

Short-Stay Study Abroad Program of SOKENDAI

Completion Report

2017.8.30 – 2017.9.13

34th Microelectrode Techniques for Cell Physiology Workshop
The Marine Biological Association of the United Kingdom
Plymouth, UK

2017.9.14 – 2017.10.04

Visiting study at Prof. Gregor Belušič laboratory
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Motivation and Preparation

To acquire the necessary skills for the electrophysiological experiments of my PhD study, our collaborators Prof. Gregor Belušič (University of Ljubljana) and the late Prof. Matti Weckström (University of Oulu) strongly recommended me to attend the workshop of microelectrode techniques for cell physiology in Plymouth, UK. It is one of the best workshops that provides intensive practical experience of microelectrode and optical measurements for cell physiology. It is my honor to be offered a place at this acclaimed workshop. The Plymouth workshop is highly competitive. Every year only 20 places are available. In fact, this was my second application for the Plymouth workshop. I was not selected in the first application two years ago. After I was noticed that I got a place, I discussed the travel plan with Prof. Kentaro Arikawa (SOKENDAI) and Prof. Gregor Belušič (University of Ljubljana). Prof. Gregor Belušič kindly invited me to visit University of Ljubljana and to work in his lab at after the workshop.

For applying the travel grants, I requested an acceptance letter from the workshop and an invitation letter from Prof. Belušič. In addition to the financial support from the short-staying study abroad program that was held by SOKENDAI (400,000 yen), I also applied our departmental travel grant (200,000 yen) and the workshop bursary (750 GBP). In my case, no VISA is required for visiting Europe. One month before leaving from Japan, I finalized the schedule and booked the flights (about 250,000 yen), coaches (about 25,000 yen) and accommodations (about 200,000 yen), and paid the workshop fee (1,500 GBP) and travel insurances (about 20,000 yen). The accommodation in Plymouth and in Ljubljana was booked by the workshop organizer and Prof. Belušič, respectively.

On the other hand, all participants got a hand book from the Plymouth workshop. We needed to finish the pre-course reading before attending the workshop. In addition, I planned to work on my PhD topic using my study material when I visited the lab of Prof. Gregor Belušič. I sent the pupae of the butterfly to his lab one month before leaving Japan.

Microelectrode Techniques for Cell Physiology Workshop

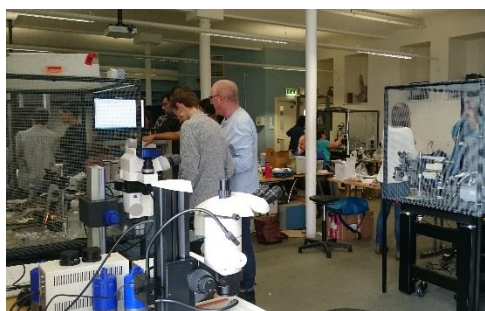


In the first two weeks, I attended the microelectrode techniques for cell physiology workshop in Plymouth. The workshop was organized by the Marine Biological Association of the United Kingdom (MBA) since 1984 and now in its 34th year. MBA is a professional body for marine biologists with some 1200 members world-wide. The MBA has earned an international reputation for excellence and innovation in research, by the resident scientific staff and visiting research workers, including several Nobel laureates. Among them, Alan Hodgkin and Andrew Huxley received the Nobel Prize for Physiology or



Medicine in 1963 for the discovery of the mechanism of action potentials in squid giant axons. They carried out this ground-breaking experiment at the Laboratory of MBA in Plymouth. It is one of the most productive and influential researches in the history of neurobiology. Every year, the MBA runs several international training workshops and advanced courses including the one I attended.

The workshop provided 25 formal lectures and 10 days of hands-on practical sessions and demonstrations, which were given by teachers from the institute of MBA as well as several inviting expert instructors. Most of students are in the last year of doctoral courses. In other words, most participants have ample experience in certain electrophysiological experiments. The workshop was especially focused on the basic theories of different microelectrode techniques. In addition to the daily lecture, the hands-on training of the



techniques was one of the core idea of the workshop. Each student selected three techniques and practiced them via one-to-one tuition during the course. One of the great things in this workshop was that the experimental space was open. Therefore, we could join other groups freely and easily. I have learned lots of new knowledge and techniques through these intensive lectures and practices.

The topics taught in morning lectures were:

- Electronics
- Patch clamp
- Whole cell clamp
- Extracellular recording
- Voltage clamp
- Slice recording
- Microelectrode injection techniques
- Photolysis
- Fluorescent indicators
- Bilayer recording
- Amperometry
- Capacitance measurement,
- Data acquisition and analysis
- Microscopy
- Transgene and viral cell labelling
- Optogenetics



The topics taught in afternoon practical sessions were: (3 blocks of 3-day experiments)

- Patch clamp
- Slice recording
- Two electrodes voltage clamp
- Discontinuous single electrodes voltage clamp
- Ion-selective microelectrodes
- fluorescent indicators
- Cell injection and labeling
- *Drosophila* neuromuscular junction and optogenetics



Every day the teaching started from 9am and ended around 7pm. We usually went to dinner around Plymouth's historical Barbican area with other classmates. We found a high quality Japanese restaurant (about 20 GBP for a meal) and a cheap Chinese restaurant (about 10 GBP for a meal). Therefore, in addition to traditional fish and chips for dinner, we preferred having Chinese and Japanese cuisine. In addition, the breakfast and hot buffet lunch provided by the workshop were fantastic. It was very interesting to have a talk with other participants during lunch and dinner. In addition to research, we talked everything including foods, weather, culture, school system, religion, sports, etc., since we all came from different countries.



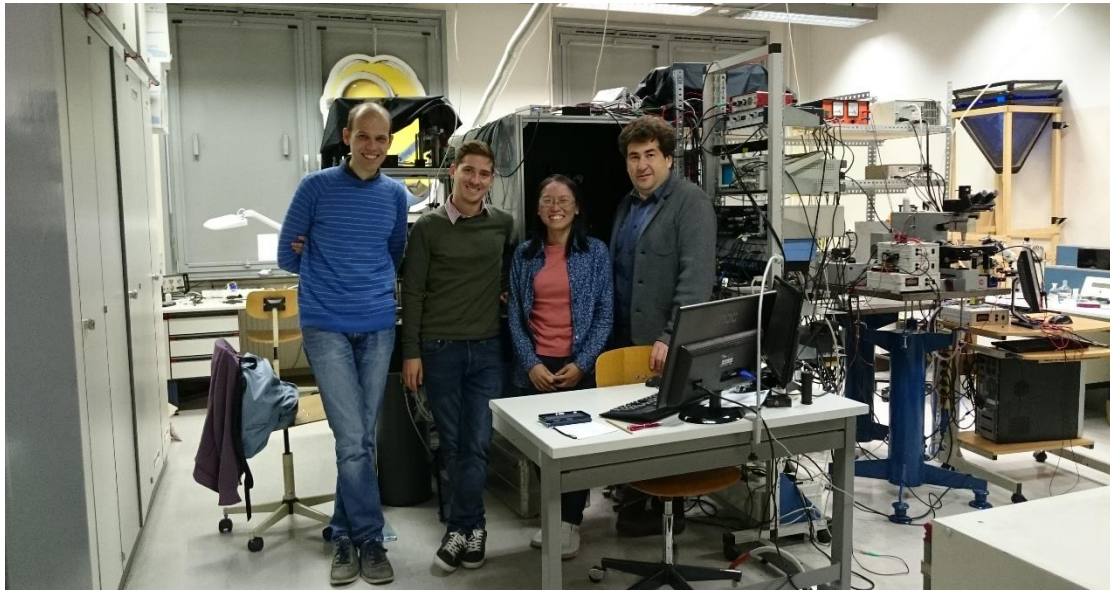
Plymouth is a lovely place; however, we didn't have enough free time for excursion. Only on September 7th morning, after the first lecture, the workshop organizer took us to visit National Marine Aquarium. The schedule of workshop was tight as mentioned above from Monday to Saturday, only Sunday was free. Unfortunately, during our stay, we always had heavy rain on Sundays. We only could spend our time in the evening after the course to have a brief exploration around the institute.



After the Plymouth workshop, I had a one-day stop in Exeter. I visited our collaborator, Dr. Natalie Hempel, in the University of Exeter. She kindly introduced her laboratory and the campus to me. I also had a nice talk with her students and post-docs.

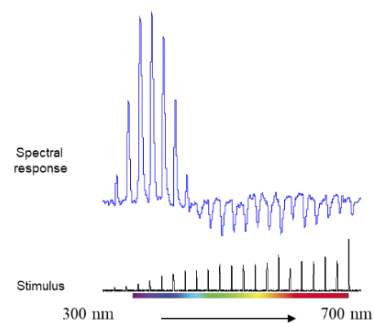


Visiting study at Prof. Gregor Belušič laboratory



In the following three weeks, I worked in the lab of Prof. Gregor Belušič at University of Ljubljana. Prof. Gregor Belušič is one of the leading experts in the field of comparative physiology of insect visual systems, especially on electrical properties of flies' photoreceptors and visual neurons. His lab is well equipped with several advanced electrophysiological and optical measurements including voltage clamp, current clamp and single electrode switching clamp. These techniques are what I need to apply in my PhD project. In fact, I have started learning the high-frequency single electrode switch clamp with Prof. Belušič during his short visits in Hayama in 2015. However, the time was far too limited to thoroughly understand the difficult matter of the switch clamp theory. Prof. Belušič was keen to invite me to his lab to learn and practice advanced techniques with him.

We were interested the hyperpolarizing responses in the lamina (the first neuropil of optic lobe) of the butterfly *Papilio xuthus*. In addition of lamina monopolar cells (LMCs), we frequently encountered spectral opponent photoreceptors that exhibited negative-going responses. Spectral opponent responses, i.e. positive at some wavelengths and negative at others, have been observed at the level of photoreceptors in some butterflies. My PhD



project aims to explain its mechanism and thus “histamine hypothesis” has been proposed. Prof. Belušič has kindly designed a new stimulation device which is suitable for asking my research question. We recorded and analyzed the physiological profile of LMCs and spectral opponent photoreceptors in the *Papilio* lamina using several electrophysiological techniques. After experiment, I analyzed the recording data in the evening. Through the intensive practice, I have developed skills and got abundant preliminary results that strongly push my PhD study progress.

In Slovenia, people usually start working before 9am and then finish around 4pm. Different to Japan, it is very rare if someone stay at the lab after 6pm. Every lunch I went to school cafeteria with other lab members. Similar as the cafeteria at Hayama campus, there are 5 menu of main dishes for chosen (4 Euro/5 Euro without/with salad and soup). I cooked dinner at the guestroom every day. I stayed at the guestroom (20 Euro per night) of the Forestry department, University of Ljubljana, which is within a short walking distance of the lab. The guestroom provided most of living groceries. In general, the daily expense in Slovenia is cheaper a bit compared to Japan. Language used during the school and on a daily basis in Slovenia is Slovenian. However, most of people here can speak English very well. Instead, the things written in Slovenian (menu, road sign, etc.) was much difficult to read and understand for me.

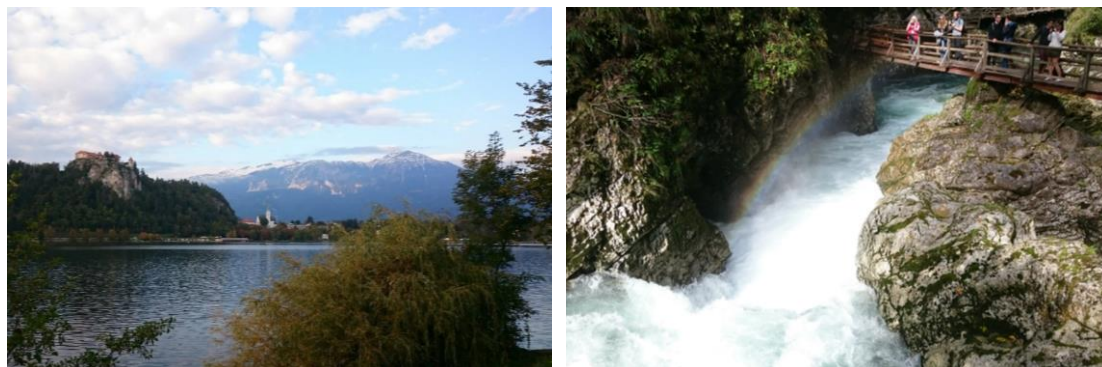


One difficulty was Internet service. In Europe, many schools use “Eduroam” (education roaming) which is a free wireless (WiFi) network system. It allows all school members from participating institutions to obtain Internet connectivity across campus and when visiting other participating institutions. However, my department (ESB of SOKENDAI) is not a member of Eduroam. I could not use WiFi at the guestroom and in the campus. It was very inconvenient. I had met the same problem several times when I attended international conferences in abroad. Since most of education institutes have joined Eduroam in the world, they do not provide additional WiFi network for attendants. I hope our institute could think about this issue seriously.

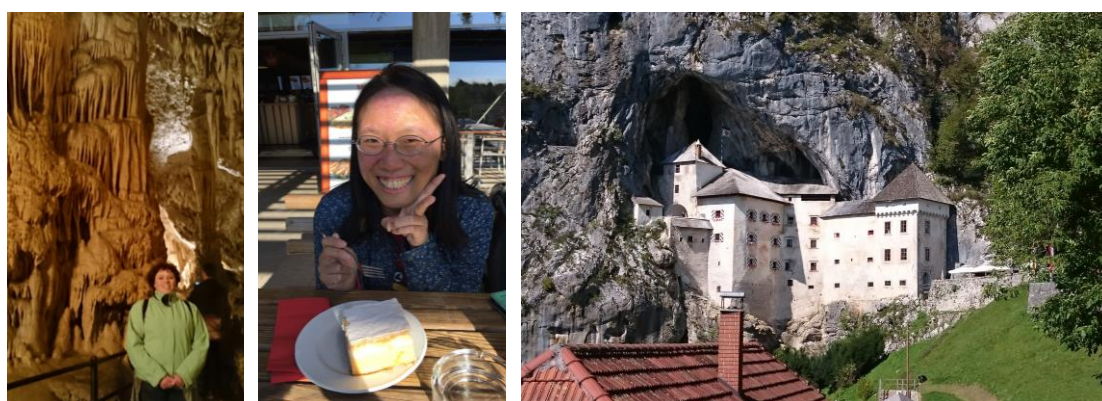
In the weekend during my stay in Slovenia, I visited some famous attractions. In the first weekend, I visited the city center of Ljubljana, within 30 minutes walking distance, which is a beautiful and peaceful place.



I went to Bled, the northern Slovenia, in the second weekend. In addition to the famous Bled Lake, I visited Vintgar gorge with amazing scenes alongside Radovna River.



In the last weekend, I hanged out with my friend, a previous lab member in Prof. Belušič lab, to the southern Slovenia. We visited Postojna Cave, the largest limestone cave in Slovenia, and Predjama Castle with a unique construction: half cave half castle! She also introduced me many traditional Slovenia food. It was really a nice trip and special experience.



Though during my stay in Slovenia, I met the worst weather of September in recent years, I enjoyed the life here a lot.

I really appreciated SOKENDAI provided the short-staying study abroad program, which is intended and beneficial for motivated and open-minded PhD students. It helped me to push myself into the academic community through sharing research and learning new knowledge with leading scientists. Of course, certain level of language ability is very important for efficient discussion in abroad. The travel not only pushed my academic progress in PhD study but also mentally. Exposing to international community is very important for PhD student to broaden their horizons by developing the confidence, feeling of independence and perspectives for future career. I strongly encourage every PhD student at SOKENDAI to experience different academic cultures at institutes outside Japan. If you have a plan to go abroad, do not forget SOKENDAI has this program for your financial assistant.