Research Experiences for Urban Science Teachers

Mortality rate of *Drosophila* following traumatic brain injury

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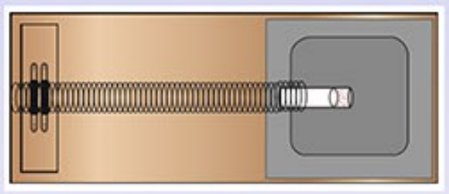
TBI in neurodegenerative *Drosophila*

Traumatic brain injury (TBI) results from a bump, jolt, or blow to the head that damages brain cells and functions.[1] Results may range from mild (concussion) to severe (extended loss of consciousness, memory loss, death).[2] Many resources are available regarding the cause, risk factors, and treatment of TBI patients; however, a limited amount of knowledge exists regarding the action of TBI on a cellular level or its effects in neuro compromised individuals. Here, we investigate severe TBI in four neurodegenerative diseased lines of *Drosophila melanogaster*.

Trauma device delivers controlled TBI

Students at Altoona Area Junior High School constructed a mechanism to deliver controllable closed brain trauma to *Drosophila.* The mechanism is based on the High Impact Trauma (HIT) device originally designed and engineered by Dr. David Wassarman of the University of Wisconsin.[2]



The HIT device delivers closed brain trauma to a large number of flies at once.

1. Flies are placed into an empty vial and confined with a cotton plug to the bottom fourth of the vial
2. Vial is loaded into spring and spring is raised to desired height
3. Spring is released causing vial to hit the foam pad delivering a blow to the flies

The user is able to control the degree of force applied by adjusting the angle to which the spring is raised as well as having the ability to deliver accurate multiple blows.

Neurodegenerative lines subjected to closed brain TBI

AAJHS students tested four fly lines to determine the effects of closed brain TBI on nerve cells predisposed to neurodegeneration. The overexpression of fly homolog genes for neurodegenerative diseases created models of Parkinson's, ALS, Huntington's, and Alzheimer's diseases in the fly lines. The expression was pan-neuronal. Students tested age, gender, and number of impacts with varying degrees of time between impacts using all four diseased lines.

Multiple TBI in Parkinson's flies shows steady decline

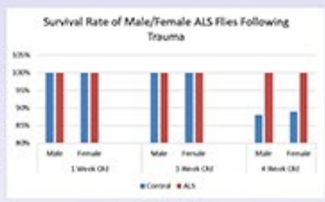
Students tested TBI affects in Parkinson's flies of three age ranges by striking with a 90° angle and delivering up to five hits with a five minute recovery time between impacts.

Students hypothesized the steady decline in survival was due to a nerve's innate ability to quickly recover from introduced trauma. Students suggested the diseased lines, already able to cope with damage, enacted a work-around to quickly recover from further trauma.

Gender shows no bearing on mortality following TBI

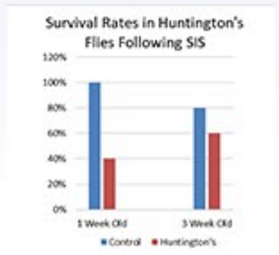
Students compared TBI in control and ALS and Huntington's flies. Males and females of young, mid, and old ages were tested independently with one strike from a 90° angle.

Students hypothesized that gender has no bearing on mortality. Older control flies were unable repair damage due to age factors independent of gender, while old aged ALS flies were better able to recover. Students hypothesized this could be due to a better repair mechanism following nerve injury.



Huntington's flies hint at neuro repair following SIS

Students investigated second impact syndrome (SIS) in Huntington's flies subjecting one and three week old flies of mixed gender to double impacts, allowing five minutes of recovery between impacts, and striking at a 45° angle.



Analysis shows an inverse relationship between age and survival. Students noted diseased flies were moving, twitching, and struggling to climb within seven minutes after the second impact. Control fly data for SIS was within 10% of human SIS data. Students suggested older neuro-damaged flies were used to damaged nerves and had established a way to stop further deterioration.

Standard protocol for future TBI trials

A standard fly protocol was developed for future student investigations. Flies of mixed ages were concussed at a 90° angle with a five minute wait time between impacts.

The vast majority of concussed flies that were incapacitated following initial trauma recovered within 10-15 minutes. Deaths shown can be attributed to TBI. Control flies hit at a 90° angle show a greater mortality rate at 24 and 48 hours. Hits at 75° did not show additional death after 24 hours.

Diseased show degeneration resistance

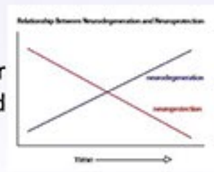
Neuro-diseased lines of mixed ages showed less mortality compared to control lines. Initial deaths increased with the number of hits. The number of flies alive at 24 and 48 hours is greater than control results. These lines may be more resistant to further degeneration because they have already been exposed to extensive stress. One cause could be a more effective neuroprotective cascade following TBI.

Future trials aimed at age and behavior

AAJHS students will investigate neurodegeneration, neuroprotection, and behavior in fly lines following TBI.

- Spinocerebellar ataxia (overexpression)
- Spastin (loss of function)
- Alzheimer's

1) Age parameters will be used to test for high neuroprotection in young flies and high neurodegeneration in older flies.



2) RING assay will be used as a model to access how motor functions and behavior changes in diseased flies after TBI.

References:

1. Katzenberger, R. J., Loewen, C. A., Wassarman, D. R., Petersen, A. J., Ganetzky, B., & Wassarman, D. A. (2013). A *Drosophila* model of closed head traumatic brain injury. *Proceedings of the National Academy of Sciences of the United States of America*, *110*(44), E4152–E4159. http://doi.org/10.1073/pnas.1316895110Muroyama
2. Katzenberger, R. J., Loewen, C. A., Bockstruck, R. T., Woods, M. A., Ganetzky, B., & Wassarman, D. A. (2015). A Method to Inflict Closed Head Traumatic Brain Injury in *Drosophila*. *Journal of Visualized Experiments : JoVE*, (100), 52905. Advance online publication. http://doi.org/10.3791/52905

Acknowledgments

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