



#ALLFORWU

BIOLOGY: DNA SEQUENCING TECHNOLOGY

Project Overview: Our project is designed (1) to sequence microorganisms from environmental samples in Kansas and (2) to examine the utility of DNA sequencing applications in Forensic Biology. In order to accomplish our goals, we are seeking funding to purchase the Oxford Nanopore MinION. The MinION is a mini, portable DNA sequencer capable of sequencing an entire genome over only a few days. Many popular applications of DNA technology (ex: kits to determine ancestry, DNA profiling, etc.) are predicated on analyzing only small portions of the genome, so much can be learned with whole genome sequencing. Previously, this type of technology required larger and more expensive instrumentation. The MinION represents a way to expose Washburn students to cutting edge sequencing technology, setting them up for future success. **Our goal is to raise \$5,000 to bring this instrumentation, and the many benefits it will give our students, to Washburn.**



How Your Donation Benefits Washburn: DNA sequencing technology represents the cutting edge of the molecular biology field. The cost generally associated with this technology often makes it inaccessible to undergraduate students. Through your donations, we can give Washburn students access to this technology that will benefit them by:

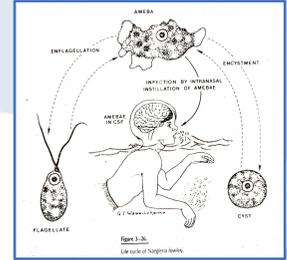
- Gaining a competitive advantage over peers in the job market or pursuing graduate studies by increasing their familiarity with the technology that is prominent in the field
- Incorporating current technology into biology courses
- Integrating computer science and biology by introducing students to the bioinformatics tools used to process the data
- Giving students a unique research experience (see below for examples)



Sequencing Free-Living Ameboflagellates Collected from Kansas Soil and Water Using MinION Technology
Dr. John Mullican, Department of Biology

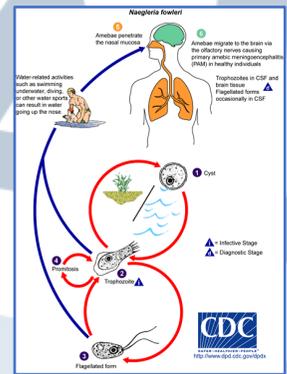
WHAT?

Free-living ameboflagellates in the genus *Naegleria* are ubiquitous in nature and have been found on all continents. They are fascinating to study because the "crawling" amoebae can transform into "swimming" flagellates or "dormant" cysts.



WHY?

While most are innocuous one species, *Naegleria fowleri*, is a potential human pathogen. Better known as the "brain-eating amoeba," *N. fowleri* causes a fatal primary amebic meningoencephalitis (PAM) that kills nearly 100% of its victims. We are interested in surveying Kansas freshwater and soils for *N. fowleri* and other *Naegleria* species to learn more about their



prevalence and ecology of Kansas environmental samples may identify new species and my help understand why some are human pathogens and most are not.



Sequencing Bacteriophage DNA from Environmental Samples Using MinION Technology
Dr. Andrew Herbig, Department of Biology



ATGCTGTGCACGCCAAATT
TACGCACCGTGCCTTTAA

What are Bacteriophages?

Bacteriophages, or phages, are viruses which specifically infect and replicate within bacteria. Many phages kill their hosts after they replicate and thus greatly impact bacterial populations.

Why are we interested?

Like living cells, phages have DNA which codes for all of its characteristics. Sequencing phage DNA will allow us to know much more about their biology. Students in Dr. Herbig's research lab have been studying phages isolated from soil samples in Kansas to learn about their replication characteristics. Sequencing the genes from these phages will greatly improve our understanding of these fascinating microbes.

How will we use the MinION?



The Use of MinION Sequencing Technology in Human and Nonhuman Forensics
Dr. Joshua Smith, Department of Biology

Human Forensics: The DNA that is most commonly evaluated for human forensic purposes includes only a small percentage of the total genome. This DNA is from noncoding regions, meaning it cannot be used to predict physical traits. As sequencing technology has improved, it is now possible to sequence more of the genome, including areas known to influence physical traits. For example, there are areas of the DNA that play a role in determining eye and natural hair color. Instrumentation such as the MinION can be used to sequence these areas, allowing for the possibility of suspect images no longer being forensic sketches, but instead

The IrisPlex System

Gene	SNP	Allele	No. of Alleles
HERC2	rs1393102	T	0 1 2
OCA2	rs1800407	A	0 1 2
SLC45A2	rs12886269	T	0 1 2
SLC45A2	rs15881862	C	0 1 2
TYR	rs1288350	T	0 1 2
RP4	rs1202592	T	0 1 2

Display Predicted Phenotype | Download Predicted Phenotype



The left image shows an example of how DNA can be used to predict eye color (from <https://irisplex.erasmusmc.nl/>). The right image shows a forensically important blowfly studied here at Washburn University (Image by Muhammad Mahdi Karim).

Nonhuman Forensics: There are nonhuman organisms that can also be important to a forensic investigation. For example, many species of blowflies are attracted to and lay eggs on a corpse. The larva that hatch will feed and stay on the corpse until they are ready to pupate. By estimating the age of the larva, an investigator can estimate how long the body has been there and presumably the minimum time since death. This estimate of the time since death is crucial in determining the validity of alibis.

Undergraduate Teaching

DNA sequencing of relatively small DNA fragments has been available at Washburn for many years but the MinION will bring a new technology to Washburn, permitting students to sequence 100% of the DNA from an organism in a day or two! Students in our teaching labs will be able to generate genome sequences from a variety of organisms, which will generate enormous data sets that can be studied for years. Knowing the genetic blueprint of life for new organisms will aid in the understanding of many

Undergraduate Research

Dr. Mullican and his undergraduate research students will sequence genomes of free-living ameboflagellates isolated from Kansas samples. Students will learn a new sequencing technology and how to organize, assemble, and interpret the DNA sequences from a variety of organisms, which will generate enormous data sets that can be studied for years. Knowing the genetic blueprint of life for new organisms will aid in the understanding of many



Undergraduate Teaching

This is an exciting opportunity to bring DNA sequencing technology into the microbiology teaching lab! Students will be able to quickly generate and analyze sequence data from phages and other microbes they have obtained from environmental samples. Since the MinION generates data in real-time, students can monitor the progress of their experiment during the lab period. Students will design their own sequencing projects, generate data, and learn bioinformatics to analyze their results.

Undergraduate Research

DNA sequencing and analysis is at the forefront of today's biological sciences. Students interested in independent research projects in Dr. Herbig's lab will be able to learn a cutting-edge technology and generate data which they can present and publish. They will be generating new knowledge in phage biology and genomics.

Undergraduate Teaching

As part of a future Forensic Biology degree, a course in Forensic Molecular Biology will soon be offered. A course topic will be phenotyping, where DNA can be used to predict physical traits (i.e. eye color). The MinION gives students the ability to conduct phenotyping and use their own results to evaluate how reliable these marker types are; thus, bringing real world application to the classroom setting. Additionally, students will be able to confidently discuss this technology with graduate schools and/or jobs after graduation.

Undergraduate Research

Students in Dr. Smith's research lab work on the genetics of forensically important blowfly species as it pertains to development rate. An example of a trait known to influence these development rates in other insects, but is not well understood for blowflies, is sex (gender). The MinION would allow for the generation of independent research projects focused on how sex influences development rate. The data generated will allow for student publications that could help develop policy in the forensic

Washburn students who use the MinION will have an opportunity that is unique among undergraduate institutions!