF, as well as its successors and assigns; and

4. promises and affirms that it has not entered, and will not enter, into any assignment, contract, or understanding that conflicts with this Assignment.

Signature: Kathy Nugent Date: 1/14/16

Kathy Nugent, Ph.D.
Managing Director, UAB Research Foundation

STATE OF ALABAMA
JEFFERSON COUNTY

I, Karen Songer, a Notary Public in and for said County in said State, hereby certify that Kathy Nugent, Ph.D., whose name as Managing Director of the UAB Research Foundation, a corporation, is signed to the foregoing Assignment, and who is known to me, acknowledged before me on this day that, being informed of the contents of the Assignment, she, as such officer and with full authority, executed the same voluntarily for and as the act of said corporation.

Given under my hand this the 14th day of January, 2016.

(Seal)

NOTARY PUBLIC
My Commission Expires:

KAREN D. SONGER
My Commission Expires
March 20, 2018

Assignee hereby acknowledges receipt of the entire right, title, and interest in and to the intellectual property referenced in Section 1 above and which is owned by UABRF.

Signature: _____ Date: 1/14/2015

Name: Frank Skidmore

Witness: _____ Date: 1/14/15

Name:

SCHEDULE 1
(ATTACH UABRF IPD)

INTELLECTUAL PROPERTY DISCLOSURE (IPD)

The UAB Research Foundation
770 Administration Building
701 20th Street South
Birmingham, AL 35294-0107

For UABRF internal use only:

IPD#: U2012-0045 Mgr: Terry Bray

Approved: Gloria Hamaker Date: 02-02-12

1. Title of Intellectual Property:
   Iterative imprinting of three dimensional brain maps

2. Discloser Contact Information:

Name: Frank M. Skidmore	Office Phone: (205) 975-3395
Email: fskidmor@uab.edu	Fax Number:

3. Type of Intellectual Property:

☐	Therapeutic	☒	Diagnostic	☐	Device
☐	Software	☐	Assay	☐	New Composition of Matter
☐	Vaccine	☐	Method of Treatment	☐	Other ____

4. Description of Intellectual Property:
   a. Please submit a brief summary of the Intellectual Property of 250 words or less describing the invention and its uses, in layman's terms.
   b. Also, attach any additional information that would complete the description of your invention. As an example, you may attach publication drafts, drawings, sketches, and photographs to help describe the Intellectual Property.

5. Please indicate the actual or approximate date when this intellectual property was first conceived. Initial concept and start of work occurred 10/21/2011, initial verification of validity or proof of concept occurred on Friday, January 20, 2012 at approximately 10:30 AM.

6. Have you already disclosed this intellectual property, either orally or in writing, to any persons or groups (conferences, meetings, seminars, proposals, etc.)? ☐ Yes ☒ No
   Are there any publications, presentations (include electronic disclosures) or other public disclosures planned in the future? ☒ Yes ☐ No
   Please describe the nature of the disclosure and the date that it has occurred or will occur.

DISCLOSURE:	DATES:
1. CCTS Pilot Grant Program	Internal submission: external reviewers are required, date uncertain
2. Other potential future grants are planned	

7. Is this intellectual property related to another intellectual property previously disclosed to this office? ☐ Yes ☒ No
   If yes, please provide the IPD number and title.

8. Were any materials, used directly or indirectly, acquired with an MTA or from a third party (either outside or within UAB)? ☐ Yes ☒ No
   Please list below.

INSTITUTION/COMPANY/PERSON:	DATES:	MATERIAL:	MTA:
1.			☐ YES ☐ NO
2.			☐ YES ☐ NO

9. Please list all sources of funds, internal and external, that gave rise to the Intellectual Property and provide the details below. Information must be complete and the total percentage support must equal 100%. (i.e., departmental funds, salary source, grant information, etc.)

AGENCY/SPONSOR:	DATES OF AGREEMENT:	GRANT/CONTRACT #:	ESTIMATED %:
1.			
2.			
			TOTAL = 100%

10. Disclosers: Please list all persons believed to have made essential contributions to the Intellectual Property during the evolution of the initial concept or reduction to practice. Include UAB as well as non-UAB persons and obtain signatures from all Disclosers. (Fax signatures of non-UAB personnel will be accepted.) Determination of inventorship is a legal matter and will be determined by legal counsel.

Lead Discloser

Printed Name: Frank M. Skidmore	Received JAN 30 2012 Office of the U.A.B. Research Foundation	Country of Citizenship: USA
Campus Address:		UAB Employee: ☒ Yes ☐ No
Email: fskidmor@uab.edu		Daytime Phone:
Home Address: 3645 Crestside Road, Mountain Brook, Alabama		
Signature: Frank M. Skidmore		Date: 01/26/2012

Discloser #2

Printed Name:	Country of Citizenship:
Campus Address:	UAB Employee: ☐ Yes ☐ No
Email:	Daytime Phone:
Home Address:	
Signature	Date:

Discloser #3

Printed Name:	Country of Citizenship:
Campus Address:	UAB Employee: ☐ Yes ☐ No
Email:	Daytime Phone:
Home Address:	
Signature	Date:

Discloser #4

Printed Name:	Country of Citizenship:
Campus Address:	UAB Employee: ☐ Yes ☐ No
Email:	Daytime Phone:
Home Address:	
Signature	Date:

Discloser #5

Printed Name:	Country of Citizenship:
Campus Address:	UAB Employee: ☐ Yes ☐ No
Email:	Daytime Phone:
Home Address:	
Signature	Date:

*If additional signature lines are required, please download the additional IPD signature form at http://main.uab.edu/Sites/UABRF/info_researchers/intellectual_property/.

11. Commercialization/Potential Licensees:

Please list any companies that may be interested in your Intellectual Property:	
COMPANY	CONTACT INFORMATION (IF KNOWN)
1. Phillips	
2. Seimens	
3.	

Do you know of companies that are using similar technologies? If so, please provide the details below:		
COMPANY	CONTACT INFORMATION (IF KNOWN)	PRODUCT
1.		
2.		
3.		

UAB RESEARCH FOUNDATION REVENUE DISTRIBUTION AGREEMENT U2012-0045

Date: 01	Lead Discloser: Frank M. Skidmore
Title of Invention: Iterative imprinting of three dimensional brain maps	
Contact Name: Frank M. Skidmore	Office Phone: (205) 975-3395
Email: fskidmor@uab.edu	Fax:

Please list all UAB Disclosers and Contributors and obtain signatures of all Disclosers and Contributors and their affiliated departments and schools. Signatures of the parties indicate that they have reviewed the Intellectual Property Disclosure Form and approve the indicated distribution of revenue. The sum of the percent contributions must equal 100%. All appropriate signature lines must be executed.

Lead Discloser

Printed Name: Frank M. Skidmore		Percent Contribution: 100
Signature: Frank M. Skidmore	Date: 1/26/2012	Social Security No:
Department: Neurology	Department Chair Signature:	Date: 1/27/2012
School: Medicine	Dean Signature: Rashell Cameron for "Dean Watts"	Date: 2/1/12

Discloser or Contributor #2

Printed Name:		Percent Contribution:
Signature:	Date:	Social Security No:
Department:	Department Chair Signature:	Date:
School:	Dean Signature:	Date:

Discloser or Contributor #3

Printed Name:		Percent Contribution:
Signature:	Date:	Social Security No:
Department:	Department Chair Signature:	Date:
School:	Dean Signature:	Date:

Discloser or Contributor #4

Printed Name:		Percent Contribution:
Signature:	Date:	Social Security No:
Department:	Department Chair Signature:	Date:
School:	Dean Signature:	Date:

Discloser or Contributor #5

Printed Name:		Percent Contribution:
Signature:	Date:	Social Security No:
Department:	Department Chair Signature:	Date:
School:	Dean Signature:	Date:

*If additional signature lines are required, please download additional RDA signature form at http://main.uab.edu/Sites/UABRF/info_researchers/intellectual_property/.

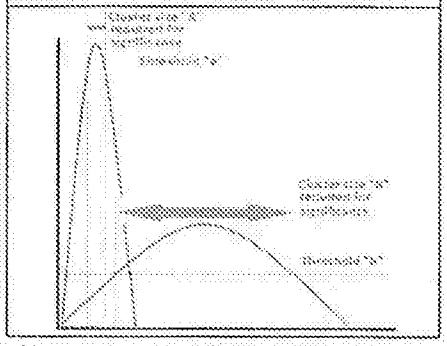
SUMMARY: This invention relates to generating a new method of brain mapping applicable to any imaging method (including structural images, functional magnetic resonance images, diffusion tensor images, PET images, or SPECT images) in which data is available from a representative group of subjects in which a particular characteristic is useful (such as presence or absence of a disease, or gradations of severity within a particular disease). Using this method of brain mapping, reliable voxels in the dataset can be identified that can be used for diagnostic purposes, or to follow disease progression or development. The same technique can be used on an individual subject basis to extract information from an individuals brain to evaluate if the subjects brain has characteristics associated with a specific imprint that has been identified. This invention is relevant to any condition in which changes in features in the brain are an aspect of the disorder. When we create an imprinted map using the method, it is similar to a "fingerprint" of the disorder in that it contains information about how that disorder reliably changes a specific imaging modality. Often times, diseases occur in the context of other diseases, and consequently it is possible for individual to have the imprint of many disorders. For example, when individuals are older than a certain age, a number of distinct disease can occur contemporaneously. An individual may have a stroke, but may also have Alzheimer disease, essential tremor, and changes in the brain associated with longstanding smoking and hypertension. Alternatively, an individual may have history of a traumatic brain injury, but is now displaying clinical signs of memory loss and has a parkinsonian tremor. A key strength of our method is the utility of the method in excluding epiphenomenon within the dataset to show the reliable change associated with a particular condition. Returning to the example of fingerprint analysis, a partial print may still provide useful information, and it may be possible to discern features of multiple overlapping imprints. An additional strength of our method is that it is possible, using this method, to identify regions to exclude from analysis because of evidence of damage or changes that cannot be accounted for by the target queried conditions. We envision, using this method, the creation of multiple maps associating changes in certain imaging modalities in association with certain conditions. These maps may be used individually for diagnostic purposes, or may be combined to improve accuracy of diagnosis.

BACKGROUND: A standard imaging dataset may contain hundreds of thousands of voxels (a voxel is a term representing a 3-dimensional region of space, analogous to the 2-dimensional "pixel" on a flat television screen – obviously with an added dimension). I will describe briefly standard imaging techniques, the PI's perceptions of problems associated with these techniques and how my innovative methods of analysis differ from those typically used in the field and are designed to address the perceived problems.

Standard statistical techniques referable to MRI analysis assume that a region of image space has a normal (or Gaussian) probability distribution, with a bell-shaped probability density function. Further analysis often uses two distinct processes to identify regions that are "significant" in a full brain analysis. First, a statistical threshold is performed. Let us say we set our statistical threshold at $p < 0.05$. Based on our assumption of a Gaussian distribution, we would then assume that 5% of all voxels would be statistically purely positive "by chance." In this case, in a dataset of 200,000 voxels we would then expect that 10,000 voxels would be statistically positive purely as a result of "chance". A second statistical analysis in then performed to identify associations that are unlikely to be chance associations. In this second analysis, the volume of interest is once again assumed to be a uniform structure governed by a pure Gaussian distribution. A random number generator is used to randomly place statistically "positive" voxels inside this container (in the above setting, 10,000 "positive"

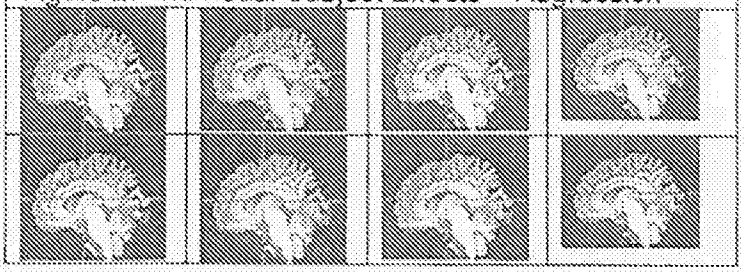
voxels would be randomly placed within the larger 200,000 voxel volume). This random process is re-iterated 1,000 times, and a counting procedure counts how many times a certain number of "statistically significant" voxels "randomly" are clustered together. A second threshold (related to number of voxels) is then used to characterize what regions are significantly different. If we set the threshold at $p < 0.05$, a certain number of voxels might need to be together to form a "significant" cluster. However, consider if I set the threshold at $p = 0.001$? In this case we would expect only 200 voxels to be "randomly" positive – a much smaller cluster is required to achieve "significance" (see figure 1). A problem immediately becomes apparent at this step. If I choose a very high threshold, I am able to capture a small highly significant cluster (represented by the tall, sharp peak), but I miss the large broad peak that is also potentially important because it does not meet our significance threshold. Conversely, if I choose the low threshold, I will miss the highly significant small region, because it is not of sufficient size to meet our cluster size threshold. In practice, data is often approached with a-priori hypotheses, and variably thresholded, a questionable statistical procedure given the possibility of introduction of bias. A second, more subtle problem is also built in to this type of analysis. The brain is not in fact a uniform set of voxels with a Gaussian distribution of importance. Some small regions (such as compact regions in the brainstem or basal ganglia) might be extraordinarily important, while in other regions (such as the cortex) more variability in size might be reasonable depending on what is being measured. Some individuals use region of interest analysis to overcome the cluster size-threshold conundrum, however this method brings in inherent bias, as the investigator must approach the data with a specific assumption (called an a-priori-hypothesis) in order to draw a region. In bringing in the assumption, the investigator not only throws out a significant portion of the data, but even in approaching a specific region may overlook important findings that do not agree with the hypothesis.

Figure 1: The Cluster Size-Threshold Conundrum



In addition to the Cluster-Size-Threshold Conundrum identified above, an additional issue, subject variability is a substantial issue in the analysis of imaging datasets. The author formally analyzed the issue of inter-subject variability with colleague Mark Yang. Mark Yang and I asked the question – is it possible that this individual variability might be important in developing ostensible "differences"in statistical maps using standard techniques? We found the answer was clearly "yes" within our dataset. We obtained a measure of low frequency fMRI blood oxygen level dependent (BOLD) signal in the sample (see Skidmore, Yang, et al. 2011a and methods below for more detail). Figure 2 shows an example: a subject is "left out" of the analysis and the analysis is repeated. With each repeated analysis, a new statistical map is created with the remaining subjects in the analysis. In this case 16 subjects were in the initial behavioral analysis (8 representative examples are shown, each color shows a specific region that is "statistically significant", either positively or negatively). In this analysis, the effect was so powerful that in 9 of 16 analyses, specific regions were deemed

Figure 2: Individual Subject Effects – Regression



"significant" that were present in only one of the "left out" 16 analyses. The effect was particularly evident in regression analyses, but was also apparent in group comparisons (figure 3 – showing a group comparison of 16 subjects with PD and 15 controls, 16 representative subjects shown, blue in this case represents *less* resting activity in the PD subjects). This circumstance is clearly not tenable if the goal is to create a predictive map with a resting fMRI scan (our goal at the time). Mark Yang and I first approached the problem of creating a statistical map by creating a map of all regions that were "significant" by cluster thresholding within a primary analysis.

We then repeated these analyses iteratively, each time leaving a subject out. The maps were overlapped, creating a "reliability" map (Figure 4 – Skidmore, Yang, et al. 2011a, 2011b). Red spectrum in the figure indicates significance in most of the analyses, while greener/bluer spectrum colors indicate statistical significance in fewer analyses. Statistically identifiable regions overlapping with "reliable" regions (identified arbitrarily as regions in which 90% or greater of the "leave one out" analyses were positive) were used to select regions used for prediction. Combining the "reliable" regions, Mark Yang and I were able to identify the "left out" subject with 92% sensitivity and 87% specificity. In a regression analysis involving the 16 subjects with PD, we were able to identify first, that most "significant" regions identified by a standard regression analysis were completely useless (e.g. no better than a coin toss) for predicting the depression or apathy score in a left out subject. We were also able to identify, however, that specific regions – the subgenual cingulate in the case of depression, and the supplementary motor cortex and right orbitofrontal regions, were able to specifically predict the representative depression or apathy score in the left out subject. The use of reliability maps, in combination with standard cluster thresholding, for the creation of diagnostic maps has been filed as a provisional patent application by the University of Florida (see attached provisional patent).

Figure 3: Individual Subject Effect – Group Comparison

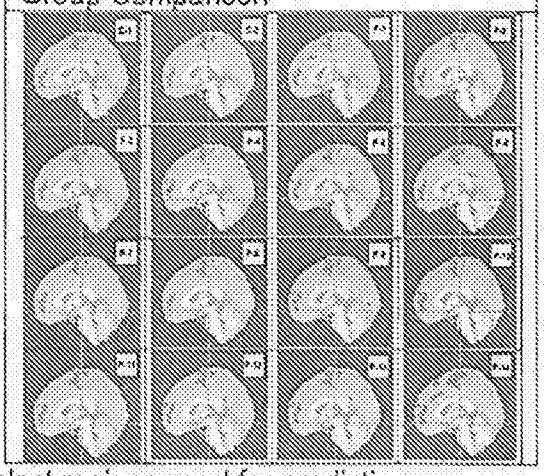
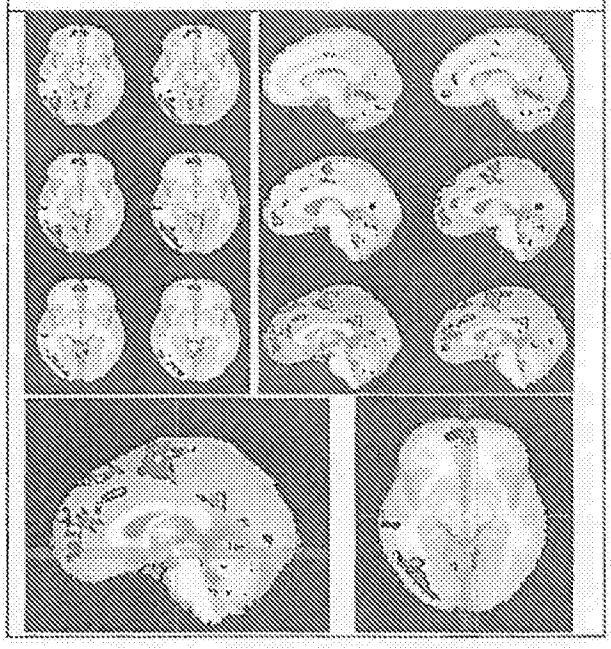


Figure 4: Reliability Map



INNOVATION: The foregoing work under provisional patent did provide some improvement over existing methods for the purposes of creating predictive templates, however I was still dissatisfied with the results. Specifically, while starting to account for individual subject variability, the method Dr. Yang and I developed did not tackle the

issue I identified at the beginning of this section (the cluster size-threshold conundrum). Mark and I had discussed this problem, but the solution eluded us. In late October of 2011, a potential solution to the cluster-threshold problem became apparent. The chief constraint in developing a statistical map not constrained by the cluster-threshold conundrum is the develop of alternative statistical means to clarify the relevance of a given voxel to a disorder in question. A method that clarifies meaning at a voxel-wise level must robustly identify individual voxels or groups of voxels within a group of hundreds of thousands of voxels that are predictive of a particular disease or situation by a robust statistical method relevant on a per-voxel basis. Since it is nearly impossible for a given voxel in a single analysis to be sufficiently different from the rest of voxels in the brain when adjusting for a groupwise comparison within several hundred thousand other voxels, it is important to generate additional analyses to evaluate the robustness of individual voxels in a particular comparison. I tackled this problem using an iterative bootstrap. A reduction to practice of this technique is shown below. In this case, we discuss an initial raw dataset of 35 fractional anisotropy maps (20 subjects with a disease – Parkinson disease, and 15 healthy controls). Using a random number generator, and leaving one subject out each time, the subject pool is "re-seeded" multiple times to create a new statistical maps. Specifically, in each of a given number of "slots" (20 for PD, 15 for controls), a random appropriate individual (PD or control respectively) was re-assigned to occupy the "slot" in question. In the new dataset of 20 PD subjects and 15 controls, subjects might therefore be not represented, represented once, or randomly repeated multiple times. The iterative maps in this case are added to create a bootstrapped map of which voxels are reliably positive, regardless of how the data is randomly re-assorted. The statistical properties of a representative bootstrapped map from this case is notable (see figure 5 above), in that the majority of voxels are statistically "positive" in 3-5 of the analyses (a purely random Gaussian curve would predict a peak "false positive curve at 1-3, although perhaps this is explainable by the random repetition of data in some bootstraps). However, very few voxels are statistically "positive" again and again as we approach the full 50 iterations in this particular run. If we secondly ask the question of which voxels predict the identity of the left out subject, we obtain an overlapping map that reduces the effects of individual variability – such as that seen in the our first analysis of resting fMRI above. Using the method above, one reduction to practice of this invention, we have created a statistical

Figure 5: Number of Positive Voxels at Threshold
(Threshold is $p < 0.05$, 50 bootstrapped iterations)

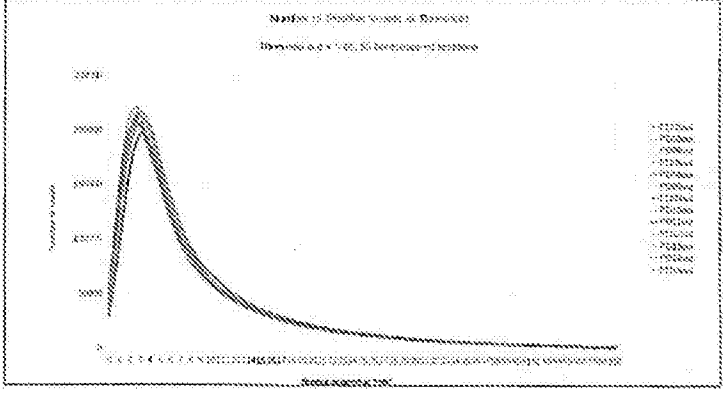
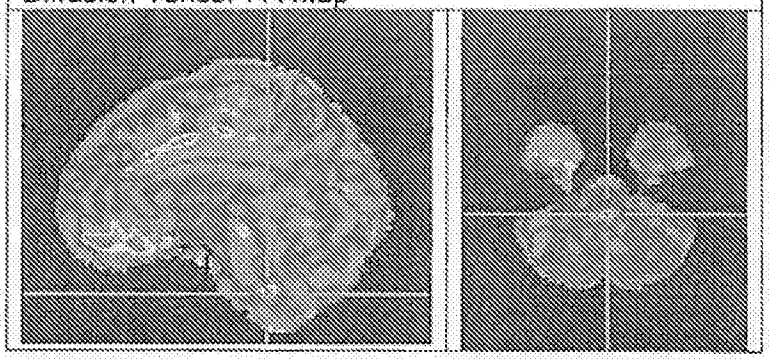


Figure 6: Predictive Template Map, PD vs. Controls, Diffusion Tensor FA Map



procedure that avoids the limitations of the standard method (the cluster size-threshold limitation). In other words, the resultant map creates a map, not subject to the cluster size limit, identifying all voxels that are predictive of subject class. This method can identify small regions (such as small important regions in the brainstem or basal ganglia associated with a particular condition), but can also identify large regions. Examples of this type of map are shown in figure 6. In this case, 1750 bootstrapped iterations were used to create a map of iteratively predictive voxels. Naturally, the map is internally consistent, predicting 100% of subjects, however testing in this fashion would involve a circular logic that is not statistically defensible. With new data, however, the constructed map predicted 7/7 new control subjects as healthy controls. A repeated analysis of the data – using a random number generator to "elect" 6 PD subjects as out of the dataset, was then constructed. In this dataset, we therefore had now 22 controls, and 14 subjects with PD. We again in this situation were able to identify 6 out of the 6 left out subjects as "PD". Notably, in all cases we identified that our regions of interest had an ***increase*** in the fractional anisotropy signal, the reverse of what might be expected. Second, the regions we found did not specifically directly overlap regions of known lewy body pathology, but were instead adjacent and around these regions. Third, our method identified "biomarker" regions highly predictive of subject class in this sample for the first time including a region close to the dorsal motor nucleus of the vagus, and the olfactory regions, as well as identifying the globus pallidus, among other regions. While olfactory regions and the dorsal motor nucleus of the vagus have not been identified in the past as regions of particular value in segregating PD from healthy controls using DTI, abnormalities in the olfactory regions and the dorsal motor nucleus of the vagus have been long recognized as being among the very first regions in the brain to develop lewy body pathology. Specifically, Braak et al. (2003) pathologically has classified lewy body pathology in the olfactory regions and dorsal motor nucleus of the vagus as "Stage I Parkinson disease" (clinical diagnosis, in Braak's pathological construct, occurs when lewy bodies occur in the substantia nigra
at Stage III disease). We therefore using this method for the first time discovered biomarker potential in regions that have otherwise been overlooked using standard analytic techniques.

A few important points in relation to this reduction to practice are important to stress. First, using a non-hypothesis directed method, we nonetheless identified multiple regions in our analysis that correlate roughly with expected regions of change associated with Parkinson disease. Second, our findings were consistent locationally with what might have been expected, but many investigators might have expected a decrease rather than an increase in FA signal, and Third, our method identified regions that had never been identified before as useful regions from the perspective of a biomarker. Left to the natural course of science, all of these independent observations might have taken years to develop using usual voxel-wise, threshold and cluster dependent, and hypothesis driven research. Our method therefore represents a shortcut to developing useful predictive diagnostic maps.

A final important step in the current invention involves recognition of the importance of the curve in figure 5. The curve in figure 5 shows that multiple regions in the brain are essentially similar in individuals with PD and healthy controls. Further work will be done to flesh out the importance of this, but in brief, regions that should be similar can also be used for the purposes of sample stratification. For example, if most regions support a particular diagnosis, but a particular additional region adjacent to a known predictive region unexpectedly shows a difference, this might present a signal regarding the utility of a particular overall region for prediction of a specific condition (e.g. if you have had a stroke in a region, the signal will be different and we can know from our

analysis that this region should not be used in our diagnostic template). Unexpected differences can also potentially be used to identify overlapping and concurrent disorders, if the remainder of the map is supportive of such a diagnostic leap.

In this particular reduction to practice, the full algorithm, including stratifying the iterative map by the predictive map, was used to create the predictions. However, simply identifying regions in the iterative map that have high reliability can also in some cases create a robust imprint of reliable regions sufficient for subsequent predictive identification. While we used an iterative bootstrapping of a specific analysis (in this case an iterative T test at a particular threshold), modifications of this technique would easily be envisioned by individuals skilled in the art. For example, using other techniques and algorithms to create overlapping maps, that can be collapsed as a single evaluation of robustness and reliability of a particular voxel, would be considered within the scope of this invention disclosure.

Our mapping technique allows the generation of a "preponderance of the evidence" analysis, that was accurate in predicting all 13 left out subjects. While we used this technique for the purposes of generating a subject classifier, there is no reasons that similar techniques could not be used for the purpose of, for example, detecting regions important for the development of disease progression, specific cognitive/behavioral changes, or mood problems within a given sample. Further, there is no factor that limits the PIs analytic technique to one disorder or another (as is the case with specific biomarkers focused on particular proteins), or one imaging modality over another (within certain limits). With sufficient sophistication, overlapping maps could be developed that might predict one or more disorders or impending clinical disorders simultaneously.

With reference to the investigator's preceding invention (development of reliability maps), this current invention uses the general concept of reliability mapping, but adds a significant inventive step that allows us, to step outside of the boundaries of current analytic techniques subject to the threshold-cluster size limitation, to create truly predictive maps useful for clinical purposes. We are fingerprinting disease specific elements visible in brain images.