CHAPTER VII

CONCLUSION AND FUTURE PERSPECTIVES

7.1 Conclusion

This study showed that designed custom peptides derived from snake venom neurotrophin molecules can protect PC-12 cells (*in vitro*) and *C. elegans* (*in vivo*) from PT-induced neurotoxicity. The custom peptides counteract PT-induced neurotoxicity by thwarting excessive ROS production, oxidative stress, MMP, and premature apoptotic death. Our findings provide a greater understanding of the altered expression of proteins involved in the neurodegeneration pathways in the PT-induced PD model of PC-12 cells and *C. elegans* and how the expression of our custom peptides can be used in treatments.

The neuroprotective activity of custom peptides against PT-induced chemotaxis behavioral defects, DAergic neurodegeneration, α-synuclein accumulation, and reduced life span may be related to their antioxidant and antiapoptotic properties. The PT group of worms displayed maximum variation in differential gene expression compared to the untreated (control) group of *C. elegans*. In contrast, the custom peptides pre-treatment group showed minimal variation compared to the untreated (control) group, indicating normalization or restoration of altered gene/protein dynamics. This study highlights custom peptides' therapeutic importance in reducing neurodegeneration against toxic chemicals.

Further study shows that in the PT group of *C. elegans*, the downregulated miRNAs target genes involved in the developmental process during embryogenesis; genes regulate the longevity of *C. elegans*, α-synuclein degradation, and oxidative stress response. Moreover, upregulated miRNAs target genes in the PT group involved in apoptotic pathways, decreased lifespan, innate immune response, and metabolic pathways are reported to increase the progression of PD.

Using the tested custom peptides as a drug prototype exclusively depends upon their non-toxic nature in preclinical studies. The acute toxicity studies in mice models showed that the peptide is devoid of toxicity in mice at a dose that is 100 times higher than its therapeutic dose determined in *C. elegans*. Further, it has no detrimental effect on the biochemical parameters of blood parameters and vital organs, suggesting the safety of the peptides for the development of drug prototypes. Furthermore, the reduced concentration of inflammatory mediators (TNF-α, IL-6, IL-1β) in the CPs-treated mice, compared to control (1X PBS-treated) mice, diminished the risk of inflammatory response post-treatment with peptides.

7.2 Future perspectives

The findings of the present study provide a fundamental basis for developing new agents that specifically target neurodegenerative diseases, especially PD, and for the successful development of safe drug prototypes through *in vivo* pharmacokinetics, and pharmacodynamics and encourage further in-depth investigation through bioavailability studies. Two uncharacterized miRNAs, cel-miR-8207-3p and cel-miR-57-3p, are upregulated with PT treatment and downregulated with mouse 2.5 S-NGF and custom peptides pre-treatment. The function of these two miRNAs is unknown and needs to be explored.

7.3 Limitations of the study

While this study provides promising evidence that custom peptides derived from snake venom neurotrophin molecules can offer neuroprotective effects, certain limitations should be acknowledged:

Limited *in vivo* Models: Although the study demonstrated neuroprotective effects in *C. elegans*, additional studies using more advanced mammalian models of PD are necessary to validate the therapeutic potential of these peptides in higher organisms.

Long-Term Effects: The long-term efficacy and safety of the custom peptides were not extensively studied, particularly regarding prolonged treatment durations. Potential side effects, bioaccumulation, or unforeseen consequences from long-term use remain unexplored.

Bioavailability and Pharmacokinetics: While *in vitro* and *in vivo* results are promising, comprehensive pharmacokinetics and bioavailability studies in larger animal models are required to understand how effectively these peptides can be absorbed, distributed, metabolized, and excreted in complex biological systems.

Dose-Response Relationship: Although the acute toxicity study indicated a lack of toxicity, further research is necessary to explore the precise dose-response relationship for therapeutic efficacy and potential side effects in humans.

Unknown miRNA Functions: The study identified two uncharacterized miRNAs (cel-miR-8207-3p and cel-miR-57-3p) affected by the treatment, but their specific roles and mechanisms in neurodegeneration and peptide interaction remain unexplored, which could limit the understanding of the complete molecular effects of the peptides.

These limitations suggest that while this research represents an important step toward developing new therapies for PD, further investigation is required to fully explore the therapeutic potential and safety of these custom peptides.