Student Free Design Activities (One Health Collaborative Training)
報告書 Report

報告者 [Reporter]

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活動報告 [Activity Report]
※活動内容が判る様な写真や図表を加えて下さい。/ Provide photos, tables and figures that clearly show the activities during the period.

タイトル [Course Title]	Study on ion channel-linked cardiovascular diseases by Voltage-clamp fluorometry
実施期間 [Periods]	08/12/2024 - 08/30/2024
共同実施者 [Other participants]	N/A
言語 [Language]	English
実施場所 [Location]	Linköping University, Sweden

申請時計画の実施報告 [Report how you carried out your plan in the application form]

Did you follow the schedule you initially planned? Did you get the outcome(s) you expected? Please describe what you did during the activity period in detail.

I tried to adhere to the schedule I initially planned, with only minor adjustments that did not significantly impact the overall flow of my training in Linköping University, Sweden. Each activity was carried out as scheduled, allowing me to achieve the expected outcomes.

Detailed Description of Activities

Mastering Voltage-Clamp Fluorometry (VCF) Technique: The whole technique is divided into a few steps. I learnt all the steps from mRNA microinjection into oocytes to the data analysis and interpretation.

Oocytes Collection: VCF is usually done by using oocytes from *Xenopus Laevis* (African Clawed Frogs). After we received oocyte batch from a single frog, we sorted good quality oocytes from the batch for the microinjection.

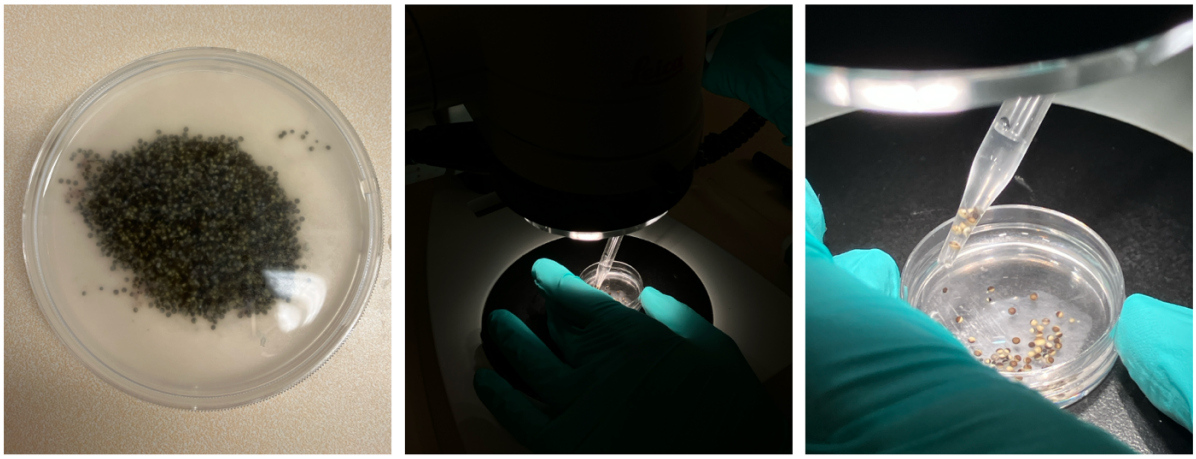

Figure 1: Sorting of healthy oocytes for mRNA microinjection.

mRNA microinjection: mRNA microinjection is a technique used to introduce messenger RNA (mRNA) directly into cells or embryos. This method is widely used in molecular biology and developmental biology to study gene function, protein expression, and cellular processes. I learnt how to perform mRNA microinjection. We injected synthesized mRNA encoding the Hyperpolarization-activated Cyclic Nucleotide-gated (HCN1) channel directly into the cytoplasm of Xenopus oocytes. This technique enables the expression of the HCN1 protein in the oocyte membrane, allowing to study its electrophysiological properties.

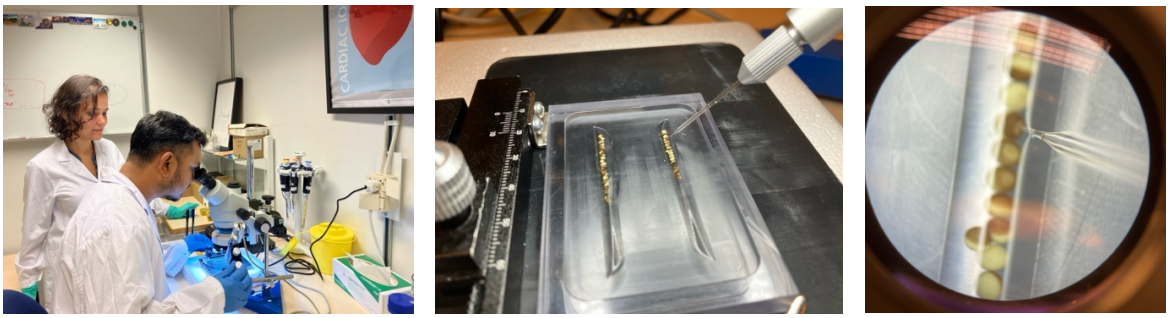

Figure 2: (Left) Veronika, a PhD student, is teaching me how to perform mRNA microinjection, (Middle) Micropipette is inserted into the oocyte for mRNA injection, (Right) Microscopic view of microinjection.

Data Recording: After 2/3 days of microinjection, we checked whether HCN1 channel is expressed in oocytes. Once it is expressed, we labeled the oocyte with Fluorophore dye probe by dipping the oocyte into the dye for about 20 minutes. After that, we measured and recorded the current and fluorescence emission from the cell.

Figure 3: (From the left) The VCF machine full set-up, Data recording is going on, The two electrodes are inserted into the oocyte for data recording, The recorded current from an oocyte.

Data Analysis: After I obtained data, I learnt how to analyze and interpret the data.

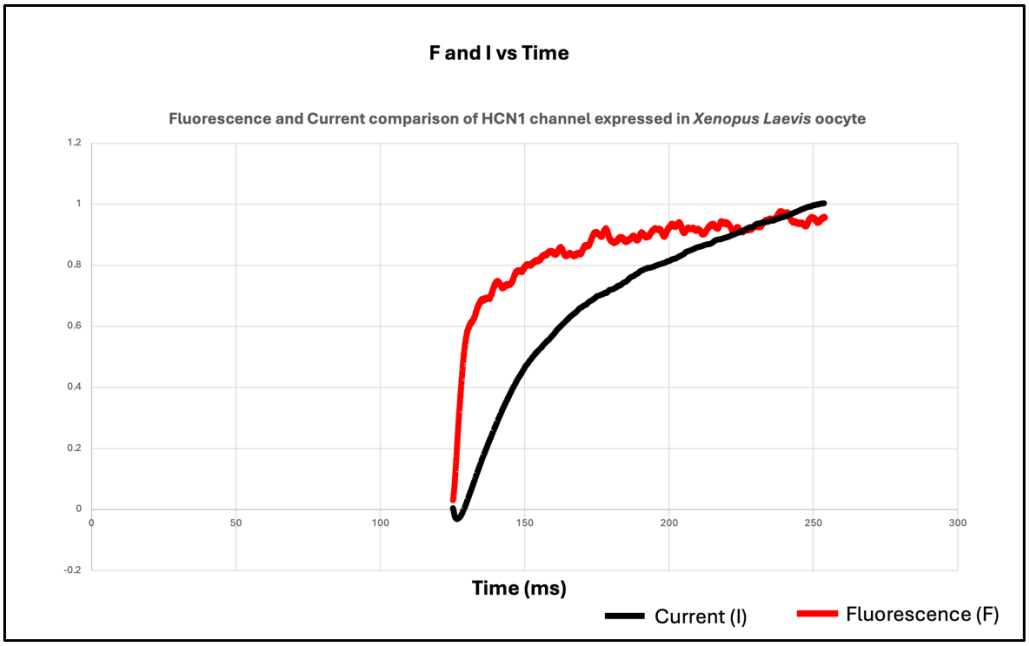

Figure 4: A representative analyzed data showing the difference between fluorescence and current of HCN1 channel.

Discussion: I also had a great discussion with the professor and his team about outcome I had through the training, and about how I can apply the earned knowledge and experience into my current research. My current research is on OTOP1 proton channel in brown adipose tissue. Since there is no VCF done in OTOP1 proton channel so far, it would be a great initiative if I could start working on it. Therefore, professor was illustrating how I can fit VCF technique into my PhD research. Additionally, we discussed about the possibility of collaborative work in future and also about the postdoc opportunity for me in his lab in the future. Overall, it was a successful discussion indeed!

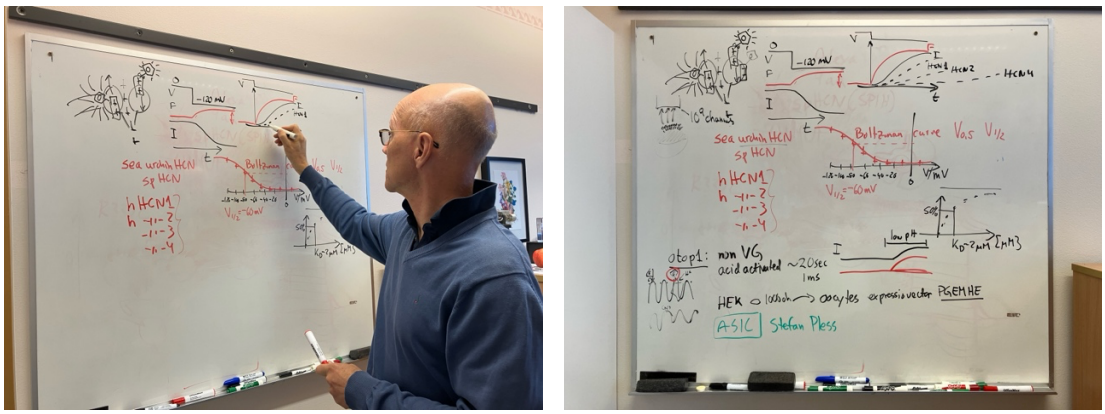

Figure 5: Professor is illustrating the possible way how I can fit VCF technique in my current research.

目的達成状況報告 [Report how you achieved your goal/objectives listed in the application form]

Did you achieve all the goals you initially planned? If not, please describe why you failed to fulfill your objectives.

I designed my training with an objective of academic learning. I was also focused to intensify the inter-disciplinary understandings of One health, to recognize diverse perspectives, research methodologies, and scientific cultures in Sweden, and to create a global scientific research network for further research opportunities. Through my three-week training in Linköping University, I believe I achieved these objectives effectively.

Learning VCF Technique: One of the core objectives of my training was to learn the Voltage-clamp Fluorometry (VCF) technique, a sophisticated method crucial for studying the conformational changes in ion channels. This technique is essential in understanding the structural dynamics that can lead to cardiac diseases. The hands-on experience I gained, coupled with expert guidance from the faculty, enabled me to master the technique. I now feel confident in applying this method to my research, particularly in exploring how these conformational changes can impact cardiac function.

Enhancing Interdisciplinary Understanding of One Health: Another key goal was to deepen my interdisciplinary understanding of the One Health concept, which recognizes the interconnectedness of human, animal, and environmental health. During my time at Linköping University, I engaged with researchers from diverse fields, which broadened my perspective on how different scientific disciplines can collaborate to address complex health challenges. This exposure has significantly enhanced my ability to integrate these diverse perspectives into my own research, making my approach more holistic and comprehensive.

Recognizing Diverse Research Methodologies and Scientific Cultures: My time in Sweden provided a unique opportunity to observe and understand the diverse research methodologies and scientific cultures that exist in a different academic environment. I was able to compare and contrast these approaches with those I am familiar with, leading to a richer understanding of how scientific research can be conducted effectively across different contexts. This experience has not only broadened my methodological toolkit but also taught me the value of flexibility and adaptability in research.

Building a Global Scientific Research Network: A significant aspect of my training was networking with other researchers, both within and outside of Linköping University. I actively participated in group discussions, collaborative projects, and informal exchanges that allowed me to establish connections with scientists from various disciplines. The department I visited was a department for human medicine. Therefore, I met various scientists who are working on the human medicine research. I also met some professors from the universities of other countries who came for a collaboration work with Linköping University. Overall, these interactions have laid the groundwork for future collaborative research opportunities, and I now have a global network of colleagues with whom I can exchange ideas and potentially work on joint projects in the future.

Reflecting on the Impact of the Training: Overall, my training at Linköping University was a transformative experience that not only equipped me with new technical skills but also enriched my understanding of the broader scientific landscape. I am returning with a deeper knowledge of the Voltage-clamp Fluorometry technique, a more integrated perspective on health research, and valuable connections that will support my future endeavors. This experience has undoubtedly advanced my research capabilities and opened up new avenues for collaboration and discovery.

One Health Approach実践報告 [Report how your activity could link to One Health Approach]

Did you have a chance to experience One Health approach (collaboration with people from other academic areas)? Please describe some of the examples of One Health approach you implemented in your activity. Otherwise, explain the possibility(ies) how you could link this activity to One Health approach for your future.

During my training at Linköping University, I had several opportunities to observe and engage in interdisciplinary collaborations that align with the One Health approach. One Health emphasizes the interconnectedness of human, animal, and environmental health, and this perspective was evident in the projects and discussions I encountered. One of my main focuses during my time at Linköping University is the study of ion channels using VCF technique, specifically the HCN1 channel, which plays a crucial role in cardiovascular diseases (CDVs). Recent research highlights the significance of ion channel mutations in causing sudden cardiac deaths in animals, such as English Springer Spaniel dogs with Long QT Syndrome. Other animals, including cats, horses, and cattle, have also shown similar cardiac disorders due to ion channel dysfunctions. Moreover, some CVDs have zoonotic potential, meaning they can be transmitted between animals and humans. For instance, Brucella endocarditis is known to be transmitted from animals to humans and is particularly common in regions with large livestock populations. The VCF technique I learnt, can be applied to study conformational changes in various voltage-gated ion channels. Ion channels regulate processes such as neural communication, muscle contraction, and nerve conduction, making them attractive targets for drug discovery. By utilizing the VCF technique, my future research could contribute to the development of new treatments for both human and animal diseases, further advancing the One Health agenda. Furthermore, environmental factors, such as pollutants and toxins, also contribute to the development of cardiac diseases in both humans and animals, which emphasizes the need for integrated environmental health strategies. The risks posed by climate change—such as extreme weather events—further exacerbate cardiovascular risks, increasing the incidence of heart attacks and strokes. By discussing with all the researchers in the lab, I could explore how environmental stressors—such as pollution or climate change—might influence cardiac health in both humans and animals. The diverse network I built during my training at Linköping University has also opened doors for future interdisciplinary collaborations. I engaged in a discussion with colleagues to explore how the techniques I learned, such as VCF, can be applied to other species, further deepening my integration of the One Health framework in my research.

備考 [Remarks]

This SFDA training significantly enhanced my overall understanding of the One Health approach, fostering interdisciplinary collaboration and expanding my knowledge of its application to cardiovascular health and research.