FUN Newsletter

May, 2016

Newsletter Staff

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President's Corner

I hope this late spring newsletter finds you in the wonderful position of finishing up a successful semester and gearing up for whatever adventures your summer might hold (be it research, vacation or sleep). The FUN Executive Committee has held two web conferences to finish business from the last, and to begin planning for the next, SfN. We have added a few new members to some standing committee: Samantha Gizerian and Kristen Frenzel are working with Veronica Acosta and Bob Calin-Jageman on the website, Marsha Penner is helping Joe Trapani with social media and Elizabeth Becker helped create this newsletter – thanks to those members for stepping into more active roles. The Exec committee has also been discussing that the Society changed our Social and Poster Session to Sun evening.

Interested in doing more for FUN? We will be electing a President-elect, Treasurer-elect and 3 Councilors this fall. Please check out the website, see what the tasks are, ask other members, those currently in positions, and then keep an eye out for an announcement from Leah Chase later in the summer to make a nomination.

Don't forget about Student Travel Awards. As you submit your SfN abstracts think about your strong student co-authors that might apply for FUN travel awards. That application deadline is May 20 and will require each student to have a faculty sponsor and letter of support. Abstracts for the Social and Poster session are not due until September, so you have time.

Please be in touch if you have ideas, thoughts, or concerns about FUN.

Amy Jo Staimer

Make your Voice Heard

Submit to the next issue of the FUN Newsletter.
We welcome submissions on any topic suitable for the FUN membership.

An undergraduate lab exercise

to explore the functional beauty of identified neurons: Fluorescent visualization of the motor neurons innervating the superficial flexor muscle of the crayfish tail

By Stephen Hauptman, Bruce Johnson, and Hadley W Horch

A search for images of neurons on the web quickly leads to breathtaking photographs and drawings of neurons of differing types and complexities (cf. Brainbow images: http://cbs.fas.harvard.edu/science/connectome-project/brainbow#). These images can inspire students (and faculty) to pursue a deeper understanding of neuroscience fundamentals. When Ron Hoy, Bob Wyttenbach and one of us (BJ) produced the Crawdad lab manual (Wyttenbach et al 1999) we described two complementary student exercises, electrophysiological and anatomical, that examined the organization of a small motor network in crayfish. We hoped that describing electrophysiological properties of individual neurons within a limited neural network along with visualizing their neuroanatomy, would provide a deeper understanding of neuronal form and function than could be achieved with either approach alone. We chose the classical crayfish abdominal nerve 3/superficial flexor (SF) muscle preparation (Atwood 2008) for this because there are only 6 motor neurons that travel though a purely motor nerve to innervate this crayfish tail postural muscle. The motor neurons are tonically active *in vivo* and *in vitro* and have different axon diameters. Thus, the six neurons can be distinguished physiologically by their extracellular action potential amplitudes amplitudes. Along with the physiology exercise, our students attempted an anatomical exercise to visualize the motor neurons' structure by backfilling the distal cut ends of nerve 3 with cobalt. Cobalt retrogradely diffuses through the individual cut axons into the ganglionic cell bodies and their dendrites, and can be precipitated in the neurons upon further processing to reveal their morphology.

However, in our students' hands, and with the normal time constraints of a few lab sessions, successful preparations were rare and at best disappointing. A successful cobalt backfilling of nerve 3 axons showed dark, dye filled cell bodies, some axons, and murky grey shadows of dendrites. The "wow" factor was weak. Consequently, only the electrophysiological characterization of the nerve 3 motor neurons has stood the test of time as a student laboratory exercise (Wyttenbach et al 2014).

Two of us (HWH and SH) have developed a neuronal staining exercise with fluorescently labeled dyes that can now fulfill the original Crawdad goal. Using the standard nerve 3 dissection from the Crawdad lab manual, we have the students perform the neuronal dye fills *in situ*. In order to isolate the nerve, a petroleum jelly well is constructed in the isolated tail preparation, the nerve is cut in the well, and biocytin is added to the well, which then fills the neurons all the way back to the cell bodies in the ganglia of the ventral nerve cord. The biocytin can be directly conjugated to either green or red fluorescence, or one can use a secondary process that uses a fluorescently tagged streptavidin. With these techniques, students are much more successful visualizing the cell bodies and their associated dendrites in the ganglia. We have had success using either standard epifluorescent microscopes, or, for additional detail and three-dimensional focus, a confocal microscope.

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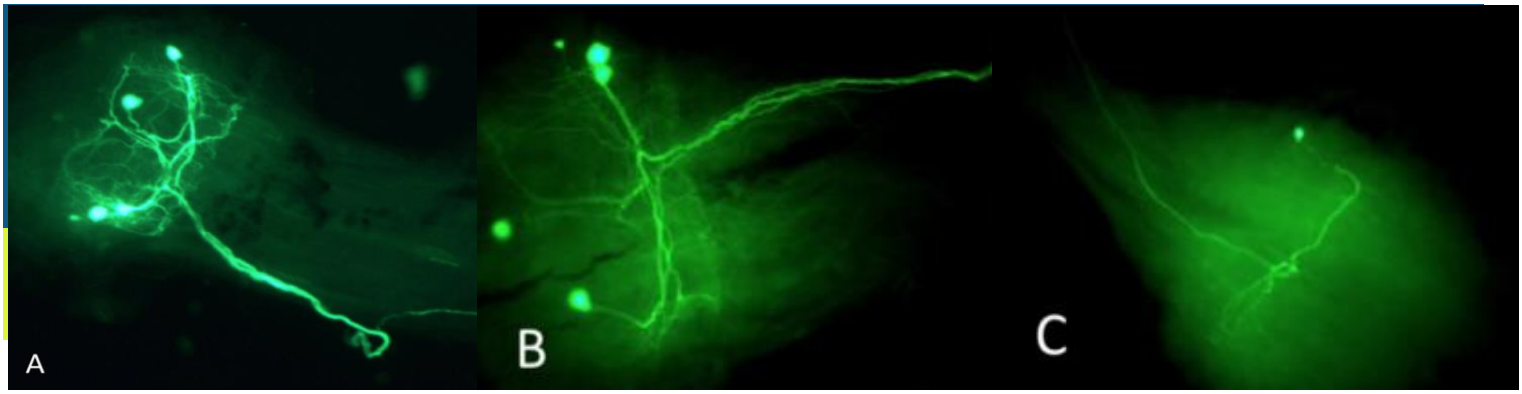

FIGURE Crayfish nerve 3 images prepared by the students in the Neurobiology class at Bowdoin College. **A**. Overview showing the proximate ganglion, five nerve 3 cell bodies and their axons, and the single axon travelling caudally to the more posterior ganglion. **B.** Anterior ganglion showing all five cell bodies. Axonal diameters can be compared. **C.** More posterior ganglion showing the single cell body located there and its axon. Images by students at Bowdoin College.

The students can more clearly count the axons in nerve 3 and match the axonal counts with their electrophysiological results. The neuroanatomical results clearly show 5 cell bodies in the ganglion of nerve 3 origin (Figure A and B) and 1 cell body in the next posterior ganglion that sends an axon rostrally to join the others (Figure C), for the full complement of 6 axons innervating the SF muscle. It is possible to compare the diameters of the axons and consider how they correspond to the amplitudes of the recorded extracellular spikes. Now the neuroanatomy and the extracellular recordings from the same neurons are truly complementary. The streptavidin technique is somewhat more involved than the conjugated biocytin approach, but it brings additional clarity, depth of focus, and details to the visualized anatomy. Both techniques are quite manageable in the undergraduate lab, and student success rate is about 80%. The quality and completeness of the fills varies from lab group to lab group, so we make all the images available to everyone in the class in order for the students to accurately reconstruct the anatomy.

Using this procedure, techniques for visualizing neurons can more easily be the focus of stand-alone labs, or can complement labs focusing on neuronal electrophysiology. The crayfish nerve 3 preparation is well-suited to the visualization of individual neurons because of the limited number of neurons in the nerve and the ease of access for a backfill of fluorescently labeled dyes. Students are very engaged in the staining technique and become excited about the aesthetically beautiful images. They also develop valuable microdissection skills during the process of preparing these images.

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Wyttenbach, RA, Johnson, BR and RR Hoy. 2014. Crawdad: An Online Lab Manual for Neurophysiology. Sinauer Associates, Inc., Sunderland, MA.

Contact Stephen Hauptman at Bowdoin College (shauptma@bowdoin.edu) for detailed staining protocols and questions. A full article on this exercise is in preparation to be submitted to JUNE.

Brain Awareness Outreach: A Liberal Arts Perspective

By Cecilia M. Fox, Director of the Neuroscience Program, Moravian College
President of the Lehigh Valley SfN Chapter

If you are a colleague at a liberal arts institution, it may not be surprising to be asked the following question: "how are the liberal arts embedded within the neuroscience program?". As we try to address the ever-changing nature of higher education and the influx of new technologies for pedagogical success, the answer to this question may seem like a moving target. My first consideration to such a question is to reflect on my interpretation of a liberal education. Of the various definitions that exist, I most closely relate to that of the Association of American Colleges and Universities that states, "a liberal education is an approach to learning that empowers individuals and prepares them to deal with complexity, diversity, and change. It provides students with broad knowledge of the wider world (e.g. science, culture, and society) as well as in-depth study in a specific area of interest."

During my thirteen years at Moravian College, I have witnessed how our liberal arts curriculum develops communication skills, analytical proficiency and social responsibility for the common good among our students. Over a decade ago, I designed our neuroscience major using the mission of our college as a guiding principle.

From the start, I envisioned an interdisciplinary program that would form meaningful connections among the humanities, social sciences and natural sciences. Our majors learn about the brain and behavior in biology, psychology, philosophy, and computer science courses. But, it is our Brain Awareness Outreach Program that highlights the extensions of neuroscience into areas such as art, music and sociology.

One of the goals of our neuroscience program is to educate our college and Lehigh Valley community about the brain and benefits of neuroscience research. Our service learning events have extended into public libraries, schools, assisted living communities, churches and non-profit organizations. As the home of the Lehigh Valley Society for Neuroscience Chapter, we have developed a strong collaborative program with the other colleges and universities in the Lehigh Valley. Service learning projects are incorporated into the curriculum of our major. Students then share their hands-on neuroscience stations or research seminars with the public. Our majors tend to double major or minor in areas outside the natural sciences. Some of our neuroscience undergraduates have pursued additional degrees in photography, Spanish, Japanese, philosophy, art and music. These students contribute unique views of the mind and brain from a multidisciplinary perspective. This enriches conversations in class as well as the development of some truly engaging service learning activities.

Brain Awareness Season in the Lehigh Valley has always focused on a theme that is designed to attract undergraduates and faculty from other disciplines. Examples of such events are the Art of Neuroscience, The Musical Brain, Brain Gender and most recently, The Neuroscience of Poverty and Inequality.

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We intentionally draw upon the liberal arts to provide seminars and films on a given topic. The creative nature of our undergraduates has only strengthened the outreach programming that we offer to our community. At a given event, we will demonstrate the typical comparative brain anatomy (a station we commonly refer to as "Cow Brains and Sheep Brains and Rat Brains, Oh My!"). However, we then tie this to another station where children use clay to build brains that may be missing a particular sense, such as smell or have an enhanced visual sense. They are asked, "what would that brain look like?". We have introduced art stations known as "Dendritic Art" where children draw neuronal connections using nothing but paint and a straw to blow through. We also have a concussion station where a "helmet" is made for a character using Legos and then the integrity of this helmet is tested (incredibly popular with children from ages 2-102!). We have even used some of the clever products from Backyard Brains to draw in those who love electrical circuitry.

Our majors have truly enjoyed and taken ownership of these programs as they see how engaged the local community becomes when learning about the brain and behavior in this accessible manner.

In an effort to instill a sense of civic responsibility, our majors are also learning how to become advocates for supporting scientific funding. With the next presidential election just around the corner, our undergraduates are beginning to educate themselves about their representatives' views on scientific funding. As an SfN Chapter we have even taken steps to invite our local representatives to attend our upcoming LVSfN Undergraduate Research Conference so they may learn of the importance of supporting this next generation of scientists, physicians and educators. It is our hope that through this type of engagement, we may be able to positively encourage our elected representatives to support funding for NIH and NSF.

As I reflect on the past thirteen years at Moravian College, it has been a wonderful educational journey for our undergraduates, faculty and local community. I work in an environment where I have permission to move outside the boundaries of the natural sciences and embrace other disciplines that only enhance my understanding of the brain. I began this piece with the question of "how are the liberal arts embedded within the neuroscience program?". At the end of the day, I find myself asking the question, "how is neuroscience embedded in the liberal arts"? We must prevent the silo approach to our disciplines and embrace the interconnectedness that can exist. It is in the liberal arts where the brain, mind and soul come together to express our humanity.

CrawFlying

By Bruce R. Johnson, Cornell University

For the last five years the Department of Neurobiology and Behavior at Cornell has been the base for evolving partnerships that continuously shape the faculty teaching workshops originally based on the Crawdad Neurophysiology Lab Manual (Wyttenbach et al 2014). Our present workshops train neuroscience educators in classical neurophysiology techniques and advanced neurogenetic approaches that can be adapted for the undergraduate neuroscience teaching laboratory. The workshops offer hands-on experience with invertebrate model preparations as an effective way to give students insights into signal transmission in the nervous system of all animals, and for students to learn and practice the skills of a working neuroscientist.

Our Cornell program of faculty neuroscience workshops began in the 1990s with an NSF grant to Ron Hoy to work with Bob Wyttenbach and me to develop the Crawdad lab Manual and organize of a series of yearly week-long faculty workshops for neuroscience educators. The workshop goals were to provide resources for neuroscience educators to run laboratory exercises addressing fundamental aspects of signal transmission in the nervous system, and to initiate a community of educators supporting each other. Until the FUN Electrophysiology Labs (IFEL) workshop at Bowdoin College in the summer of 2006, our busy professional lives mostly distracted us from this mission after the NSF workshops ended.

The IFEL workshop brought together Carol Ann Paul, Steve Hauptman, Wes Colgan from ADInstruments (ADI), and me to teach in an intense and inspiring FUN event. This became the seed and model and for reviving the CrawDad workshops, and for the initiation of other intensive regional faculty workshops with an academic and industry partnership. The official partnership between Cornell and ADI to co-sponsor the modern Crawdad-based faculty workshops began at Cornell in June 2011 with a traditional CrawDa workshop. Cornell supplied faculty (Ron Hoy, Bob Wyttenbach [now at Emory University] and me), the teaching lab and electrophysiological equipment. ADI supported Wes Colgan as a faculty member (with ADI engineers rotating through), training for participants, and competitive tuition scholarships, especially for young investigators, women and under-represented minorities, and supplied hardware and software. Also around this time Stefan Pulver (the lead in developing new fruitfly optogenetics student lab exercises [see Pulver and Berni, 2012; Pulver et al, 2011]) began teaching with me at Cornell. Shortly after, CrawFly was born in 2012 as a new Cornell/ADI faculty workshop, with the additional partnerships of the Howard Hughes Medical Institute (HHMI) supporting Stefan as faculty, and additional equipment support from A-M Systems. CrawFly's goal was to provide complementary approaches to teaching integrative neuroscience for international neuroscience teaching faculty.

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The 2016 JUNE CrawFly Faculty Workshop will be held Wed., June 22 through Sun., June 26 at Cornell in Ithaca NY

https://www.eventbrite.com/e/crawfly-invertebrate-neurophysiology-course-tickets-21385675130

Scholarships for tuition and other support are available to encourage participant diversity and the interests of beginning faculty and graduate students/post-docs in teaching practical laboratory courses in neuroscience. Note scholarship application deadline: April 21, 2016, with awards announcement by May 1, 2016. Contact Bruce Johnson (brj1@cornell.edu) for questions and more information.

Other upcoming co-sponsored faculty workshops:

https://www.eventbrite.com/e/intro-to-neurophysiology-workshop-tickets-22118624401

http://www.adinstruments.com/events/training/snailfest-invertebrate-neurophysiology-workshop

The Craw section teaches basic electrophysiological recording and signal transmission in the nervous system with time-tested crayfish, snail and plant preparations. The Fly section teaches principles of neurogenetics, and behavioral assays to examine remote control of neural circuits in behaving animals and remote control of synaptic transmission in *in vitro* preps through optogenetic manipulations. The general workshop outcomes for participants include: 1) hands-on experience with invertebrate teaching modules for the undergraduate laboratory that teach basic principles of genetics, nervous system physiology and animal behavior, 2) facility with electrophysiological equipment and data acquisition and analysis systems, and 3) interaction with a broader and international neuroscience teaching community. Past participants include senior educators, lab coordinators, beginning faculty, post-docs and advanced graduate students interested in teaching careers.

Evaluations of previous CrawFly workshops demonstrate our success in accomplishing our workshop outcomes and promoting new teaching at participants' home institutions. A full Crawfly article including our workshop assessments will be presented in a future JUNE issue.

More recently, Stefan left the US for a faculty position in neuroscience at the University of St Andrews in Scotland, and he is focusing on his research and teaching there. New partnerships for the workshop include additional Fly faculty: Ilya Vilinsky (University of Cincinnati), Karen Hibbard (HHMI at Janelia Research Campus- continuing the HHMI partnership), and David Deitcher (Cornell University). The shift in the Fly faculty has added new insect ERG exercises (Stowasser et al., 2015) as well as continued development and inclusion of new fly lab exercises (Titlow et al., 2015) without the constraints required with vertebrate preparations.

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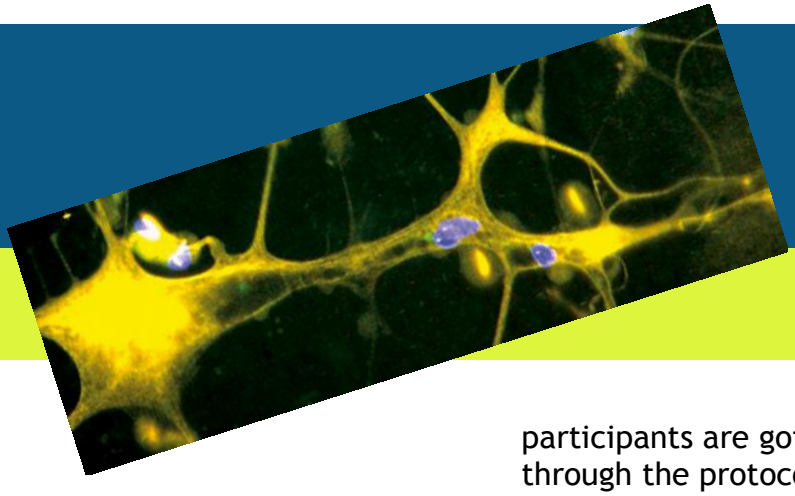

Responsible Research Planning?

By: Bob Calin-Jageman
Dominican University

An important part of training the next generation of neuroscientists is teaching ethical research conduct (e.g. Frantz, DeHaan, Demetrikopoulos, & Carruth, 2006) . Some topics in research conduct are familiar and are already well integrated into many neuroscience curricula: protecting confidentiality, ensuring data integrity, etc. One emerging theme, though, is in the need for *responsible research planning*, a topic that springs from the surprising confluence of inferential statistics and research ethics.

What is the ethical dimension to research planning? It's all about selecting an ethically defensible sample size. Sample sizes that are too small relative to the research question will be unlikely to detect expected effects—this means that animal or human subjects are being put through a research protocol with high odds of futility. Sample sizes that are too large, though, can also be ethically questionable because in this case more participants are going through the protocol than reasonably necessary to answer the research question. Good planning from the start, then, is needed to plan a sample size likely to be just right.

Currently, responsible research planning is not often part of our research practices. Imagine supervising an enthusiastic undergraduate who wants to study sex differences in water maze performance. You'd spend time and effort helping the student develop a proposal outlining their research hypotheses, experimental procedures, and (hopefully) analysis strategies. In addition, you'd work together to obtain IACUC approval. What about sample size planning? Often, this is left vague, with students running their studies until they run out of time, money, and/or interest. Clearly, though, this is problematic, and is likely to lead to the use of research animals for purposes that are either futile or wasteful. Even when sample sizes are planned, it is often in reference to studies in the topic.

Unfortunately, previous reports are likely to have inadequate sample sizes (Button et al., 2013), so following precedent may also be problematic. Incorporating responsible research planning into student training is not very difficult. There are a number of free tools that can be used to help plan sample sizes for adequate power (e.g. Dupont & Plummer, 1998) or precision (e.g. Cumming, 2011). Unfortunately, planning a sample size in advance can be sobering. For example, the student in the example above would find from previous research that sex differences in water maze performance typically yield Cohen's d = 0.49 (Jonasson, 2005). In most research contexts this is considered a fairly substantial effect. Unfortunately, this effect size requires a whopping 66 animals *per group* to achieve power of 0.8 (and many more to try to find a contrasting interaction with a drug treatment). For a truly depressing set of sample size recommendations, check out this article by Lakens and Evers (2014).

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“Engaging in responsible research planning is likely to require more expense, more time, and/or more ingenuity. But this is likely to become the new norm, as granting institutions and ethical review boards are becoming increasingly cognizant about these issues.”

How can we cope with the sample sizes required for responsible research? One good option is to conduct cumulative research projects in which each student contributes additional data towards an overall research question. Data sharing can even allow this to happen on a large scale (Lakens, 2013), enabling students to become part of crowd-sourced projects that answer important research questions (Grahe et al., 2012).

Engaging in responsible research planning is likely to require more expense, more time, and/or more ingenuity. But this is likely to become the new norm, as granting institutions and ethical review boards are becoming increasingly cognizant about these issues. Fortunately, planning reasonable sample sizes provides more than just ethical peace of mind—it should also make your research projects much more likely provide precise and interpretable results.

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Upcoming FUN events!

FUN workshop—save the dates:

July 28-30, 2017 at Dominican University (River Forest, Illinois)

There will be a preworkshop intensive laboratory experience as well as a Best Practices poster session

2016 Equipment Loan Program call for proposals now open

- A complete list of available equipment is on the FUN website http://www.funfaculty.org
- Review of applications will begin on Oct 1
- Questions? Contact Jeff Smith jsmith12@svsu.edu

Deadline for submissions for next FUN Newsletter – Aug 1

2016 Travel Award application online submissions now open (deadline May 20!)

Questions? Contact Leah Chase chase@hope.edu

SFN changes you need to know about:

FUN Social and Poster Session – Sunday Nov. 13, 6:45-8:45 pm, location TBA

FUN Business Meeting – Sunday Nov. 13, 7-8 am, location TBA