C. elegans and Alcohol – A Physiological Response

Document Overview:
Teacher Handout (including background information)
Student Handout (incuding materials list)

Minnesota State Science Standards:
9.1.1.1.2 Understand that scientists conduct investigations for a variety of reasons, including: to discover new aspects of the natural world, to explain observed phenomena, to test the conclusions of prior investigations or to test the predictions of current theories.
9.1.1.2.1 Formulate a testable hypothesis, design and conduct an experiment to test the hypothesis, analyze the data, consider alternative explanations and draw conclusions supported by evidence from the investigation.
9.1.3.3.2 Communicate, justify and defend the procedures and results of a scientific inquiry or engineering design project using verbal, graphic, quantitative, virtual or written means.
9.1.3.4.3 Select and use appropriate numeric, symbolic, pictorial, or graphical representation to communicate scientific ideas, procedures and experimental results.
9.1.3.4.6 Extension: Analyze the strengths and limitations of physical, conceptual, mathematical and computer models used by scientists and engineers.
9.4.1.1.1 Extension: Explain how cell processes are influenced by internal and external factors, such as pH and temperature, and how cells and organisms respond to changes in their environment to maintain homeostasis.
9.4.1.1.2 Extension: Describe how the functions of individual organ systems are integrated to maintain homeostasis in an organism
9.4.4.2.4 Extension: Explain how environmental factors and personal decisions, such as water quality, air quality and smoking affect personal and community health.
9C.1.3.3.1 Extension: Explain the political, societal, economic and environmental impact of chemical products.

Objective:
Students will be able to design, conduct, and analyze a controlled experiment testing the effects of alcohol on different strains of the roundworm C. elegans in regards to movement.

Type of Activity: Lab

Duration: 1-2 Class Periods (55-110 min)

Connection to Nobel speakers:
Paul W. Glimcher, Professor of Neural Science, Economics, and Psychology, Center for Neural Science, and Director, Center for Neuroeconomics, New York University
Working with Professor David Sparks at the University of Pennsylvania in the early '90s researching the brainstem and those nuclei that control eye rotations, Paul Glimcher uncovered evidence that structures participating in the execution of saccadic eye movements might be involved in planning those movements as well. Consequently, his lab has focused on the identification and characterization of signals that intervene between the neural processes that engage in sensory decoding and those that engage in movement generations. These are the signals that must, in principle, underlie decision-making. Glimcher and his lab study these processes using tools drawn from the fields of neuroscience, economics, and psychology, with methodologies ranging from single-neuron electrophysiology to game theory.

Larry J. Young, William P. Timmie Professor, Department of Psychiatry and Behavioral Sciences, Emory University School of Medicine, and collaboratory leader, Center for Behavioral Neuroscience, Atlanta, Ga.

Larry Young has spent his professional life trying to understand the relationship between genes, brain, and innate behaviors. As a postdoctoral fellow he began investigating the molecular mechanisms underlying social attachment in prairie voles, which differ from other species of voles in forming lifelong social bonds. Earlier research had identified two hormones in the regulation of pair bond formation in prairie voles and, using comparative molecular approaches, Young investigated the molecular mechanisms underlying the species’ differences in behavior. His lab is now using interdisciplinary approaches to understand how specific genes regulate the expression of innate behaviors, with a continuing focus on social attachment and social behavior in general. By understanding the mechanisms underlying social attachment, he and his colleagues hope to gain insight into human disorders characterized by social impairments, including autism spectrum disorders and schizophrenia.

Connection to Other Activities: “Understanding the Ones”
C. elegans and Alcohol - A Physiological Response
Teacher Handout

Background Information*:
Segment taken directly from http://www.vcu.edu/pharmtox/faculty/faculty_bios/davies.htm

As occurs with humans, C. elegans display dose-dependent alterations in behaviors when intoxicated by alcohol. By concentrating on the depressive effects of alcohol it is possible to identify multiple genetic mutants that display altered sensitivity to the behavioral effects of alcohol. Several of the identified mutations affect a single gene, slo-1, that encodes the C. elegans voltage and calcium-dependent large conductance potassium (BK) channel. Electrophysiology and further genetic studies showed that alcohol appears to activate the SLO-1 potassium channel, an action that would result in decreased neuronal activity and could explain aspects of the behavioral depression associated with alcohol intoxication. C. elegans genetics may be used to identify other molecular targets of alcohol. The identification of genes that mediate alcohol’s effects in C. elegans will provide candidates for genes that play similar roles in humans. Genes that affect an individual’s acute sensitivity to the drug may be important in predisposing an individual to alcoholism because there is a strong correlation between a naive drinker’s level of tolerance to alcohol and their likelihood of abusing alcohol later in life.

* Idea - Students will diagram the above paragraph to understand the differences in the two strains of C. elegans regarding neural structure, potassium channels, and the affect of alcohol.

Concepts, Connections, and Terms:

- Addiction
- Alcohol
- Genes
- Mutation
- Proteins

Acknowledgements:
Modified from Brain U at http://brainu.org/c-elegans-and-alcohol
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Engage –
What do you know about C. elegans? What did they look like? How did they move?
Movement Video Clip - http://youtu.be/olrkWpCqVCE (this address IS correct)
Or completion of “Understanding the Uncs” lab activity

Ask the class to think about and share what might happen if the worms were placed into alcohol? Why? Record this list on the whiteboard or overhead.

*Develop Questions – Experimental Design*

Discuss* with students how they should watch the worms’ behavior and record any changes they see. What ways could you (students) measure movement? Qualitatively? Quantitatively?

*Discussion may lead to watching thrashing. This is a good behavior to track.*

One way to quantify locomotor activity (i.e. thrashing)

1. Decide how you are going to define “thrashing” behavior and describe it.
2. Decide which person will keep time and who will count the thrashes.
3. Set the timer for 30 seconds.
4. Have one partner look at the worms from one petri dish or depression in the slide under the microscope. Focus on one worm and start counting the number of thrashes or movements it makes after the timer is on.
5. Stop for 2-3-minutes, then once again, set the timer for 30 seconds, and follow step
6. Do this step one more time.
7. After counting the thrashes, do steps B and C for the worms on the second petri dish or slide well in the slide.

*Lab Activity* - See Student Handout - “C. elegans and Alcohol - A Physiological Response”

Materials may be obtained from [http://www.cbs.umn.edu/CGC/](http://www.cbs.umn.edu/CGC/).

- petri dish 1—contains water
- petri dish 2—contains water
- petri dish 3—contains alcohol
- petri dish 4—contains alcohol
- N2 worms (2 petri dishes)
- Slo-1 strain worms (2 petri dishes)
- distilled water (4 ml)
- pipettes (2)
- small cap vials (4)
- stable magnifiers and/or microscopes

*Explore 1 – Concentrating the Worms*

Materials: *C. elegans* on agar, a pipette or eyedropper, and a snap cap vial.

1. Give each pair a petri dish with *C. elegans* on agar, a pipette or eyedropper, and a snap cap vial.
2. Have students pipette out 1 ml of distilled water and gently put it into the petri dish with
worms.
3. Have them gently swirl the petri dish, while keeping it horizontal, to get the worms into the water.
4. Have student tilt the dish at a 30 degree angle and use the pipette/eyedropper to GENTLY draw up the water with worms.

*You might get students first to practice keeping the water at the end of the dropper by drawing up a little distilled water.*
5. The worm-containing water should be gently squeezed into the snap cap vial.
6. The snap vials should be capped, labeled with initials, and put upright in ice for 5-10 minutes; a cloud of cold worms will be form on the bottom of the vial.
Ask students: What will happen to the worms when the ice cools them?
(The worms will stop moving and settle to the bottom of the vial).
Why might the C. elegans die if they are cooled too much?
(The enzymatic reactions will cease to occur).
7. Give each pair a petri dish with only water and one with only alcohol for each strain of C. elegan.
*Follow Steps 1-6 for each strain of C. elegans. In the end there should be 2 petri dishes with water, 2 petri dishes with alcohol, 2 small capped vials with N2 strain C. elegans and 2 small capped vials with Slo-1 strain C. elegans.*

Explore 1 - Observations - Try looking at how the worm strains behave in water.

1. Use a pipette to suction up the bottom (where the worms are concentrated) of the small capped vial of the Slo-1 worms.
2. Place the Slo-1 worms in the petri dish with the water.
3. Place the petri dish with the Slo-1 worms under the microscope or magnifying glass. Record your observations.
4. Repeat steps 1-3 for the N2 worms.

• How does each strain of worm move in/on water?

N2s? __________________________________________

Slo - 1 Strain? ______________________________________

•Using a Venn Diagram, compare and contrast the movement and behavior of the worms on each petri dish.
Explore 2 – Now try looking at how the worm strains behave in alcohol

1. Use a pipette to draw up the bottom (where the worms are concentrated) of the small capped vial of the Slo-1 worms.

2. Place the Slo-1 worms in the petri dish with the alcohol.

3. Place the petri dish with the Slo-1 worms under the microscope or magnifying glass. Record your observations.

4. Repeat steps 1-3 for the N2 worms.

• How does each strain of worm move in/on alcohol?

N2s? ________________________________

Slo - 1 Strain? ________________________________

• Using a Venn Diagram, compare and contrast the movement and behavior of the worms on each petri dish.

Explore 2—Observations

• What do you notice about how the worms move in alcohol compared to water?

• What do you think is happening to the worms?

• Looking at the N2 and the Slo-1 strains of worms, what do you notice is similar about the two types of worms?

• Looking at the N2 and the Slo-1 strains of worms, what do you notice is different about the two types of worms?

• Using a Venn Diagram, compare and contrast the movement and behavior of the worms on each petri dish.
Assessment Ideas:
- Formal Lab Report including Observation Answers
- Student Handout
- Extensions
- Class Presentation of results

Extensions:
1. Analyze the strengths and limitations of using the *C. elegans* as a model.

2. Create a lab to observe the changes in *C. elegan* behavior as the alcohol is processed by the organism or with varying concentrations of alcohol.

3. Research the following questions:
   - How does alcohol affect the human body?
   - How is alcohol processed by a human?
   - How do these affects and processes help to maintain homeostasis?

4. If one’s genetic make-up predispose one to becoming an alcoholic, would you choose to be tested? Should testing be mandatory? Should only people that do not carry the genes for alcoholic predisposition be issued an “alcohol licence” (meaning they can consume alcohol while for others it is illegal)?

5. Discuss the political, societal or economic of alcohol use and/or abuse.

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**C. elegans** and Alcohol - A Physiological Response

Student Handout

*C. elegans* is a nematode—a member of the phylum Nematoda: The roundworms and threadworms, a phylum of smooth-skinned, unsegmented worms with a long cylindrical body shape tapered at the ends; includes free-living and parasitic forms both aquatic and terrestrial. *(Academic Press Dictionary of Science and Technology)*

*C. elegans* is a non-hazardous, non-infectious, non-pathogenic, non-parasitic organism. It is small, growing to about 1 mm in length, and lives in the soil—especially rotting vegetation—in many parts of the world, where it survives by feeding on microbes such as bacteria. It is of no
economic importance to man (http://www.cbs.umn.edu/CGC/what.html)

Materials:
- petri dish 1—contains water
- petri dish 2—contains water
- petri dish 3—contains alcohol
- petri dish 4—contains alcohol

and
- N2 worms on agar plates (2)
- Slo-1 strain worms on agar plates (2)
- distilled water
- alcohol
- pipettes (2)
- snap cap vials (4)
- stable magnifiers and/or microscopes

Procedure: Concentrating the Worms
1. Obtain a petri dish with C. elegans on agar, a pipette or eyedropper, and a snap cap vial.
2. Pipette 1 ml of distilled water and gently put it into the petri dish with worms.
3. Swirl the petri dish for one min., while keeping it horizontal, to get the worms into the water.
4. Tilt the dish at a 30 degree angle and use the pipette/eyedropper to GENTLY draw up the water with worms.
5. The water containing the worms should be gently squeezed into the snap cap vial.
6. The snap vials should be capped, labeled with initials, and put upright in ice for 5-10 minutes; a cloud of cold worms will be form on the bottom of the vial.
7. Follow Steps 1-6 for each strain of C. elegans. You should have two vials of each strain of C. elegans and four vials total..

Answer the following questions while you are waiting:
What happen to the worms movement when the ice cools them?

Explain your thoughts...

Explore 1 – Observations
1. Transfer the N2 strain worms from the bottom of the capped vial to petri dish 1- water using a pipette.
2. Transfer the Slo-1 strain worms from the bottom of the capped vial to petri dish 2- water using a pipette.
• How does each strain of worm move in/on water in petri dish 1 and 2?

N2s? ____________________________________________________________

Slo - 1 Strain? __________________________________________________

• Using a Venn Diagram, compare and contrast the movement and behavior of the worms on each petri dish.
Now try looking at how the worm strains behave in alcohol

*Explore 2 – Observations*

1. Transfer the N2 strain worms from the bottom of the second capped vial to petri dish 3-alcohol using a pipette.
2. Transfer the Slo-1 strain worms from the bottom of the second capped vial to petri dish 4 using a pipette.

- How does each strain of worm move in/on the alcohol in petri dish 3 and 4?

  N2s? ________________________________
  Slo - 1 Strain? ________________________________

- What do you notice about how the worms move in alcohol compared to how they move in water?

- What do you think is happening to the worms?

- Looking at the N2 and the Slo-1 strains of worms, what do you notice is similar about the two types of worms?

- Looking at the N2 and the Slo-1 strains of worms, what do you notice is different about the two types of worms?

- Using a Venn Diagram, compare and contrast the movement and behavior of the worms on each petri dish.

*Assessment:*

- Formal Lab Report including Observation Answers
- Class Report
- Extensions
Extensions:
1. Analyze the strengths and limitations of using the *C. elegans* as a model.

2. Create a lab to observe the changes in *C. elegan* behavior as the alcohol is processed by the organism or with varying concentrations of alcohol.

3. Research the following questions:
   ○ How does alcohol affect the human body?
   ○ How is alcohol processed by a human?
   ○ How do these affects and processes help to maintain homeostasis?

4. If one’s genetic make-up predispose one to becoming an alcoholic, would you choose to be tested? Should testing be mandatory? Should only people that do not carry the genes for alcoholic predisposition be issued an “alcohol licence” (meaning they can consume alcohol while for others it is illegal)?

5. Discuss the political, societal or economic issues surrounding alcohol use and/or abuse.