

# **Pennsylvania Department of Health Final Performance Summary Report Non-Formula Grants**

## **Overview of the Health Research Project Performance Review Process and Criteria**

An applicant that receives a health research grant under Tobacco Settlement Act / Act 77 of 2001, Chapter 9, is subject to a performance review by the Department of Health upon completion of the research project. The performance review is based on requirements specified by Act 77 and criteria developed by the Department in consultation with the Health Research Advisory Committee.

As part of the performance review process, each research project contained in a grant is reviewed by at least three experts who are physicians, scientists or researchers. Reviewers are from the same or similar discipline as the research grant/project under review and are not from Pennsylvania. Reviewers use the applicant's proposed research plan (strategic plan), the annual progress reports, interim review reports, corrective action plan, and final progress report to conduct the review. A grant that receives an unfavorable performance review by the Department may be subject to a reduction in funding or become ineligible for health research funding in the future. The overall grant evaluation rating is based on the ratings for the individual research projects contained in the grant.

This performance review report contains the outcome of the review for the grant as a whole (outstanding, favorable, or unfavorable), strengths and weaknesses of each research project, as well as recommendations for future improvement.

The following criteria were applied to information submitted by research grant recipients:

- **Criterion 1 - How well did the project meet its stated objectives? If objectives were not completely met, was reasonable progress made?**
  - Did the project meet the stated objectives?
  - Consider these questions about the data and empirical results: Were the data developed sufficiently to answer the research questions posed? Were the data developed in line with the original research protocol?
  - If changes were made to the research protocol, was an explanation given, and, if so, is it reasonable?
  - Consider (only for clinical research grants) the extent of laboratory and clinical activities initiated and completed and the number of subjects relative to the target goal.
  - Were sufficient data and information provided to indicate or support the fact that the project met its objectives or made acceptable progress?
  - Were the data and information provided applicable to the project objectives listed in the strategic plan?

- **Criterion 2 - What is the likely beneficial impact of this project? If the likely beneficial impact is small, is it judged reasonable in light of the dollars budgeted?**
  - Consider any changes in risk factors, services provided, incidence of disease, death from disease, stage of disease at time of diagnosis, or other relevant measures of impact and effectiveness of the research being conducted.
  - Consider any major discoveries, new drugs and new approaches for prevention, diagnosis and treatment, which are attributable to the completed research project.
  - What are the future plans for this research project?
  
- **Criterion 3 - Did the project leverage additional funds or were grant applications submitted?**
  - If leveraging of funds were expected, did these materialize?
  - Are the researchers planning to apply for additional funding in the future to continue or expand the research?
  
- **Criterion 4 - Did the project result in any peer-reviewed publications, licenses, patents, or commercial development opportunities? Were any of these submitted / filed?**
  - If any of the above listed were expected, did these materialize?
  - Are the researchers planning to submit articles to peer-reviewed publications, file for any licenses or patents or begin any commercial development opportunities in the future?
  - Consider the number/quality of each and what was proposed in the original application.
  
- **Criterion 5 - Did the project enhance the quality and capacity for research at the grantee's institution?**
  - If any improvements in infrastructure were expected, were they made?
  - Were any new investigators added or were any researchers brought into the institution to help carry out this research?
  - Were funds used to pay for research performed by pre- or post-doctoral students?
  
- **Criterion 6 - Did the project lead to collaboration with research partners outside the institution, or new involvement with the community?**
  - Are the researchers planning to begin any collaborations as a result of the research?
  - For clinical research only: consider the number of hospitals and health care professionals involved and the extent of penetration of the studies throughout the region or the Commonwealth.

## **Overall Evaluation Rating**

An overall evaluation rating is assigned to each research project. The rating reflects the overall progress the project attained in meeting the stated goals and objectives. The rating is based on a scale of 1–3, with 1 being the highest. An average rating is obtained from all the reviews (minimum of 3) of each project and is the basis for the determination of the final overall rating for each project as follows:

1.00 – 1.33 = *Outstanding*

1.34 – 2.66 = *Favorable*

2.67 – 3.00 = *Unfavorable*

The grant level rating is an average rating from all projects as above. The numerical rating appears in parentheses for the grant and each project in the ***Overall Grant Performance Review Rating*** section of the report.

***Overall Grant Performance Review Rating***

**Grant Rating:** Outstanding (1.29)

**Project Rating:**

<b>Project</b>	<b>Title</b>	<b>Average Score</b>
08863	CHOP/Penn Center of Excellence for Autism Research	Outstanding (1.29)

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**Project Number:** 08863  
**Project Title:** CHOP/Penn Center of Excellence for Autism Research  
**Investigator:** Robert T. Schultz, PhD

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## Section A. Project Evaluation Criteria

**Criterion 1 - How well did the project meet its stated objectives? If objectives were not completely met, was reasonable progress made?**

### *STRENGTHS AND WEAKNESSES*

#### Reviewer 1:

This program of research is divided into 6 projects.

**PROJECT 1: CHARACTERIZING THE COMMON INTERGENIC VARIANT AT THE 5p14 locus IN ASD. (PI: HAKON HAKONARSON, MD, PhD)**

This project was based on the premise that there is a significant GWAS signal at 5p14 intergenic locus for autism spectrum disorder (ASD). The goal of the current project was 1) to characterize the locus further by resequencing the region to capture the functional variant(s) involved and 2) to genotype candidate SNPS in 3000 ASD and 3000 control samples.

#### Strengths:

A total of 3199 cases and 2992 controls were genotyped in total. Initial Quality Control (QC) of samples and data resulted in 148 cases and 123 controls being removed for poor genotyping quality. This left 2954 cases and 2869 controls with good genotyping.

Following a variety of quality control and analytic steps with data from two platforms the researchers found that no custom-based SNP genotype using the Veracode/Goldengate panels at the 5p14 intergenic locus reached significance over and above the GWAS SNPs, suggesting that DNA variation may not be the underlying cause for the GWAS signal.

The researchers speculate that the original signal may be due to alterations in over expression of a noncoding RNA antisense to moesin at 5p14.1.

Germane to project 3, the researchers found that there were no significant SNPs either in the CDH9 or CDH10 exon sequences generated and genotyped.

#### Weaknesses:

While this failure to replicate the initial GWAS was probably unanticipated, it would have been optimal to have alternate hypotheses to evaluate during the program period.

**PROJECT 2: UNDERSTANDING THE CONTRIBUTION OF RARE VARIANTS. (PI: MAJA BUCAN, PhD)**

This project focused on detection of rare variants in autism subjects recruited to the Center for Autism Research. This project evolved from initially proposing to evaluate 100 candidate genes to the implemented strategy of carrying out whole genome sequencing (WGS) on a subset of highly phenotyped individuals.

**Strengths:**

The researchers have performed WGS analysis on 70 subjects with autism to investigate the role of rare genetic variants in the etiology of this disorder. Sequence data for 20 of the subjects were obtained near the end of the project period and data analysis is ongoing. A very large database of information has been gathered. The analysis identified 11.1 million sequence variants within the 97% called genome fraction. The overall rate of novel SNP variants was approximately 3.4%. Among previously reported ASD-associated genes, the researchers detected novel variants in *CACNA1H*, *CACNA1C*, *MADCAM1*, *CDH22*, *HTR2A* and *BZRAP1*.

**Weaknesses:**

Despite the proposed correlation of genetic variation with both phenotypic variables and neuroimaging results, these more complex inter-modality analyses have apparently not yet been carried out. This may be due, in part, to the ongoing recruitment of subjects into Project 4.

Given that Project 4 was able to recruit and phenotype over 400 subjects with autism spectrum disorder, it is unclear why the WGS of the last 20 subjects was carried out at the very end of the project funding period.

**PROJECT 3: ANIMAL MODELS OF GENETIC INFLUENCES ON BRAIN AND BEHAVIOR. (PI: EDWARD BRODKIN, MD; CO-PI, EDWIN ABEL, PhD)**

This project has gone through substantial evolution of goals during the course of the program. The original goal for Specific Aim 3.1 was to generate a *Cdh9-Cdh10* intergenic region deletion however this was deemed to be too risky. The researchers then decided to generate a mouse in which the *Cdh10* gene itself is flanked with loxP sites and then conditionally deleted. This was outsourced to a company (GenOway) that took much longer than expected to accomplish the task. But, in year 4 of the program floxed *Cdh10* on a pure C57BL/6 genetic background arrived at University of Pennsylvania, and mice were bred at University of Pennsylvania. While there were some behavioral alterations in these mice, they did not show impairments in social behavior.

Generation of the *Cdh9* conditional knockout mouse using the cre-lox system was initiated during Year 2. However, chimera mice from this line only produced wild type pups, i.e. germline transmission of the conditional knockout allele was not achieved, despite multiple attempts to do so, extending into the last year of the award.

Specific aim 3.2 dealt with *Pcdh10* knockout mice. This mouse strain did show a significant decrease in sociability only in males. A similar male only deficit was observed in long term fear

memory. The investigators also used c-fos immediate early gene staining to identify differences in social neurocircuitry and identified differences in some nuclei of the amygdala and other brain regions.

**Strengths:**

The *Pcdh10* knockout mice were subjected to a very thorough phenotyping that included analyses of morphogenesis, motor behavior, memory, olfactory function, anxiety and social behavior. Electrophysiological studies of the hippocampus were also carried out.

**Weaknesses:**

This was a comprehensive behavioral and neurobiological analysis of one mouse strain that may be of relevance to the etiology of autism spectrum disorder. No weaknesses were detected.

**PROJECT 4: CHARACTERIZING THE PHENOTYPES OF THOSE CARRYING RARE AND COMMON RISK ALLELES. (PI: ROBERT SCHULTZ, PhD; CO-PI, DAVID MANDELL, ScD)**

**Strengths:**

The goal of this project was to recruit and to deeply phenotype a large cohort of children with autism spectrum disorder (ASD) and a smaller group of age-matched typically developing individuals. While the initial pace of recruitment was slower than anticipated due to the limitations of clinical staff, this project ultimately met its goal and recruited 419 children with confirmed ASD and 153 typically developing controls. This is a major accomplishment for the program period.

In addition to the recruitment and assessment of the cohorts described above, there were two specific aims for this project:

Aim 4.1: To characterize the relationship between the degree of social impairment and genetic status in a large cohort of children with ASD

Aim 4.2: To characterize the impact of the risk SNP on treatment outcome...

Since recruitment of the cohort continued until the end of the funding period, these analyses had not been carried out by the time this final progress report had been submitted.

**PROJECT V: CHARACTERIZING THE BRAIN ENDOPHENOTYPES OF THOSE CARRYING RARE AND COMMON RISK ALLELES. (PI: ROBERT SCHULTZ; CO-PIs: PhD RAGINI VERMA, PhD, TIMOTHY ROBERTS, PhD)**

The goal of Project 5 as outlined in the original grant application was to study with MRI and magnetoencephalography (MEG) 80 youth with ASD and 80 matched typically developing controls (TDCs) characterized in Projects 1 and 4.

**Strengths:**

Overall, recruitment was more difficult and slower than anticipated; nevertheless the researchers were able to meet their MRI scanning goals, and successfully studied a total of 164 participants (81 with ASD and 83 with TDC). MEG recruitment and scanning fell slightly short (n=150 with

79 ASD and 71 TDC) since new equipment was even further delayed in its arrival and installation compared to MRI.

**Weakness:**

The researchers expanded the age range of subjects from 6-10 to 6-13 because they were having difficulty getting the younger children to remain still during the MRI scan. This is unfortunate because there are many examples of brain changes that occur during the preadolescent to adolescent period. The analyses that the investigators will carry out may be confounded by the pubertal changes.

There are no reports of findings since subjects were being recruited until the end of the funding period. However, preliminary data assessments could have been carried out on the substantial subset of subjects that were recruited up to the last year of the project. The lack of any project-based results is a weakness.

Both MEG and fMRI datasets are at early stages of analysis.

**PROJECT 6: MINORITY TRAINING PROGRAM (PI: JAMES CONNELL, PhD)**

This project had as its goal the establishment of the Program in Autism Research Training (PART) to recruit undergraduate and graduate students from under-represented groups. The project goals of recruiting 4 undergraduates and 4 graduate students was met. While this seems like a very modest goal, all of the participants are involved in education or work efforts that are related to health or ASD.

**Reviewer 2:**

Project 1: The 5p14 locus remains a promising target, and Pennsylvania Department of Health funded work characterizing this locus has been helpful.

Project 2: The successful whole genome sequencing of 70 ASD subjects will continue to provide a valuable database for the scientific community.

Project 3: The successfully characterized mouse model of ASD will continue to have relevance for the autism research community.

Project 4 (phenotyping) and 5 (brain imaging): Project 4 successfully recruited a massive sample of 164 participants with MRI scanning and 150 participants with MEG scanning. This is a remarkable dataset with 9 reported and planned manuscripts arising from the effort, which is sure to increase in the coming months and years.

Project 6: Minority training was successfully facilitated with 8 students recruited.

**Reviewer 3:**

**Strengths:**

The investigators met ambitious recruitment goals for nearly all of the projects, and have completed most of the data collection for the proposed experiments. Interim analyses have been

performed where possible and yielded several manuscripts and additional funding. An impressive battery of behavioral tasks was implemented for the PDCH10 mouse model, with some interesting results that speak to some ASD-relevant behaviors, and a good design that separated testing environment from social stimuli. The investigators made careful re-evaluations about human subjects recruitment and the decision to expand the age range to 6-13 was a strength. The integration across the projects, particularly 4 and 5, was also a strength.

#### Weaknesses:

Data analysis has lagged because the entire project period has been devoted to data collection; thus, it is difficult to evaluate whether the hypotheses in the original proposal were supported by the data gathered for the project. There were some discrepancies on reported recruitment for Project 2 (20 or 50?). Some data in Study 3.2 were not really interpreted, such as increased speed to find buried food in the heterozygous mutant mice. The second aim of Project 4 was only able to re-recruit about 30% of the targeted former participants. There were issues with several of the figures, including one being 50% cut off by text, others being cut off such that axes were unreadable or appeared unlabeled, missing or scrambled (Fig 5.11) labels. There is also not a clear thematic link between the brain imaging and the genetic studies, although the deep phenotyping helps to link them.

#### Reviewer 4:

This project is to support the Children's Hospital of Philadelphia Center of Excellence for Autism Research with the central theme of understanding the relationship between genetic risk factors, brain and behavior in ASD. The project contains 5 science projects and a training project. Overall, the Center has made impressive progress in both building infrastructure to enhance research and to support training. This award helped to launch multiple funded projects, to create new collaborations, to expand research capacities, and to train numerous students. No major weakness was identified with the Center overall. For the 5 research projects, the original goals set for each study have largely been met although the progress is uneven in which Projects IV and V are the "stars" and the progress of Projects I, II and III were hindered by various difficulties (see details below).

**Project I:** Built upon their previous novel findings, this study was designed to identify and characterize the causal variant(s) in the *CDH9/CDH10* intergenic region on 5p14 to further address the roles of the variants in ASD pathogenesis. The study proceeded with the plan but failed to identify the causal variant. However, new markers (*CDH26*) have been identified that may be associated with ASD. Functional studies to address the impact of these variants on RNA expression and gene function in cell-based assays are underway. So, the proposed goals were largely met although the results are bit disappointing. No paper was published and researchers are planning to submit two NIH grants.

**Project II:** Originally, the researchers proposed a candidate-gene approach. With the advancement of sequencing technology, changes were made and WGS was chosen to replace the old approach. However, for reasons that are not well explained, only 70 sequencings have been done (20 with data analyzed) with regard to 400 in the original proposal. As a result, only partial goals have been achieved for this project. It is unclear whether a sample size of 70 is sufficient to address the original question. One paper has been published and one NIH grant is in planning.

Project III: Due to technical difficulty, modification has been made to the proposal: instead of generating a mouse model with *Cdh9-Cdh10* intergenic region deletion, a mouse model with conditionally deleted *Cdh10* gene was generated. This change caused much delay in the project; only partial goals can be accomplished. No paper was published and several grants are in preparation.

Project IV: Although there was recruitment delay, excellent progress has been made on this project, which is designed to assess relationships between phenotypic dimensions (e.g., the relationship between social motivation and face perception) and to create a rich phenotypic database for analyses of brain-behavior relationships (in collaboration with Project 5), genotype-phenotype relationships (in collaboration with Project 1), and genotype-phenotype-endophenotype (brain imaging) relationships. Researchers have been able to perform many analyses with smaller subsets of data and generated ~5 peer-reviewed papers (4 submitted) and conference presentations focused on the social motivation theme of this project.

Project V: The goal of this project was to study with MRI and magnetoencephalography (MEG) in 80 youth with ASD and 80 matched typically developing controls (TDCs) characterized in Projects 1 and 4. Recruitment for Project 5 was delayed by MRI and MEG equipment orders that failed to arrive on time and recruitment was slower than anticipated. Nevertheless the researchers were able to meet their MRI scanning goals. Data analysis is still ongoing. ~10 peer reviewed papers have been published on this project.

Project VI: Excellent progress has been made in training records, lecture series, course development and workshops. There was no noted weakness.

#### Reviewer 5:

The project met all human subjects research stated objectives with the exception of a slight reduction in the overall number of subjects receiving MEG testing. Overall, the human subjects objectives were successfully met without problem. The data developed across human and animal study were in line with data proposed to be generated in the original research plan. The animal work did deviate due to difficulty making an initially proposed mouse line. The research team went with a different mouse model that initially proposed, a conditional KO *Cdh10* model mouse. This resulted in significant delays in the animal program. Despite these delays, the research team did generate a significant amount of applicable mouse data in the time available following delay. The only compromise to the animal program resulting from this was an insufficient number of animals in some, but not all, experiments. Animal experiments with insufficient numbers include those under Project 3 Figures 2 and 3. The data and information provided were applicable to the project objectives in the strategic plan.

#### Reviewer 6:

Project I: Project 1 was successful in the collection of case and control samples and the identification of GWAS signals at the 5p14 locus. Re-sequencing at this locus for the case and control samples proceeded successfully, but no significant SNPs were identified. The investigators hypothesized that DNA variation may not be the source of the GWAS signal, but do note that the results of a recent paper show a biological and functional mechanism potentially

explaining the GWAS signal (noncoding RNA antisense to moesin). Further research into the characterization of these phenomena are suggested. SNPs were not found in the targeted cadherin/procadherin loci (CDH9/CDH10), but a different locus (CDH26) exhibited 3 SNPs significantly associated with ASD, a novel finding. Functional studies on these SNPs are underway.

**Weaknesses:**

Although not all study hypotheses were proved, interesting and potentially significant results in line with the study aims were generated. It is not clear how the ancestral component of Specific Aim 1.3 was addressed in the stated results.

**Project 2:** The focus of this study switched from focusing on a panel of 100 ASD candidate genome regions to whole genome sequencing on a smaller subset of ASD individuals (70, 50 of which were ready for analysis at the time of this report). This was done due to advances in next generation sequencing technology, and was a justifiable change in protocol. The purpose of the analysis was to identify rare ASD variants and compare to publically available data. Lists of SNPs were derived from whole genome sequencing in the 50 subjects and compared to non-ASD sequences. The authors noted that not one of the SNPs was common to all ASD individuals, supporting genetic heterogeneity in ASD. Gene annotation uncovered 6 novel variants in ASD associated genes. Work is ongoing to integrate data from the remaining 20 subjects. Analysis of non-coding regions using estimated brain regulatory potentials uncovered 6 novel SNPs, although these results require additional study. A notable feature of this project has been the development of computational tools for annotation and whole gene sequencing which will serve future projects and has provided data from ASD subjects for future analysis.

**Weaknesses:**

Results from this project have yet to see publication, although data from 20 additional samples is forthcoming.

**Project 3:** Investigators successfully developed CDH10 knockout mice and were able to run behavioral tests. No differences in locomotor activity or social choice were observed. Pcdh10 knockout mice were obtained and social, behavioral, and functional tests run. Juvenile male Pcdh10<sup>+/-</sup> mice showed reduced social approach and investigation behaviors as well as long-term fear memory. Other results were null. The sociability deficit in males was partially recovered pharmacologically with d-cycloserine. An immunohistochemical study identified a potential mediator of social deficits in the form of the basilateral amygdala. The phenotypic characterization of Pcdh10<sup>+/-</sup> mice as a mouse model relevant to ASD was a notable achievement.

**Weaknesses:**

There were issues in generating the CDH9-CDH10 region deletion, so investigators focused on separate CDH9 and CDH10 deletion. Generation of CDH9 knockout mice was unsuccessful. Many of the behavioral/functional studies were of particularly small sample size; conclusions regarding non-significant comparisons should be more muted than as stated in the report (failure to reject the null does not signify equality). The veracity of many of the results quoted in the narrative is difficult to judge due to several poorly formatted and truncated tables and figures in

the document (Table 2, Figures 5, 7, 9, 11, 13, 15, 16, 17). Publication of these preliminary findings has yet to follow.

Project 4: Recruitment goals were adequately met. Changes to the goals of this project were made due to valid concerns regarding the generalizability of the risk variant findings on chromosome 5 from previous studies. Rather than association studies confined to ASD individuals, more comparative studies were conducted of ASD individuals vs. TDC individuals. These changes were justifiable. Data collection and cleaning has been recently completed. Five manuscripts have been published and 4 additional submitted.

**Weaknesses:**

Analytical objectives were not completely met at the time of the report, since data entry and cleaning was completed just before the reporting deadline. Several of the manuscripts appear to be incremental in scope. Due to procedural and recruitment hurdles, the specific aims for this study – relating social impairment and genetic status and characterizing the impact of risk SNPs on the outcome of the STAR treatment protocol – were not met. The investigators did make good use of the social and behavioral data collected as part of this study, but further analyses to advance the specific aims should be forthcoming. Several papers use the “forward entry” method in fitting linear models, which can result in biased estimates of targeted effects (although the likelihood of that here is low given the non-complexity of most of the linear models).

Project 5: Recruitment and MRI scanning goals were met. The publication of interim results based on subsets of the full data is noteworthy. Six methodological papers regarding diffuse tensor imaging (DTI) were published, and the methods developed and described will be applicable in the completion of Project 5 aims. Impressive progress has been made with regard to using fMRI and MEG to characterize social brain functioning and ASD risk factors, although publication of these results are awaiting additional data.

**Weaknesses:**

Image processing and statistical analysis were not completed as the grant period ended – analyses are ongoing and results are speculated to be forthcoming by the time the review response is due. None of the specific aims for project 5 were technically met, although the investigators were able to complete recruitment and data collection and publish interim results while full data analyses were forthcoming.

Project 6: Objectives were met. 4 undergraduate and 4 post-baccalaureate students from under-represented minorities were enrolled in the PART training program. The training program was extensive and the post-baccalaureate enrollees successfully progressed to graduate school or employment. In addition, numerous other undergraduate and graduate researchers participated outside the umbrella of the PART program.

Weakness: Information on outcomes for the 4 undergraduate students would have been helpful.

Reviewer 7:

This project is actually an interrelated group of six sets of scientific aims and objectives, as follows:

- I. Characterizing a postulated common inter-genetic variant in ASD (Hakonarson – PI).
- II. Understanding the contributions of rare variants utilizing single nucleotide polymorphisms (SNPs) and Copy Number Variants (CNVs) as risk-variants for autism (Bucan – PI).
- III. Developing animal (mouse) knockout models of these genetic influences on brain and behavior (Brodkin – PI and Abel – CoPI).
- IV. Characterizing the phenotypes of individuals carrying rare and common risk alleles (Schultz – PI, Mandell – CoPI).
- V. Characterizing the brain endophenotypes of subjects carrying rare and common risk alleles for ASD (Schultz, Verma, and Roberts – CoPIs)
- VI. Training in ASD diagnosis, treatment, and research for minority and non-minority students (Doehring – PI)

This set of projects included partnerships with Lincoln University, Temple University, and the School District of Philadelphia. The overriding purpose was to establish a new Center of Excellence for Autism Research located within the Children’s Hospital of Philadelphia (CHOP) and the University of Pennsylvania. This new research center is planned to work synergistically with the Center for Autism Research (CAR) which had previously engaged in only clinical research projects, allowing integration with basic science studies aimed at causal mechanisms of Autism Spectrum Disorders (ASD). CAR is also a close collaborator with the Center for Applied Genomics (CAG) for genetic studies of autism. The projects themselves focused on developing a better understanding of the relationship between genetic risk, brain, behavior, and treatment response. The peak single nucleotide polymorphism (SNP) and five other SNPs with strong signals lie in an area between two genes thought to encode membrane cell-adhesion molecules, cadherin 10 (CDH10) and cadherin 9 (CDH9), and so these were a focus for the animal models planned. The study designers proposed that different combinations of rare and common polygenetic risk factors produce the substrate for ASD disabilities. It is hoped that more effective treatments for individuals with ASD (and presumably the ultimate prevention of such disorders) would likely best arise from a more complete understanding of the specific underlying causal mechanisms involved. In addition, the project envisioned the creation of undergraduate and graduate level training programs for underrepresented minorities in collaboration with the University of Pennsylvania, Temple University and Lincoln University as a part of Project VI, with students working within the labs of researchers on the grant and receiving additional group and individual teaching and mentorship to prepare them for future professional work in the area of ASD.

The proposed work was directed towards the stated objectives.

The empirical questions to be answered within this interrelated set of research protocols were complex, but the studies were designed to address the results in detailed and precise ways. The changes to the original, anticipated study designs are addressed below, but none of the changes appear to have diminished the overall quality of the work.

A change was made within Project III involving deletions in the intergenetic regions Cdh9-Cdh10. Completing both deletions was found to be impractical and it was decided to generate a conditional deletion of the mouse genome at Cdh10 only, enlisting the assistance of an outside company to do so.

Within Project V, recruitment was slow initially but progressed by the conclusion of the project – the clinical portion of the project originally anticipated 480 subjects and although slow to roll out, the study ultimately reports enrolling 678 subjects, according to the final progress report. (The age range was expanded by two years to allow for more subjects.)

It appears that the program met its objectives or made significant progress in each of the six project areas. There was a question raised in an earlier review as to whether the goals for Project VI were sufficiently robust at the outset, but the grant was approved based on the plans outlined. It may be necessary to come up with a different paradigm altogether in order to recruit larger numbers of underrepresented minorities in any such academic setting in a truly meaningful manner. Work was on-going on several of the projects up to the final report, so although data was collected, analysis and publication, while planned, has not yet been completed.

The data and information provided were applicable to the project objectives listed in the strategic plan.

**Strengths:** The development of a clinical and research center for ASD at Children’s Hospital of Philadelphia and the University of Pennsylvania is a worthy goal in and of itself, especially given the increased numbers of people affected with ASD in recent years. This set of interrelated projects demonstrated a very multipronged approach to the issues and this center's work should be able to provide a strong basis for future discoveries. The imaging dataset particularly should yield much useful information over time. Despite a slow start on recruitment, ultimately a greater number of subjects were recruited than originally planned for the clinical studies.

**Weaknesses:** None.

**Criterion 2 - What is the likely beneficial impact of this project? If the likely beneficial impact is small, is it judged reasonable in light of the original proposal and the dollars budgeted?**

### ***STRENGTHS AND WEAKNESSES***

#### Reviewer 1:

The major benefit from this program is the recruitment and phenotyping of a very large cohort of children with ASD and age-matched controls. The CURE funding has been used as Core support for the Center for Autism Research and the recruitment of the subjects has enabled many current and future studies that will tap into this cohort. Thus, the beneficial impact of this effort will no doubt materialize over the next several years.

The use of phenotypic and genotypic information to help predict treatment outcomes (one of the specific aims in project 4) could potentially have enormous impact once implemented. These are complex studies and are not possible until a large cohort of individuals are gathered for analysis. It is likely that genetic information from this cohort will be added to much larger collections that will be used to understand the genetic architecture of ASD.

The mouse model may prove beneficial for the development of new pharmacological agents to treat the core or co-morbid features of ASD.

Reviewer 2:

Each of the sub-projects met goals of the proposed specific aims, with high impact for genetic, animal modeling, and neurophysiological characterization of autism, meeting or exceeding all expectations for the project.

Reviewer 3:

Strengths:

The main beneficial impact of this study is the power that it achieves through massive sample sizes relative to many other studies. Collecting and analyzing this much phenotypic, genetic, and neuroimaging data in the same sample is likely to provide extraordinarily novel insights into the mechanisms of behavior differences in ASD. A strength of projects 1 and 2 is a thorough and methodical approach to better understand genetic targets that have been uncovered by previous work. A strength of Project 5 is the dimensional approach to ASD symptom severity and its application to DTI data. The PUNCH approach is likely to improve our ability to attribute neural circuits to differing levels of impairment, and facilitate comparison with other clinical groups or subclinical populations with similar traits such as BAP.

Weaknesses:

Because much of the analyses have yet to be completed, it is unknown how much of the (considerable) potential impact will translate into actual impact. Another weakness noted by this reviewer is that the investigators may not be completely objective with regard to the social motivation hypothesis. They appear to have bought into this hypothesis rather wholesale, interpreting even negative results to support it. There is evidence that some percentage of individuals with ASD are highly socially motivated, but lack the skills, and a positive feedback loop of failure and anxiety serves to promote disengagement from social stimuli despite interest and motivation. This kind of scenario could equally explain the lack of “audience effect” seen in the ASD group in the JADD paper. The lack of group difference in eye tracking might also be consistent with this view, rather than overly-engaging nonsocial stimuli that render the test less sensitive, as might the superior sensitivity of interacting faces compared to static and dynamic, but the investigators seem unwilling to entertain the possibility that ASD may not be universally associated with diminished motivation for social stimuli.

Reviewer 4:

This funding helped to support the Center of Excellence for Autism Research at Children’s Hospital of Philadelphia, with partnerships at Lincoln and Temple Universities, the University of Pennsylvania, and the Philadelphia Public School System. Proposed studies are motivated by their own discoveries and entail studying ASD genomics intensively, and relating it to clinical presentation. Excellent progress has been made on this front. The next step would be to translate knowledge gained from this study into clinical practice or to develop effective treatments for individuals with autism.

Reviewer 5:

The social motivation focus on the human imaging work should help focus future treatment development in the autism field. The eye tracking and imaging modalities employed in the human subjects portion of this grant should be employed in the future as outcome measures in treatment trials and/or as means to subgroup individuals at baseline to potentially predict treatment response. The animal work including novel finding regarding cycloserine treatment results holds promise for targeted treatment development in a rare genetic form of ASD. There is less enthusiasm for the impact Projects 1 and 2 will have on future treatment and future research in the autism field.

Reviewer 6:

Probably the most significant impact of the project was the establishment of a substantial and impressive autism research infrastructure at the host institution (including the CAR), which will foster research at the institution for some time to come. The identification of novel variants related to autism heredity may give rise to novel therapies, but further research is to come. Development of the Pchd10+/- mouse model opens up many research opportunities as does the discovery of a potential pharmacologic agent for the restoration of sociability. Although these developments directly affect only animal research, translational research in the future will likely result from these discoveries.

Weaknesses:

Many of the significant aims were left incomplete due to the ambition of the projects and procedural, administrative, and recruitment delays, so the beneficial impact of this research is less than what was originally proposed. This concern is mitigated by the establishment of the CAR as well as the large amount of additional funding secured by the researchers to continue the incomplete work done under this study.

Reviewer 7:

The future benefits of such a center for research, embedded in an on-going clinical setting such as Children's Hospital of Philadelphia, should be substantial.

Such a center should have a favorable impact on early diagnosis within the city of Philadelphia, the state of Pennsylvania, and the surrounding region. Enhanced understanding of the substrate for ASD symptomatology may ultimately assist in decreasing the incidence and levels of disability, although that result remains in the future.

Although no new patent applications have been made so far, there are potential medication approaches to some of the abnormalities studied of which the research team is already aware and plans are being made to explore these further.

The future plans for this research as described in the final progress report are as follows:

Project I: The researchers plan to: 1) Characterize the role of MSN antisense function (MSNP1AS; moesin pseudogene 1, antisense) at the 5p14 locus in interfering with regulation and action of the moesin protein on the X chromosome, in cell-based assays and animal models.

2) Characterize GABA and glutamate gene networks related to ASD in preparation for clinical trials. The research group reports that there is a drug candidate (NS-105) that appears to be able to restore the glutamine function in persons with ASD and disruption of the glutamine-signaling pathway. In addition, there are drug candidates such as Topamax that may restore the GABA pathway function in persons with ASD who have GABA-network disruption.

Project II: The researchers' immediate plan is to finalize analyses integrating genetic findings with the phenotypic data for publications (collaboration between Drs. Bucan, Hakonarson and Schultz).

Project III: The researchers plan to continue phenotype characterization of the *Cdh10* conditional knockout mouse line, and to elucidate the mechanisms of reduced sociability in the *Pcdh10*<sup>+/-</sup> mouse line, and mechanisms of sociability rescue in this line. Based on the data generated in this project, they hypothesize that sociability deficits of *Pcdh10*<sup>+/-</sup> mice are attributable to deficits in glutamatergic signaling in neural circuits involving the basolateral amygdala. The researchers plan to carry out future studies to test this hypothesis and to test the efficacy of pharmacologic agents that modulate glutamatergic signaling in rescuing social behaviors in this mouse line. The researchers also plan to extend this project to translational studies in humans with ASD, testing the association of protocadherin and cadherin gene variants in human ASD, and testing the efficacy of pharmacologic agents that modulate glutamatergic signaling, such as d-cycloserine, in treatment of social behavior deficits in ASD.

Project IV & V: The researchers plan to continue data analyses and publication of the large amount data collected in Projects IV and V. Effort over the first 4 years of this work focused more on data collection than analysis, so an effort has been planned to finish the analyses and generate additional publications. The phenotypic and brain imaging databases established by Projects IV and V are very valuable scientific resources that will be utilized for several years. Data from Project IV will be utilized to focus in genotype-phenotype analyses and from Project V to integrate genetic data into unimodal analyses. Project V will also focus on multimodal pattern analyses for publication. In addition to generating new peer review publications, the data from Projects IV and V will be used for preliminary data in support of new grant applications.

Project VI: This training program project does not have scientific goals as such; however, the minority-training infrastructure will continue at the researchers' home institutions for other training programs.

Strengths: The research teams involved have multiple plans for additional work based on the discoveries and techniques utilized in these DOH CURE-funded projects, and much of the anticipated impact has yet to be realized.

Weaknesses: None.

**Criterion 3 - Did the project leverage additional funds or were additional project applications submitted?**

***STRENGTHS AND WEAKNESSES***

Reviewer 1:

The investigators associated with this multi-part project have submitted a large number of grant applications to both public and private funders. Many of these applications have been successful. Notable contributions have come from the Lurie Family Foundation. Other successful grant applications include the following:

1. Neural and behavioral mechanisms of sociability deficits in Pcdh10 mutant mice (PI: Brodtkin) \$1,250,000
2. Comprehensive phenotyping of mouse models of autism (PI: Abel) \$507,000
3. Characterizing IQ Impairments in ASD and testing their genetic foundations (PI: Schultz) \$481,886
4. Toward Outcome Measurement of Anxiety in Youth with Autism Spectrum Disorders (Subcontract to CHOP; PI: Schultz) \$902,300
5. Autism Treatment Network\* \$420,000
6. Novel computational methods for higher order diffusion MRI in autism (PI: Verma) \$3,561,199

Reviewer 2:

Numerous additional funding is proposed or submitted with the benefit of the DOH support. Infrastructure was a critical development due to DOH funds that will raise competitiveness for future NIH funding.

Reviewer 3:

Strengths: The team successfully competed for foundation grants for Project 1, 3, and 4, as well as industry and federal funding for Project 4, and federal funding for Project 5. Several other grant applications are planned across the projects to continue and expand the work, with the exception of Project 6.

Weaknesses: None noted.

Reviewer 4:

All projects except projects II and VI managed to leverage additional funding, especially Project IV and V which generated multiple successful grant applications. All projects, except Project VI, have grants in preparation.

Reviewer 5:

Projects 1, 3, 4, and 5 all successfully leveraged funds for additional related work. Clearly this grant project enabled these investigators to generate significant data that was key in receiving additional funding. This additional funding includes 1 million in foundation support of GABA and mGluR gene network analysis, 500k in foundation support of mouse phenotyping, a 2 million dollar industry biobehavioral marker project grant, an almost 1 million dollar NIH

outcome measures in ASD-associated anxiety grant, and a 3.5 million NIH grant for higher order MRI analysis in autism. The investigators also intend to apply for significantly more related funded across project areas in the near future. Overall, the leveraged funding received and plans for additional funding applications are impressive.

Reviewer 6:

The Center of Excellence for Autism Research was established just prior to the Pennsylvania Department of Health funding; so leveraging of additional funding was substantial. An additional \$10M in funding was secured from various sources including the Lurie Autism Foundation, Simons Foundation, Shire Pharmaceuticals, Autism Speaks, and three NIH R01 grants. Concrete plans for additional funding applications are provided and one NIH R01 is in review.

No weaknesses.

Reviewer 7:

Additional funds were leveraged.

The following additional related funded grants were listed in the final project report:

<b>Title of research project</b>	<b>Funding agency</b>	<b>Amount awarded:</b>
<b>Project I</b>		
Role of GABA and mGluR gene networks in ASD pathogenesis	Lurie Foundation	\$,1,000,000
<b>Project III</b>		
Comprehensive phenotyping of mouse models of ASD	Simons Foundation	\$507,710
<b>Project IV</b>		
Characterizing IQ impairments in ASD and testing their genetic foundations	Simons Foundation and Nancy Lurie Marks Foundation	\$481,886
Biobehavioral markers for anxiety in ASD	Shire Pharmaceuticals	\$2,000,000
Toward outcome measurement of anxiety in youth with ASD	NIH (Subcontract)	\$902,300
Autism Treatment Network	Autism Speaks	\$420,000
<b>Project V</b>		
Novel computational methods for higher order diffusion MRI in ASD	NIH( R01)	\$3,561,199
Quantifiable markers of ASD via multivariant MEGDTI combination	NIH (R01)	\$275,000

The additional grants listed as funded totaled \$9,148,095 in funding leveraged through the Pennsylvania Department of Health CURE grant. (In addition, there was one pending [\$600,000 made through NIH] grant application, as well as six non-funded grant applications, made to various entities that were listed.)

The researchers are planning to apply for additional funding in the future to continue or expand the research.

Project I: The researchers are planning to submit two grants to the NIH: 1) to characterize the role of MSN antisense function (MSNP1AS; moesin pseudogene 1, antisense) at the 5p14 locus in interfering with regulation and action of the moesin protein on the X chromosome, in cell based assays and animal models, and 2) to study autism clinical development through GABA and glutamate/mGluR mechanisms.

Project II: The researchers plan to submit a proposal in response to the NIH Program Announcement PAR-13-231 entitled, “The role of essential genes in ASD,” and will include experimental validation of rare exonic and regulatory (non-coding) variants identified during the CURE grant.

Project III. The researchers on this project intend to apply for additional NIH and private foundation grants to study amygdala development and function in protocadherin 10 +/-mice, and the role of amygdala functioning in social behavior development relevant to autism, as well as grants to identify pharmacologic agents that can rescue sociability and amygdala function in this mouse line. There is also intent to apply for NIH and private foundation grants to continue study of the Cdh10 conditional knockout mouse line.

Projects IV and V: Studying the relationships between genetics and neurobiology of ASD via imaging, behavioral and clinical features methods will remain the scientific focus of these researchers’ work. The core mission of the Center for Autism Research is to characterize the causes of ASD so that more effective treatments can be devised. Most of the researchers funded through this CURE Grant are either fully or largely dedicated to ASD research. To effectively compete for grant funds for research through the NIMH and other granting agencies with similar priorities, the researchers will continue this line of work. Project IV led to submission of an R01 in February of 2012, which was scored and will be resubmitted once more papers from Project 4 are in press. The researchers also plan to submit grants on genetics and brain imaging based on results from Project V. The work begun with this center grant was originally intended to provide the data, publication track record, and scientific motivation needed for future ASD grant applications.

Project VI: While the researchers have no immediate plans to apply for new grant funding to continue the minority training program, the training model developed as part of the CURE Center Grant will be implemented as a continued component of CAR’s future (minority and non-minority) undergraduate, post-baccalaureate, and graduate training on ASD. Moreover, the researchers who participated in this CURE Center grant also participate in a number of NIH funded graduate and post-doctoral training programs via NIH’s T32 grant mechanism. Experience with this minority-training program has been valuable and can provide a model to

enhance elements of minority training in these other programs, which also emphasize the need for aggressive new strategies in order to expand the pool of minority participants in research. This is a critical mission in light of the poor representation of certain minorities, and it requires that research organizations be as proactive as possible. It is intended that the consortium of undergraduate and graduate student research training programs that includes LEND, McNair, PennPREP, Temple University, and Lincoln University can continue to serve in this goal.

Strengths: This set of projects generated an additional \$9,148,095 in extramural funding from federal, industrial, and private foundation sources. This is a remarkable effort and bodes well..

Weaknesses: None.

**Criterion 4 - Did the project result in any peer-reviewed publications, licenses, patents or commercial development opportunities? Were any of these submitted/filed?**

***STRENGTHS AND WEAKNESSES***

Reviewer 1:

There are many peer-reviewed articles that have arisen from research related to these projects. Many of these papers deal with technical issues that have arisen in the conduct of the proposed studies. Many other papers do not directly address the specific aims laid out in the original grant but make access of the cohort of subjects that have been recruited or to some of the data that have been acquired. There is no doubt that the number of papers that should emerge from the first five years of this program should increase dramatically over the next several years as the voluminous genetic and imaging datasets are mined for results.

Reviewer 2:

More than 20 manuscripts have either been submitted or published, which noted grant support from the Pennsylvania Department of Health, and included several high-impact publications.

Reviewer 3:

Strengths: Ten papers are accepted or published in a variety of clinical and neuroscience journals.

Weaknesses: Many of these are review articles, rather than primary literature. Very little has been published on the first two aims of the project at this point, although several "in preparation" manuscripts are listed.

Reviewer 4:

Projects IV and V generated impressive numbers of publications, many of which are in high impact journals. One paper was published for Project II. It is unclear if any papers were published for Projects I and III.

Reviewer 5:

A number of peer reviewed papers related to this work have been published in high impact journals. Many more publications appear "in the works". No patents have been filed resulting

from this work have been filed which is somewhat surprising given the diagnostic potential of the ROC curve analysis and given the novel treatment implications of the animal work. It could be that future patents will be filed prior to publication/public dissemination.

#### Reviewer 6:

No licenses or patents were filed, but one – the reported CDH26 discovery is planned. 21 publications have been submitted, of which 11 have published/accepted status. Concrete plans to submit seven additional manuscripts were outlined and several studies under projects 4 and 5 are in progress, the results of which will result in several additional publications. Each of the publications was in line with the goals of each project.

Weakness: Projects 1 and 2 are under-represented in the publication list, but this is justifiable. The acceptance of a Project 1 manuscript submitted to JAMA would be a major step forward. The manuscripts (particularly those accepted/published) are in journals of varying quality, but are all in line with project goals.

#### Reviewer 7:

Peer-reviewed publications were listed in the final progress report as follows:

1. Varol, E., Gaonkar, B., Erus, G., Schultz, R., Davatzikos, C. (2012). “Feature ranking based nested support vector machine ensemble for medical image classification” Proceedings - International Symposium on Biomedical Imaging (ISBI), 146-149.
2. Varol, E., Gaonkar, B., Davatzikos, C. (2013). “Classifying medical images using morphological appearance manifolds” Proceedings - International Symposium on Biomedical Imaging (ISBI), 740-743.
3. Kohls, G., Chevallier, C., Troiani, V., & Schultz, R.T. (2012). “Social ‘wanting’ dysfunction in autism: Neurobiological underpinnings and treatment implications” Journal of Neurodevelopmental Disorders, 4(7), 1-20.
4. Bloy, L., Ingalhalikar, M., Eavani, H., Schultz, R.T., Roberts, T.P.L., & Verma, R. (2012). “White matter atlas generation using HARDI based automated parcellation” Neuroimage, 59(4), 4055-63.
5. Bloy, L., Ingalhalikar, M., Batmanghelich, N.K., Schultz, R.T., Roberts, T.P.L., & Verma, R. (2012). “An integrated framework for HARDI-based investigation of structural connectivity” Brain Connectivity, 2(2), 69-79.
6. Bloy, L., Ingalhalikar, M., Eavani, H., Roberts, T., Schultz, R.T., Verma, R. (2011). “HARDI based pattern classifiers for the identification of white matter pathologies” Medical Image Computing and Computer Assisted Intervention, 14(Pt 2), 234-41.
7. Ghanbari, Y., Herrington, J., Gur, R.C., Schultz, R.T., & Verma, R. (2013). “Locality preserving non-negative basis learning with graph embedding” in J.C. Gee, S. Joshi, K.M. Pohl, W.M. Wells, & L. Zollei. (Eds.), Information Processing in Medical Imaging 2013, Lecture Notes in Computer Science, Vol. 7917, pp. 316–327. Springer, Heidelberg.

In addition, the following were listed as being in various stages of pre-publication:

1. Ghanbari, Y., Smith, A., Schultz, R.T., Verma, R. “Connectivity subnetwork learning for pathology and developmental variations” Lecture Notes in Computer Science (in press).
2. Tunc, B., Ghanbari, Y., Smith, A., Pandey, J., Browne, A., Schultz, R.T., & Verma, R. PUNCH: Population Characterization of Heterogeneity (in preparation).

Two of the seven publications listed appear to be reports of presentations done at an international meeting and the team also listed two non-published works, one of which was in press and the other at some level of pre-publication preparation at the time of the final project report. The final report also notes that the group may have “mistakenly neglect[ed] to cite funding” from the Pennsylvania Department of Health in some resulting publications but does not report how many or where they may have been published.

There are mentions in the reports of planned publications, and there appear to be some opportunities for commercial (new medication) development that have been generated by this set of projects. The final report does discuss following up on them.

Other publications were included with the documentation which elevate the level of published productivity, as follows:

1. “The Impact of the Metabotropic Glutamate Receptor and Other Gene Family Interaction Networks on the Autism Spectrum Disorders” by Dexter Hadley, Zhi-liang Wu, Charlly Kao, Akshata Kini, Alisha Mohamed-Hadley, Kelly Thomas, Lyam Vazquez, Haijun Qiu, Frank Mentch, Renata Pellegrino, Cecilia Kim, AGP Consortium, Joseph Glessner, Hakon Hakonarson – which is listed as a “letter” and has no publication source included.
2. “Brief communication: The Role of mGluR Network Genes in Genetic and Environmental Forms of Syndromic Autism Spectrum Disorder” by Tara L. Wenger, Charlly Kao, Donna M. McDonald-McGinn, Elaine H. Zackai, Alice Bailey, Robert T. Schultz, Bernice E. Morrow, Beverly S. Emanuel, and Hakon Hakonarson, which looks like a draft submission.
3. “Whole-Genome Sequencing in an Autism Multiplex Family” by Lingling Shi<sup>1</sup>, Xu Zhang, Ryan Golhar, Frederick G Otieno, Mingze He, Cuiping Hou, Cecilia Kim, Brendan Keating, Gholson J Lyon, Kai Wang<sup>1</sup>, and Hakon Hakonarson which appears to have been published in Molecular Autism.
4. “From Mouse to Human: Evolutionary Genomics Analysis of Human Orthologs of Essential Genes” by Benjamin Georgi, Benjamin F. Voight, and Maja Buc’an reproduced from an “Open Access” research journal entitled PLOS Genetics.
5. “The Basolateral Amygdala (BLA) Is Activated during Social Approach and Investigation in Juvenile C57BL/6J Mice: BLA Activation during Juvenile Social Behavior” by Arati S. Kreibich, Matthew Torre, Cara T. Piccoli, Hongzhe-Li, Ruben Gur, Ted Abel, and Edward S. Brodtkin which appears to be a draft publication submission.
6. “Footprints in the Sand: Children with Autism do not Show Sequence Effects with Auditory Stimuli by Catherine Molesworth, Coralie Chevallier, and Francesca Happé which appears to be a manuscript from work done at King’s College London, prior to Coralie Chevallier’s move to the CAR at CHOP.
7. “Levels of Autistic Traits in Anorexia Nervosa: A Comparative Psychometric Study” by Annaig Courty, Anne Solène Maria, Christophe Lalanne, Damien Ringuenet, Christine Vindreau<sup>(3)</sup> ; Coralie Chevallier<sup>(4,5)</sup> ; Lydia Pouga<sup>(4)</sup> ; François Pinabel, Anne Philippe, Jean-Louis Adrien, Caroline Barry, and Sylvie Berthoz which also appears to be a draft submission done by Coralie Chevallier prior to her move to CAR at CHOP.

8. “Visual Attention to Dynamic Faces and Objects Is Linked to Face Processing Skills: A Combined Study of Children with Autism and Controls” by Julia Parish-Morris, Coralie Chevallier, Natasha Tonge, Janelle Letzen, Juhi Pandey and Robert T. Schultz published in Frontiers in Psychology.
9. “The Social Motivation Theory of Autism” by Coralie Chevallier, Gregor Kohls, Vanessa Troiani, Edward S. Brodtkin, and Robert T. Schultz, a review published in Trends in Cognitive Sciences.
10. “Reward Associations Modulate Awareness of Novel Objects” by Vanessa Troiani, Coralie Chevallier, and Robert T. Schultz which appears to be another draft manuscript.
11. “The Broad Autism Phenotype Predicts Child Functioning in Autism Spectrum Disorders” by Christina R. Maxwell, Julia Parish-Morris, Olivia Hsin, Jennifer C. Bush, and Robert T. Schultz which is another draft submission.
12. “Salient Social Cues are Prioritized in Autism Spectrum Disorders Despite Overall Increase in Social Attention” by Coralie Chevallier, Pascal Huguet, Francesca Happe´, Nathalie George, and Laurence Conty published in Journal of Autism and Developmental Disorders.
13. “Susceptibility to the Audience Effect Explains Performance Gap between Children with and without Autism in a Theory of Mind Task” by Coralie Chevallier, Julia Parish-Morris, Natasha Tonge, Loan Le, Judith Miller, and Robert T. Schultz, another draft submission.
14. “A Signal Detection Approach to Quantifying Social Motivation: Developing Tools for the Research Domain Criteria Framework” by Coralie Chevallier, Natasha Tonge, Vanessa Troiani, Gregor Kohls, Judith Miller, and Robert T. Schultz submission to The American Journal of Psychiatry.
15. “Social Rejection Enhances Preconscious Processing of Faces” by Coralie Chevallier, Vanessa Troiani, and Robert T Schultz submitted to Elsevier for publication as a “brief report.”
16. “Joint Analysis of Band-Specific Functional Connectivity and Signal Complexity in Autism by Yasser Ghanbari, Luke Bloy, J. Christopher Edgar, Lisa Blaskey, Ragini Verma , and Timothy P. L. Roberts from the CHOP Radiology Department, published in Journal of Autism and Developmental Disorders.

Strengths: The listed output of 12 publications seems quite reasonable for this level of grant funding, especially since an additional 13 journal submissions seem to have been generated.

Weaknesses: Some of the submitted papers listed may or may not ultimately see publication. One or two papers appear to have been included that originated from collaborations that may have been established prior to this project but involved faculty subsequently recruited to CAR. The oversight in omitting credit for the contributions of the Pennsylvania Department of Health grant in some papers seems like a very surprising flaw in such an otherwise ambitious and, as described at least, seemingly well-orchestrated project.

**Criterion 5 - Did the project enhance the quality and capacity for research at the grantee's institution?**

***STRENGTHS AND WEAKNESSES***

Reviewer 1:

This project was initiated soon after Dr. Schultz was recruited to establish the Center for Autism Research. A number of recruits have been brought to the University of Pennsylvania and the funding provided by this project is the single largest impetus to developing a research infrastructure and team to carry out modern, multidisciplinary autism research.

CURE funding was extremely helpful for bringing together the autism research community at Children's Hospital of Philadelphia and for the recruiting of new faculty (e.g., Drs. Herrington, Yerys, and Miller) at Children's Hospital of Philadelphia and the University of Pennsylvania.

Reviewer 2:

There was extensive support for training and infrastructure that led to a large recruitment effort. These factors definitively enhanced the research capacity of the institution.

Reviewer 3:

Strengths: The investigators note new collaborations, publications, resource development, and increased intra-institutional collaboration among the investigators involved in the project.

Weaknesses: None noted.

Reviewer 4:

This CURE grant was instrumental to the Center. 13 investigators were added to the Center, mostly through Project 4. In addition, 60 trainees were listed.

Reviewer 5:

Clearly this project enhanced the quality, breadth, and depth of this institution's work in the autism field. It is clear that this project came at a great time when the Director of the Children's Hospital of Philadelphia autism program arrived. Many new investigators were brought into this group to participate thus enhancing the overall research product. Many students were involved effectively in this work.

Reviewer 6:

Three pieces of equipment were purchased and used for the grant aims, and conceivably will see future research use. Pennsylvania Department of Health funds aided in the establishment of the center, consolidated autism research efforts at the host institution and fostered collaboration within and outside the institution. 14 new investigators were brought to the institution and 66 baccalaureate, master's, pre- and post-doctoral researchers participated.

No weaknesses.

Reviewer 7:

Overall it appears that the project enhanced the quality and capacity for research at the grantee's institution.

The development of the new Center itself is an improvement in infrastructure. An Agilan 2100 Bioanalyzer was purchased with grant funds of \$16,285.15 for Project II. It was anticipated that this piece of equipment might be used for other projects as well. Avisoft Bioacoustics software was purchased for \$4,265.25 (this represents half of the total cost of \$8,530.50 since the cost could be shared with another funded project) to measure ultrasonic (mouse) vocalizations in support of Project III, as was a computer for data analysis acquired for \$699.50.

New investigators were added or brought into the institution to help carry out this research. Benjamin Georgi, Ph.D., Predoctoral researcher, formerly with the Max Planck Institute in Berlin, Germany, was recruited for Project II. The following were recruited from elsewhere for Project IV: Preeti Prabhakar, M.S., Data Analyst, Hoffmann La Roche Inc., Nutley, New Jersey; Suzannah Ferraioli, M.S., a student from Rutgers University in New Brunswick, New Jersey; Stephanie Colantonio, B.A., a student from Yale University in New Haven, Connecticut; Kieran Rump, Ph.D., a Pre-doctoral Fellow from the University of Miami in Miami, Florida; Julianne Fretz, B.A., a student from the University of Maryland in College Park, Maryland; Lisa Guy, Ph.D., Staff Psychologist from the Marcus Institute in Atlanta, Georgia; Leandra Beery, Ph.D., a Pre-doctoral Intern from Chicago, Illinois; and Caitlan Stone, Ph.D., a Pre-doctoral Intern from Boston, Massachusetts. Project V included the recruitment of Harini Eavani, M.S., a graduate student from Ann Arbor, Michigan; Janelle Letzen, B.A., a student from the University of Florida in Gainesville, Florida, as well as a minority post-baccalaureate student funded via Project VI. In addition, Judith Miller, a clinical psychologist, was recruited from the University of Utah in Salt Lake City, Utah, to serve as CAR's new Director of Clinical Training to work with Dr. Doehring (Project VI).

Funds were used to pay for research performed by pre- or post-doctoral students as can be seen from the listing above.

**Strengths:** The establishment of the center is a significant advancement. The equipment purchases would appear to have been highly useful additions to the infrastructure of the center, and the recruitment of additional faculty is an outstanding strength.

**Weaknesses:** None.

**Criterion 6 - Did the project lead to collaboration with research partners outside of the institution, or new involvement with the community?**

***STRENGTHS AND WEAKNESSES***

Reviewer 1:

This funding led to an integration and consolidation of faculty interested in the basic biology, genetics, behavior and neurology of autism spectrum disorder. It was directed first and foremost

at developing capacity at Children's Hospital of Philadelphia and the University of Pennsylvania in autism research that will enable future multi-institutional collaborations.

Reviewer 2:

Over a dozen out-of-state researchers were recruited to participate in the research, a remarkable tally. Projects 4 & 5, in particular, led to extensive community outreach efforts.

Reviewer 3:

**Strengths:** The investigators list new collaborative relationships that emerged from Projects 1-5. Detailed description of community involvement associated with Projects 4 and 5 is given, with outreach and educational activities targeted to families as well as health and educational professionals. A successful public lecture series and educational workshop are also described.

**Weaknesses:** The nature of the collaborative relationships is not really described in much detail, particularly for Project 1.

Reviewer 4:

There was an excellent effort to collaborate. Multiple collaborations were initiated, both within and without of the institution.

Reviewer 5:

The project has resulted in new collaborations with investigators in Australia, Ireland, Connecticut, Washington DC, Jefferson University, Philadelphia, Pittsburgh, and Bucknell. Truly, the collaborations are international in scope and project driven collaborations span across the Commonwealth.

This appears to be a strength of the project.

Reviewer 6:

14 out-of-state researchers were recruited to participate in this project. Local, national, and international collaborations were established via 5 of the 6 program projects. Community outreach was a significant and notable component of the grant – attendance at over 350 community events, participation in Huddle Up for Autism, the Distinguished Lecture Series, participation in Next Steps, and the online autism Roadmap were noteworthy achievements.

No weaknesses.

Reviewer 7:

The project led to collaborations within the community and with other research institutions.

The final progress reports states that, as a result of this grant, Dr. Hakonarson and colleagues have established research collaborations in autism at The Children's Hospital in Melbourne Australia, and Queens University of Belfast in Northern Ireland, as well as The University of Connecticut Health Center in Farmington, Connecticut. The researchers have also enhanced collaborative efforts with the AGP consortium as a result of this research. Dr. Bucan has established collaboration with Dr. Toru Takumi (Riken Institute for Brain Research, Japan) and

with Dr. Matthew Dalva (Jefferson University, Philadelphia) on the experimental validation of variants found in Project II. This research has led to a new collaboration with Dr. Joshua Corbin, an Associate Professor of Pediatrics, Pharmacology, and Physiology at George Washington University School of Medicine and Health Sciences, who plans to work with the Brodtkin lab in assessing amygdala development in Pcdh10<sup>+/-</sup> mice. The researchers in Projects IV and V have begun collaborations with researchers in the new Geisinger-Bucknell Autism and Developmental Medicine Center in Lewisburg, Pennsylvania, based on data gathered from Projects IV and V. Drs. David Ledbetter, David Evans and colleagues are beginning new imaging and phenotypic studies at the Geisinger-Bucknell Autism Center, with assistance from researchers at CAR. This project has also led to new collaborations on phenotypic-genetic relationships with Dr. Bernie Devlin at the University of Pittsburgh, who has funding from a grant from the Simons Foundation.

According to the final progress report, 16 hospital and healthcare professionals were involved in this set of research projects.

Strengths: A number of new collaborations were reportedly initiated, some in Pennsylvania and others at distant venues.

Weaknesses: None.

## ***Section B. Recommendations***

### **SPECIFIC WEAKNESSES AND RECOMMENDATIONS**

#### Reviewer 1:

1. There were essentially no results presented for either Project 4 or Project 5. While it is appreciated that the recruitment and imaging was a major undertaking, there appeared to be little effort placed in analyzing the acquired data in real time. One concern is that there may have been a systematic problem in some aspect of the data collection which would only be appreciated after the project was over and not allowing for any mid-course corrections. These projects should be completed and the data presented in peer-reviewed publications.
2. It would be excellent to see further integration between the genetic data of Project 2 with the neuroimaging data of Project 4.
3. The real benefit of this first 4 year project will be the usefulness of the resource over the next several years. I would encourage the investigators to reach out to investigators even beyond their institution to analyze the neuroimaging and genetic databases that have been acquired.

#### Reviewer 2:

None.

#### Reviewer 3:

1. The oversight with regards to the figures in the report is concerning. When I see utter failure to proofread a final document like this it always brings up questions for me about how

thorough the investigators are in other aspects of the scientific enterprise. In the future, the investigators should be more attentive to the final product.

2. The most significant weakness across the projects is the fact that data collection and management has filled the entire project period, leaving many of the planned analyses unfinished at the time of the report. Although there is substantial record of productivity in publications that use parts of the dataset or that report methods development that will be applied to the dataset, it will be important to see the results of the planned analyses in the final sample to assess the success of the project.
3. The investigators appear unwilling to consider alternatives to the social motivation hypothesis of autism (see above). It is important for negative results to be acknowledged and alternative explanations to be considered for the observed data.

Reviewer 4:

There was no major weakness noted for the overall progress of the Center. The next step would be to translate knowledge gained from this study into clinical practice or to develop effective treatments. There are minor concerns with individual projects (see above), in which efforts need to be made to accomplish the original goals.

Reviewer 5:

There were no clear major weaknesses noted. The animal work needs completion on some tests given the delay in model development.

Reviewer 6:

Many of the unmet objectives were supplemented by additional research goals. Although these goals were worthwhile and resulted in advancements in knowledge and publications, the investigators are encouraged to further pursue studies in line with original study objectives where applicable and possible.

Reviewer 7:

None.

### **Generic Recommendations for Children's Hospital of Philadelphia**

Reviewer 1:

For a typical NIH grant, this program would be considered problematic since many of the specific aims have not yet been accomplished during the funding period. So, there has been very little integration of data across projects to answer questions such as the genetic basis of certain patterns of altered brain development. However, a major benchmark that was accomplished is the establishment of a large, and well phenotyped cohort of individuals which has produced a large database of various types of information that can be mined for years to come.

Reviewer 5:

The results from this project seem appropriate for the level of funding and the complexity of the work. While not 100% successful as a few MEG scans were missed, there was delay involving

the animal model, and less than earth shattering genetics results, the work is solid and really accomplished a great deal.

Reviewer 7:

This appears to have been a well-developed, interrelated set of research projects targeting ASD that will have a substantial beneficial impact over time.

***ADDITIONAL COMMENTS***

Reviewer 5:

Strengths include novel animal model development, successful MRI and MEG scanning of difficult populations, dissemination of scientific results in a timely manner, and generation of several new promising research collaborations resulting from this work.