

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
Follow this format for each person. **DO NOT EXCEED FIVE PAGES.**

NAME: Rakhi Rajan

eRA COMMONS USER NAME (credential, e.g., agency login): RRAJAN

POSITION TITLE: Associate Professor of Chemistry and Biochemistry

EDUCATION/TRAINING

INSTITUTION AND LOCATION	DEGREE (if applicable)	Completion Date MM/YYYY	FIELD OF STUDY
Kerala Agricultural University (KAU), India	B.S.	06/1998	Agriculture
Tamil Nadu Agricultural University (TNAU), India	M.S.	10/2000	Biotechnology
The Ohio State University, Columbus, OH	Ph.D.	08/2007	Biophysics
Northwestern University, Evanston, IL	Postdoc	03/2013	Biochemistry and Structural Biology

A. Personal Statement

My research focuses on biological processes mediated by protein-nucleic acid complexes. The current focus of my group at the University of Oklahoma is the structural and functional characterization of the bacterial and archaeal immune system, CRISPR-Cas, that comprises protein-RNA-DNA complexes. The specific questions that we address are: (i) RNA-mediated activation mechanisms of Cas9 and Cas12a proteins; and (ii) development of high fidelity Cas proteins for gene editing applications by modulating protein-RNA-DNA interactions; (iii) characterizing the protein-nucleic acid interactions essential for CRISPR adaptation, a process to develop immunity against new intruding genetic materials; and (iv) the mechanisms by which CRISPR-Cas systems perform non-canonical functions such as catalysis of guide RNA-independent DNA cleavage.

During my graduate studies (under Dr. Charles Bell) and postdoctoral research (under Dr. Alfonso Mondragón), I used biochemistry, biophysics, and structural biology tools to characterize Recombinase A (RecA) and Topoisomerase V (Topo V). I was introduced to CRISPR-Cas systems during the last couple of years of my postdoctoral research to perform biochemical characterization of *Neisseria meningitidis* Cas9 for Dr. Erik Sontheimer's lab (Zhang *et al.*, 2020). Since starting my independent research, we have been using a multi-dimensional approach to understand the enzymatic mechanisms of Cas proteins and CRISPR adaptation machinery. Our research has identified a previously unknown RNA-independent DNA cleavage activity by Cas proteins that can potentially impact the safety of CRISPR-Cas mediated gene editing applications (Sundaresan *et al.*, 2017). We have reported on methods to develop high-specificity Cas9 variants by engineering bridge helix, a helix that is conserved in several Cas enzymes commonly used for biotechnology applications (Babu *et al.*, 2019). Our contribution to the field of CRISPR adaptation, a mechanism by which bacteria develop CRISPR-based immunity, included distinct mechanisms for type II-A CRISPR adaptation with unique sequence specificities (Van Orden *et al.*, 2020). We collaborate with other scientists to enable a comprehensive understanding of our research questions [e.g., Dr. Peter Qin at the University of Southern California for the use of Electron Paramagnetic Resonance (EPR) spectroscopy; Dr. Jin Liu at the University of North Texas Health Science Center for Molecular Dynamics (MD) simulations; Dr. Yihan Shao in my department for computational studies to characterize catalytic mechanisms of Cas9 and Cas12a. The Biochemistry and Structural Biology groups in the Department of Chemistry and Biochemistry at the University of Oklahoma provides an excellent research environment with several core facilities to help our research goals.

This proposal is seeking user time through the National Center for CryoEM Access and Training (NCCAT) at the New York Structural Biology Center to advance our ongoing cryo-EM studies with Cas9, Cas12a, and Cas1-Cas2 nucleic acid complexes. We are requesting Block Allocation Access (BAG) for Krios data collection

time for three different projects that will each look at different types of Cas protein-nucleic acid complexes. PI Rajan and the graduate student Chhandosee Ganguly will be the key personnel involved in this proposal. Since the University of Oklahoma currently lacks electron microscope infrastructure needed for single particle analysis, BAG access will be highly advantageous for progressing our research goals as well as to acquire preliminary data for proposal submissions.

[*indicates corresponding author]

1. Zhang, Y.[†], **Rajan, R.**[†], Seifert, H.S., Mondragón, A., Sontheimer, E.J.* (2015). DNase H activity of *Neisseria meningitidis* Cas9. *Mol Cell*, **60** (2): 242-255. PMID: PMC4609032. ([†]equal contributors).
2. Sundaresan, R., Parameshwaran, H.P., Yogesha, S.D., Keilbarth, M.W., and **Rajan, R.*** (2017). RNA-independent DNA cleavage activities of Cas9 and Cas12a. *Cell Rep*, 21: 3728-3739. PMID: PMC5760271.
3. Babu, K., Amrani, N., Jiang, W., Yogesha, S.D., Nguyen, R., Qin, P.Z., **Rajan, R.*** (2019) Bridge helix of Cas9 modulates target DNA cleavage and mismatch tolerance. *Biochemistry*, 58(24): 1905-1917. PMID: PMC6496953.
4. Van Orden M.J., Newsom S., **Rajan R.*** (2020). CRISPR type II-A subgroups exhibit phylogenetically distinct mechanisms for pre-spacer insertion. *J Biol Chem*. 295(32):10956-10968. PMID: PMC7415960.

Ongoing and recently completed projects that I would like to highlight include:

U.S. Department of Defense, Congressionally Directed Medical Research Progs, DOD-CDMR (HT94252310256)

Rajan, R. (PI) ; co-PIs: Ji Hwan Park, Carolyn Ibberson

06/01/2023 to 05/31/2025

Identifying protein motifs in Cas9 essential for bacterial virulence

NSF MCB-1716423

Rajan, R. (PI), Qin, P.Z., (Co-PI, USC)

09/15/2017-08/31/2023 (2-year no cost extension)

Collaborative Research: Mechanisms of RNA-directed activation of a Cas9 nuclease competent for DNA interrogation

Oklahoma Center for the Advancement of Science and Technology (OCAST), HR20-103

Rajan, R. (PI)

10/01/2020-12/31/2023

Protein engineering to develop stringent CRISPR-Cas tools

NIH R21, 1R21GM144860-01

Liu, J. (PI), Rajan, R. (collaborator)

01/01/2022-12/31/2023

De novo development of small CRISPR-Cas proteins using artificial intelligence

University of Oklahoma Faculty Investment Program

Rajan, R. (PI)

04/15/2022-08/31/2023

Atomic-resolution structures of Cas protein complexes by cryo-Electron Microscopy

University of Oklahoma Collaborative Research Faculty Fellowship

Rajan, R. (PI), co-PI: Wilhelm, S.

06/01/2022-05/31/2024

Combining Cas enzyme platform and inductively coupled plasma mass spectrometry (ICP-MS) for early-stage disease diagnosis

NIH Oklahoma Center of Biomedical Research Excellence in Structural Biology (OCSB) P30GM145423

West, A.H. (PI), Somalinga, V. (Pilot project leader), Rajan R. (co-PI)

08/15/2022 to 05/31/2024

Structural and functional characterization of proteins essential for metabolism and virulence in *Streptococcus sanguinis*

NIH Oklahoma Center of Biomedical Research Excellence in Structural Biology (OCSB) P30GM145423
West, A.H. (PI), Shao, Y. (Pilot project leader), Rajan R. (co-PI)

08/15/2022 to 05/31/2024

A combined computational and experimental study of CRISPR Cas9/12a Enzyme reaction mechanisms

NIH Centers of Biomedical Research Excellence (COBRE) in Structural Biology, P20GM103640

West, A.H. (PI), Rajan R. project leader

09/06/2017-12/31/2019

Mechanistic characterization of CRISPR-Cas complexes that mediate pathogenicity in the bacterium *Francisella tularensis novicida* (Phase II)

B. Positions, Scientific Appointments, and Honors

Positions and Scientific Appointments

07/2020– present	Associate Professor, Department of Chemistry and Biochemistry, University of Oklahoma (OU), Norman, OK
08/2014–06/2020	Assistant Professor, Department of Chemistry and Biochemistry, University of Oklahoma (OU), Norman, OK
04/2013–07/2014	Research Associate with Dr. Alfonso Mondragón, Department of Molecular Biosciences, Northwestern University, Evanston, IL
10/2007–03/2013	Postdoctoral fellow with Dr. Alfonso Mondragón, Department of Molecular Biosciences, Northwestern University, Evanston, IL
06/2002–08/2007	Graduate student with Dr. Charles Bell, Department of Biological Chemistry and Pharmacology, The Ohio State University, Columbus, OH

Other Experience and Professional Memberships

Associate Editor: *Frontiers in Cellular and Infection Microbiology*, *Specialty section: Molecular Bacterial Pathogenesis* (2018–current)

Proposal Reviewer, National Science Foundation, Molecular and Cellular Biosciences, Molecular Biophysics (ad-hoc reviewer and panelist), The Wellcome Trust-DBT India Alliance Fellowship, Dutch Research Council (NWO) Talent Programme – Veni Domain Science

Member, American Chemical Society (2018-2022)

Member, American Society for Biochemistry and Molecular Biology (2017, 2020)

Member, Biophysical Society (2004–07, 2019)

Member, American Heart Association (2012, 2015, 2017)

Member, American Association for the Advancement of Science (2006–07, 2010-13)

Member, American Crystallographic Association (2010–11)

Manuscript Reviewer for *Nucleic Acids Research*, *Molecular Therapy*, *Viruses*, *Cell Reports*, *PLOS One*, *Scientific Reports*, *ACS Synthetic Biology*, *Frontiers in Genome Editing*, *Current Opinion in Structural Biology*, *Journal of Biomolecular Structure & Dynamics*, *Microbial Biotechnology*, *RNA Biology*, *Microbial Cell Factories*

Honors

2016	Poster abstract selected for the late-breaker poster at the American Society for Microbiology Microbe 2016 meeting
2012	Best poster award, Northwestern University Biophysics Symposium
2010–13	American Heart Association Postdoctoral Fellowship, Northwestern University
2006	Outstanding Student Research Achievement Award, The Ohio State University Biophysics program
2004	Best poster award, The Ohio State University Molecular and Cellular Biochemistry annual retreat
2000–02	University Grants Commission Junior Research Fellow National Eligibility Test Scholarship for pursuing a Ph.D. degree in India
1998–2000	Jawaharlal Nehru University (M.S.) Scholarship from the Department of Biotechnology (India)
1996–97	Chinnamma Thomas Memorial Endowment for highest GPA (B.S., 3 rd yr., KAU, India)

C. Contributions to Science

[*indicates corresponding author]

1. Conformational and mechanistic characterizations of DNA cleavage by Cas9.

One of the research interests of my laboratory is to characterize the mechanisms by which Cas9 targets and cleaves DNA. An in-depth mechanistic characterization will enable increasing stringency with which Cas proteins carry out DNA cleavage, leading to the development of protein variants with high fidelity for gene editing and gene therapy applications. Our lab reported for the first time, an unprecedented, RNA-independent DNA cleavage by Cas proteins, which warrants further characterization of CRISPR-Cas systems for therapeutical applications (Sundaresan *et al.*, 2017). Our lab also reported for the first time that manipulating the interactions of a helix, called the bridge helix, that is highly conserved across several Cas9s can serve as a method to develop stringent protein variants with low off-target DNA cleavage for gene editing (Babu *et al.*, 2019, Babu *et al.*, 2021). In addition, we collaborate with other CRISPR experts to understand the mechanisms of Cas protein-guide-RNA-mediated DNA cleavage of Cas9 (Zuo *et al.*, 2019) and Cas12a.

- a. Sundaresan, R, Parameshwaran, H.P., Yogesha, S.D., Keilbarth, M.W., and **Rajan, R.*** (2017). RNA-independent DNA cleavage activities of Cas9 and Cas12a. *Cell Rep*, 21: 3728-3739. PMID: PMC5760271.
- b. Babu, K., Amrani, N., Jiang, W., Yogesha, S.D., Nguyen, R., Qin, P.Z., **Rajan, R.*** (2019) Bridge helix of Cas9 modulates target DNA cleavage and mismatch tolerance. *Biochemistry*, 58(24): 1905-1917. PMID: PMC6496953.
- c. Babu, K., Kathiresan, V., Kumari, P., Newsom, S., Parameshwaran, H.P., Chen, X., Liu, J., Qin, P.Z., **Rajan, R.*** (2021). Coordinated actions of Cas9 HNH and RuvC nuclease domains are regulated by the bridge helix and the target DNA sequence. *Biochemistry*. 60(49):3783-3800. PMID: PMC8675354.
- d. Zuo, Z., Zolekar, A., Babu, K., Lin, V.J.T., Hayatshahi, H.S., **Rajan, R.**, Wang, Y.C.,* Liu, J.* (2019). Structural and functional insights into the *bona fide* catalytic state of *Streptococcus pyogenes* Cas9 HNH nuclease domain. *Elife*, 8:e465000. PMID: PMC6706240.

2. Conformational and mechanistic characterizations of DNA cleavage by Cas12a.

Research in the CRISPR field has revealed similarities and differences between Cas9 and Cas12a, the two Cas protein families that are widely used for genome applications. We take a parallel route by comparing and contrasting DNA cleavage mechanisms of Cas9 and Cas12a. We showed that Cas12a also possesses the unprecedented, RNA-independent DNA cleavage and that it can use diverse DNA substrates for this activity compared to Cas9 (Sundaresan *et al.*, 2017). We established that the method of modulating the interactions of bridge helix with RNA and DNA to improve DNA cleavage stringency is conserved in Cas12a (Parameshwaran *et al.*, 2021, Martin *et al.*, 2023). We also collaborate with other CRISPR experts to understand the mechanisms of guide-RNA-mediated DNA cleavage of Cas12a (Jiang *et al.*, 2019).

- a. Sundaresan, R, Parameshwaran, H.P., Yogesha, S.D., Keilbarth, M.W., and **Rajan, R.*** (2017). RNA-independent DNA cleavage activities of Cas9 and Cas12a. *Cell Rep*, 21: 3728-3739. PMID: PMC5760271.
- b. Jiang, W., Singh, J., Allen, A., Li, Y., Kathiresan, V., Qureshi, Q., Tangprasertchai, N., Zhang, X., Parameshwaran, H.P., **Rajan, R.**, Qin, P. Z.* (2019), "CRISPR-Cas12a Nucleases Bind Flexible DNA Duplexes without RNA-DNA Complementarity." *ACS Omega*, 4, 17140-17147. PMID: PMC6811856.
- c. Parameshwaran, H.P., Babu, K., Tran, C., Guan, K., Allen, A., Kathiresan, V., Qin, P.Z., and **Rajan, R.*** (2021). The bridge helix of Cas12a imparts selectivity in *cis*-DNA cleavage and regulates *trans*-DNA cleavage. *FEBS Lett*, 595(7):892-912. PMID: PMC8044059.
- d. Martin, L., Rostami, S., and **Rajan, R.*** (2023). Optimized protocols for the characterization of Cas12a activities. *Methods Enzymol*, 679:97-129. PMID: 36682874.

3. Characterization of CRISPR adaptation mechanisms.

Adaptation is the process by which bacteria and archaea develop immunity against a new phage infecting them. This involves excising short DNA fragments and inserting into the CRISPR locus precisely, to avoid self-targeting of the genome and disruption of essential genes during the insertion process. We conducted bioinformatic studies to locate DNA motifs that may contribute to site-specific insertions of phage DNA into bacterial genome. We identified conserved 3'-leader motifs in type II-A CRISPR systems (Van Orden *et*

et al., 2017). Following this bioinformatic study, we characterized the role of DNA sequences in sub-group specific DNA insertion that revealed distinct mechanisms for DNA insertion in the seemingly related CRISPR type II-A subtypes (Van Orden *et al.*, 2020).

- a. Van Orden, M., J., Klein, P., Babu, K., Najar, F.Z., **Rajan, R.*** (2017). Conserved DNA motifs in the type II-A CRISPR leader region. *PeerJ*, 5:e3161. <https://peerj.com/articles/3161/>. PMID: PMC5382924.
- b. Van Orden M.J., Newsom S., **Rajan R.*** (2020). CRISPR type II-A subgroups exhibit phylogenetically distinct mechanisms for prespacer insertion. *J Biol Chem* 295(32):10956-10968. PMID: PMC7415960.
- c. Flusche, T., **Rajan, R.** Molecular details of DNA integration by CRISPR-associated proteins during adaptation in bacteria and archaea. (2023). *Adv. Exp. Med. Biol.* 1414:27-43. PMID: 35852729.

4. The structure and mechanism of Topoisomerase V (Topo V).

Topoisomerases enable supercoiling and relaxing DNA. Topo V present in *Methanopyrus kandleri*, a hyperthermophilic archaeon, is unique because it has both DNA relaxation and DNA repair activities in the same polypeptide. I determined the structures of several fragments of Topo V encompassing different active sites: structure of an N-terminal 44 kDa fragment of Topo V that showed conformational changes essential for binding DNA (Rajan *et al.*, 2010); biochemical characterization of the topoisomerization reaction, (Rajan *et al.*, 2014); crystal structures of N-terminal 78 kDa and 94 kDa fragments of Topo V that illustrated the architecture of the AP lyase sites (Rajan *et al.*, 2013, Rajan *et al.*, 2016).

- a. **Rajan, R.**, Taneja, B., and Mondragón, A.* (2010). Structures of minimal catalytic fragments of topoisomerase V reveals conformational changes relevant for DNA binding. *Structure*, **18** (7): 829-838. PMID: PMC2907367.
- b. **Rajan, R.**, Prasad, R., Taneja, B., Wilson, S.H., and Mondragón, A.* (2013). Identification of one of the apurinic/apyrimidinic lyase active sites of topoisomerase V by structural and functional studies. *Nucleic Acids Res*, **41** (1): 657-666. PMID: PMC3592480.
- c. **Rajan, R.**, Osterman, A.K., Gast, A.T., Mondragón, A.* (2014). Biochemical characterization of the topoisomerase domain of *Methanopyrus kandleri* topoisomerase V. *J Biol Chem*, **289** (42): 28898-28909. PMID: PMC4200249.
- d. **Rajan, R.**, Osterman, A., Mondragón, A.* (2016). *Methanopyrus kandleri* topoisomerase V contains three distinct AP lyase active sites in addition to the topoisomerase active site. *Nucleic Acids Res*, **44** (7): 3464-3474. PMID: PMC4838376.

5. Structural and functional characterization of RecA.

RecA protein catalyzes the strand exchange reaction in bacterial homologous recombination. *Deinococcus radiodurans* (Dr) is an extremophilic bacterium resistant to ionizing radiations, and the extreme resistance is attributed to its efficient DNA repair mechanism catalyzed in part by RecA. During my graduate study in Dr. Bell's laboratory at The Ohio State University, I determined the first structure of Dr RecA by X-ray crystallography (Rajan *et al.*, 2004). The Dr RecA structure showed features that supported the "inverse strand exchange pathway" in Dr. In addition, I used a set of biochemical assays to better understand the DNA sequence preferences exhibited by Ec RecA (Rajan *et al.*, 2006).

- a. **Rajan, R.**, and Bell, C.E.* (2004). Crystal structure of RecA from *Deinococcus radiodurans*: insights into the structural basis of extreme radioresistance. *J Mol Biol*, **344** (4): 951-963. PMID: 15544805.
- b. **Rajan, R.**, Wisler, J.W., and Bell, C.E.* (2006). Probing the DNA sequence specificity of *Escherichia coli* RECA protein. *Nucleic Acids Res*, **34** (8): 2463-2471. PMID: PMC1459065.

Complete List of Published Work in My Bibliography:

<https://www.ncbi.nlm.nih.gov/myncbi/1vWGwzbnGbQys/bibliography/public/>

BIOGRAPHICAL SKETCH

Provide the following information for the Senior/key personnel and other significant contributors.
Follow this format for each person. **DO NOT EXCEED FIVE PAGES.**

NAME: CHHANDOSEE GANGULY

eRA COMMONS USER NAME (credential, e.g., agency login): Chhandosee

POSITION TITLE: Graduate Student Research/ Teaching Assistant

EDUCATION/TRAINING *(Begin with baccalaureate or other initial professional education, such as nursing, include postdoctoral training and residency training if applicable. Add/delete rows as necessary.)*

INSTITUTION AND LOCATION	DEGREE (if applicable)	Start Date MM/YYYY	Completion Date MM/YYYY	FIELD OF STUDY
University of Calcutta	B.Sc.	07/2014	05/2017	Microbiology
St. Xavier's College Kolkata	M.Sc.	07/2017	05/2019	Microbiology
University of Oklahoma	Ph.D.	01/2021	05/2026 (Expected)	Chemistry and Biochemistry

A. Personal Statement

Understanding the mechanism underlying biological phenomena and translating them to problem-solving applications in our daily lives has always fascinated me.

While working on my master's thesis at Sea6 Energy Pvt. Ltd., I came across the concept of natural product isolation from microorganisms and applying it to generate plant bio-stimulants. There I worked with the R & D department of the company to identify and characterize the fungal strains which were able to produce carragenase enzyme with better lysis potential than acid hydrolysis. The company plans to manufacture various products, including algal biofuel with those isolated enzymes. During the five years of my bachelor's and master's program in Microbiology, I participated in multiple research projects that grew my interest in learning more about enzymes and their function. By the end of my master's study, I knew I wanted to pursue a long-term career in research. Finally, I decided to move into biochemistry and structural biology for my doctoral training at the University of Oklahoma. Currently, under the guidance of Dr. Rakhi Rajan, my research interest is understanding the structure-function relationships of the CRISPR-Cas proteins. I am a second-year graduate student who contributed to a publication and aiming to submit the manuscript as a first author within next year. Working in the lab, I have acquired skills in performing various advanced biochemical and analytical techniques and is learning macromolecular x-ray crystallography and cryo-electron microscopy (cryo-EM) sample preparation and data analysis methods. Collecting high-resolution cryo-EM data for various Cas9-nucleic acid complexes is critical in deciphering the molecular mechanism of the high-fidelity variants that we developed in the Rajan lab. This is a major research goal of my Ph.D. dissertation. I have successfully completed my Ph. D. candidacy examination and majority of the graduate coursework. In addition to academics,

I also contribute to departmental service by serving as a student representative in various committees in my department, such as the Graduate Curriculum Committee and the Graduate Student Admissions and Recruitment Committee. I am the International Student Representative of the OU's Chemistry and Biochemistry Student Research Organization. Coming from a middle-class Indian family, I am the first to receive doctoral training, and I am committed in investing my efforts to attain the end goals of our research. Overall, I believe that my current research goals, training plan, and service experiences will help me reach the zenith of my career and enable me to achieve my dream of becoming a successful independent investigator.

B. Positions, Scientific Appointments and Honors

2014 – 2017	Bachelors Student, University of Calcutta
2017 – 2019	Masters Student, St. Xavier's College Kolkata
2021 – Present	Graduate Student, University of Oklahoma, Norman
2022 – Present	Student representative, Graduate Curriculum Committee, Chemistry & Biochemistry, University of Oklahoma, Norman
2022 – Present	Student representative, Graduate Student Admissions and Recruitment Committee, Chemistry & Biochemistry, University of Oklahoma, Norman
2022 – Present	International student representative, Chemistry & Biochemistry Student Research Organization, University of Oklahoma, Norman

Honors

2011	Proficiency, Rajya Purashkar (State level award) from Bharat Scouts and Guides
2012	Scholarship, Certificate of Merit from Central Board of Secondary Education
2017	Poster presenter, Modern trends in Microbiology
2019	Oral presenter, Modern trends in Microbiology
2021	Scholarship, Center for Antibiotic Discovery and Resistance

C. Contributions to Science

Bachelor's Research: Ashutosh College, Kolkata (Summer Project) April 2017-June 2017

- Comparative studies on dose dependent inhibitory effect of aqueous extracts of garlic and lemon on coliform bacterial isolates from Bagjola canal water"

Role- Designed antimicrobial assays and performed data analysis using MS-excel under guidance of Dr. Kuntal Goswami.

Master's Research: St. Xavier's College Kolkata (August 2017- May 2019) projects carried under guidance of Dr. Arup Mitra

Chhandosee Ganguly (2019) A Novel Biopesticide Formulation for Environmental Sustainability, International Journal of Innovative Science, Engineering & Technology, Vol. 6 Issue 8, August 2019 ISSN (Online) 2348 – 7968

- Formulation of novel biopesticides using entomopathogenic fungi and bacteria for an industry: Organic Agro India (Aug 2017- May 2019)

Role- Performed microbial synergistic tests, determined the suitable composition for preparation of biopesticide, applied the formulation on group of test plants, and compared the results with the control group to assess the effect of the biopesticide formulation.

Graduate Research: University of Oklahoma

The main goal of my graduate research is to derive molecular mechanisms by which Cas9 variants developed by modulating protein-RNA-DNA interactions enable high selectivity while cleaving target DNA such that unwarranted off-target effects are minimized during gene editing. I also collaborate with other research laboratories in mechanistic characterization of related Cas proteins for improving their biotechnological applications. I use a combinatorial approach including biochemistry, structural biology (x-ray crystallography and cryo-EM) and work collaboratively for molecular dynamics simulations to derive molecular mechanisms.

- Zhicheng Zuo, Kesavan Babu, **Chhandosee Ganguly**, Ashwini Zolekar, Sydney Newsom, Rakhi Rajan, Yu-Chieh Wang, Jin Liu. (2022). Rational Engineering of CRISPR-Cas9 Nuclease to Attenuate Position-dependent Off-target Effects. *CRISPR J.*, **5**(2):329-340. PMCID: PMC9271410
Role- Performed *invitro* plasmid cleavage assays to determine the efficacy of the newly developed protein with respect to the high-fidelity variants available in the field.
- Valentin V. Rybenkov, Helen I. Zgurskaya, **Chhandosee Ganguly**, Inga V. Leus, Zhen Zhang, and Mohammad Moniruzzaman. (2021). *Chemical Reviews*, **121** (9), 5597-5631. PMCID: PMC8369882
Role- Contributed majorly in writing the section 3- 'OM Porins and Translocators'.

D. Scholastic Performance[#]

YEAR	COURSE TITLE	GRADE
University of Calcutta (BSc)		
2014	Biomolecules	A
2014	Biophysical Chemistry and Biometry	A
2014	General Chemistry	A
2014	Principles of organic chemistry	A
2014	Basics of Inorganic chemistry	A
2014	General Microbiology	A
2015	Microscopy and biochemistry (Practical)	A
2015	Plant Kingdom	A
2015	Cellular and Molecular biology	A
2015	Metabolism and bioenergetics	A
2015	Environmental and food microbiology	A
2015	Basic Physical Chemistry	A
2015	Principles of qualitative inorganic analysis	A
2016	Microbial Genetics	A
2016	Immunology	A
2016	Industrial Microbiology	A
2016	General Environmental studies	A
St. Xavier's College Kolkata (MSc)		
2017	Biophysical Techniques & Microscopy & Macromolecules	A

YEAR	COURSE TITLE	GRADE
2017	Cell Biology, Cell signaling and function	A
2017	Taxonomy, Morphology & Ecology	A
2017	Thermodynamics & Reaction Kinetics of Enzymes	A
2018	Molecular Biophysics & Crystallography, General and Microbial Metabolism	A
2018	Molecular Biology	A
2018	Environmental Microbiology and Agricultural biology	A
2018	Recombinant DNA technology, Gene therapy and Biostatistics	A
2018	Plant Physiology, Breeding and biotechnology & Tissue culture	A
2018	Immunology, Cancer biology, Biosafety and Industrial Microbiology	A
2018	Bioinformatics, Genomics and Proteomics	A
2019	Medical microbiology	A
2019	Food technology & Virology	A
2019	Project and Dissertation	A
2019	Grand Viva and Critical Review	A
University of Oklahoma		
2021	Laboratory Rotation	A
2021	Molecular Biology	A
2021	Fundamentals I	A
2021	Principles of Biochemistry	A
2021	Macromol Structure & Function	A
2022	Spec Topics in Struct Biology (RNA biology)	A
2022	Fundamentals II	A
2022	Biochem and Biophys Methods	A
2022	Macromolecular Crystallography	A
2022	Spec Topics in Struct Biology (Cryo EM)	A
2022	Practicum in Struct Biology (Crystallography)	A

#only courses related to my graduate research are listed for the bachelors and masters studies.