Timothy Klein

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Professional Summary

Biochemistry Ph.D. with two years of postdoctoral research experience at the interface of protein biochemistry, structural biology, and molecular biology. I have seven years of experience in protein expression, purification, characterization, and structural determination. I care deeply about leadership (mentored ten young scientists), communication (published five first-author publications), and collaboration (providing biochemistry support for multiple lab mate's projects).

Research Experience

Postdoctoral Scholar (January 2023 – present), Bondy-Denomy Lab, UCSF

- Currently investigating DNA-targeting anti-phage systems of *Pseudomonas aeruginosa*.
- Collaborated on a project that determined the factors required for protein import into a phage nucleus which is published in *Nature*.
- Responsible for managing the lab's protein biochemistry workflow including maintaining and managing FPLC devices, training lab members on all facets of protein expression/purification, and consulting on the lab's various protein-focused projects.

Graduate Researcher (September 2017 – December 2022), Whitney Lab, McMaster University

- Studied the type VII secretion system of Gram-positive bacteria and initiated protein-protein interaction and structural biology studies of this system.
- Published five first-author publications including four research publications (PNAS, mBio, Structure, JBC) and one review article (Trends in Micro.).
- Served as one of the original members of the Whitney lab. Contributed to building the lab by setting up equipment/infrastructure, managing critical resources, training multiple junior members of the lab, and initiating collaborations with scientists locally and abroad.
- Awarded the prestigious Alexander Graham Bell Canada Graduate Scholarship worth over $100,000. My research inspired a successful national grant application (which I helped write) worth over $650,000 in funding for the lab.
- My work spawned research projects for three junior graduate students. Mentored four undergraduate students, all of whom were included as authors on my publications. Taught dozens of undergraduate students essential topics in biochemistry, structural biology, and microbiology as a TA.
- Highly involved with the graduate community and served as the president of the department's Graduate Student Association for three years. Was directly responsible for planning departmental events, organizing our membership, chairing group meetings, and handling funds. Created and chaired the inaugural two editions of our biochemistry research symposium.

Skills

Biochemistry:

- **Protein engineering** of tagged constructs for solubility (SUMO, MBP) and purification/visualization (His, VSV-G, FLAG, Myc).
- **Protein expression** using bacterial expression models (*E. coli*, *Pseudomonas*).
- **Purification of proteins** and protein complexes with Ni-NTA gravity columns and FPLC (AKTA - size-exclusion, ion-exchange, HisTrap), including purification of proteins for antibody generation in rabbits.
- **Protein-protein interaction** studies by co-immunoprecipitation/pulldown assays, bacterial two hybrid assays and **microscale thermophoresis**.
- Protein size and stability analysis using **SEC-MALS, mass photometry**, and **CD spectroscopy**.

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- **Protein quantification** by UV-spectroscopy.
- **Protein structure determination by X-ray crystallography** including purification of high-purity protein, screening and optimizing crystallization conditions, collecting X-ray data, and determining and analyzing protein structures. Structure model prediction by **Alphafold3**.
- Protein analysis using **SDS-PAGE electrophoresis, Coomassie stain, silver stain, SYPRO red stain, and western blot**.
- Sample preparation and analysis of **mass spectrometry** data.
- **Fluorescent labeling** of purified proteins for MST and membrane topology mapping.
- **Optimization of enzymatic assays** for luciferase (luminescence) and NAD hydrolysis assays.
- RNA purification and analysis by **RT-qPCR**.

Molecular biology/genetics:

- **Molecular cloning** of bacterial and phage genes including primer design, PCR, DNA-purification, restriction digests, Gibson assembly or ligation, and transformation (heat shock, electroporation), conjugation.
- Analysis of DNA molecules by **agarose gel electrophoresis**.
- Cloning of mutant alleles through **site-directed mutagenesis**.
- Cloning of engineered genes using **SOE (splicing by overlap extension) PCR**.
- Making mutant bacterial strains through **allelic exchange**.
- **DNA/RNA quantification** by spectrophotometric and fluorometric quantification.
- Purification of DNA and setup of **whole genome sequencing** for bacteria and phages.

Software:

- **Microsoft Office suite** for writing, data analysis, and presentations.
- **Adobe Illustrator** for making publication-quality figures.
- **Geneious, Benchling** for genome analysis and primer design.
- **Graphpad Prism** for data visualization.
- **UNICORN 7** for AKTA FPLC.
- **PHENIX, Coot** for protein structure determination.
- **ChimeraX, Pymol, PDBePISA, ConSurf** for protein structure analysis.
- **DALI, Foldseek** for protein structure homology search.
- **JackHMMER, Clustal, BLAST, ProtParam** for protein sequence analysis.

Education

Ph.D. Biochemistry (September 2017 – July 2022), McMaster University, Ontario, Canada
B.Sc. Biomedical Sciences (September 2012 – May 2017), University of Waterloo, Ontario, Canada

Selected Publications

Kokontis C, **Klein TA**, Silas S, Bondy-Denomy J. 2025. Multi-interface licensing of protein import into a phage nucleus. *Nature*.

Klein TA, Shah PY, Gkragkopoulou P, Grebenc DW, Kim, Y, Whitney JC. 2024. Structure of a tripartite protein complex that targets toxins to the type VII secretion system. *PNAS* 121(3): e2312455121.

Klein TA, Grebenc DW, Shah PY, McArthur OD, Dickson BH, Surette MG, Kim Y, Whitney JC. 2022. Dual targeting factors are required for LXG toxin export by the bacterial type VIIb secretion system. *mBio* 13(5): e02137-22.

Full publications list: https://scholar.google.com/citations?user=X5qURHgAAAAJ&hl=en&oi=ao