Final Progress Report for Research Projects Funded by Health Research Grants

Instructions: Please complete all of the items as instructed. Do not delete instructions. Do not leave any items blank; responses must be provided for all items. If your response to an item is "None", please specify "None" as your response. "Not applicable" is not an acceptable response for any of the items. There is no limit to the length of your response to any question. Responses should be single-spaced, no smaller than 12-point type. The report **must be completed using MS Word**. Submitted reports must be Word documents; they should not be converted to pdf format. Questions? Contact Health Research Program staff at 717-783-2548.

1. **Grantee Institution:** Drexel University

2. **Reporting Period (start and end date of grant award period):** 1/1/2009-12/31/2010

3. **Grant Contact Person (First Name, M.I., Last Name, Degrees):** Anne Martella

4. **Grant Contact Person's Telephone Number:** (215) 895-6471

5. **Grant ME Number or SAP Number:** 4100047631

6. **Project Number and Title of Research Project:** 2 - *Developing Therapies for Treating Hereditary Spastic Paraplegia*

7. **Start and End Date of Research Project:** 1/1/2009 - 12/31/2009

8. **Name of Principal Investigator for the Research Project:** Peter Baas, PhD

9. **Research Project Expenses**.

   9(A) Please provide the amount of health research grant funds spent on this project for the entire duration of the grant, including any interest earned that was spent:

   $ 112,500

   9(B) Provide the last names (include first initial if multiple individuals with the same last name are listed) of **all** persons who worked on this research project and were supported with health research funds. Include position titles (Principal Investigator, Graduate Assistant, Post-doctoral Fellow, etc.), percent of effort on project and total health research funds expended for the position. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name	Position Title	% of Effort on Project	Cost
Baas	Professor	5%	$6,929
Lin	Research Associate	50%	$18,500
Sudo	Research Associate	50%	$18,481

9(C) Provide the names of **all** persons who worked on this research project, but who *were not* supported with health research funds. Include position titles (Research Assistant, Administrative Assistant, etc.) and percent of effort on project. For multiple year projects, if percent of effort varied from year to year, report in the % of Effort column the effort by year 1, 2, 3, etc. of the project (x% Yr 1; z% Yr 2-3).

Last Name	Position Title	% of Effort on Project
None		

9(D) Provide a list of **all** scientific equipment purchased as part of this research grant, a short description of the value (benefit) derived by the institution from this equipment, and the cost of the equipment.

Type of Scientific Equipment	Value Derived	Cost
None		

10. Co-funding of Research Project during Health Research Grant Award Period. Did this research project receive funding from any other source during the project period when it was supported by the health research grant?

Yes_____ No_____x____

If yes, please indicate the source and amount of other funds:

11. Leveraging of Additional Funds

11(A) As a result of the health research funds provided for this research project, were you able to apply for and/or obtain funding from other sources to continue or expand the research?

Yes___x_____ No_____

If yes, please list the applications submitted (column A), the funding agency (National Institutes of Health—NIH, or other source in column B), the month and year when the application was submitted (column C), and the amount of funds requested (column D). If you have received a notice that the grant will be funded, please indicate the amount of funds to be awarded (column E). If the grant was not funded, insert "not funded" in column E.

Do not include funding from your own institution or from CURE (tobacco settlement funds). Do not include grants submitted prior to the start date of the grant as shown in Question 2. If

you list grants submitted within 1-6 months of the start date of this grant, add a statement below the table indicating how the data/results from this project were used to secure that grant.

A. Title of research project on grant application	B. Funding agency (check those that apply)	C. Month and Year Submitted	D. Amount of funds requested:	E. Amount of funds to be awarded:
I have a subcontract on an NIH R01 awarded to Dr. Gerardo Morfini of UIC. The title is: Axonal transport deficits during heredistary spastic paraplegia The amounts listed here are for my contract only.	☐NIH ☐ Other federal (specify:_____ _____) ☐ Nonfederal source (specify: _____)	July 2009	$100,000 direct cost per year for 5 years	$84,000 direct costs per year for 5 years

11(B) Are you planning to apply for additional funding in the future to continue or expand the research?

Yes____x____ No_____

If yes, please describe your plans:

I hope to submit a small foundation grant to the Hereditary Spastic Paraplegia Foundation and also to submit an R01 to the NIH.

12. Future of Research Project. What are the future plans for this research project?

Continue the research essentially as started here.

13. New Investigator Training and Development. Did students participate in project supported internships or graduate or post-graduate training for at least one semester or one summer?

Yes_____ No_____x____

If yes, how many students? Please specify in the tables below:

	Undergraduate	Masters	Pre-doc	Post-doc
Male				
Female				
Unknown				
Total				

	Undergraduate	Masters	Pre-doc	Post-doc
Hispanic				
Non-Hispanic				
Unknown				
Total				

	Undergraduate	Masters	Pre-doc	Post-doc
White				
Black				
Asian				
Other				
Unknown				
Total				

14. Recruitment of Out-of–State Researchers. Did you bring researchers into Pennsylvania to carry out this research project?

Yes_____ No_____x___

If yes, please list the name and degree of each researcher and his/her previous affiliation:

15. Impact on Research Capacity and Quality. Did the health research project enhance the quality and/or capacity of research at your institution?

Yes____x____ No_____

If yes, describe how improvements in infrastructure, the addition of new investigators, and other resources have led to more and better research.

The CURE award helped by providing a major translationally-directed component of one of our major research programs, which is instrumental for future funding from NIH.

16. Collaboration, business and community involvement.

16(A) Did the health research funds lead to collaboration with research partners outside of your institution (e.g., entire university, entire hospital system)?

Yes____x____ No_____

If yes, please describe the collaborations:

I wrote a grant application to the NIH with Dr. Gerardo Morfini (P.I. on the grant) of University of Illinois at Chicago (UIC) and it was funded. I have a subcontract on the grant. See 11(A) above.

16(B) Did the research project result in commercial development of any research products?

Yes_____ No_____x____

If yes, please describe commercial development activities that resulted from the research project:

16(C) Did the research lead to new involvement with the community?

Yes_____ No_____x____

If yes, please describe involvement with community groups that resulted from the research project:

17. Progress in Achieving Research Goals, Objectives and Aims.

List the project goals, objectives and specific aims (as contained in the grant application's strategic plan). Summarize the progress made in achieving these goals, objectives and aims for the entire grant award period. Indicate whether or not each goal/objective/aim was achieved; if something was not achieved, note the reasons why. Describe the methods used. If changes were made to the research goals/objectives/aims, methods, design or timeline since the original grant application was submitted, please describe the changes. Provide detailed results of the project. Include evidence of the data that was generated and analyzed, and provide tables, graphs, and figures of the data. List published abstracts, poster presentations and scientific meeting presentations at the end of the summary of progress; peer-reviewed publications should be listed under item 20.

This response should be a DETAILED report of the methods and findings. It is not sufficient to state that the work was completed. Insufficient information may result in an unfavorable performance review, which may jeopardize future funding. If research findings are pending publication you must still include enough detail for the expert peer reviewers to evaluate the progress during the course of the project.

Health research grants funded under the Tobacco Settlement Act will be evaluated via a performance review by an expert panel of researchers and clinicians who will assess project work using this Final Progress Report, all project Annual Reports and the project's strategic plan. After the final performance review of each project is complete, approximately 12-16 months after the end of the grant, this Final Progress Report, as well as the Final Performance

Review Report containing the comments of the expert review panel, and the grantee's written response to the Final Performance Review Report, will be posted on the CURE Web site.

There is no limit to the length of your response. Responses must be single-spaced below, no smaller than 12-point type. If you cut and paste text from a publication, be sure symbols print properly, e.g., the Greek symbol for alpha (α) and beta (ß) should not print as boxes (□) and include the appropriate citation(s). DO NOT DELETE THESE INSTRUCTIONS.

Summary of Research Completed

Introduction. Neurons express at least two enzymes that sever microtubules, namely katanin and spastin. Spastin is especially interesting because its mutations are the chief cause of hereditary spastic paraplegia (HSP) in humans. HSP results in gradual degeneration of the corticospinal tracts, leading to weakness and spasticity of the lower limbs. HSP is an autosomal dominant disease that typically displays no developmental abnormalities, with symptoms typically appearing in adults around the age of 35. The prevailing view that the degeneration is caused by haploinsufficiency, meaning that the lower levels of functional spastin are insufficient to carry out the necessary degree of microtubule-severing. However, the question arises as to why there are no ill effects during the development of the nervous system, and why the disease only afflicts certain tracts in the adult. Another possibility is that the mutant forms of spastin may have "gain of function" effects that cause certain axons to degenerate through cytotoxicity. However, again, the question remains as to why no such cytotoxic effects are apparent during development, in the peripheral nervous system, or in other tracts of the CNS. Resolving this mystery is critical for designing appropriate strategies for treating the disease, and to obtain a better understanding of the normal physiology of the severing proteins in neurons.

The SPG4 gene which encodes spastin has two start codons, and hence produces a longer protein called M1 and a slightly shorter protein called M87 in humans (or, in the case of rodents, M85). Both M1 and M87 can sever microtubules, although M87 is more effective. Interestingly, M87 is the only isoform detectable throughout the nervous system during development, and throughout almost all of the nervous system in the adult. M1 is only detectable in the adult spinal cord (where it comprises about 20% of the total spastin), and is virtually absent everywhere else. Thus, it is our hypothesis that when the SPG4 gene is mutated, the altered M1 and M87 proteins may or may not be able to sever microtubules, but this is irrelevant to the disease because the levels of functional spastin and katanin are perfectly sufficient for the vitality of the axon. Moreover, mutant M87 is harmless. By contrast, mutant M1 becomes abnormally folded and is cytotoxic. During aging, the proteasome system becomes gradually less effective at disposing of mutant M1, so that sufficient levels accumulate in the adult to elicit nerve degeneration. In order to make progress toward developing therapies for HSP, the first step is to resolve the manner by which spastin mutations actually cause axons to degenerate. This report represents our progress over the 12 months of funding.

Progress. We have published a paper in Human Molecular Genetics on the proposed experiments (J. Solowska, J.Y. Gabern, and P.W. Baas, 2010, Human Molecular Genetics 19: 2767-2779). This paper takes the approach of investigating a small number of mutations to human spastin that give rise to HSP but do not lie within regions of the molecule essential for function.

About 200 different mutations have been detected in the SPG4 gene in patients with HSP and most of these mutations occur within the AAA-domain, and hence almost certainly destroy the microtubule-severing activity of the spastin protein. The haploinsufficiency model assumes that the lower levels of functional spastin are insufficient to carry out the necessary degree of microtubule-severing. However, there are some reasons to doubt this model, such as the fact that enzymes are typically synthesized in amounts that are higher than needed, and their levels are often regulated by demand, such that one functional spastin gene may be sufficient to produce enough spastin. Second, there are other microtubule-severing proteins in neurons that could compensate for lower levels of spastin, the main one being katanin. Third, development of the nervous system is much more dependent upon adequate microtubule-severing than is the maintenance of adult axons, but there are no known developmental abnormalities in most HSP patients. In addition, spastin knockout mice do not suffer any developmental problems. Finally, although most pathogenic spastin mutations cause loss of spastin function, there are a small number of mutations outside of the AAA-domain that would not be expected to prohibit the protein from severing microtubules, and yet still cause the disease with equally strong symptoms as the other pathogenic spastin mutations. We previously explored the possibility that the lack of functional spastin can be compensated for by katanin. We also investigated the possibility that HSP may be caused by cytotoxicity that is specific to an isoform of spastin (termed M1) that is present in the adult spinal cord but not elsewhere in the adult or anywhere in the developing nervous system. Indeed, mutated M1, when experimentally expressed in cultured cortical neurons proved to be harmful to the axon. It is our hypothesis that mutant M1 accumulates in the corticospinal tracts over the patient's lifetime and reaches levels that are pathogenic in adulthood. This, we hypothesize, is due to the proteasome system being gradually unable to dispose of mutant M1. This "gain of function" scenario is not unlike the mechanisms that give rise to other neurodegenerative diseases, but is completely different from the prevailing haploinsufficiency model.

We have prepared truncated mouse constructs specifically to inhibit microtubule-severing, rather than to faithfully reproduce mutations observed with human disease. Although the mouse sequence is quite similar to human, we feel that the truest test would be to use the precise human sequence for our constructs and to introduce the precise mutations known to cause the disease in human patients. Toward this end, we have been working to prepare the necessary human constructs to carry out the proposed studies (see table 1). Among the mutations associated with pure adult HSP, we chose two different missense mutations that lie outside of the AAA-domain and would not be expected to destroy the microtubule-severing properties of the expressed spastin protein. We have also prepared spastin constructs with mutations that are identical to the mutations in HSP patients that either inactivate the AAA domain or result in synthesis of a truncated spastin protein that completely lacks the AAA-domain. To express M1 and M87 isoforms separately for microtubule-severing and

cytotoxicity studies, we have cloned wild type (WT) or mutated M1 cDNA as well as WT or mutated M87 cDNA into mammalian expression vector pCMV-Tag. The vector provides a perfect Kozak's sequence and allows high levels of transcription and translation of myc-tagged spastins. To study degradation/accumulation ratios of WT spastin isoforms as well as mutated spastin isoforms, we prepared constructs in which the 5'untranslated region (5'UTR) was included and translation of both isoforms is controlled by endogenous M1 or M87 Kozak's sequences. These constructs simultaneously express moderate levels of untagged M1 and M87. For the proposed axonal transport studies, we will use bicistronic vector with IRES (internal ribosome entry site) to express mCherry and untagged spastins from the same promoter. Constructs like this allow fluorescence imaging without directly tagging spastins to anything larger than myc such as GFP or mCherry, which we believe runs the risk of altering the relevant properties of the expressed protein. In fixed samples, we can detect expressed spastins with two new highly effective rabbit anti-human spastin antibodies that we have developed in our laboratory, or with myc antibodies. One of these is directed against amino acids 339-467 located within AAA domain and second is directed against amino acids 90-335.

In one set of experiments, we are testing the microtubule-severing properties of the various constructs that we have generated. Our hypothesis is that the mutations outside of the AAA-domain should not adversely affect the severing properties of the protein whereas the mutations within the AAA-domain should. Of course, the truncated constructs with no AAA-domain should have no severing properties whatsoever. These studies are being conducted precisely as in our published work on the mouse spastin constructs. The constructs are being introduced into RFL-6 fibroblasts using the Amaxa nucleofector, and then the cells are being fixed and immunostained 28 hours after plating for microtubules and either spastin or myc. With fibroblasts, we generally stain for spastin (rather than myc) because there is virtually no endogenous spastin in these cells (whereas for neurons, we stain for myc; see later). For quantitative analysis, we select cells that are expressing spastin within a defined range as assessed by fluorescence intensity, and then quantify the levels of microtubules relative to control cells expressing myc alone. An additional benefit to these studies is that the fibroblasts are very flat, and therefore allow for a great deal of resolution so that the degree to which various expressed spastins tend to aggregate is quite obvious and potentially quantifiable. Using this procedure in our published study on mouse spastins, we showed that M1 has a stronger tendency to aggregate than M85, even when the two proteins do not bear any mutations. M1 also is considerably less effective at severing microtubules compared to M85. We understand that the fibroblasts are different from neurons with regard to microtubule-associated proteins as well as factors that may influence protein processing and degradation, so we are also performing these studies in cultured primary rat cortical neurons. If we are correct that the pathogenic mutations outside of the AAA-domain do not adversely affect microtubule-severing, this will confirm one of the strongest lines of reasoning against the haploinsuffiency model. Indeed, preliminary studies with our new human constructs indicate that the mutant M87 E112K has severing activity undistinguishable from that of WT M87 when overexpressed in RFL-6 cells.

Table 1. List of human spastin mutants associated with HSP-SPG4 and prepared in our laboratory

1. Mutation Ser44Leu (S44L) is located in the N-terminal region of M1 isoform in exon one. This mutation when present homozygously is associated with one case of pure HSP with mild symptoms first noticed at the age of 60 years. When present in addition to some other mutations in a second spastin allele this mutation is associated with early childhood onset of HSP. In our studies, we will compare degradation/accumulation ratio of M1S44L mutant spastin and other mutated M1 isoforms to determine if the differences in mutants stability might explain the different outcomes observed in patients. It is also possible that this mutation localized outside M87 coding sequence might affect M87 expression by changing the sequence of a cryptic M87 promoter. To test this possibility, we will compare the ratio of M87 to M1 expression using WT M1/M87 and M1/M87S44L constructs. **2.** Mutation 46STOP in exon 1 is associated with one case of pure HSP. This mutation is located outside of M87 coding region and should only truncate M1. It is however possible that mutated region contains a cryptic M87 promoter and therefore the mutation might affect M87 expression. As in case of S44L mutation we will test this possibility by comparing the ratio of M87 to M1 expression using WT M1/M87 and M1/M8746STOP constructs. We will also test the possibility that a short 46 amino acid N-terminal fragment of M1 is expressed and it is toxic to neuronal cells. **3.** Mutation Glu112Lys (E112K) in exon one affects a moderately conserved residue just upstream from MIT domain. This mutation is associated with one case of pure HSP that started at the age of 41 and had a severity score 3 on a 4-point scale. **4.** Mutation Leu195Val (L195V) in exon three affects a conserved residue in MIT domain. This mutation is associated with two cases of pure HSP with the onset of symptoms at the age of 37 and 55 years. Severity score was 2 and 3 on a 5-point scale where 1 indicates normal or very slight stiffness in the leg and 5, wheel chair bound. **5.** Mutation 245STOP resulting in an expression of truncated spastin without AAA domain is associated with two cases HSP with an age of onset of 39 and 44 years and a severity score 1 and 3 on 4-point scale. **6.** Mutation F381L located in AAA domain is associated with three cases of HSP with an age of onset of 32, 35 and 45 years and a severity score 1 and 2 on 4-point scale. **7.** Mutation D444Q located in AAA domain is associated with at least one case of HSP with an age of onset of 2 years and severity score 4 on 4-point scale. **8**. We have also mutated the first ATG sequence in the upstream open reading frame (uORF) located -28 nucleotides from M1 initiation codon, to test if this mutation affects the expression levels of M1 isoform. It has been reported that uORFs might restrict expression of toxic proteins.

The published paper also includes studies on the expression levels of the WT and mutated spastin isoforms, both with and without proteasome inhibitors. The studies suggest that the M1 mutants have more of a tendency to form aggregates and that in some cases this can create a partial dominant/negative situation wherein the severing properties of M85 are partially compromised. Thus while haploinsufifiency does not make sense as a mechanism for the nerve degeneration, these studies suggest that there might be a “loss of function”

component, at least in the case of some of the mutants, resulting from a partial dominant/negative function of the mutants.

Over the past few months, we have launched studies on rat cortical neurons. These are being done in two ways. First, we are depleting endogenous rat spastin with siRNA that targets rat but not human spastin, and then expressing the human spastin constructs. This will test the severing properties of the human spastin proteins apart from any potential dominant-negative effects of the dysfunctional mutants. Then, we are repeating the studies on neurons without depleting the endogenous spastin to ascertain whether the constructs result in diminished severing relative to controls. Because the rat neurons are not flat like the fibroblasts, the length of individual microtubules are not as discernable but ascertaining microtubule mass is sufficient as an indirect indication of the degree of microtubule-severing. In addition, the branching of the axons is directly correlated with spastin activity, and hence this is being used as an indication of the enhancement of severing by functional mutants or potential inhibition of severing by dysfunctional mutants. Once having completed this work, we will begin the proposed axonal transport studies in neurons expressing the various spastin mutants.

An additional paper dealing with the role of another microtubule-severing protein, namely katanin, in tauopathic degenerative diseases in now in press in Human Molecular Genetics (H. Sudo and P.W. Baas, 2010, Human Molecular Genetics, in press). This paper was partially funded by the CURE funds, given the mechanistic connection between severing proteins and disease.

Also partially funded by the CURE funds were a paper on the basic biology of the severing proteins (L. Qiang, W. Yu, M. Liu, J.M. Solowska, and P.W. Baas, 2010, Mol. Biol Cell 21: 334-344), and a book chapter dealing with the subject matter of the CURE grant (A. Falnikar and P.W. Baas, 2009, Results Probl Cell Differ 48: 47-64).

In terms of securing external funding for this work to continue, I participated with Dr. Gerardo Morfini of University of Illinois-Chicago on an R01 application dealing with the mechanisms of HSP. Dr. Morfini is P.I., and I have a sub-contract of $100,000 direct costs per year for five years. The grant was successful, and funding has begun.

18. Extent of Clinical Activities Initiated and Completed. Items 18(A) and 18(B) should be completed for all research projects. If the project was restricted to secondary analysis of clinical data or data analysis of clinical research, then responses to 18(A) and 18(B) should be "No."

18(A) Did you initiate a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

_____Yes
___x__No

18(B) Did you complete a study that involved the testing of treatment, prevention or diagnostic procedures on human subjects?

_____Yes
___x__No

If "Yes" to either 18(A) or 18(B), items 18(C) – (F) must also be completed. (Do NOT complete 18(C-F) if 18(A) and 18(B) are both "No.")

18(C) How many hospital and health care professionals were involved in the research project?

_____Number of hospital and health care professionals involved in the research project

18(D) How many subjects were included in the study compared to targeted goals?

_____Number of subjects originally targeted to be included in the study
_____Number of subjects enrolled in the study

18(E) How many subjects were enrolled in the study by gender, ethnicity and race?

Gender:
_____Males
_____Females
_____Unknown

Ethnicity:
_____Latinos or Hispanics
_____Not Latinos or Hispanics
_____Unknown

Race:
_____American Indian or Alaska Native
_____Asian
_____Blacks or African American
_____Native Hawaiian or Other Pacific Islander
_____White
_____Other, specify:_____
_____Unknown

18(F) Where was the research study conducted? (List the county where the research study was conducted. If the treatment, prevention and diagnostic tests were offered in more than one county, list all of the counties where the research study was conducted.)

19. Human Embryonic Stem Cell Research. Item 19(A) should be completed for all research projects. If the research project involved human embryonic stem cells, items 19(B) and 19(C) must also be completed.

19(A) Did this project involve, in any capacity, human embryonic stem cells?
_____Yes
___x__ No

19(B) Were these stem cell lines NIH-approved lines that were derived outside of Pennsylvania?
_____Yes
_____ No

19(C) Please describe how this project involved human embryonic stem cells:

20. Articles Submitted to Peer-Reviewed Publications.

20(A) Identify all publications that resulted from the research performed during the funding period and that have been submitted to peer-reviewed publications. Do not list journal abstracts or presentations at professional meetings; abstract and meeting presentations should be listed at the end of item 17. **Include only those publications that acknowledge the Pennsylvania Department of Health as a funding source** (as required in the grant agreement). List the title of the journal article, the authors, the name of the peer-reviewed publication, the month and year when it was submitted, and the status of publication (submitted for publication, accepted for publication or published.). Submit an electronic copy of each publication, listed in the table, in a PDF version 5.0.5 format, 1,200 dpi. Filenames for each publication should include the number of the research project, the last name of the PI, the number of the publication and an abbreviated research project title. For example, if you submit two publications for PI Smith for the "Cognition and MRI in Older Adults" research project (Project 1), and two publications for PI Zhang for the "Lung Cancer" research project (Project 3), the filenames should be:

Project 1 – Smith – Publication 1 – Cognition and MRI
Project 1 – Smith – Publication 2 – Cognition and MRI
Project 3 – Zhang – Publication 1 – Lung Cancer
Project 3 – Zhang – Publication 2 – Lung Cancer

If the publication is not available electronically, provide 5 paper copies of the publication.

Note: The grant agreement requires that recipients acknowledge the Pennsylvania Department of Health funding in all publications. Please ensure that all publications listed acknowledge the Department of Health funding. If a publication does not acknowledge the funding from the Commonwealth, do not list the publication.

Title of Journal Article:	Authors:	Name of Peer-reviewed Publication:	Month and Year Submitted:	Publication Status (check appropriate box below):
1. Evaluation of the haploinsufficiency model for SPG4-based hereditary spastic paraplegia	Joanna M. Solowska, James D. Garbern, and Peter W. Baas	Human Molecular Genetics	December 2009	☐Submitted ☐Accepted ☒Published
2. Strategies for diminishing katanin-based loss of microtubules in tauopathic neurodegenerative diseases.	Haruka Sudo and Peter W. Baas	Human Molecular Genetics	October 2010	☐Submitted ☐Submitted ☒Published
3. Basic fibroblast growth factor elicits formation of interstitial axonal branches via enhanced severing of microtubules	Liang Qiang, Wenqian Yu, Mei Liu, Joanna M. Solowska, and Peter W. Baas	Molecular Biology of the Cell	September 2009	☐Submitted ☐Accepted ☒Published
4. Critical Roles for Microtubules in Axonal Development and Disease	Aditi Falnikar and Peter W. Baas	Cell Biology of the Axon Results and Problems in Cell Differentiation	2009	☐Submitted ☐Accepted ☒Published

20(B) Based on this project, are you planning to submit articles to peer-reviewed publications in the future?

Yes___x_____ No_____

If yes, please describe your plans:

The work is ongoing. We are currently conducting studies to ascertain the stability of the M1 and M87 isoforms of spastin, both wild-type and mutant.

21. Changes in Outcome, Impact and Effectiveness Attributable to the Research Project. Describe the outcome, impact, and effectiveness of the research project by summarizing its impact on the incidence of disease, death from disease, stage of disease at time of diagnosis, or other relevant measures of outcome, impact or effectiveness of the research project. If there were no changes, insert "None"; do not use "Not applicable." Responses must be

single-spaced below, and no smaller than 12-point type. DO NOT DELETE THESE INSTRUCTIONS. There is no limit to the length of your response.

Given that the rest of the community is focused on haploinsuffiency as the cause of hereditary spastic paraplegia, our studies focusing on toxicity of the mutants are the only studies anywhere in the world that have a chance of leading to effective therapies (assuming our model is correct, which mounting evidence suggests that it is).

22. Major Discoveries, New Drugs, and New Approaches for Prevention Diagnosis and Treatment. Describe major discoveries, new drugs, and new approaches for prevention, diagnosis and treatment that are attributable to the completed research project. If there were no major discoveries, drugs or approaches, insert "None"; do not use "Not applicable." Responses must be single-spaced below, and no smaller than 12-point type. DO NOT DELETE THESE INSTRUCTIONS. There is no limit to the length of your response.

Given that the rest of the community is focused on haploinsuffiency as the cause of hereditary spastic paraplegia, our studies focusing on toxicity of the mutants are the only studies anywhere in the world that have a chance of leading to effective therapies (assuming our model is correct, which mounting evidence suggests that it is). Once we have made some additional strides along these lines, we will begin testing potential therapies. Our published paper dealing with strategies for ameliorating the impact of tauopathies may suggest some avenues that could be pursued with regard to HSP as well.

23. Inventions, Patents and Commercial Development Opportunities.

23(A) Were any inventions, which may be patentable or otherwise protectable under Title 35 of the United States Code, conceived or first actually reduced to practice in the performance of work under this health research grant? Yes_____ No___x_____

If "Yes" to 23(A), complete items a – g below for each invention. (Do NOT complete items a - g if 23(A) is "No.")

a. Title of Invention:

b. Name of Inventor(s):

c. Technical Description of Invention (describe nature, purpose, operation and physical, chemical, biological or electrical characteristics of the invention):

d. Was a patent filed for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?
Yes_____ No___

If yes, indicate date patent was filed:

e. Was a patent issued for the invention conceived or first actually reduced to practice in the performance of work under this health research grant?
Yes_____ No____
If yes, indicate number of patent, title and date issued:
Patent number:
Title of patent:
Date issued:

f. Were any licenses granted for the patent obtained as a result of work performed under this health research grant? Yes_____ No____

If yes, how many licenses were granted?_____

g. Were any commercial development activities taken to develop the invention into a commercial product or service for manufacture or sale? Yes___ No____

If yes, describe the commercial development activities:

23(B) Based on the results of this project, are you planning to file for any licenses or patents, or undertake any commercial development opportunities in the future?

Yes_____ No_____x____

If yes, please describe your plans:

24. Key Investigator Qualifications. Briefly describe the education, research interests and experience and professional commitments of the Principal Investigator and all other key investigators. In place of narrative you may insert the NIH biosketch form here; however, please limit each biosketch to 1-2 pages. *For Nonformula grants only – include information for only those key investigators whose biosketches were not included in the original grant application.*

BIOGRAPHICAL SKETCH

NAME Baas, Peter W.	POSITION TITLE Professor of Neurobiology and Anatomy
eRA COMMONS USER NAME PBAAS1	

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, and*

INSTITUTION AND LOCATION	DEGREE *(if applicable)*	YEAR(s)	FIELD OF STUDY
Michigan State University, East Lansing, MI	B.S.	1979-1981	Zoology
Michigan State University, East Lansing, MI	M.S.	1981-1983	Zoology
Michigan State University, East Lansing, MI	Ph.D.	1983-1987	Physiology
Temple University, Philadelphia, PA	Postdoc	1987-1990	Cellular Neuroscience

Positions and honors

1991-1996	Assistant Professor of Anatomy, The University of Wisconsin
1992-1997	Recipient of Research Career Development Award from NIH 1997 Recipient of the Jordi Folch Pi Memorial Award for outstanding young neuroscientist from the American Society for Neurochemistry
1997-2000	Associate Professor of Anatomy, The University of Wisconsin
2000-present	Professor of Neurobiology and Anatomy, Drexel University College of Medicine
2001, 2003	Graduate Student Association Award for Teaching, Drexel University College of Medicine
2002	Dean's Award for Outstanding Research Contributions, Drexel University College of Medicine
2001-present	Reviewer for Study Sections of NIH, including regular participation in a Neuroscience-related Study Section that reviews Predoctoral and Postdoctoral NRSA applications as well as one that reviews MD/PhD NRSA applications
2003- Present	Director, Graduate Program in Neuroscience, Drexel University College of Medicine

Selected Publications:

Qiang, L., W. Yu, A. Andreadis, M. Luo, and P.W. Baas. 2006. Tau protects microtubules in the axon from severing by katanin. J. Neurosci. 26: 3120-3129.

King, M.E., H.-M. Kan, P.W. Baas, A. Erisir, C.G. Glabe, and G.S. Bloom. 2006. Tau-dependent microtubule disassembly initiated by pre-fibrillar β-amyloid. J. Cell Biol. 175: 541-546.

King, M.E., H.-M. Kan, P.W. Baas, A. Erisir, C.G. Glabe, and G.S. Bloom. 2006. Tau-dependent microtubule disassembly initiated by pre-fibrillar β-amyloid. J. Cell Biol. 175: 541-546.

Takahashi, D., W. Yu, P.W. Baas, R. Kawai-Hirai, and K. Hayashi. 2007. Rearrangement of microtubule polarity orientation during conversion of dendrites to axons in cultured pyramidal neurons. Cell Motil. Cytoskel. 64: 347-359.

Katsetos, C.D., E. Draberova, B. Smejkalova, G. Reddy, L. Bertrand, J.P. de Chadarevian, A. Legido, J. Nissanov, P.W. Baas, and P. Draber. 2007. Class III beta-tubulin and gamma-tubulin are co-expressed and form complexes in human glioblastoma cells. Neurochem. Res. 32: 1387-1398.

Myers, K.A., and P.W. Baas. 2007. Kinesin-5 regulates the growth of the axon by acting as a break on its microtubule array. J. Cell Biol. 178: 1081-1091.

Yu, W., Qiang, L., and P.W. Baas. 2007. Microtubule-severing in the axon: implications for development, disease, and regeneration after injury. J. Environ. Biomed. 1: 1-7.

Solowska, J.M, G. Morfini, A. Falnikar., B.T. Himes, S.T. Brady, D. Huang, and P.W. Baas. 2008. Quantitative and functional analyses of spastin in the nervous system: implications for hereditary spastic paraplegia. Journal of Neuroscience 28: 2147-2157.

Yu, W., L. Qiang, J.M. Solowska, A. Karabay, S. Korulu, and P.W. Baas. 2008. Spastin and katanin participate differently in the formation of axonal branches. Molecular Biology of the Cell 19: 2147-2157.

Nadar, V.C., A. Ketschek, K.A. Myers, G. Gallo, and P.W. Baas. 2008. Kinesin-5 is essential for growth cone turning. 2008. Current Biology 18: 1972-1977.

Tint, I., D. Jean, P.W. Baas, and M.M. Black. 2009. Doublecortin associates with microtubules preferentially in regions of the axon displaying actin-rich protrusive structures. J. Neurosci. 29: 10995-11010.

Qiang, L., W. Yu, M. Liu, J.M. Solowska, and P.W. Baas. 2010. Basic fibroblast growth factor elicits formation of interstitial axonal branches via enhanced severing of microtubules. Molecular Biology of the Cell 21: 334-344.

Tang-Schomer, M.D., A.R. Patel, P.W. Baas, and D.H. Smith. 2010. Mechanical breaking of microtubules in axons during dynamic stretch injury underlies delayed elasticity, microtubule disassembly, and axon degeneration. FASEB J. 24: 1401-1410.

Sudo, H., and P.W. Baas. 2010. Acetylation of microtubules influences their sensitivity to severing by katanin in fibroblasts and neurons. J. Neurosci. 30: 7215-7226.

Solowska, J.M., J.Y. Garbern, and P.W. Baas 2010. Evaluation of loss of function as an explanation for SPG4-based hereditary spastic paraplegia. Human Molecular Genetics 19: 2767-2779.

Liu, M., V.C. Nadar, F. Kozielski, M. Kozlowska, W. Yu, and P.W. Baas. 2010. Kinesin-12, a mitotic microtubule-associated motor protein, impacts axonal growth, navigation, and bundling. J. Neurosci. 30: 14896-14906.

Sudo, H., and P.W. Baas. 2010. Strategies for diminishing katanin-based loss of microtubules in tauopathic neurodegenerative diseases. Human Molecular Genetics, in press.

Research Funding

ACTIVE

NIH R01 NS28785 (Baas- P.I.) 09/01/07-08/31/12
NIH
Title: Microtubule dynamics and axon growth
Description: This grant proposes to determine the roles of various molecular motor proteins in the axonal transport of short microtubules as well as the participation of long microtubules in growth cone turning.

NSF (Baas-P.I.) 04/01/09-03/30/13
NSF
Title: Axonal microtubules cut and run. This grant involves experiments on the basic biology of microtubule-severing proteins in the nervous system.

NIH R01 NS066942 (Morfini-P.I.) 03/01/10/02/28/15
Title: Axonal transport deficits during hereditary spastic paraplegia R01 to Dr. Gerardo Morfini, from the University of Illinois at Chicago. The grant deals with the mechanisms underlying neurodegeneration resulting from spastin mutations. Dr. Baas has a sub-contract on the grant.