Section IV: Discussion

The significant increase in the rate of regeneration of *Schmidtea mediterranea* after LiCl and bFGF were exogenously applied suggests that both factors, whether used separately or in combination, work synergistically to modulate the Wnt signaling pathway, thereby inducing regeneration. Since the Wnt signaling pathway is similar across many species, it may also help human dental pulp stem cells (hDPSCs) grow and develop more effectively, as it has been shown to do in the past (Angelova Volponi et al., 2018). The method for regeneration could thus pose a promising solution to dental pulp necrosis.

One of the key objectives of this study was to determine whether bFGF, when used alongside LiCl, could amplify the regenerative response in *Schmidtea mediterranea*. The data supports this hypothesis, since worms treated with both factors exhibited a significantly higher regeneration rate than those exposed to LiCl alone, as evidenced by blastema growth rate. The statistical analysis confirms this relationship; the Student's t-test yielded a p-value of 0.01, indicating strong statistical significance.

A major strength of this study is its use of Schmidtea mediterranea as a model organism. Due to its regenerative abilities, such as the ability to regenerate from even 1/279th of a fragment, and the presence of signaling pathways such as the Wnt/β-catenin pathway, *Schmidtea mediterranea* is an excellent organism to study the process of tissue regeneration (Lobo et al., 2012).

That said, no study is without its limitations. One potential limitation is the inherent variability in regeneration rates among individual planarians. To account for this, repeated trials were conducted to ensure a sufficient sample size to minimise random variation. Additionally, while there is strong evidence that Wnt pathway modulation plays a role, we have not directly measured Wnt activity in our samples. Future studies should include direct assays of Wnt pathway activation, such as β-catenin localization or expression analysis of downstream targets.

Despite these challenges, our research builds upon and extends prior work in significant ways. Previous studies have established that LiCl enhances Wnt signaling and promotes regeneration (Ishimoto et al., 2015), while bFGF has been widely studied for its role in tissue repair (ten Berge et al., 2008). However, this study is among the first to demonstrate that their combined application yields a more substantial regenerative effect. This synergy could provide a more effective strategy for tissue engineering and stem cell-based therapies.

Future Research

Moving forward, this study creates many new avenues for future research. A key next step is to investigate whether the regenerative effects observed in *Schmidtea mediterranea* can be replicated in human dental pulp stem cells (hDPSCs). Since the Wnt signaling pathway is similar across many species, it may also help hDPSCs grow and develop more effectively. Future experiments should explore the optimal concentrations and timing of LiCl and bFGF application in mammalian cells. Additionally, investigating potential side effects or unintended consequences of prolonged Wnt signaling activation will be crucial for ensuring the safety of any clinical applications. With further development, this approach could significantly improve treatments for dental injuries, pulp necrosis, and even broader applications in regenerative medicine. Furthermore, since increased Wnt activation has been shown to induce regeneration, it may be beneficial to explore inhibiting this pathway in cancer patients by suppressing bFGF production.

Section V: Conclusion

The study aimed to determine whether the combined application of lithium chloride (LiCl) and basic fibroblast growth factors (bFGF) could enhance regeneration in *Schmidtea mediterranea* by modulating the Wnt signaling pathway. Using a controlled experimental approach, regeneration rates were observed and quantified in planarians treated with LiCl alone versus those treated with both LiCl

and bFGF. The findings suggested that these two factors worked synergistically to increase the rate of regeneration. The results highlight a potential strategy for enhancing tissue regeneration beyond model organisms. Given the evolutionary conservation of Wnt signaling, this approach may have valuable applications in regenerative medicine, particularly in promoting the proliferation and differentiation of human dental pulp stem cells (hDPSCs).

Regenerative medicine is constantly evolving, with the potential to significantly improve how damaged tissues are repaired. The study adds to that progress by identifying a way to speed up regeneration, which could have meaningful applications in medical treatments. If future studies show that LiCl and bFGF work similarly in human cells, this approach could open doors to major advancements in dental and regenerative medicine, overall changing how tissue repair is approached in clinical settings.