SOAR Research Proposal - Summer 2014

Project title — Creating a model for the neurological disease PCCA in *Drosophila melanogaster*
Faculty — Christopher Jones, Associate Professor of Biological Sciences
Student — Caitlin Hyland
Project duration — 10 weeks, Tuesday May 27 through Monday August 4

Description of the project —

Progressive cerebellocerebral atrophy (PCCA) is an inherited neurological disease first described a decade ago; symptoms include mental retardation, spasticity, seizures, and progressive microcephaly. In 2010, researchers in Israel showed that two distinct mutations in the gene *SepSecS* were responsible for this condition in 4 patients; they noted, however, that in other families with affected children there was no mutation in this gene, suggesting that at least one other gene remained to be identified. Two weeks ago a paper published in the *Journal of Medical Genetics* demonstrated that two mutations in the gene *VPS53* are causative in the remaining known cases.

The fruit fly D*rosophila melanogaster* is a powerful model organism for the genetic analysis of human diseases; of the thousands of genes that have been linked to diseases in man, approximately three-quarters have a homologous counterpart in the fly. Creating mutant flies carrying such a disease mutation enables researchers to study the molecular effects of the aberration and to test potential therapeutics quickly and efficiently. This project will use the CRISPR system (which I am currently piloting with students in BIOL 365 to confirm its utility and efficiency) to precisely target the Drosophila genome to create the equivalent of PCCA mutations in the homologous fly genes. The resulting mutants will be studied to determine the effects (if any) of these changes on morphology, longevity, fecundity, and behavior.

Roles and responsibilities —

Caitlin plans to conduct a "behavioral" project with me next year for her Honors research, and so wants to do something more "molecular" this summer. She will be responsible for background research (reviewing the basic molecular genetics she learned in BIOL210 this past fall, as well as learning about PCCA and the CRISPR mutagenesis system), determining the mutations needed, planning and carrying out the molecular experiments, and assessing possible phenotypes of the flies created.

My role will be to guide Caitlin's background research, coordinate the various aspects of the project (I will have a better idea than Caitlin how much time will be required for the different stages and so will need to plan for how to best optimize the limited time we'll have) and to serve as a voice of experience, having conducted many similarly "molecular" projects myself.

Weeks 1–3: Literature research and review; plan out precise mutations to be engineered into flies, order oligonucleotides.
Weeks 4–6: make molecular constructs for injection into eggs; send them to an outside facility to carry out the injections
Weeks 7–10: Confirm successful mutation by DNA sequencing; begin characterization of mutant phenotypes; begin to prepare for presentation at Scholars Day, the NCUR conference, and the Drosophila Research Conference (if appropriate).

Student engagement —
Caitlin is currently a junior who took Genetics with me last fall (2013); she was one of the best students in the class. Caitlin took the initiative of asking me to mentor her in a SOAR project, and I agreed after talking with her about her interests and goals. Caitlin is considering a career in life science research, and would like to gain some research experience to help her decide whether and how to pursue that goal. Her involvement in all phases of the project, from planning the mutations through analyzing and interpreting his results, will strengthen her command of the research process. As a result, Caitlin will be much better prepared to continue on to graduate school, should she decide that that's what she wants to do after Moravian.

In order to fully understand the impact of her work, Caitlin will need to learn a bit about a range of different areas in biology, including amino acid metabolism, neural function, and development; this sort of learning on a "need-to-know" basis is increasingly common in modern biology, and will stand her in good stead wherever she finds herself. Her several roles in this project: carrying out background research, designing the molecules to create the desired mutations, synthesizing those molecules, and analyzing the resulting mutant animals, are all fundamental aspects of research in molecular genetics.

Student contributions —
If the project is successful, I expect that it will lead to additional experiments, which will ultimately be published; as a newly described disease, there are currently no models available for the study of PCCA. Although it's very unlikely that the results of this SOAR project will, by themselves, be publishable, I anticipate that they will be more than sufficient to merit presentation at regional and national conferences. In years past my SOAR students have presented their work at the regional Beta Beta Beta convention (Tri-Beta is the undergraduate biology honor society), the National Council for Undergraduate Research conference, and at the national Drosophila Research Conference. Next spring the "Fly Meeting" will be held in Chicago, a relatively accessible city from Bethlehem, so I foresee no problem with Caitlin's participation there, if all goes well this summer.

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Caitlin Hyland, Biology, 2015

Dr. Christopher Jones

No On-Campus Housing

I am interested in participating in the SOAR program because I would like to gain as much research experience as possible. After I graduate from Moravian, I am planning on going to graduate school for research in biology-particularly genetics. Therefore, I want to have as much research experience as I possibly can.

In addition to SOAR, I am participating in the Honors Program next fall and spring semesters. My honors project has a behavioral genetics focus- I am going to be studying the effects of an Alzheimer's-related mutation on learning in fruit flies. Since I am doing a behavioral genetics project for honors, I decided that it would be best to concentrate on the other major focus in genetics, molecular genetics, for SOAR. Therefore, I am planning on using the SOAR program with the Honors program to gain the most experience possible in genetics-based research. My SOAR project will also provide a useful transition into my honors project, since they are both relatively similar projects.

In addition to research experience, I think SOAR will give me an opportunity to present my work. Since I want to do research, as a profession, presenting work will be a major component of my career. SOAR will allow me to perfect my presentation skills and allow me to be more comfortable presenting my work.

In conclusion, I am expecting to gain a variety of experiences by participating in SOAR. I am expecting to gain experience in research and learn variety of laboratory techniques that were not covered in class and/or to perfect laboratory techniques that were covered in class. I also think that SOAR will help me in my honors project. In addition, I think that SOAR will provide me with confidence in my presentation skills.

Application, Part 4: Expense Proposal

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Most of the molecular reagents necessary for this project will be available through acquisitions made for my course BIOL365 (Molecular Genetics) this spring. However, there will be two items which are unique to each project: custom oligonucleotides to create the desired mutation(s) in the fly genome, and injection of the resulting molecular constructs into appropriate *Drosophila* eggs.

Oligonucleotides are readily available from a variety of sources; the one I regularly use would charge $5 to $7 per oligo, depending on length and sequence. I estimate that we will need 10 to 15 of these for the project, or approximately $100.

To do injections in-house would be a significant amount of work, with no guarantee of success; like DNA sequencing, this is a specialized task that most labs hire an outside specialty firm to conduct. Injection will cost approximately $200.

Additional expenses (e.g. routinely-used laboratory supplies, fly food ingredients, additional reagents) will be covered by the Department of Biological Sciences.