BIO299 – Effects of Notch inhibitor on nervous system regeneration in *Nematostella vectensis*

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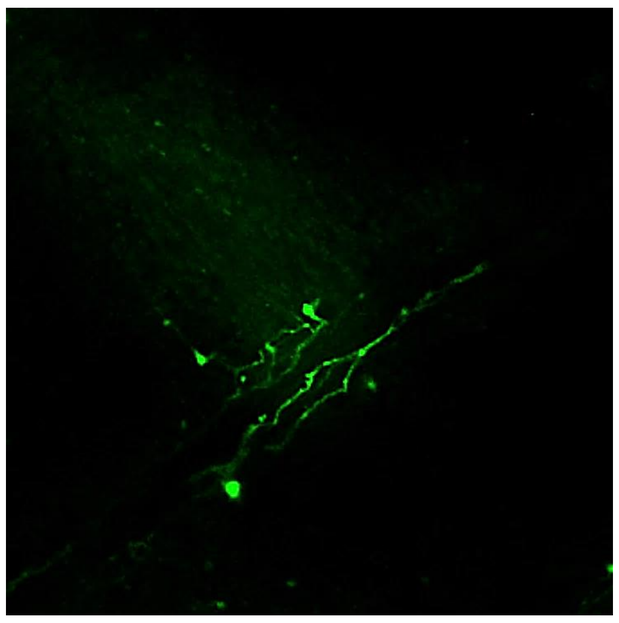
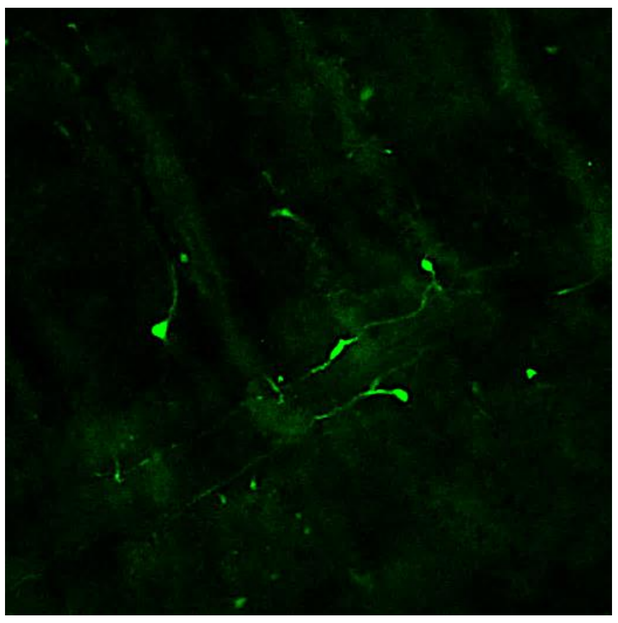
Why the starlet sea anemone?
It can regenerate any part of its body completely. We are interested in its ability to regenerate its nervous system after it is destroyed.

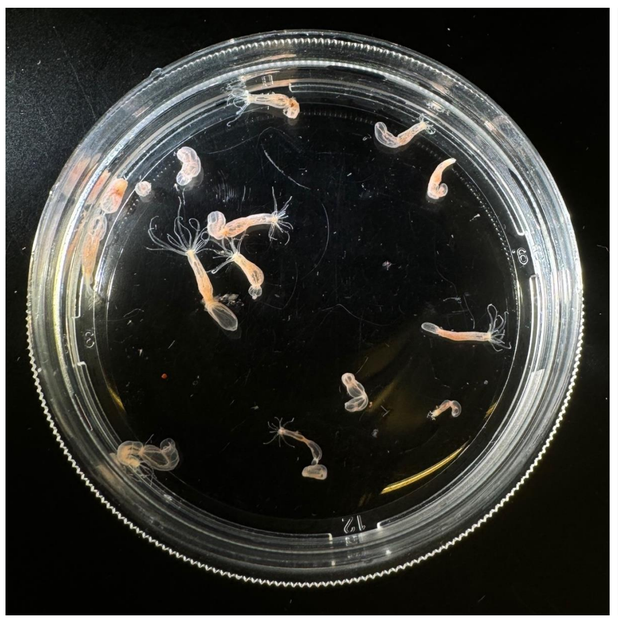
Why Notch inhibition?
Disrupting Notch signaling has shown to influence nervous system development in embryogenesis.
We were therefore interested in studying its effects on older animals ability to regenerate.

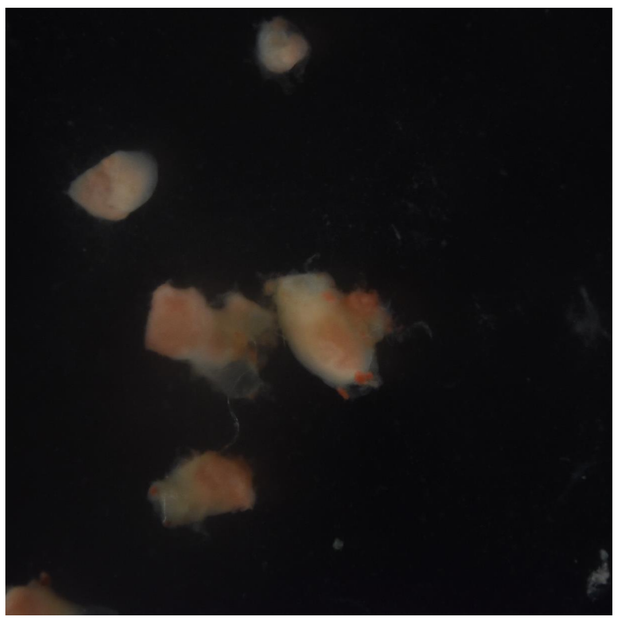
What did we learn? (days post ablation)
4 days: No significant difference
8 days: No significant difference
14 days: Animals subjected to notch inhibitor were **unhealthy or dead**.

Suggesting that a gentler approach of subjecting them to the notch inhibitor for only the initial days of nervous system regeneration would be more suitable for further study.

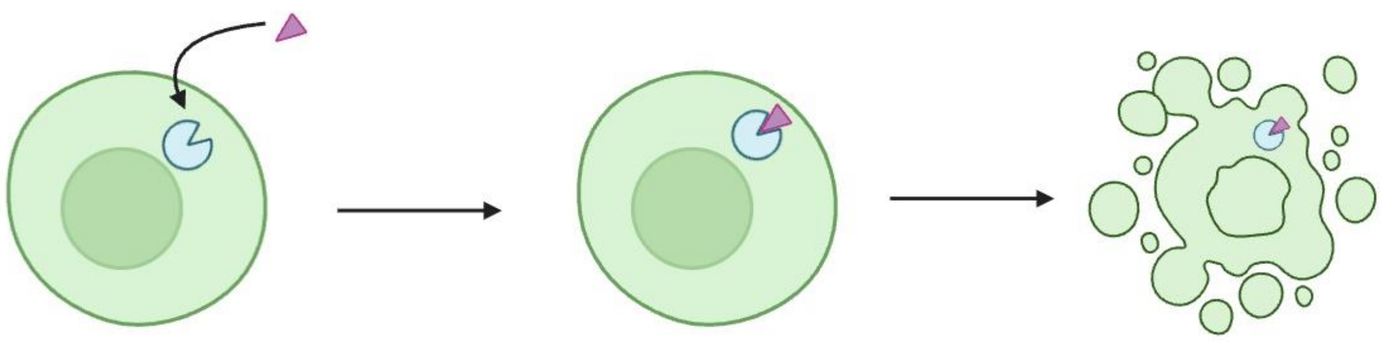

8 Days post ablation, no notch inhibitor


8 days post ablation, with notch inhibitor


Healthy animals


Unhealthy / Dead animals

A method enabling us to destroy the specific cells we want to whenever we want to.


NTR containing cell subjected to NFP

NTR reacts with NFP, triggering cell death

We used a regulatory element (**ElaV**) almost **exclusively active in the nervous cells** of *Nematostella.* This was used to control the expression of the two proteins **NTR-Cerulean and mOrange.** These proteins are therefore only expressed in the nervous cells of the animals.

NTR-Cerulean encodes for a two-part protein:
1. **Nitro reductase (NTR):** this protein triggers programmed **cell death** when subjected to **Nifurpirinol (NFP).**
2. **Cerulean:** fluorescent protein to show presence of **NTR**

mOrange encodes a fluorescent protein used to visualize the nervous stem under **fluorescent microscopy.**

At 6 weeks old, **half** the animals were **subjected to NFP** and the other half kept as control.

Post ablation
These groups were then divided into being subjected or not to the **notch inhibitor LY-411575.**
Inhibited and control groups were then **fixated** at **4 and 8 days** post ablation and analyzed using **confocal microscopy**.

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