## Vita

Candidate's name: Jaewook Hwang

Universities Attended:

University of New Brunswick (2023)
Bachelor of Science
Honours Medicinal Chemistry

University of New Brunswick (2025)
Masters of Science

## Publications

Hwang, J.; Kirshner, J.; Deschênes, D. A. R.; Richardson, M. B.; Fleck, S. J.; Guo, J.; Perley, J. O.; Shahsavarani, M.; Garza-Garcia, J. J. O.; Seveck, A. D.; Doiron, S. S.; Mai, Z.; Silliphant, S. N.; Calhoun, L.; Gao, D.; Lian, J.; Deslongchamps, G.; Albert, V. A.; Qu, Y. Ancient Gene Clusters Initiate Monoterpenoid Indole Alkaloid Biosynthesis and C3 Stereochemistry Inversion. In revision.

Hwang, J.; Richardson, M.; Kirshner, J.; Albert, V.; Deslongchamps, G.; Qu, Y. Yohimban Acetylation in Rauvolfia is Mediated by a Leaf-Specific Acetyltransferase in Reserpine Biosynthetic Gene Cluster. In preparation.

Plant Canada Conference 2024 Poster Presentation

Phytochemical Society of North America conference 2025 poster presentation

# Discovery, Functional Genomics, and Biochemical Characterization of the Reserpine Biosynthetic Gene Cluster Involved in Monoterpenoid Indole Alkaloid Biosynthesis and Diversification

UNIVERSITY OF NEW BRUNSWICK

THESIS DEFENCE AND EXAMINATION

in Partial Fulfillment

of the Requirement for the Degree of
Master of Science

by

**Jaewook Hwang**

in the Department of Chemistry

U.N.B., Fredericton, N.B.

**Friday, August 15th, 2025**
**10:00 a.m.**

Toole Hall, Room 303 & via MS TEAMs

Examining Committee

| | |
|---|---|
| Dr. Yang Qu | Supervisor |
| Dr. Barry Blight | Internal Examiner |
| Dr. Bryan Crawford | Int-Ext Examiner |
| Dr. Adam Dyker | Chair of Oral Examination |

# Abstract

Monoterpenoid indole alkaloids (MIAs) are structurally complex plant metabolites with profound pharmacological applications. However, their biosynthetic mechanisms and evolutionary origins remain incompletely understood. This thesis integrates biochemical and phylogenomic approaches to unravel the enzymatic and genetic basis of MIA diversification in the order of Gentianales. In collaboration with our partners, we discover an ancient geissoschizine synthase (GS) biosynthetic gene cluster (BGC) and its segmentally duplicated derivative, the reserpine BGC, in *Catharanthus roseus* and *Rauvolfia tetraphylla*. Synthetic studies suggest that the GS BGC originated at the base of the Gentianales order (~135 million years ago), while the reserpine BGC emerged more recently in the Rauvofioid clade of the Apocynaceae family.

Within the reserpine BGC, I biochemically characterized an oxidase-reductase pair, the heteroyohimbine/yohimbine/corynanthe C3-oxidase and C3-reductase (HYC3O/HYC3R). The HYC3O/HYC3R pair, conserved across MIA-producing Gentianales species, catalyzes a previously enigmatic C3 stereochemical inversion step critical for accessing the 3R-MIA scaffold. This discovery elucidates the biosynthetic origin and mechanism underlying the formation of a group of structurally distinct MIAs, including the antihypertensive drug reserpine and spirooxindole alkaloids.

Together, the discoveries allowed us to reconstruct the evolutionary trajectory of strictosidine, GS, and reserpine BGCs, demonstrating how genomic architecture and enzyme evolution underpin chemical diversity. This study also demonstrates that tissue-specific expression patterns within a BGC can further refine spatiotemporal alkaloid accumulation. By linking enzyme function, BGC evolution, and phylogenetic history, this thesis illuminates how nature generates chemical complexity, offering insights for drug discovery and plant biotechnology while advancing synthetic biology strategies to engineer high-value MIAs.