

Pennsylvania Department of Health Final Performance Summary Report 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 report and final progress reports 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?
 - Were the research design and methods adequate in light of the project objectives?
 - Consider these questions about 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 projects) 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 research 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?**
 - What is the significance of this project for improving health?
 - Consider the value of the research completed towards eventual improvement in health outcomes.
 - 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 any additional grant applications submitted as a result of this project?**
 - 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.

- **Criterion 5 - Did the project enhance the quality and capacity for research at the grantee's institution?**
 - Were there improvements made to infrastructure?
 - 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: Favorable (2.00)

Project Rating:

Project	Title	Average Score
0991601	Modeling the Epigenetic Changes in Alternative Promoters of Cancer Genes	Favorable (2.00)
0991602	Characterizing Mechanisms of Transcriptional Activation Using Live Cell Imaging	Favorable (2.00)
0991603	Elucidation of the Integrator Composition and Function	Favorable (2.00)

Project Number: 0991601
Project Title: Modeling the Epigenetic Changes in
Alternative Promoters of Cancer Genes
Investigator: Davuluri, Ramana V.

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:

To date, the PI has made progress on the stated objectives of (1) the development of computational tools for annotating alternative promoters and pre-mRNA isoforms, that are differentially used in different human cancers; (2) identifying differentially regulated alternative promoters in cancer genomes, pre-mRNAs, and associated local epigenetic modifications in various cancer genomes by integrative ChIP-seq and bioinformatics approaches; and, (3) performing bioinformatics analysis to discover tissue- and cancer-specific promoters, modeling altered chromatin structures, and identifying epigenetic signatures of cancer cells.

There are some questions with regards to the research design and methods that are not provided in great detail but are highly relevant to the proposed work, such as: 1) How comparable are the data collections in the public domain? 2) Were there any normalization steps taken? 3) What are the in-depth methods taken to process the data? This becomes more important with RNA-seq, since there is greater variability between sequencing centers, machines, etc. Also, the Web link to the Web interface was not working, <http://bioinformatics.wistar.upenn.edu/CancerPromDb/>, therefore making it hard to evaluate.

What kind of evaluation was done comparing RNA-seq with exon arrays? How consistent are the findings?

The notion that isoforms have improved ability to distinguish tumor versus normal samples is very interesting, and the PI shows some compelling examples. However, a more in-depth description and systematic analysis on the events would be insightful. What clustering method was used, how was this selected, and how robust was this difference at the gene vs. isoform level if the clustering method was altered?

With regards to empirical results, the data developed was in line with the original research protocol; however, more in-depth results could have been provided summarizing the findings from such a diverse sequencing data collection.

No changes were made to the research protocol requiring an explanation.

Reviewer 2:

The investigators completed research that addressed some of the stated objectives. With regard to aim one, the researchers were successful in developing a database that identifies promoter elements in cancer and non-cancer tissues. However, it does not appear that RNA data have been integrated into this database, although the researchers report plans to do so.

With regard to aims two and three, the main finding reported by the researchers is that isoform-level measures of expression can be more effectively used to discriminate cancer and non-cancer cell lines than can gene-level measures of expression. They provide interesting examples where changes at the isoform level cancel each other out, making these changes invisible when looking at the gene level. This is an interesting finding. However, the researchers did not seem to address effectively their goals of finding cancer-associated epigenetic modifications or modeling altered chromatin structures to define unique and common epigenetic signatures in different cancer cells.

Reviewer 3:

The project has three specific aims that address important issues in gene regulation in cancer. The investigators made progress in all three stated objectives. A database has been developed and is open to the public. Grants have been submitted based on the research. Papers have been submitted. Overall, the project has met the stated objectives.

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?

STRENGTHS AND WEAKNESSES

Reviewer 1:

The proposed work will not be of any immediate translation impact for improving cancer research. The proposed work is a resource for the community, and any findings made from leveraging this resource will take time to translate. Therefore, I would consider the value of the research as a longer-term investment for improving health outcomes.

The proposed research would be greatly strengthened by processing, analyzing, and integrating more data collections with clinical information in order to generate new approaches for prevention, diagnosis and treatment of cancer. However, to date there is nothing along these lines attributable to the completed research project.

While the PI states that a novel method to classify cancers (e.g., gliomas) based on isoform-level gene expression signatures is currently under development, there is insufficient description to make any evaluation of progress.

Reviewer 2:

The database developed to address aim one may facilitate progress by other researchers by elucidating genomic regions involved in controlling the expression of genes. The work described in response to aims two and three may lead to a better understanding of the differences between transcriptional patterns in cancer and normal tissues. Findings that show that isoform

expression levels provide better information for classification of cancer and non-cancer samples than do gene-level measures of expression may provide deeper insight into differences between cancer and non-cancer tissues and may lead to some diagnostic tools that might provide useful measures of cancer severity to practitioners. If their research is ultimately published, the likely impact is probably in line with the dollars awarded.

Reviewer 3:

Most of the current genomics studies focus on gene expression without paying sufficient attention to the alternative promoter driven transcription that generates alternative transcripts, which can be critical, since they will produce protein isoforms that may even have opposite functions. Investigations at this level will likely uncover newer deregulation important for cancer. The investigators already found some examples in this regard. Further studies along this line have potential in finding better diagnosis and prognosis markers and identifying novel mechanisms for cancer hallmarks and for targets of therapeutics.

Criterion 3 - Did the project leverage additional funds or were any additional grant applications submitted as a result of this project?

STRENGTHS AND WEAKNESSES

Reviewer 1:

The PI leveraged additional funds and intends to apply for more, both of which are pending.

Reviewer 2:

Two grants were submitted to NIH. Two future NIH submissions were planned. The outcome of the funding applications is unknown.

Reviewer 3:

The investigators indicated co-funding of the American Cancer Society, although it is not clear who the PI is, since it is not shown in the biosketch of the PI. The investigators have submitted two grants to NIH pending review.

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:

The PI submitted an article to *Genome Medicine*. There are no licenses, patents or any commercial developments planned.

Reviewer 2:

One manuscript was submitted to *Genome Medicine* in August, 2011. I was unable to find evidence that this paper has been published. This is a relatively low level of productivity in terms of peer-reviewed publication. No other activities involving licenses, patents or commercial development were reported.

Reviewer 3:

Publication is relatively weak. Only one paper was submitted to a relatively small journal. This causes some concern regarding the productivity of this research team.

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

STRENGTHS AND WEAKNESSES

Reviewer 1:

Presuming the proposed Web site works, this should be a resource not only to the grantee's institution but to the larger research community.

The funds were used to cover the costs of post-doctoral fellows but not any pre-doctoral students.

Reviewer 2:

Funding increased the capability to conduct large-scale ChIP-seq and RNA-seq studies. Three post-doctoral fellows were partly supported by the project. No students were supported by the project. There was no reported recruitment of out-of-state researchers.

Reviewer 3:

A database was developed that should be important for the infrastructure. The PI is involved in several grants directed by other PIs, thus it is apparent that this team is important for various research efforts at the institute.

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:

The researchers have initiated two collaborations, one at the University of Pennsylvania and the other with Weill Cornell Medical College.

Reviewer 2:

Two new collaborations were formed with Dr. Donald O'Rourke, Director of the University of Pennsylvania Brain Tumor Tissue Bank, neurosurgeon, the University of Pennsylvania; and Dr. Vivek Mittal, an internationally recognized cancer researcher and the Director of the Lehman Brothers Lung Cancer Laboratory at Weill Cornell Medical College, Cornell University, New York, NY.

Reviewer 3:

The investigators listed two new collaborations, although it is not clear what the nature of collaboration is, since there are no joint publications.

Section B. Recommendations

SPECIFIC WEAKNESSES AND RECOMMENDATIONS

Reviewer 1:

1. It would be beneficial to see more in-depth analysis on the methods behind CancerPromDb. The ability to accurately analyze Chip-Seq and particularly RNA-Seq is a rapidly developing area of research that would require more in-depth descriptions. Furthermore, understanding the technical comparisons between exon array and RNA-Seq would be beneficial for interpreting the results.
2. What are the technical limitations of evaluating differential gene expression, both at gene and isoform-level, between (i) all normal and all cancer cell lines, (ii) normal breast (HMEC) and breast cancer (MCF7), and (iii) normal melanocytes and melanoma cell lines? What noise is introduced by integrating these data sets? And how do the researchers rule out that the consistently up-/down-regulated genes are not artifacts in the data? What about tumor specific alterations?
3. The researchers describe analyzing 160 cell lines of tumor tissue origin. However, when they conduct the pathway analysis they identify a breast-cancer regulation by stathmin1 as a significantly enriched pathway. What is the interpretation of this finding? Perhaps more work could be placed on determining, via in silico and experimental methods, the accuracy of such an approach. It is unclear whether this is relevant or a false positive.
4. The annotation resource will be valuable to the community, but the inability to access the web site is concerning.
5. It would be beneficial to see more of the progress made looking at specific cancer cohorts such as TCGA to assess the significance of this approach in a cancer context.

Reviewer 2:

1. One manuscript was submitted to Genome Medicine in August, 2011. I was unable to find evidence that this paper has been published. This is a relatively low level of productivity in terms of peer-reviewed publication. No other activities involving licenses, patents, or commercial development were reported. The researchers should do a better job of promptly publishing the results of their funded research.
2. The investigators should either carry out all the proposed research or explain why some of the planned research was not completed. Specifically in question is the work on finding cancer-associated epigenetic modifications and modeling altered chromatin structures to define unique and common epigenetic signatures in different cancer cells.

Reviewer 3:

The productivity of this research group in terms of publication is relatively weak. With this relatively novel research area and the amount of funding available to this group, one would expect better and more publications, which would allow reviewers to better evaluate the success

of this group. Currently, this review has to take the investigator's words for it and relies more on the potential of the project rather than the tangible outcome for the rating.

Project Number: 0991602
Project Title: Characterizing Mechanisms of Transcriptional
Activation Using Live Cell Imaging
Investigator: Janicki, Susan M.

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 was a very ambitious project, one that challenged the limits of light microscopy. The project did not meet all its stated objectives, but the PI made changes to the study design to accommodate the insufficient fluorescent signal from the PI's new construct.

The strengths of the proposal were the logical development of the aim and the impressive preliminary data. However, experimental findings demonstrated that several underlying assumptions in the experimental design could not be overcome. The PI designed a construct that would allow for detection of green fluorescent protein (GFP)-tagged transcription factor assembly at a transcription binding site. Aim 1 failed because it was assumed that transcription factor assembly was required at a DNA binding site. This assembly process was anticipated to provide temporal dissociation sufficient to do kinetic analysis of factor assembly; so selective depletion of regulatory factors failed to result in any sort of kinetic delay that would be analyzable. The underlying assumption for Aim 2 was that a p53 activation domain construct would have a signal similar to the VP15 activator. Unfortunately this was not the case, in that the p53 activation domain had half the activity of the VP15 activator. This resulted in a fluorescent signal that was too weak for unambiguous detection.

Upon recognizing these failings, the PI redirected the research to more fruitful areas, such as a way to analyze gene silencing. One aspect of the work not considered by the PI is that her imaging system may not be sensitive enough for the studies she proposed. There are newer, more sensitive cameras that have better sensitivity in low light level conditions whose operational characteristics are superior to the Hitachi Orca cameras. (Andor is one company that comes to mind.) It is also possible that the Hitachi Orca camera has not been set to its highest sensitivity just out of the box.

Reviewer 2:

The first aim of the project is to *Interrogate by shRNA depletion the requirement of known regulatory factors on (i) chromatin decondensation; (ii) pre-initiation complex assembly; and (iii) RNA synthesis, using kinetic, live cell imaging of single cells.* The original goal was to employ the transgene array to understand the recruitment timing of the transcriptional activation

machinery. The assay used, unfortunately, does not have the temporal resolution needed to discern the near synchronicity of the recruitment of both the activators and the activation factors. The investigators decided not to pursue it further. In addition, several knockdown experiments have failed to produce phenotype in transcriptional activity, suggesting that there is an ample redundancy in the system. The investigators have altered their study to focus on this system as a model of how replication-independent histone assembly regulates gene silencing at a region of repetitive DNA. This is a reasonable change in direction considering the exploratory nature of the proposal.

The strength of the project is that live cell imaging technique is appropriately leveraged to study these very challenging biological events. There are, however several weaknesses in the execution of the assays. First, the dynamic changes of the fluorescent intensity should be translated into actual rates (preferably initial rates) to tease out subtle differences in biological processes. Second, the investigators missed an opportunity to study the turnover rates of these proteins on the complexes. While the apparent molecular recruitment may not show appreciable distinction in dynamics, the actual turnover rate and therefore the dissociation constants may differ, and this can be teased out easily using simple techniques such as fluorescence recovery after photobleaching (FRAP).

Use p53 as a model transcriptional activator to interrogate the timing of its activation kinetics using live cell imaging analysis. The results are less than ideal, and the conclusions are weak. The investigators did not achieve the proposed goal, and no alternative plan was pursued.

Reviewer 3:

The project made some progress in terms of the establishment of a system to perform live cell imaging and visualization of the HAT dynamics. However, it was impossible to address the timing of transcriptional activation events and to significantly reduce transcriptional activation in the transgene array system. Also, it was impossible to study the activation kinetics of p53 using the single-cell imaging because of its lower binding to the transcription site when compared with the VP16 activator. Nevertheless, the established system should facilitate future research in these areas.

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?

STRENGTHS AND WEAKNESSES

Reviewer 1:

The original premise of the proposal was that transcription factors could be visualized to analyze how transcription forks are controlled. The PI has clearly demonstrated that this technology can be used to visualize assembly of transcription factors and histone remodeling in real time. While there is no direct result on improving health, this work will further our understanding of this basic biological process.

No drug discoveries are anticipated from this work.

The research has been redirected to identify mechanisms of gene repression, an area that is understudied and will have important consequences for cancer research. One can envision therapies in the future that shut down oncogenes in tumor cells.

Reviewer 2:

It is highly doubtful that the project will result in any discernible scientific impact. The investment cannot be fully justified.

Reviewer 3:

This support has enabled the setup of an advanced imaging system to investigate the mechanisms of transcriptional activation using live cell imaging. This will be useful for studying chromatin assembly in the PI's lab.

Discoveries made so far have no immediate impacts on human health.

Criterion 3 - Did the project leverage additional funds or were any additional grant applications submitted as a result of this project?

STRENGTHS AND WEAKNESSES

Reviewer 1:

No additional funds have been obtained in support of the project. The PI intends to submit a grant for R01 funding on her work on gene repression, which is a new direction for the project.

Reviewer 2:

The project did not leverage further funding.

Reviewer 3:

No additional funds have been leveraged, and no grant application has been submitted. However, the PI plans to submit an R01 proposal to NIH to study the functions of the chromatin assembly machinery using the established imaging system.

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:

One manuscript was submitted to the Proceedings of the National Academy of Sciences (PNAS); review is pending.

Reviewer 2:

One manuscript was in review at the time this report was submitted.

Reviewer 3:

The project has resulted in one manuscript submitted to PNAS in July 2011. However, it appears that it has not been published yet according to the PubMed search. No invention, patent or commercial products have been generated.

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

STRENGTHS AND WEAKNESSES

Reviewer 1:

Funds were used to pay for pre- and post-doctoral students.

Reviewer 2:

There was an upgrade made to the PI's imaging system. Overall, nothing significant has been made to the grantee's institution.

Reviewer 3:

The project enabled the purchase of Laser MM Upgrade. This should enhance the capability of the PI and other investigators on campus to use the system for live cell imaging. Pre- and post-doctoral students have also been supported by the fund.

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:

There are no current plans to begin collaborations.

Reviewer 2:

The investigator did not present any plan to begin collaborations.

Reviewer 3:

None

Section B. Recommendations

Specific Weaknesses and Recommendations

Reviewer 1:

1. I believe that Aim2 is still salvageable with a better fluorescence detector/camera. I recommend that the PI be given additional funds to support the purchase of a new detector for the microscopy system she has in the laboratory. She should seek advice from experts in light microscopy or fluorescence detectors (James Pawley, University of Wisconsin-Madison comes to mind) before purchasing a new detector.
2. I believe the gene silencing work is very exciting with great potential for cancer research. This work should be given additional funds for development.

Reviewer 2:

There are several weaknesses in the execution of the assays. First, the dynamic changes of the fluorescent intensity should be translated into actual rates (preferably initial rates) to tease out subtle differences in biological processes. Second, the investigators missed an opportunity to study the turnover rates of these proteins on the complexes. While the apparent molecular recruitment may not show appreciable distinction in dynamics, the actual turnover rate and therefore the dissociation constants may differ, and this can be teased out easily using simple techniques such as fluorescence recovery after photobleaching (FRAP).

Reviewer 3:

1. The proposed research is innovative and exciting. However, the PI encountered several technical hurdles that prevented the PI from pursuing the original goals. This may be related to the short funding period. One strategy is to allow the PI to have a no-cost extension to solve the technical challenges. For example, the established transgene array did not permit the proposed research. If time permits, the PI could modify the transgene array or establish different arrays.
2. The funding appears mainly to provide salary support to the PI (74% effort). Is the funding designed to relieve the PI from other duties (teaching, service, etc.)? Also the effort for the PI seems to be too high.

Project Number: 0991603
Project Title: Elucidation of the Integrator Composition and Function
Investigator: Shiekhattar, Ramin

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:

The research direction was altered, and interesting new avenues of research were pursued. The research design and methods were adequate. The data was developed sufficiently to answer the research questions posed. However, the data was not entirely in line with the original research protocol. There was ample explanation for the decision to pursue the integrator complex in the transcription of protein coding genes; however, there was no explanation for the decision to abandon previously proposed experiments. The progress report was quite complete. The data and information all pertained to the characterization of the integrator complex.

The proposed project was centered upon the characterization of the integrator multi-protein complex and its role in snRNA processing. The initial aims of the project were 1) to further analyze the subunits of the complex and to reconstitute the complex using recombinant proteins, and 2) to identify the components of the complex that mediate association with the carboxyl-terminal domain (CTD) of RNA polymerase II. The investigators made great progress in Aim 1 of the proposal. They were able to purify and sequence the integrator associated proteins and to develop an *in vitro* RNAase protection assay to assess the processing of snRNAs. Further, they were able to show that ints1, 2, 3, 4, 9, and 11 are important for the processing of the U2 snRNA as well as for transcription.

While part of Aim 1 was completed, the focus of the research veered from the originally planned research based on findings in early experiments. While characterizing the complex, the laboratory found that the integrator complex was strongly associated with the chromatin fraction, leading the investigators to suspect that the integrator complex played a broader role in RNA Pol II transcription. In subsequent experiments, investigators showed that the integrator complex does indeed associate with the 5' ends of protein coding genes (as compared to the 3' ends of the snRNA genes) and plays an important role in the activation of transcription.

Reviewer 2:

The majority of the work described relates to Aim 2, which makes it difficult to assess the extent of progress on Aim 1. Aim 2 and the expansion of investigation of how the integrator complex might globally affect transcription are interesting and potentially of high scientific impact, but

the project report does not give a clear description of how the work accomplished relates to the original goals.

The data provided appear to be high quality, but linkage to the aims is not well explained. Also, given that the project was co-funded by NIH, it is difficult to assess what aspects of the work are the outcomes of DOH vs. NIH funding.

Reviewer 3:

The project proposed to investigate the function of integrator, a protein complex involved in the processing of the 3'-end and snRNA and perhaps mRNAs. The specific objectives were to characterize and reconstitute the complex using recombinant proteins and to identify which components mediate association with the CTD of Pol II. As is sometimes the case, however, the research has wandered in a somewhat different and much more functional direction, rather than the more biochemical characterization of the components of integrator and its interaction with the polymerase CTD that was proposed in the specific aims. Instead of the reconstitution of the complex, the focus was on integrator purified from chromatin which was then used to characterize its location on the genome. These experiments led to the discovery of a new role for integrator in transcriptional activation and chromatin architecture, wider than was previously believed, by extending beyond snRNA transcription and RNA processing. In addition, the experiments led to a new set of hypotheses on how this protein complex functions in regulating transcription, perhaps in suppressing aberrant transcripts. The research design and methods, as described in the project reports, were in line with this new direction in the project. Mention was made in the report of the original more biochemical goals of characterizing the complex components and reconstituting it, specifically with regards to the analysis of subunits (although less on reconstitution), but the focus of the report was largely on a functional rather than biochemical topic that was somewhat distinct from the original specific aims but no less interesting. Thus, experiments were extended to the *in vivo* functional analysis, but Specific Aim 2 appears to have been substantially neglected.

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?

STRENGTHS AND WEAKNESSES

Reviewer 1:

Understanding the regulation of snRNA processing and transcription regulation is key to understanding alterations in normal processes that can lead to disease.

It is quite interesting that the integrator complex plays a dual role in transcription and processing of snRNA genes, as well as in transcription of protein coding genes. The finding is novel and the data robust. The future directions of the research project were minimally discussed.

Reviewer 2:

The project is likely to give new insight into an important aspect of transcriptional regulation in mammalian cells, as well as into the mechanisms for regulation of the splicing machinery. While specific health-related impact is not obvious at this point, the project will provide

important information about fundamental cellular processes that are likely to impact cancer. Future plans are not clearly described, making this difficult to evaluate.

Reviewer 3:

The value of the project lies entirely in understanding the fundamental mechanism of gene expression and its regulation. Transcription and RNA processing are of course fundamental steps of gene expression, and all biological processes and diseases depend on their correct execution. Therefore, understanding how a new complex of proteins with activity in RNA processing also participates in activation of transcription, is potentially very important for its long-term impact on understanding basic human biology. There were, however, no new drug discoveries, and none should have been expected, given the fundamental character of the project. Concerning future plans, the investigator makes a generic statement that they plan to gain understanding of how the integrator functions in transcriptional activation, as they should, but provides no specific plans of how this will be done and from where resources will come. I was surprised, however, to read that the project was awarded in excess of \$900K, and I have concerns about the value for money resulting from such a large award over such a short period of time.

Criterion 3 - Did the project leverage additional funds or were any additional grant applications submitted as a result of this project?

STRENGTHS AND WEAKNESSES

Reviewer 1:

During the tenure of funding, Dr. Shiekhattar secured an R01 from NIH entitled, “Elucidation of Integrator’s Function.”

Reviewer 2:

The project was co-funded by an NIH grant. No leveraging of funds was attempted. No information is provided on how funds from each grant were used.

Reviewer 3:

No additional funds were secured or pursued, surprisingly; although I strongly suspect this work will be included in any NIH-funded grants that the investigator currently has or will apply for, and indeed NIH funds were used during the execution of the project.

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:

The researchers are planning to submit articles for peer-reviewed publication. A submission was planned for late 2011. If a manuscript was submitted, it has not yet become available to the public.

Reviewer 2:

No publications or development opportunities have resulted from the work; one publication is planned.

Reviewer 3:

No publication has been completed, which is perhaps not surprising given the relatively short time of the award; but the investigator states that a publication is forthcoming. This statement is credible given the data presented. There are no plans for commercialization or patents.

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

STRENGTHS AND WEAKNESSES

Reviewer 1:

There were no improvements made to infrastructure. There were no investigators added or researchers brought into the institution to help carry out this research. Funds were used to pay for research performed by four post-doctoral students. The funds were utilized to support an existing team of scientists.

Reviewer 2:

It does not appear that any improvements in infrastructure were made or that new investigators or researchers were brought into the project. One pre-doctoral and several post-doctoral trainees were supported. A significant amount of funds was used to provide salary for the PI. Overall, the ability to fund these researchers probably enhanced the research capacity at the institution, although this is not explicitly explained.

Reviewer 3:

It does not appear that any significant improvement was made at the Wistar Institute in regards to infrastructure or the involvement of new investigators, but funds were used to support pre- and post-doctoral students.

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:

No new collaborations were described.

Reviewer 2:

No collaborations or involvement with the community appear to have been attempted.

Reviewer 3:

No new collaborators were involved, and no community outreach initiatives were undertaken.

Section B. Recommendations

SPECIFIC WEAKNESSES AND RECOMMENDATIONS

Reviewer 1:

1. The novel roles for the integrator complex that were identified are important and are the major strength of the project. The project made acceptable progress, but failed to meet most of its stated objectives. The reasons for not pursuing the stated objectives should be clearly outlined. The replacement research should be well-rationalized and outlined, as well.
2. Efforts should be made to recruit new scientists and to be involved in trainee education. Note: Though the application cites support for post-doctoral trainees, Question 13 was answered, “No.” The four post-doctoral trainees should be included in the table, and the question should have been answered, “Yes.”
3. When the work is published, the Health Research CURE Program should be appropriately cited.
4. This was a strong basic science research project that yielded unexpected but interesting data. Similar research should continue to be supported, but training and recruitment should be fostered as part of the projects.

Reviewer 2:

1. Publication of the data is an important goal. Leveraging the results for future research outcomes is crucial, and future goals are not clearly outlined.
2. A clearer explanation of the progress on Aim 1, including how and why the project direction has apparently changed, would have been helpful in evaluation of the overall progress in the project.
3. There should be a better explanation of the roles of individual investigators. In addition, how the funds were used to improve the research capacity of the institution should be included.

Reviewer 3:

1. The execution of the project veered away in part from the original proposal. This is not a substantial weakness because that is how science works, and the end results were no less interesting or important.
2. The value for money and benefits seem to be out of line with the investment, if the figure of \$907K in 18 months is correctly understood by this reviewer.