Soumita Dutta, Ph. D.

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EDUCATION

2015	**Ph.D. (Microbiology)**, University of Calcutta, Kolkata, India *Thesis title*: "**Functional characterization of a novel phosphoesterase encoded by mycobacteriophage D29**". *Supervisor*: Prof. Sujoy Kumar Das Gupta, Dept. of Microbiology, Bose Institute, Kolkata, India.
2008	**Post Graduate one year advanced diploma course in Bioinformatics** University of Calcutta, Kolkata, India
2007	**Master of Science (M.Sc.) in Biotechnology,** Jadavpur University, Kolkata, India
2005	**Bachelor of Science (B.Sc.) in Microbiology (Major)**, University of Calcutta, Kolkata, India
2002	**Higher Secondary Examination (10+2)**, West Bengal Board of Higher Secondary Education, West Bengal, India
2000	**Madhyamik Pariksha (10th)**, West Bengal Board of Secondary Education, West Bengal, India

POSITION and EMPLOYMENT

Nov. 2015-Present	**Post-Doctoral fellow,** Dept. of Anatomy and Cell Biology, University of Kansas Medical Center, Kansas, USA Supervisor: Prachee Avasthi, Ph. D. **Project 1**: MAPK activated ciliary length regulation in Chlamydomonas. **Project 2**: Role of actin phosphorylation in ciliary dynamics. **Project 3**: Actin redundancy in *Chlamydomonas reinhardtii.* **Project 4**: Study of flagella-regeneration, a proteomics-based approach
Jan. 2009-June 2015	**Junior and senior research fellow** (Ph.D. student) in Dept. of Microbiology, Bose Institute, Kolkata, India Supervisor: Prof. Sujoy Kumar Das Gupta **Project 1**: Isolation and characterization of a Calcineurin like protein phosphatases of mycobacteriophage D29 and mycobacteriophage Wildcat **Project 2**: Investigating the role of protein phosphorylation and dephosphorylation in the growth of mycobacteriophages

Sep. 2014-June 2015	**Guest lecturer** for graduate and post graduate course in the Dept. of Microbiology, Bidhannagar Govt. College, West Bengal State University, West Bengal, India.
May 2008-Nov. 2008	**Summer project trainee,** Dept. of Botany, Bose Institute, Kolkata, India. Supervisor: Prof. Dibyendu N. Sengupta **Project work:** Analysis of codon usage pattern in *Solanum lycopersicum* (tomato)-a computational approach
July 2006-June 2007	**Laboratory trainee,** Dept. of Life Science and Biotechnology, Jadavpur University, Kolkata, India. Supervisor: Dr. Ratan Gachhui **Project work:** Study of nitrogen metabolism and regulatory component of *Glucanacetobacter kombuchae*, a nitrogen fixing and cellulose producing bacteria

HONOURS and AWARDS

2008	Qualified **National Eligibility Test (CSIR-NET)** for **Junior Research Fellowship** in Life Science, organized by Council of Scientific and Industrial Research (CSIR-HRDG), Govt. of India
2008	Qualified **Junior Research Fellowship(DBT-JRF)**, category B, organized by Dept. of Biotechnology (DBT), Govt. of India
2008	Qualified **Junior Research Fellowship (ICMR-JRF),** organized by Indian Council of Medical research (ICMR), Govt. of India
2008	Qualified Graduate Aptitude test in Engineering **(GATE)** in Life Science with 99.14 Percentile; All India Rank 113
2007	Ranked **First with first class in M.Sc.** (Biotechnology) from Jadavpur University, Kolkata.
2002	Ranked 96 in state (West Bengal) and secured national scholarship in class XII (Higher Secondary Examination)
2000	Ranked 44 in state (West Bengal) and secured national scholarship in class X (Madhyamik Pariksha)

PUBLICATIONS

2017	**Soumita Dutta** and Prachee Avasthi. **Flagellar synchronization is a simple alternative to cell cycle synchronization for ciliary and flagellar studies.** *mSphere 2: e00003-17*.http://doi.org/10.1128/mSphere.00003-17.
2014	**Soumita Dutta,** Niketa Bhawsinghka and Sujoy K. Das Gupta**. Gp66, a calcineurin family phosphatase encoded by mycobacteriophage D29, is a 2', 3' cyclic nucleotide phosphodiesterase that negatively regulates phage growth.** *FEMS Microbiology Letters*, 361 (2014) 84-93.
Manuscript under preparation	**Soumita Dutta** and Prachee Avasthi. **Identification of the novel pathways for ciliary length regulation in *Chlamydomonas* during MAPK phosphatase inhibition.**

CONFERENCES

Dec 3rd-7th, 2016

Speaker in the mini-symposium entitled "Actin Dynamics" under the topic Cytoskeleton, Motility, and Cell Mechanics, at the 2016 American Society for Cell Biology (ASCB) Annual Meeting in San Francisco, California, USA.
Title: **Investigating the role of ERK-mediated actin phosphorylation in ciliary dynamics.**

Poster Presenter at the session entitled "ciliary signaling and ciliopathies", at the 2016 American Society for Cell Biology (ASCB) Annual Meeting in San Francisco, California, USA.
Title: **Investigating the role of ERK-mediated actin phosphorylation in ciliary dynamics.**

Poster Presenter at the session entitled "ciliary formation and trafficking", at the 2016 American Society for Cell Biology (ASCB) Annual Meeting in San Francisco, California, USA.
Title: **Flagellar length synchronization in *Chlamydomonas*.**

Aug 14th-19th, 2016

Poster Presenter at 'Plant and Microbial Cytoskeleton meeting', Gordon Research Conference, on 'The Mechanics of Building Cells in Plants and Microbes', New Hampshire, USA
Title: **Role of actin phosphorylation in eukaryotic flagellar dynamics.**

Nov 8th -11th, 2012

Poster presenter at 81st annual meeting of The Society of Biological Chemists (India) at Science City, Kolkata, India
Title: Characterization of a novel phosphoesterase protein encoded by gene 66 of mycobacteriophage D29

Jan 8th -11th, 2012

Poster presentation at 2nd International Conference on 'Perspectives of Cell Signaling and Molecular Medicine', Bose Institute, India.
Title: **Characterization of a novel phosphoesterase protein encoded by gene 66 of mycobacteriophage D29**

Nov 24th -28th, 2008

Poster presentation at International Conference on 'A Journey from Plant Physiology to Plant Biology', Bose Institute, India.
Title: **Expression of le-mads 'rin' gene in tomato fruit is required for the expression of ethylene producing genes and many other genes essential for the fruit ripening in tomato**

SKILL and RESEARCH

Area of specialization

Molecular biology, Microbiology, Biochemistry, Cell Biology

Current area of research

- Flagellar dynamics using *Chlamydomonas reinhardtii* as a model organism.
- Actin dynamics based on Chlamydomonas flagellar dynamics.
- MAPK signaling and ciliary length regulation in *Chlamydomonas reinhardtii.*
- Protein expression study in Chlamydomonas during flagellar regeneration

Experimental techniques known

- **General techniques** including isolation, characterization as well as preservation of bacteriophage, bacterial and algal strains.
- **Molecular biology techniques:** DNA isolation (genomic, plasmid, phage), PCR amplification (primer designing and optimization), cloning and expression of genes, chemical transformation and electroporation, RNA isolation, RT-PCR, Southern and northern hybridization, site-directed mutagenesis, DNA sequencing, EMSA, etc.
- **Protein-related techniques:** Biochemical estimation of protein content, spectrophotometric analysis of enzyme activity, protein purification by affinity chromatography, Phos-Tag SDS-PAGE, gel-filtration chromatography, etc., protein profiling by 1D/2D SDS-PAGE, protein identification by MALDI-TOF, Mass Spectrometry, size-exclusion chromatography, pulse-chase radiolabeling, Silver Staining.
- **Cell biology techniques:** MTT assay, study of apoptosis, DAPI staining, FACS, DIC microscopy, Confocal microscopy, Fluorescence microscopy, TIRF microscopy.
- **Immunological techniques:** Raising antibody in rabbits, immunoprecipitation, agar diffusion method, rocket electrophoresis, western blotting, immunocytochemistry.
- **Analytical methods:** UV-Visible spectroscopy, fluorescence spectroscopy, Thin Layer Chromatography (TLC), High Performance Liquid Chromatography (HPLC).
- **OS/Softwares known:** Windows, MS Office, Photoshop, CorelDraw, Graph Pad Prism5, Image J, Adobe Illustrator.
- **Sequence/structural alignments and phylogenetic studies:** BLAST, ClustalW, Protein structural alignment software (Dali), MEGA, Bio-edit, Geneious, Espript, Prot-param, Psi-pred.
- **Homology modelling and structural analysis:** Swiss-PdbViewer, I-Tasser, Pymol.

REFEREES

Brief description of current and previous research work:

Post-doctoral research:

Characterization of a simple and effective method for flagellar length synchronization in *Chlamydomonas*:

Chlamydomonas reinhardtii, a unicellular alga, has served as a model organism for studies of flagellar assembly and function. As flagellar length is regulated by the cell cycle, synchronous culture is advantageous for minimizing variability when studying the biological and biochemical effects of several factors on flagellar length. We have developed a method in which 100 % of cells are synchronized with respect to their flagellar length but not synchronized with respect to the cell cycle. The method requires the regeneration of cells for 3 hours following deflagellation, at which point the flagellar length becomes more tightly distributed and maximally homogeneous. We found that time-limiting new protein synthesis during flagellar synchronization restricts the variability of flagellar length by limiting the variability of unassembled pool of flagellar protein. We have also observed that the method is very effective and efficient in detecting minor changes in flagellar length following chemical and genetic perturbation. The method is easy to perform, requires no changes of media, is less time-consuming than other methods and requires no special equipment. Using flagellar length synchronization, we have increased our ability to study ciliary length regulation and related disease etiologies.

Identification of the novel pathways for ciliary length regulation in *Chlamydomonas* during MAP kinase phosphatase inhibition:

Eukaryotic cilia and flagella play essential roles in signal transduction, cell motility and development. The assembly and maintenance of cilia and flagella requires intraflagellar transport (IFT), a bidirectional, evolutionary conserved motility process. Understanding the regulation of ciliary assembly-disassembly are vital for detecting sources of cilia related disorders. Recent evidences support that the regulation of the ciliary dynamics is controlled by homologues of mitotic proteins and other several kinases and phosphatases. Using *Chlamydomonas reinhardtii* as a model organism, we have found that flagellar dynamics is regulated by a mitogen activated protein kinase phosphatase (MKP). Genetic and chemical perturbation of MKP results in steady-state flagellar shortening. Importantly, flagellar assembly is completely inhibited at regenerating state after MKP-inhibition and accumulation of motor proteins near the basal body can be observed. we have also detected fewer IFT proteins in flagella at steady state in inhibitor-induced MKP condition. Now, we are engaged in investigating the mechanism by which MKP inhibition affects flagellar assembly by disrupting motor-IFT protein interaction. Understanding the ciliary length regulation via MAPK signaling will help us to establish a connection between mitogenic pathway and basic ciliary dynamics. This eventually provides a broader context of understanding into the mechanism of cilia related diseases.

Understanding the role of actin modification in *Chlamydomonas* flagellar dynamics:

Chlamydomonas has two actin proteins, one conventional actin (Ida5) and another highly divergent actin (NAP). Previously, we have identified the relationship between *Chlamydomonas* flagellar dynamics and actin dynamics. During screening of small molecule inhibitors for flagellar length regulator, we have found that a MKP3 inhibitor BCI shortens the length of flagella, activates extracellular regulated kinase (ERK) and leads to increased actin phosphorylation. Our current findings reveal that ERK phosphorylation as well as flagellar shortening started within 15 minutes of BCI treatment and continued up to 120 minutes. Phosphorylated actin appeared within 30 minutes and then decreased gradually in a time-dependent manner. Surprisingly, mutants lacking conventional actin made the BCI-mediated flagellar shortening

more severe. This suggests that the role of actin modification may be to resist the disassembly rather than mediate it. We are working on identifying the phosphorylated residues in ERK activated actin to establish their roles in flagellar dynamics both *in vitro* and *in vivo.*

In a separate phospho-proteomic based study during flagellar regeneration, we have found that NAP is one of the protein that becomes phosphorylated during regeneration. Currently, our lab is engaged in studying the relationship between NAP protein and flagellar dynamics. Our research approach will address the novel role of a fundamental protein's modification to a basic biological process and lead us to identify the therapeutic targets for cilia related disease.

Doctoral research:

Isolation and characterization of a Calcineurin-like protein phosphatases of mycobacterio-phage D29 and mycobacteriophage Wildcat:

Mycobacteriophages are the cornerstones of mycobacterial-genetics but still a clear picture of mycobacteriophage infection pathway is not available. One of the approaches might be characterizing the genes of mycobacteriophages to get a molecular view of how these phages might operate inside the host. During my doctoral research, I have studied a phage-encoded protein named Gp66, a novel calcineurin-like phosphoesterase protein of mycobacteriophage D29. In that study, we have purified the recombinant version of the protein to near homogeneity and characterized it biochemically and functionally. The main finding is that the protein along with other related mycobacteriophage proteins forms a novel distinct clade among the calcineurin-like phosphoesterase proteins. Moreover, Homology modeling and sequence alignment reveals that Gp66 shares considerable sequence and structural similarities with phosphoesterases from *Aquifex*, a deep-rooted eubacterium and *Methanococcus*, an archaeon. The protein was found to be highly active on cyclic nucleotide monophosphates particularly 2', 3'-cAMP, a product that formed during RNA degradation and repair. Gp66 is actively synthesized during the mid-phase of D29 growth cycle and a surge of 2', 3' cyclic phosphodiesterase activity was found approximately in the late stages of growth in phage-infected cells. This suggests that the protein may play a key role in phage development. On the contrary, it appears that when Gp66 activity reaches its peak, growth of the host and phage halts, though transiently. Inhibition of Gp66 activity by dominant negative mutant (Gp66N80A) resulted in increased phage as well as host proliferation. Thus, it is indicated that the protein negatively regulates phage growth.

I have also identified another calcineurin-like phosphoesterase protein (WCGp137) from a singleton mycobacteriophage Wildcat, which shares only <50% sequence similarities with other mycobacteriophages. In that study, we have found that this protein fits to the same clade where Gp66 belongs. After biochemical characterization of WCGp137, we concluded that the protein has some unique properties and thus considered as a novel calcineurin-like phosphoesterase protein.

Investigating the role of protein phosphorylation and dephosphorylation in the growth of mycobacteriophages:

Phosphorylation and dephosphorylation play a significant role in cellular metabolism as well as development. Bacteriophages are known to regulate the activity of their hosts through phosphorylation and can take full control of the host metabolic pathways. Phosphorylation and subsequent dephosphorylation can be achieved by various phage-encoded proteins. Similar phenomenon may also be happened in mycobacteriophage. During my Ph.D., I was involved in studying the proteome profile of mycobacterium host before and after phage infection. Our study was mainly focused on investigating the role of mycobacteriophage phosphatases in details to establish some novel mechanisms by which mycobacteriophages manipulate their hosts to satisfy their own needs.