## Research Activities Report - 2019 Overseas Research Experience Program -



Research Topic	Molecular mechanism of stress response in HPA axis and its applied to pain research	Graduate School of Engineering
Host University	Western University / London, Ontario / Canada	Applied Chemistry, Environmental and Biomedical Engineering
Duration	From August 27 to October 31, 2019	KAWAGOSHI Ai

## **Summary of the Research Activities**

Stress necessitates an immediate engagement of multiple neuronal and endocrine system. Stress response is regulated by corticotropin releasing hormone (CRH), adrenocorticotropic hormone (ACTH), and cortisol through the hypothalamic-pituitary-adrenal (HPA) axis [Fig1].

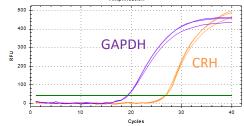
In this research, I analyzed neuron- or glia-specific CRH mRNA expression under the stress condition by using tagged ribosomal subunits (RiboTag). RiboTag method allows for immunoprecipitation of ribosome-associated RNA from specific cells within complex tissues by expressing RiboTag in desired cell types. Cell type-specific promoter restricts the RiboTag expression.

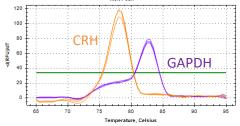
Transgenic mice expressing cell-specific RiboTag are established.

I performed RNA extraction from mouse hypothalamus by using Aurum Total RNA Fatty and Fibrous Tissue kit. Then, I synthesized cDNA by using High-Capacity cDNA Reverse Transcription kits. Expression of CRH mRNA was analyzed by using real-time PCR [Fig2]. CRH and internal control GAPDH mRNAs were detected by Real-time PCR, and the data are little variation within each three samples [Fig3].

I am planning to use the cell-specific gene expression analysis using RiboTag in my pain research.







pituitary gland

Hypothalamus

CRH

**ACTH** 

Fig1. Central

stress response

Fig2. Real-time PCR machine

Fig3. Gene amplification curve and gene melting curve

## **College Life, Friends and Others**

Western University is a Canadian state university with 11 faculties and 400 different specializations. People from different countries learned in the university, and therefore I was able to experience the culture of many countries. I made friends with the laboratory members, and we went to Toronto for sightseeing on a holiday.



## 指導教員講評

新しい研究手法の取得のため、研究室の 方々と積極的にコミュニケーションをとり、 実験に取り組んでいました。

今後の研究の発展を期待しています。

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