



University of Louisville  
Department of Chemistry

**Jing Feng**  
**Literature Seminar**

When: October 14, 2024

Time: 3:00 p.m.

Location: CBL-16

## **Characterization of Modified RNA by LC-MS/MS**

Epitranscriptomics is the study of RNA modifications in an epitranscriptome, a collection of all RNAs in a biological sample. RNA modification is regulated by specific modifying enzymes and the modification process is dynamic and reversible.<sup>1</sup> RNA modification acts as a critical regulator in cell biology, including generation, transportation, function, and metabolism. With the help of advanced high-throughput technologies, more than 170 types of RNA modifications have been discovered, and the biological functions of these modifications are getting increased attention in the studies of human diseases. Currently, liquid chromatography-tandem mass spectrometry (LC-MS/MS) is considered a gold standard in RNA modification research, and different analysis methods have been developed and applied to biological studies. In this presentation, I will introduce three different LC-MS/MS-based methods for the analysis of RNA modification at three levels: nucleoside, oligonucleotide, and intact RNA.<sup>2-4</sup> Each of these methods can achieve different goals in epitranscriptomics, and I will discuss the pros and cons of each method in my presentation.

### **References**

- (1) Qiu, L.; Jing, Q.; Li, Y.; Han, J. RNA Modification: Mechanisms and Therapeutic Targets. *Mol Biomed* **2023**, *4*, 25. <https://doi.org/10.1186/s43556-023-00139-x>.
- (2) Su, D.; Chan, C. T. Y.; Gu, C.; Lim, K. S.; Chionh, Y. H.; McBee, M. E.; Russell, B. S.; Babu, I. R.; Begley, T. J.; Dedon, P. C. Quantitative Analysis of Ribonucleoside Modifications in tRNA by HPLC-Coupled Mass Spectrometry. *Nat Protoc* **2014**, *9* (4), 828–841. <https://doi.org/10.1038/nprot.2014.047>.
- (3) Hagelskamp, F.; Kellner, S. Chapter Six - Analysis of the Epitranscriptome with Ion-Pairing Reagent Free Oligonucleotide Mass Spectrometry. In *Methods in Enzymology*; Jackman, J. E., Ed.; RNA Modification Enzymes; Academic Press, 2021; Vol. 658, pp 111–135. <https://doi.org/10.1016/bs.mie.2021.06.024>.
- (4) Huang, T.-Y.; Liu, J.; McLuckey, S. A. Top-Down Tandem Mass Spectrometry of tRNA Via Ion Trap Collision-Induced Dissociation. *Journal of the American Society for Mass Spectrometry* **2010**, *21* (6), 890–898. <https://doi.org/10.1016/j.jasms.2009.12.007>.