

This report should be submitted within 2 weeks after you return to Japan.

(Abroad • Domestic) Internship report form (Student)

_____ (Year/Month/Day)

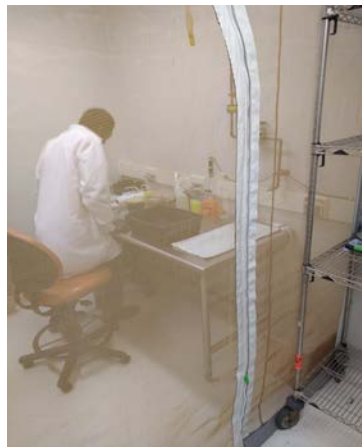
Name	Alex Gaithuma
Laboratory	Division of collaboration and Education, CZC
Year (Grade)	D3
Internship institution	Yale School of Public Health, Yale University, USA
Internship period	Internship period: 11/03/2017 - 12/3/2017 (Departure Date from Sapporo: MM/DD/YYYY, Arrival Date in Sapporo: MM/DD/YYYY)
Purpose	•To learn modern molecular techniques used in tsetse fly genomics and epidemiology research in order to improve the quality of my Ph.D. research output •To acquire new skills important for a future career in trypanosomiasis research •To establish a career network with researchers and scientists in the USA for future postdoctoral study prospects and research collaborations

- The reason why you chose this institute

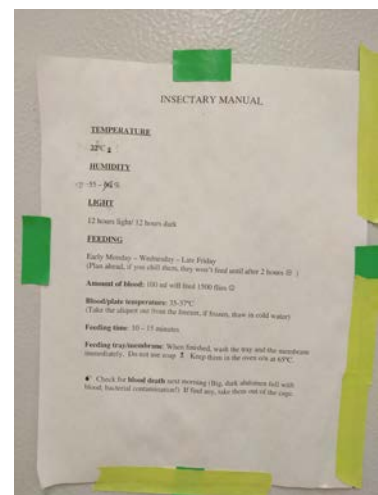
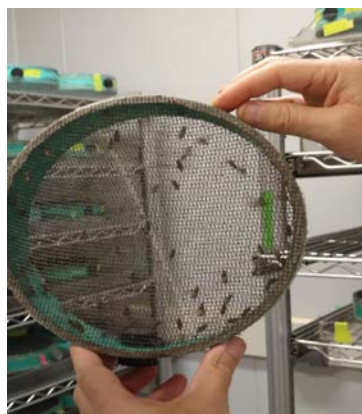
Yale University is a top university in the world ranked among the top 15 universities in the world and The Yale School of Public Health is rated the 3rd best school in the USA, for its doctoral program in Epidemiology. Prof. Askoy a professor of Epidemiology in The Yale School of Public Health and was among the project leadership and the conception team that published the first genome of the tsetse fly *Glossina morsitans* in 2014 and is the corresponding author of the paper. Her laboratory has the only tsetse breeding colony in North America is involved in an international training program to expand research capacity in tsetse-transmitted diseases in East Africa. She is also the editor in chief of PLOS Neglected Tropical Diseases. I am interested in her laboratory because it has a lot of resources, capacity and various people working in different fields of biology. It is, therefore, a laboratory with a multidisciplinary research experience.

- Result of the activity

1. Insectary Training: Undertook biosafety training on how to maintain a tsetse fly colony, feeding and handling of flies during experiments, safety and containment measures in the insectary.



Set 1: Insectary fly containment barriers, safety and emergency measures in the Insectary.



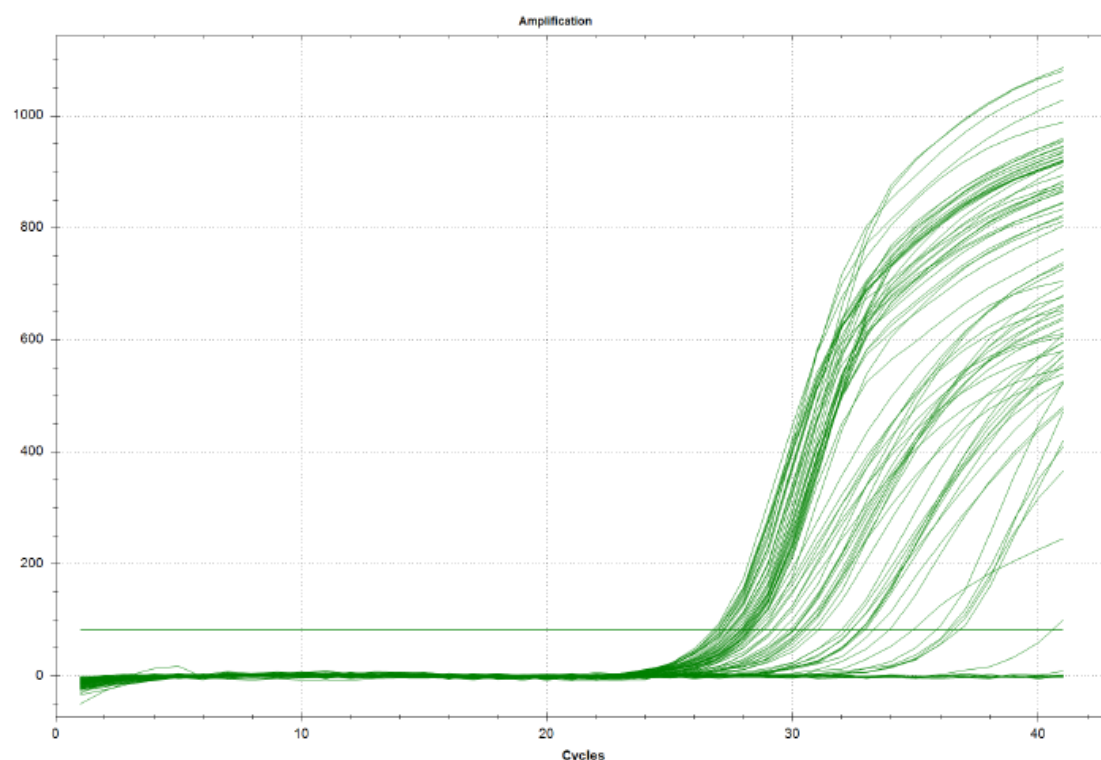
Set 2: Fly maintenance, feeding and propagation.

2. Tsetse experiments training: Differentiation and sorting of males from females, dissection of a tsetse fly and extraction of the gut, reproductive organs and other internal organs. Sample preparation like crushing of whole flies/tissues and extraction of midgut, bacterium and reproductive organs (ovaries), and antennae for DNA or RNA extraction.



Set 3: Inactivation of flies, dissection and harvesting tissue (antennae, mid-gut, bacterium and ovaries) for RNA and DNA extraction (In dry ice and liquid Nitrogen).

3. Trypanosome quantitation by qPCR: DNA extraction from blood of infected mice. qPCR for quantitation of trypanosomes using *Sodalis* (tsetse fly symbiont bacteria) DNA as in-house control gene.



Run Setup

Run Information

Run Date: 11/16/2017 12:35 PM
Run User: admin
Run Type: User-defined
Plate File: 111617_TrypTubulin & SodFlic_DNA.pltd
ID:
Notes:
Sample Volume: 10
Temperature Control Mode: Calculated
Lid Temperature: 105
Base Serial Number: CC009683
Optical Head Serial Number: 785BR3686

Protocol

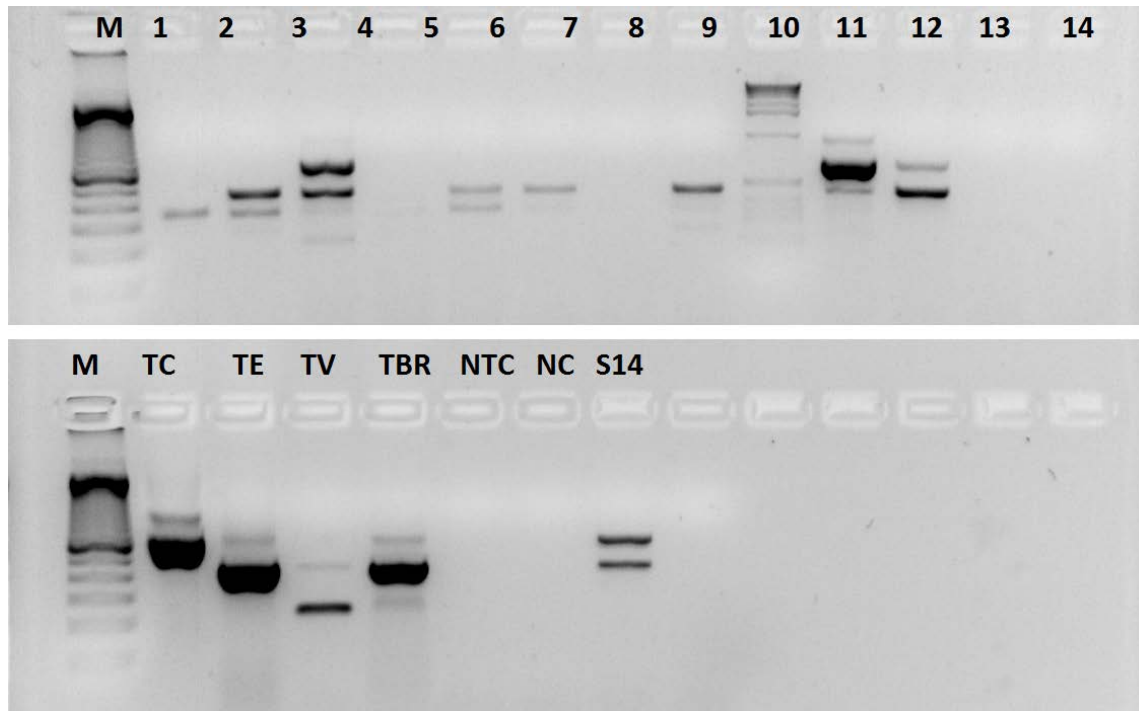
1: 95.0°C for 8:00
2: 95.0°C for 0:15
3: 55.0°C for 0:30
4: 72.0°C for 0:30
Plate Read
5: GOTO 2, 40 more times
6: 95.0°C for 1:00
7: 55.0°C for 1:00
8: Melt Curve 55.0°C to 95.0°C: Increment 0.5°C 0:30
Plate Read

Plate Display

	1	2	3	4	5	6	7	8	9	10	11	12
A	Unk Sod500_Try pTu	Unk Sod500_Try pTu	Unk Sod1000_T rypTu	Unk Sod1000_T rypTu	Unk Sod5000_T rypTu	Unk Sod5000_T rypTu	Unk Sod10000_T rypTu	Unk Sod10000_T rypTu			Neg N/C_TrypTu bulin	Neg N/C_TrypTu bulin
B	Unk Tryp250_Tr ypT	Unk Tryp250_Tr ypT	Unk Tryp500_Tr ypT	Unk Tryp1000_Tr ypT	Unk Tryp1000_Tr rypT	Unk Tryp1000_Tr rypT	Unk Tryp2000_Tr rypT	Unk Tryp2000_Tr rypT	Unk Tryp4000_Tr rypT	Unk Tryp4000_Tr rypT		
C	Unk Tryp250S_Tr ypT	Unk Tryp250S_Tr ypT	Unk Tryp500S_Tr rypT	Unk Tryp1000S_Tr rypT	Unk Tryp1000S_Tr rypT	Unk Tryp1000S_Tr rypT	Unk Tryp2000S_Tr rypT	Unk Tryp2000S_Tr rypT	Unk Tryp4000S_Tr rypT	Unk Tryp4000S_Tr rypT		
D	Unk SGM500_1 S_TrypT	Unk SGM500_1 S_TrypT	Unk SGM500_2 S_TrypT	Unk SGM500_2 S_TrypT	Unk SGM1000_1S_Tryp	Unk SGM1000_1S_Tryp	Unk SGM1000_2S_Tryp	Unk SGM1000_2S_Tryp	Unk SG500_Try pT	Unk SG500_Try pT	Unk SG1000_Tr ypT	Unk SG1000_Tr ypT

Set 4: Output of qPCR for quantitation of trypanosomes in infected mice using tsetse fly symbiont bacteria DNA (*sodalis*) as in-house control.

4. Trypanosome detection system testing: ITS1 PCR for detection of trypanosomes in tsetse flies using our newly designed primers on cDNA derived from wild caught flies whose infection was validated by microscopy.



Set 5: Gel results of ITS PCR on cDNA from samples previously showing inconclusive results with KIN primers. Parasite infection was previously confirmed by microscopy. Positive controls are the bottom gel.

5. Blood meal DNA experiment: PCR detection of sheep DNA from whole flies and midguts from tsetse flies fed with sheep blood to find out how long host DNA persists in flies after a blood meal.



Set 6: Extraction of DNA (Qiagen Kit) from whole flies and gut tissues after flies were fed with sheep blood. I carried these samples for PCR analysis.

- What do you think the positive impact of the activity will have on your further career path?

The internship at Yale will have a profound impact in my future post-doctoral studies since I discussed with Prof. Serap on possible projects in the future in Kenya and Uganda especially on tsetse fly symbiont bacteria studies and their use in controlling trypanosomiasis. Additionally, I was included in the Yale University School of public health database and was given a Yale ID domain. This will ensure I communicate with professors in the school of Public health (Epidemiology) for collaborations and ease of communication. This is also where they advertise post-doctoral and other job openings internally. I therefore have a greater chance and opportunity to join Yale school of Public health as a postdoc.

- Advice for your junior fellows

1. To be courageous, direct to the point and brief in your approach to senior scientists especially in the USA.
2. Don't be afraid to approach senior scientists even by cold calling via email (use the university email domain to your advantage).
3. Always spare time for unforeseen eventualities but always keep a positive attitude.
4. Be open to ideas, challenges and most importantly learn from every situation. Always spare time to enjoy and interact outside of scientific topics and share your experiences and aspirations (personality impression) it's the key to building healthy relationships.

Approval of supervisor	Institution • Official title • Name
	Research center for zoonosis control (CZC)
	Professor
	Chihiro Sugimoto

- ※1 Send the electronic file to the Leading School section, International Affairs Office
- ※2 Attach a copy certificate of the content of internship activity that is prepared by the counterpart at the internship institution (any form with a signature of the counterpart).
- ※3 The Steering Committee of the Leading Program will first confirm the content of this report and report will be forwarded to the Educational Affairs Committee for credits evaluation.

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