

TUHIN K. GUHA, Ph.D.

Postdoctoral Associate

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*Molecular Biology | Cell line Development | Transfection |
Viral Vector Construction | CRISPR-Cas Genome Editing |
Recombinant protein production | Flow cytometry*

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SUMMARY

Dynamic research individual with more than 8 years of extensive research experience in both standard and advanced molecular biology techniques ranging from DNA and RNA manipulations, recombinant protein expression and purification, biochemical characterization.

Experienced with generating stable and transient mammalian cell lines, DNA transfection, electroporation, microinjection, along with viral vector design for gene repair studies using modified CRISPR system.

Currently investigating the genome editing efficiencies of modified CRISPR-Cas9 system through plasmid and protein-oligo based transfections, electroporation in human cells, microinjections in frog embryos and analyzing efficiencies using flow cytometry.

TECHNICAL EXPERTISE (RELEVANT)

- Mammalian cell / tissue culture maintenance using sterile techniques
- Generation of transient and stable mammalian cell lines for protein expression, Flow cytometry
- Molecular biology techniques DNA / RNA isolation from mammalian cells and bacterial samples
- RNA biology, *in vitro* transcription and translation, cDNA synthesis (RT-PCR), qPCR
- Molecular cloning strategies including GoldenGate assembly, Mutagenesis, DNA Sequencing
- Recombinant protein expression (prokaryotic system, eukaryotic expression system)
- Protein purification using HIC, IEC, HA, SEC Affinity both manually, and using Akta-FPLC
- Western Blot, Coimmunoprecipitation (for protein-protein interaction study)
- DNA / RNA quantification and detection assay, RNA splicing assays, *in vitro* assays
- CRISPR-Cas tool for gene knockout / gene repair; Modified Cas9 for effective gene therapy
- Hybrid Cas9 and Cas9 protein-guide oligo transfection and electroporation in mammalian cell

WORK EXPERIENCE

Postdoctoral Associate **Mar. 2017 – Nov. 2017**
Department of Biochemistry, Schulich School of Medicine and Dentistry
University of Western Ontario, Canada

- Testing genome editing efficiencies of modified CRISPR/Cas9 system in human cells
- Evaluating the off-target and on-target of a hybridized Cas9 protein system
- Experienced with purifying Cas9/modified Cas9 proteins and ribonucleoprotein complex and performing microinjections in frog embryos and transfection/electroporation in human cells

Research Associate **Sep. 2016 – Feb. 2017**
Department of Microbiology, University of Manitoba, Canada

- Conducted protein expression and purification of 2 DNA-cutting enzymes
- Performed DNA extraction and sequence analysis to identify 8 unknown fungal species producing antibiotics (collaborated with the Bioengineering Department, University of Manitoba)
- Contributed towards scientific publications (2 peer-reviewed articles and an invited review)

**INDUSTRIAL WORK
EXPERIENCE****Laboratory Assistant**

Jul. 2006 – Nov. 2008

**A.S. Herbal & Hygiene Products, Bureau of Indian Standards,
Government of West Bengal, Calcutta, India**

- Performed standardized microbial and biochemical lab tests to verify fitness of products, analyze and record data
- Responsible for formulation of products and maintained laboratory equipments

EDUCATION**Ph.D., Microbiology, University of Manitoba, Canada**

2011 – 2016

Biochemical characterization of homing endonucleases encoded by fungal mitochondrial genomes (Supervisor: Dr. Georg Hausner)

- Developed an innovative “on / off switch” for temporal regulation of a DNA-cutting enzyme
- Biochemically characterized a DNA-cutting protein for future biotechnological applications
- Designed an alternative method to investigate the activity of a “difficult-to-express” protein
- Heavily published: 6 Research articles; 1 Review article (invited); 2 Book chapters (invited)

M.Sc., Microbiology, University of Manitoba, Canada

2009 – 2011

Purification of subset of *Saccharomyces cerevisiae* peroxisomal proteins (Supervisor: Dr. Peter C Loewen)

- Effectively cloned, expressed, and purified 2 soluble and 2 insoluble yeast proteins using SEC, IEC and metal (nickel and cobalt) affinity chromatography for downstream assays

M.Sc., Genetics, University of Calcutta, India

2004 – 2006

B.Sc., Zoology (Major), University of Calcutta, India

2001 – 2004

**INSTRUMENTATIONS
& SOFTWARES
(SELECTED)**

- Protein crystal screening: Crystal tray setting using Gryphon robotic arm
- High-content microscopy (Confocal, Fluorescence microscopy)
- Circular Dichroism (CD) spectroscopy
- Microsoft Office suite, CorelDRAW software

PUBLICATIONS

Guha TK, Edgell DR. Applications of alternative nucleases in the age of CRISPR/Cas9. *Int. J. Mol. Sci.* 2017, **18**:2565. doi:10.3390/ijms18122565.

Guha TK^a, Wai A^a, Mullineux ST, Hausner G. The intron landscape of the mtDNA cytb gene among the Ascomycota: introns and intron-encoded open reading frames. *Mitochondrial DNA A DNA Mapp Seq Anal.* 2017, **20**:1-10. doi: 10.1080/24701394.2017.1404042. ^a = equal contribution.

Bilto IM, **Guha TK**, Wai A, Hausner G. Three new active members of the I-Onul family of homing endonucleases. *Can J Microbiol.* 2017, **63**:671-681. doi: 10.1139/cjm-2017-0067.

Guha TK, Wai A, Hausner G. Programmable genome editing tools and their regulation for efficient genome engineering. *Comput Struct Biotechnol J.* 2017, **15**:146-160. doi: 10.1016/j.csbj.2016.12.006.

Tian M, Wai A, **Guha TK**, Hausner G, Yuan Q. Production of cellulase and xylanase from food waste by solid-state fermentation. *Waste and Biomass Valorization.* **2017**.doi: 10.1007/s12649-017-0192-7.

Guha TK, Hausner G. Using group II introns for attenuating the in vitro and in vivo expression of a homing endonuclease. *PLoS One*. 2016, **11**:e0150097. doi: 10.1371/journal.pone.0150097.

Guha TK, Hausner G. A homing endonuclease with a switch: Characterization of a twintron encoded homing endonuclease. *Fungal Genet Biol*. 2014, **65**:57-68. doi: 10.1016/j.fgb.2014.01.004.

Hafez M, **Guha TK**, Hausner G. I-Omil and I-Omill: two intron-encoded homing endonucleases within the *Ophiostoma minus rns* gene. *Fungal Biol*. 2014, **118**:721-731. doi: 10.1016/j.funbio.2014.05.002.

Hafez M, **Guha TK**, Shen C, Sethuraman J, Hausner G. PCR-based bioprospecting for homing endonucleases in fungal mitochondrial rRNA genes. *Methods Mol Biol*. 2014, **1123**:37-53. doi: 10.1007/978-1-62703-968-0_3.

CONFERENCE PRESENTATIONS (SELECTED)

Guha TK*, and Hausner G. Controlling a DNA chopper: Group II introns as attenuators for homing endonuclease expression. Biophysical Society of Canada. 1st - 3rd June 2016; University of Manitoba, Winnipeg, Canada.

Guha TK*, and Hausner G. On-switch regulators in action: Regulation of a Homing endonuclease by group II introns. Canadian Society of Microbiologists, 15th - 18th June 2015; University of Regina, Saskatoon, Canada.

Guha TK*, and Hausner G. A twintron encoded homing endonuclease with an on-switch. Keystone Symposia, Keystone Symposia- Precision genome engineering and synthetic biology. 11th - 16th January 2015; Big Sky, Montana, USA.

Guha TK*, and Hausner G. A homing endonuclease with a switch: Characterization of a twintron encoded homing endonuclease. Canadian Society of Microbiologists, 17th - 20th June 2013; Carleton University, Ottawa, Canada.

SCHOLARSHIPS/ AWARDS (SELECTED)

- International Graduate Student Entrance Scholarship, University of Manitoba (2009 - 2010, CAD 8000)
- Faculty of Science Graduate Scholarship, University of Manitoba (2011 - 2013, CAD 10000 / year)
- International Graduate Student Scholarship, University of Manitoba (2012 - 2013, CAD 8000)

VOLUNTEERING ACTIVITIES (SELECTED)

- Volunteered at numerous science fairs, "Info days", Science Rendezvous
 - Engaged children and youth with interactive scientific displays and hands-on activities
- Involved in social committees
 - Fund-raising activities