Annotated Bibliography

Heather Betz – BIOL 463

1 Tanaka, E. M., Reddien, P. W. (2011) The cellular basis for animal regeneration. *Developmental cell* **21**:172-185.

This review article details the basics of animal regeneration, including the wide spread of organisms who have some sort of regenerative capability. It describes how regenerative capabilities are currently not very well understood, though now advances in cell fate tracking are starting to help advance the field. This article was used in my project to give a background on regeneration in organisms, as well as the organisms that are known to have regenerative capabilities.

2 Borgens, R. B. (1982) Mice regrow the tips of their foretoes. *Science* **217**:747–750.

This article details the extent of mammalian regeneration, specifically the capability of mammals to regenerate distal digit tips as opposed to full limbs. The methods used in the article are somewhat outdated, as it was published 36 years ago, but the background information that it provides remains relevant. It was used in my project to show the extend of mammalian regeneration, and how that differs from other species such as salamanders or fish.

3 King, B. L., Yin, V. P. (2016) A Conserved MicroRNA Regulatory Circuit Is Differentially Controlled during Limp/Appendage regeneration. *PLoS ONE* **11**(6): e0157106.

This article was the formative article that drives this question. In it, they identified 5 miRNA that were commonly upregulated and 5 that were commonly downregulated during forelimb/fin regeneration in axolotls (Ambystoma mexicanum), zebrafish (Danio rerio), and bichir (Polypterus senegalus). To examine the function of these miRNAs, they created a network of 1550 commonly differentially expressed miRNA between the species that had functional relationships to 11 blastema-associated gene, then found a gene network for common miRNA target genes for 3 of the differentially regulated genes. They then (with the most highly upregulated miRNA, miR-21) validated the expression of its known target genes. Overall, it revealed for the first time a conserved miRNA/mRNA network for blastema formation that is involved in the regeneration process.

4 Tornini, V. A., Poss, K. D. (2014) Keeping at arm's length during regeneration. *Developmental Cell* **29**:139-145.

This review article details the formation of blastemas and the developmental patterning that goes into their formation. It also describes recent advances in research on the regeneration process in amphibians and fish. In my project, this source provided me with information on what exactly a blastema is, and how its formation functions in the regeneration process as a whole.

5 Lim L. P. et al. (2005) Microarray analysis shows that some microRNAs downregulate large numbers of target mRNAs. *Nature* **433**:769–773

In this paper, the authors conducted an experiment in which miRNAs were transfected into human cells, and the mRNA expression profile determined using microarrays. They found that adding some miRNAs caused the expression profile of the cell to shift towards that of the specific tissue in which that miRNA is normally expressed, indicating that the miRNA can control or regulate a wide variety of genes. I used this source in my paper to discuss how miRNAs aid gene regulation by acting on many targets, showing how one miRNA addition may lead to a larger-scale phenotypic change.

6 Yan, L. X. et al. (2011) Knockdown of miR-21 in human breast cancer cell lines inhibits proliferation, in vitro migration and in vivo tumor growth. *Breast Cancer Res* **13**:R2.

This source was useful in demonstrating the potential function of miR-21 in humans, as well as how it has been linked to cancer. In this study, miR-21 was knocked down in human breast cancer cell lines using RNAi. They found that when miR-21 was knocked down, there was less cancerous proliferation, growth and migration, indicating that miR-21 plays an important role in cancer growth in humans. That, combined with the fact that it is highly upregulated in blastemas, gave rise to my idea that it could possibly aid with healing, as it appears to be linked to cell growth.

7 Holman, E. C. et al. (2012) Microarray Analysis of microRNA Expression during Axolotl Limb Regeneration. *PLoS ONE* **7(**9):e41804.

This article details the miRNA expression in axolotl limb regeneration. In this experiment, it was found that miR-21 was the most highly upregulated miRNA that was present in axolotl limb regeneration. It also discussed the sequence similarity between axolotl miR-21 and hsa-miR-21, including how they may very well be identical. I used this source in my project to show how miR-21 appears to be very similar between axolotl and humans, as well as to provide a technique for RT-PCR to quantify miR-21 expression.

8 Kirkland et al. (1998) The impact of surgical-site infections in the 1990s: attributable mortality, excess length of hospitalization, and extra costs. *Infect Control Hosp Epidemiol* **20**:725-30.

This source details the impact that surgical-site infections have in recovery in patients, as well as their indirect impacts on the hospital and healthcare. SSIs can lead to longer stays in hospital, incurring extra costs and decreasing hospital resource availability, as well as increasing wound severity and mortality rates. I used this source in my project to describe the potential impacts that miRNA treatment to increase wound healing would have, in that SSIs could be decreased due to faster and better healing.

9 Barbosa, T. M., Levy, S. B. (2011) The impact of antibiotic use on resistance development and persistence. *Drug Resist. Updat*. **3**:303-311

This source discusses how antibiotic use (and its overuse) leads to quicker development and persistence of bacterial antibiotic resistance. It also discusses how the development of antibiotic resistance is difficult to deal with as long as high amounts of antibiotic use continues, as more and more strains will continue to become resistant, even though resistance cannot be reduced by simply lowering antibiotic use. I used in my project to discuss the potential impacts of improving wound healing through miRNA, and how decreasing use of antibiotics through this miRNA use would be a important impact.

10 Wellcome Sanger Institute (2007) RT-PCR Protocol. Retrieved from ftp://ftp.sanger.ac.uk/pub/resources/mouse/sigtr/RTPCR.pdf

This source was used in my project to explain the RT-PCR process, and provide a sample protocol that could be used in my experiment. I included RT-PCR in my experiment to quantify the levels of miR-21 present in control groups, as well as to monitor the functionality of my siRNA and control for TRIS.

11 Merlo, G., Altruda, F., Poli, V. (2012) Mice as Experimental Organisms. *eLS*, doi: 10.1002/9780470015902.a0002029.pub2

This review article describes the utility of mice as experimental models, including why they may be chosen and possible disadvantages to using them. I used it in my experiment to describe why I chose mice as a potential model organism, specifically because they have similar genome structure and development to humans.

12 Mulisa, E., Asres, K., Engidawork, E. (2015) Evaluation of wound healing and anti-inflammatory activity of the rhizomes of Rumex abyssinicus J. (Plygonaceae) in mice. *BMC complementary and Alternative Medicine* **15**:341

This paper was used in my project for the techniques it described, though its topic of investigation was dissimilar to my own. It investigated the quality of wound healing in mice when the rhizomes of Rumex abyssinicus was added. My experiment deals also with wound healing, and so the techniques of wound creation, and the evaluation of wound healing used in this paper would be applicable to mine as well. From it, I learned how wound creation is performed, as well as how tensile strength measurement is performed, and how it is a good measure of wound healing.

13 Thermo Fisher Scientific. Overview of RNAi and requirements for a typical RNAi experiment. Retrieved from https://www.thermofisher.com/ca/en/home/references/ambion-tech-support/rnai-sirna/tech-notes/rnai-how-to-for-new-users.html

This source details the process of RNAi production, and how it functions in the cell to knock down its RNA target. I used this source to discuss how RNAi works, and learn how RNAi would be created, as I needed to use it in my experiment.

Figure Sources

1.Thermo Fisher Scientific. The Basics: In Vitro Translation. Retrieved from https://www.thermofisher.com/ca/en/home/references/ambion-tech-support/large-scale-transcription/general-articles/the-basics-in-vitrotranslation.html

This source is a figure on the basics of in vitro translation, a technique I used in my experiment to translate amplified axolotl miR-21 DNA into transcript to be injected into wounds. I used this figure to more easily describe the in vitro translation process.

3. Thermo Fisher Scientific. Overview of RNAi and requirements for a typical RNAi experiment. Retrieved from https://www.thermofisher.com/ca/en/home/references/ambion-tech-support/rnai-sirna/tech-notes/rnai-how-to-for-new-users.html

This source is a figure describing RNAi, and how siRNAs are added into the cell in order to knock down their RNA targets. I used RNAi in my experiment to knock down miR-21. This figure was included to give a more easily understood visual representation of how RNAi works to knock this down.

2. Mulisa, E., Asres, K., Engidawork, E. (2015) Evaluation of wound healing and anti-inflammatory activity of the rhizomes of Rumex abyssinicus J. (Plygonaceae) in mice. BMC complementary and Alternative Medicine 15:341

This image visually shows the wound and tensile strength measurement that I used. It displays exactly where the wound would be created, as well as how tensile strength is performed, to further understanding for the reader on the techniques used.