Proposal Strategy/Structure II
Significance

Page Limits

Section of Application	Section of Application
Introduction to Revision or Resubmission Applications	1 page
Introduction to Revision or Resubmission Applications For each project and core of multi-component applications	1 page
Specific Aims	1 page
Research Strategy (Item 5.5.3 of Research Plan) For Activity Codes R03, R13/U13, R21, R36, R41, R43, Fellowships (F), SC2, SC3	6 pages
Research Strategy (Item 5.5.3 of Research Plan) For Activity Codes R01, single project U01, R10, R15, R18, U18, R21/R33, R24, R33, R34, U34, R42, R44, DP3, G08, G11, UH2, UH3, SC1, X01	12 pages
Research Strategy (Item 5.5.3 of Research Plan) For all other Activity Codes, including Cs, Ps, Ss, Ts, Us, etc.	follow FOA instructions *
Biosketch (per person) For all Activity Codes except DP1 and DP2	4 pages
Biosketch (per person) For DP1 and DP2	2 pages
Appendix **	No page limits, but content limitations. See relevant section of instructions and FOA

Templates

- **Mechanics of document preparation (L02-mechanics.pdf) and the LaTeX proposal template (proposal-latex-template.zip)**

Fillable Individual PHS 398 Forms

*(These forms are to be used only with paper submissions using the PHS 398. Do **not** use the PDF samples provided below in an SF424 (R&R) application. These are fillable PDF forms which will cause an error in the electronic submission of an SF424 (R&R) application. See the SF424 (R&R) application page for appropriate formats to be used for electronic submission.)*

Form Page 1: Face Page	MS Word (88 KB)	PDF (310 KB)
Form Page 1-continued: Additional form for use ***only*** if Multiple PD/PIs are proposed. Do ***not*** include if submitting a single-PD/PI application.	MS Word (76 KB)	PDF (254 KB)
Form Page 2: Summary, Relevance, Project/Performance Sites, Senior/Key Personnel, Other Significant Contributors, and Human Embryonic Stem Cells	MS Word (117 KB)	PDF (369 KB)
Project/Performance Site Format Page - use only if additional space is needed.	MS Word (92 KB)	PDF (269 KB)
Form Page 3: Research Grant Table of Contents	MS Word (79 KB)	PDF (701 KB)
Form Page 4: Detailed Budget for Initial Budget Period	MS Word (89 KB)	PDF (309 KB)
Form Page 5: Budget for Entire Proposed Project Period	MS Word (86 KB)	PDF (573 KB)
Biographical Sketch Format Page	MS Word (38 KB)	PDF (599 KB)
Biographical Sketch Sample	MS Word (72 KB)	PDF (63 KB)
Resources Format Page	MS Word (39 KB)	PDF (240 KB)
Checklist Form Page	MS Word (84 KB)	PDF (506 KB)
Continuation Format Page	MS Word (36 KB)	PDF (202 KB)

```
% -*-latex-*-
% Document name: proposal.tex, a template for the PhD Proposal
%
% Authors: Rob MacLeod
%
% Last update: Mon Jan 30 07:10:25 2012 by Rob Macleod
%    - created
%
%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
\documentclass[11pt]{report}
\usepackage{nih-proposal}

%%%%%%%%%%%%%%%%%%%%%%%%% New commands

% Change the footnotes from numbers to a series of symbols
%\renewcommand{\thefootnote}{\fnsymbol{footnote}}
\newcommand{\etal}{{\em et al.}}
\newcommand{\etc}{{\em etc.}}
\newcommand{\eg}{{\em e.g.,}}
\newcommand{\ie}{{\em i.e.,}}
\newcommand{\cpp}{C{\raisebox{0.5ex}{\tiny++}}}
\newcommand{\degrees}{{$^{\circ}$}}
\newcommand{\splitline}{\begin{center}\rule{\columnwidth}{.7mm}\end{center}}
\newcommand{\muv}{${\rm \mu V}$}
\newcommand{\ohm}{$\Omega$}
\newcommand{\sft}{${\rm ft^2}$}

%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%%
\begin{document}

%%%%%%%%%%%%%%%%%%%%%%%% The text
\setcounter{secnumdepth}{3}


%%%%%%%%%%%%%%%%%%%%%%%%%% 1) Title
\thispagestyle{empty}
\input{title}
\newpage

%%%%%%%%%%%%%%%%%%%%%%%%%% 2) Table of Contents
\tableofcontents
\newpage
```

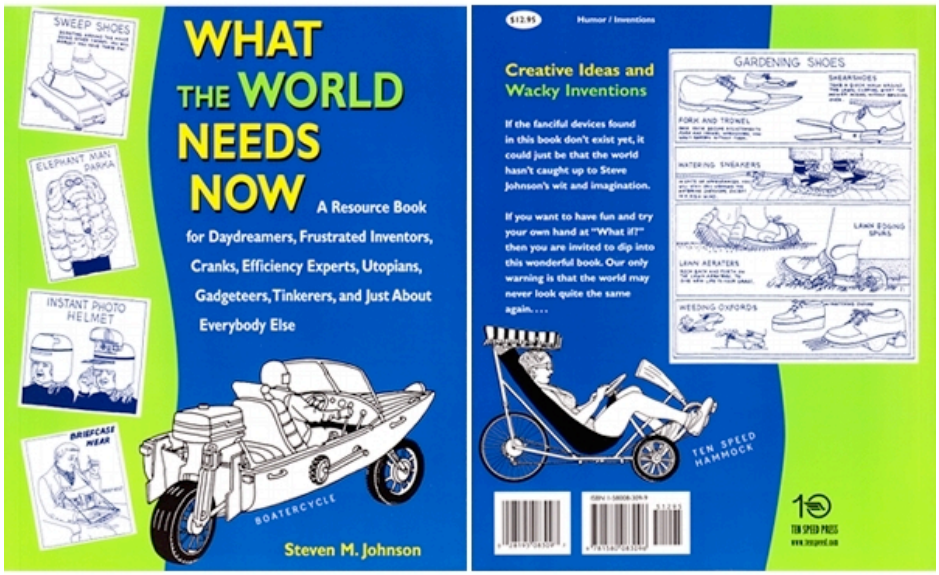

Who is the Audience?

Don't assume too much!!

NIH Research Strategy Structure

- Significance
- Innovation
- Approach
- Investigator(s)
- Environment
- Impact

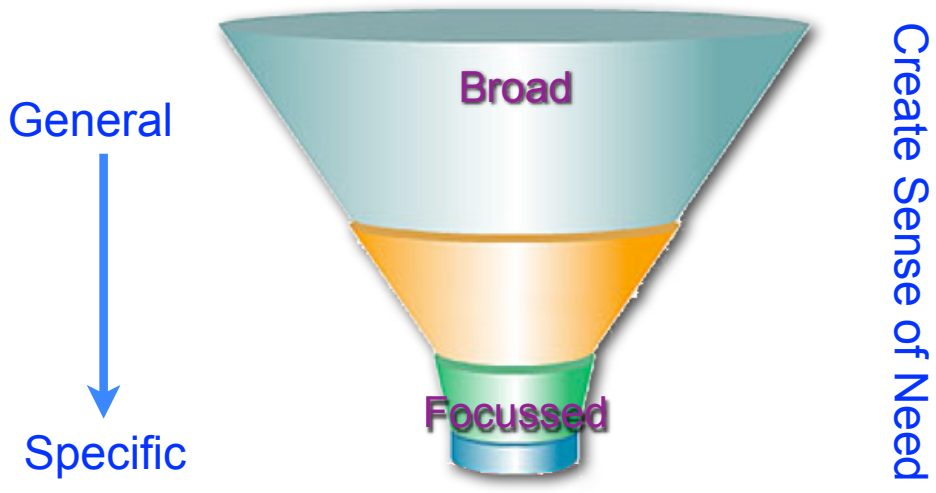
Significance



Significance

- Explain the importance of the problem or critical barrier to progress in the field that the proposed project addresses.
- Explain how the proposed project will improve scientific knowledge, technical capability, and/or clinical practice in one or more broad fields.
- Describe how the concepts, methods, technologies, treatments, services, or preventative interventions that drive this field will be changed if the proposed aims are achieved.

Background Funnel



Background Tips

- Describe what is known, but only what the reader needs to know to appreciate your proposal.
- Create need and also rationale for your research.
- Be objective, do not criticize others.
- Develop clear structure, with subsections.
- Use literature extensively.
- Background funneling to unknown.
- Be direct and use signaling words: “unknown”, “problem”, “impediment”, “challenge”.
- Link the need to the proposed research

Background funneling to unknown

- “Global warming is arguably one of the most pressing concerns of our time. It has been linked to the rapidity of observed climate change--the fact the the Earth’s temperature rose by approximately 0.7C over the last century (the most dramatic increase documented in historic times)[1-3] and the attendant threat posed by melting polar icecaps, rising seal levels, and potentially, more severe weather patterns. “
- “We do not know yet what proportion of this global warming is due to human activity and what is due to natural variations. More important, we lack an effective model to predict precisely by how much the temperature will rise as a consequence of the increase of levels of CO2 and other greenhouse gases in the atmosphere of the earth.”

Link Problem to the Research

"Because the consequences of global warming for life on earth greatly depend on where the actual warming lies within the predicted range, it is critical to narrow the range by improving our understanding of the uncertain component of the climate models with utmost urgency. We will investigate these key sources of uncertainty in this proposal by…."

Another Example

"These treatments are only moderately effective and are often accompanied by severe side effects and viral resistance. Thus, there remains a need for new therapies for this serious disease. We propose to use a novel class of experiments to test…."

Possible Elements of Significance

- Claim timeliness
- Propose practical solution to a problem
- Identify a large population affected
- Address a Gap in knowledge
- Outline implications over a wide range of practical problems
- Seek improvement to instrumentation
- Open new research directions
- Improve quality of life
- Bridge from theoretical to practical knowledge

Useful Signal Words/Phrases

- Background
  - "X is"
  - "X affects"
- Unknown/Need
  - "...is unknown"
  - "…is unclear"
  - "...has not been determined"
- Objectives/Aims
  - "We propose to…"
  - "Our objective is…"
  - "We will examine the hypothesis that…"
- Significance/impact
  - "...may result in..."
  - "...will contribute to..."
  - "...may be used to…"

Most Common Problems

Poor organization

Lack of objectivity

Amount of detail (too much or too little)

- Checklist
  - Are all components present?
  - Is writing adjusted to the audience?
  - Do components follow logically and is the flow obvious?
  - Is the writing objective, statements backed up by citations?

Image Analysis of C. Elegans Proposal Score: 10, Percentile: 2

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Associate Professor in Quantitative Microscopy,
Centre for Image Analysis, Uppsala University, Sweden

The C. Elegans Imaging Example: The Background and Need

- "The NIH is committed to translating basic biomedical research into clinical practice and thereby impacting global human health[1], and Francis Collins identifies high-throughput technology as one of five areas of focus for the NIH's research agenda[2]. For many diseases, researchers have identified successful novel therapeutics or research probes by applying technical advances in automation to high-throughput screening (HTS) using either biochemical or cell-based assays[3–6]. Researchers are using genetic perturbations such as RNA interference or gene overexpression in cell-based HTS assays to identify genetic regulators of disease processes as potential drug targets[7–9]. However, the molecular mechanisms of many diseases that deeply impact human health worldwide are not well-understood and thus cannot yet be reduced to biochemical or cell-based assays."

The C. Elegans Imaging Example: Tie to PI's Recent Research

- "Ideally, researchers could approach disease from a phenotypic direction, in addition to the traditional molecular approach, by searching for chemical or genetic regulators of disease processes in whole model organisms rather than isolated cells or proteins. Moving HTS towards more intact, physiological systems also improves the likelihood that the findings from such experiments accurately translate into the context of the human body (e.g., in terms of toxicity and bioavailability), simplifying the path to clinical trials and reducing the failure of potential therapeutics at later stages of testing. In fact, for some diseases, a whole organism screen may actually be necessary to break new therapeutic ground; in the search for novel therapeutics for infectious agents, for example, it is widely speculated that the traditional approach of screening for chemicals that directly kill bacteria in vitro has been largely exhausted[10]. Our work recently identified six novel classes of chemicals that cure model organisms from infection by the important human pathogen E. faecalis through mechanisms distinct from directly killing the bacterium itself[11]. Anti-infectives with new mechanisms of action are urgently needed to combat widespread antibiotic resistance in pathogens."

The C. Elegans Imaging Example: Justify Basic Methodology

- "Enabling HTS in whole organisms is therefore recognized as a high priority (NIH PAR-08-024)[12,13]. *C. elegans* is a natural choice. Manually-analyzed RNAi and chemical screens are well-proven in this organism, with dozens completed[14–16]. Many existing assays can be adapted to HTS; instrumentation exists to handle and culture *C. elegans* in HTS-compatible multi-well. Its organ systems have high physiologic similarity and genetic conservation with humans[17,18]. *C. elegans* is particularly suited to assays involving visual phenotypes: physiologic abnormalities and fluorescent markers are easily observed because the worm is mostly transparent. The worms follow a stereotypic development pattern that yields identically-appearing adults[19,20], such that deviations from wild-type are more readily apparent."

C. Elegans Example: Establish the Problem, Point to a Solution

- "The bottleneck that remains for tackling important human health problems using C. elegans HTS is image analysis (NIH PA-07-320)[21,22]. It has been recently stated, "Currently, one of the biggest technical limitations for large-scale RNAi-based screens in *C. elegans* is the lack of efficient high-throughput methods to quantitate lethality, growth rates, and other morphological phenotypes"[23]. **Our proposal to develop image analysis algorithms to identify regulators of infection and metabolism in high-throughput C. elegans assays would bring image-based HTS to whole organisms, and have the following impact.**"

C. Elegans Example: Impact linked to Aims

- "**Identifying novel modulators of infection by the NIH priority pathogen Microsporidia** (Aim 1). Microsporidia are emerging human pathogens whose infection mechanisms are almost completely unknown. Further, they inflict agricultural damage and are on the EPA list of waterborne microbial contaminants of concern[24,25]. Identifying anti-microsporidian therapeutics is a special challenge because they are eukaryotes. Moreover, they are obligate intracellular pathogens so they are not amenable to traditional antibiotic screens; screening for drugs to kill them requires the presence of a validated, infectible host whose immune system is homologous to mammals, such as *C. elegans* [26,27]. This screen could identify not only useful chemical research probes and compounds that kill these pathogens outright, but also those that block microbial virulence, are modified by the host for full efficacy (prodrugs), or enhance host immunity."

C. Elegans Example: Impact linked to Aims

- "**Identifying novel regulators of fat metabolism** (Aim 2). Disregulation of metabolism results in many common and expensive chronic health conditions; diabetes alone affects 24 million Americans[28]. Energy centers must receive and integrate nutritional information from multiple peripheral signals across multiple tissues and cell types to elicit appropriate behavioral and metabolic responses; screening in a whole organism is important. In particular, screening with a strain of *C. elegans* with an RNAi-sensitive nervous system will likely reveal novel energy regulators of therapeutic and research value."

Link back to human health.

C. Elegans Example: Impact linked to Aims

- "**Identifying novel regulators of infection by the pathogen Staphylococcus aureus** (Aim 3). *S. aureus* is life-threatening for immune-compromised patients. Recently, antibiotic-resistant MRSA strains have created an urgent need for therapeutics with a new mechanism of action[29]. We will identify genetic regulators of the *C. elegans* host's response to infection by *S. aureus*[30]. These will lead to potential drug targets useful for boosting humans' innate immunity."

States significance to human health

C. Elegans Example: Explicit Statement of Significance

- **Enabling the automated analysis of a wide variety of C. elegans screens.** Because C. elegans has proven to be an excellent model for many human organs and processes, the impact of algorithms for automated scoring for currently intractable *C. elegans* image-based screens on our understanding and treatment of a variety of human diseases will be substantial. Adding novel C. elegans algorithms to existing open-source software will create a flexible toolbox that can be applied to other types of assays (including alternative formats such as microfluidics chambers; see Yanik support letter) with minimal modification:
- Aim 1: **The algorithms developed** for Aim 1 will enable scoring viability and other body morphology assays probing a number of biological processes. Our collaborators plan several RNAi and chemical screens using live/dead assays to identify modulators of many other clinically relevant pathogens (see Ausubel and Mylonakis support letters).
- Aim 2: **The algorithms developed** for the fat metabolism assay can also be used to quantify the levels of any stain within worms, to measure protein expression levels, the degree of staining by fluorescent dyes or antibodies, and promoter activity in reporter assays probing a wide range of biological processes.
- Aim 3: Where localization patterns are of interest, **the algorithms developed** for the gene expression pattern assay will often be directly applicable, especially given the proposed machine learning capabilities.
- **Many benefits come** from the automation of image analysis for such screens: (a) increased throughput so as to enable genome-scale RNAi and large-scale chemical screens in whole animals; (b) quantitative results amenable to data mining31–33; (c) increased objectivity and consistency; and (d) increased sensitivity to subtle phenotypes, which often can not be scored reliably by eye. The requisite automation of sample preparation and image acquisition has the welcome side effect of improving consistency and providing a permanent record of the experiment.

C. Elegans Example: Explicit Statement of Significance

- **"Creating open-source software for the C. elegans community.** *C. elegans* is used for studying complex multicellular biological processes by more than 11,000 researchers in 750 laboratories worldwide (http://www.WormBase.org, January 2010), and the close-knit community rapidly shares methods[17,18,34,35]. Based on our experience developing the CellProfiler software system (see Preliminary studies), packaging automated image analysis algorithms in user-friendly software encourages their use by the broader research community. Although we developed CellProfiler solely for high-throughput screening, 70% of studies citing it actually used it to quantify low-throughput assays (fewer than 100 samples). In this proposal we focus on developing algorithms that are robust and efficient for large-scale experiments, but we anticipate they will become an everyday tool for many researchers in the *C. elegans* community, a good investment since many of these are funded by the NIH."

Identify need, solution, impact

The C. Elegans Example: Explicit Statement of Significance

- "Thus, in addition to the discovery of potential drugs and drug targets related to metabolism and infection, which could significantly impact the global burden of human disease, our aims will yield open-source software for automated, accurate, quantitative scoring for a wide range of *C. elegans* image-based assays that are currently intractable. The impact will be multiplied by *C. elegans* laboratories worldwide using the resulting software to study a wide variety of pathways relevant to basic biological research and human disease, in both low-throughput and high-throughput experiments"

Summary paragraph

The C. Elegans Example: Innovation

- “In response to the strong demand for C. elegans screening, we propose to build on our technological innovations in sample preparation and imaging and our computational innovations for cells and brains to now create a novel technology for C. elegans. Our proposed work to develop novel algorithms for identifying and characterizing worms in microscopy images will bridge the final gap, for the first time enabling widespread identification of genetic and chemical regulators of human biological processes and diseases via whole-organism screening.”
- In response to the strong demand for C. elegans screening, we propose to build on our technological innovations in sample preparation and imaging and our computational innovations for cells and brains to now create a novel technology for C. elegans. Our proposed work to develop novel algorithms for identifying and characterizing worms in microscopy images will bridge the final gap, for the first time enabling widespread identification of genetic and chemical regulators of human biological processes and diseases via whole-organism screening.

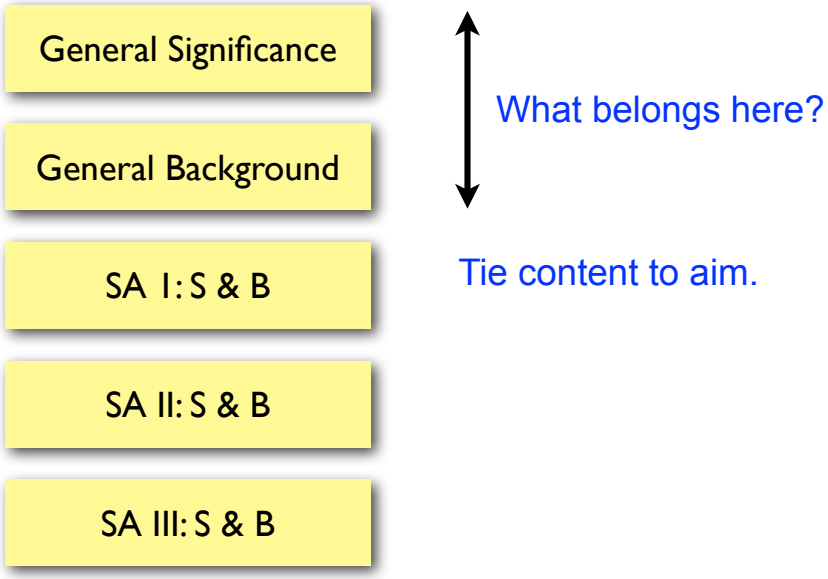
Describes Innovation and shows impact on the field.

The C. Elegans Example: Innovation

- “Several algorithmic innovations are necessary in order to quantify a variety of C. elegans phenotypes and attain the robustness required for routine high-throughput screening. We propose a novel, simplified represen- tation for worm shapes that lends itself to a probabilistic interpretation. This allows us to adapt shape models to identification of worms in a high-throughput context, and leads to a novel algorithm for detangling worms by morphology-guided graph search. We will also build upon methods from our work in deformation analysis54 and per-cell classification of cellular phenotypes by machine learning55 to quantify phenotypic variation and fluorescence localization in individual worms.”

Highlights novel approach

How to Structure With Aims

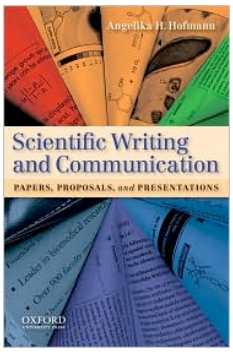


Assignment

Required: First complete draft of Specific Aims + Significance + Innovation

Optional: Outline for feedback

Information



Rob's Grant Information Page

A list of granting sources and links to grant applications. The choices reflect my biomedical bias and is in no way comprehensive.

Granting Agency Policy and Program Information

Other good grants sites

- University of Utah Health Science Resources for Basic Scientists including Research grant information
- College of Engineering grant information

NIH General Information

http://www.sci.utah.edu/~macleod/grants/

Grant Writing Tips

Some of these are specific to grants, others simply useful for any writing project.

- Rob's Writing page
- Rob's Latex Page.
- NIH Insider Guide. A set of tips from former NIH study section chairs.
- Proposal Writing: The Business of Science (pdf) by Wendy Sanders. Great advice for any grant writer.
- NIH Giudlines (also for review)
  - Grant writing tips from NIH
  - General reviewer guidlines
  - Reviewer guidance on the shortened applications
  - FAQ for review of short grants.
- RO1 Grant writing tips
- AHA Grant Writing Tips
- Proposal Writer's Guide by Don Thackrey
- The SCI text markup scheme