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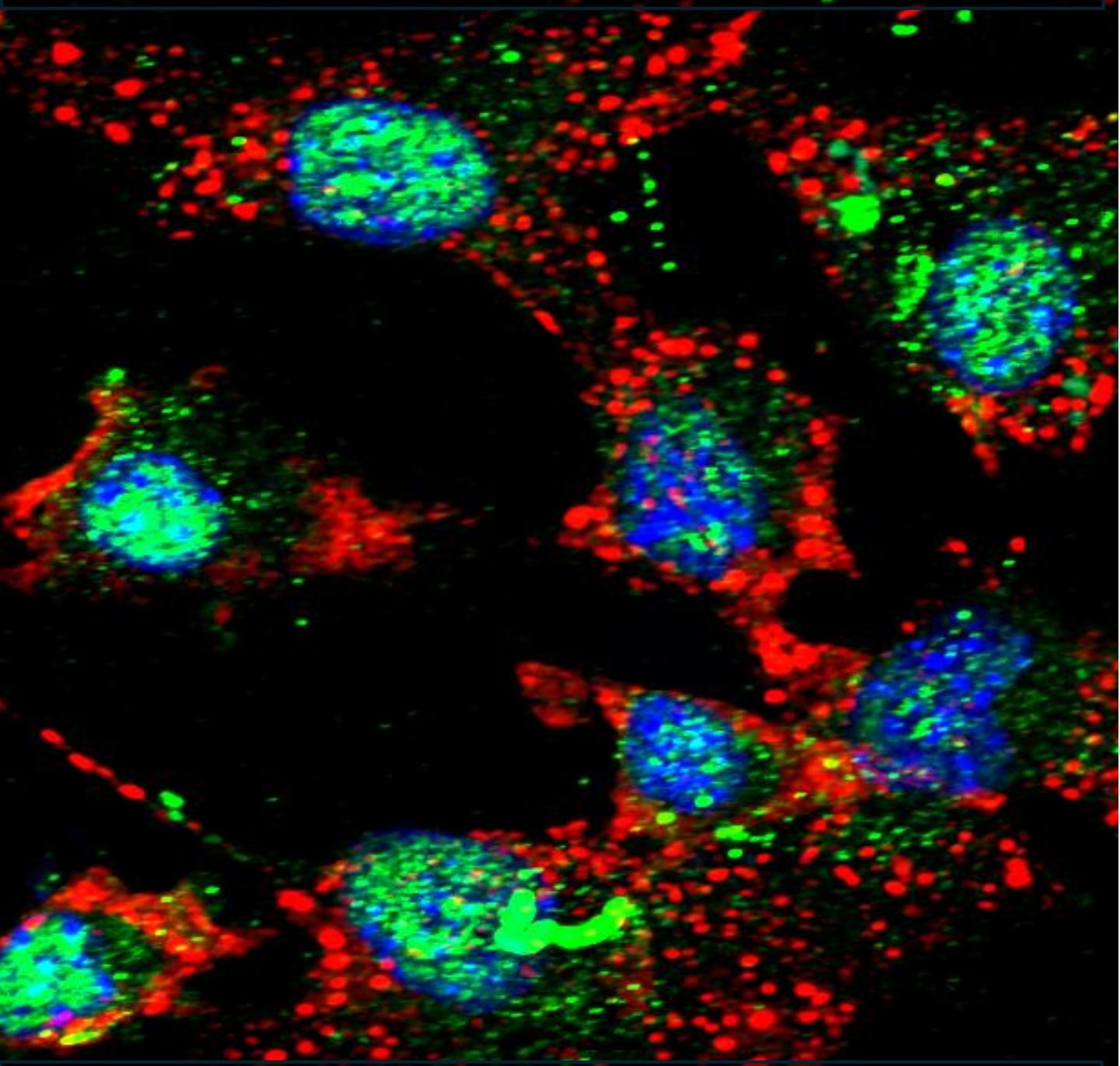
ISCB NEWSLETTER



Indian Society of Cell Biology (Regd.)

Table of contents

Welcome Note by the President

Dear Colleagues, Fellow Cell Biologists, and Young Students,

It gives me immense pleasure to address you through this newsletter—my first as the President of the Indian Society of Cell Biology (ISCB). I am both honoured and humbled to take on this responsibility, standing on the shoulders of the many illustrious mentors and distinguished scientists who have shaped the legacy of this society over the decades.

The ISCB has always been a vibrant forum for nurturing scientific exchange, encouraging young talent, and promoting excellence in cell biology research across India. As I step into this new role, I am deeply aware of our responsibilities: to keep our community inclusive, scientifically bold with high ethical standards, and future-ready. The success of the 47th All India Cell Biology Conference at Bhubaneswar underscored the power of shared inquiry and dialogue. We witnessed outstanding scientific advancements through the National symposium, collaborative workshops, and fruitful partnerships.

This issue of the ISCB newsletter introduces our newly elected office bearers and celebrates the diversity of research and voices within our community. From insightful articles from scientists, who embody the spirit of inquiry and innovation, to thought-provoking puzzles, each contribution reflects the energy and commitment that defines ISCB. We are also announcing the upcoming **48th All India Cell Biology Conference and Symposium at IIT Kanpur**, which has garnered considerable excitement within the academic community. This event convenes a diverse group of researchers, ranging from early-career scientists to established leaders, fostering an engaging environment for exchanging innovative and scholarly ideas.

As we move forward, we hope to foster greater engagement among members, strengthen national and international collaborations, and expand our outreach to school and college students and early career researchers. The field of cell biology continues to evolve rapidly, and so should we—as individuals, institutions, and as a society.

I look forward to working closely with our executive team and each one of you to build on ISCB's legacy and steer it towards an even more impactful presence.

With warm regards,
B.K. Thelma
President, Indian Society of Cell Biology

Foreword note by the Secretary

Dear Fellow Cell Biologists,

It is our pleasure to present the second issue of the ISCB Newsletter for the year 2025.I extend my heartfelt thanks to our President, Prof. Thelma, for graciously sharing her journey as a teacher and researcher, along with her insightful reflections on the Society. The full interview is featured on Page 7. I also thank all the contributing authors whose articles enrich this issue. In the "Research News" section, we feature several articles highlighting recent findings from various research groups across the country. The "*Classroom Resources*" section includes a valuable article on teaching methodologies. Special thanks to my colleagues Rashmi and Deepashree for conducting the interview with the President, and to Arveen and Kavikumar for their delightful cartoons, sketches, and puzzles featured in the "Fun Time" section. I am confident that this collection will make for both an informative and enjoyable read.

We are pleased to announce the renewal of our partnership with **Cactus Communications Private Limited (CACTUS)**, for the Society's outreach activities. As part of the **ISCB–CACTUS collaboration**, we plan to organize webinars and science communication workshops, detailed on Page 4. These events are open to all interested participants, and I encourage you to share this information with your friends and colleagues. We believe these initiatives will be especially beneficial to our student community. Our sincere thanks to CACTUS for their continued support and contributions, including two articles in this issue that serve as a preview of the upcoming activities.

We are also delighted to announce the **48th All India Cell Biology Conference and Symposium**, the Society's flagship annual event, which will be hosted by the Indian Institute of Technology Kanpur from **December 7–9, 2025**. With the theme "From Cell to Therapeutics," the conference will emphasize the pivotal role of fundamental biology in driving innovation. In addition to invited talks by leading experts, the conference will feature dedicated student sessions and ample time for poster presentations to foster active engagement. Several awards will be presented to students for both oral and poster presentations. We look forward to welcoming you to this inspiring gathering. Registration is now open, and further details can be found on Page 5.

Finally, I would like to express my sincere gratitude to Deepashree for her editorial assistance and for designing this issue of the Newsletter.

Warm regards,
S.Ganesh
Secretary, Indian Society of Cell Biology

Email: Sec.ISCB@gmail.com

New Office Bearers of the Society (2025-27)

Dr. Bittianda K Thelma
President

Dr. Sathees Raghavan
Vice President

Dr. Girish Ratnaparkhi
Vice President

Dr. S. Ganesh
Secretary

Dr. Bushra Ateeq
Joint Secretary

Dr. Nitin Mohan
Treasurer

Dr. Madhu G Tapadia
Executive Secretary

Dr. Debasmita P. Alone
Executive Member

Dr. Shaida Andrabi
Executive Member

Dr. Monisha Banerjee
Executive Member

Dr. Rashna Bhandari
Executive Member

Dr. Uma Dutta
Executive Member

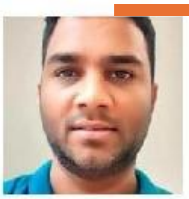
Dr. Nishigandha Naik
Executive Member

Dr. Utpal Nath
Executive Member

Dr. Gaurav Kumar Pandey
Executive Member

Dr. Mahak Sharma
Executive Member

Dr. Anand K. Tiwari
Executive Member



Dr. Anand Kumar Singh
Executive Member

Dr. Bhavana Tiwari
Executive Member

ISCB and Cactus Communications Join Forces to Empower India's Scientific Community!

The **Indian Society of Cell Biology (ISCB**) is proud to announce a dynamic partnership with **Cactus Communications Private Ltd**, a global leader in science communication, to launch a series of academic webinars designed to enrich and empower India's research ecosystem. This collaboration marks a significant step toward bridging scientific excellence with impactful communication, offering ISCB members and the broader research community access to world-class insights, mentorship, and professional development.

From October 2025 through February 2026, this joint initiative will host four engaging online events featuring renowned scientists, thought leaders, and communication experts. The series will spotlight cutting-edge research, career guidance, and the art of scientific storytelling—creating a vibrant space for learning and exchange.

🗓️ **Webinar Schedule**

•**Seminar 1**: *Prof. S.C. Lakhotia* (BHU, Varanasi)

🗓️ 13th October 2025 | 🕔 5:30–6:30 PM IST

•**Seminar 2**: *Prof. Jyotsna Dhawan* (CCMB, Hyderabad)

🗓️ 15th November 2025 | 🕔 5:30–6:30 PM IST

•**Panel Discussion**: *Navigating the Early Career Maze*

🗓️ 21st January 2026 | 🕔 5:30–6:30 PM IST

🎙️ Panelists:

- Dr. Rashna Bhandari (CDFD, Hyderabad)
- Dr. Indranil Banerjee (IISER Mohali)

•**Seminar 3**: *Visualizing Science: Enhancing Research Communication Through Visuals*

🗓️ 18th February 2026 | 🕔 5:30–6:30 PM IST

🎤 Presented by Cactus Communications

🎓 **Who Should Attend?**

Open to all, the series is especially curated for undergraduate and postgraduate students, postdoctoral fellows, and early-career researchers seeking inspiration, guidance, and community.

🌟 **Why It Matters**

This initiative reflects ISCB's enduring commitment to advancing cell biology education and outreach. By joining hands with Cactus Communications, ISCB is expanding its impact—fostering a culture of scientific excellence, collaboration, and communication across India's research landscape. Stay tuned for registration details and updates. Let's shape the future of science, together.

[Moderator: Dr Debdeep Dutta: *ddutta@iitk.ac.in*]

Indian Society of Cell Biology (ISCB)

48th All India Cell Biology Conference and Symposium

"From Cell to Therapeutics"

Date: December 7 to 9, 2025

Venue: Indian Institute of Technology Kanpur

Deadlines:
Abstract Submission: August 1 to September 15, 2025
Registration: August 1 to September 15, 2025

Satellite Workshop on Advanced Microscopy

December 10 to 11, 2025 | For details, check website

More information

Organizers

S Ganesh
Nitin Mohan
Rakesh K. Majhi

Email: cellbio.iitk@gmail.com

https://home.iitk.ac.in/~sganesh/aicbcs/

48th All India Cell Biology Conference and Symposium

December 10 to 11, 2025 | Venue: IIT Kanpur

Invited Speakers

Thelma B K, DUSC Delhi
Kaustuv Sanyal, Bose Institute Kolkata
Santosh Chauhan, CCMB Hyderabad
Chayan Kanti Nandi, IIT Mandi
Manish Jaiswal, TIFR Hyderabad
Kalika Prasad, IISER Pune
Indranil Banerjee, IISER Mohali
Amitabha Bandyopadhyay, IIT Kanpur
Jyothilakshmi Vadassery, NIPGR Delhi
Subramaniam K, IIT Madras
Rajiva Raman, BHU Varanasi
Utpal Nath, IISc Bengaluru
Pradip Sinha, IIT Kanpur
Prem K Yadav, CDRI Lucknow
Chandrasekhar K, Gothenburg Univ
Jomon Joseph, NCCS Pune
Gaurav Ahuja, IIIT Delhi
Mahipal Ganji, IISc Bengaluru
Nihar Ranjan Jana, IIT Kharagpur
Bratati Kahali, CBR IISc Bengaluru
Alok Krishna Sinha, NIPGR Delhi
Kartik Sunagar, IISc Bengaluru
Samir K Maji, IIT Bombay
Sivaprakash K Ramalingam, IIT Kanpur
Manoj Prasad, DUSC Delhi
Mukesh Jain, JNU Delhi
Dhiraj Kumar, ICGEB Delhi
Murty Srinivasula, IISER TVM
Ganesh P Namasivayam, Kyoto Univ.
Mathivanan Jothi, NIMHANS, Bengaluru
Rita Mulherkar, BKLW Hospital, Ratnagiri

Student Awards

[for Oral and Poster Presentation]

Dr B R Seshachar Memorial Prize
Dr C.M.S. Dass Prize
Dr S.R.V. Rao Prize
Dr V.C. Shah Prize
Dr A.S. Mukherjee Memorial Prize
Dr A.N. Bhisey Prize
Dr Manasi Ram Memorial Prize
CACTUS Prize
ISCB Prize
Organizer's prize
and a many more!

Don't miss!

A few abstracts submitted by non-student members for the poster presentation will also be selected for oral presentation!

This conference will serve as adynamic hub for networking, skill-building workshops, and engaging product demonstrations, providing attendees with invaluable opportunities to learn, forge connections, and explore new scientific advancements — all within one of the finest technology institutes in the country!

https://home.iitk.ac.in/~sganesh/aicbcs/

Interview with Professor B.K. Thelma: Key Insights

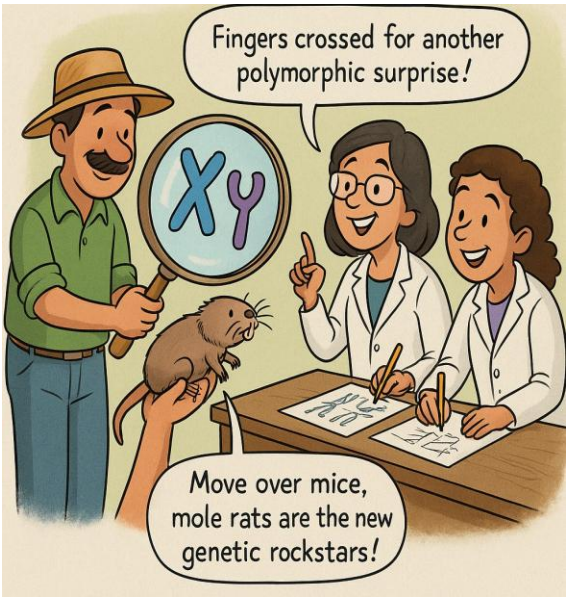
Prof. B.K. Thelma
President, ISCB
National Science Chair, Department of Genetics, University of Delhi
Email: *thelmabk@gmail.com*

Interviewers: Rashmi Parihar and Deepashree Sheshadri

From an Aspiring Doctor to a Genetics Pioneer: The Journey Begins

Rashmi Parihar: Thank you very much, Professor Thelma, for giving us your time. Let's begin at the very start. Could you take us back to your student days—was there a particular moment or experience that sparked your interest in genetics and cell biology?

Professor Thelma: When I was a schoolgirl, I yearned to be a medical doctor. Being underage when I finished Pre-University, I couldn't get into medical college. That led me to pursue a B.Sc and as per everyone's advice a Master's thereafter, and the natural progression was to a Ph. D. By then, I already had a clear idea of the area I wanted to work in—human genetics—but in those days, mid 70s, labs working in that area were quite rare. After my master's from Bangalore University, I got a fellowship at the Department of Zoology at Delhi University, which had a cytogenetics lab led by Professor S. R. V. Rao. When I expressed my desire to work in human cytogenetics or any medical genetics-related field, to Professor Rao, he advised me that clinical collaboration was challenging due to sample scarcity—even his own MD student struggled with that. Instead, he encouraged me to train thoroughly and build my foundation in cytogenetics using an animal model—a mole rat with interesting highly polymorphic X and Y chromosomes, then pursue human genetics during my postdoctoral work. That's how I started my Ph. D and continued into research—shifting gradually from mammalian cytogenetics to other areas as the field evolved, new technologies became available, and opportunities came my way.



Deepashree: What were the major challenges you faced as a young researcher? How did you overcome them?

Professor Thelma: To be honest, I didn't face any major challenges in those early years. The Zoology Department at Delhi University was among the best at the time. I had an excellent mentor, and the lab was already strong and self sufficient in animal cytogenetics. Our responsibility was to identify contemporary scientific questions and pursue them. The environment was very conducive to doing good science.

Rashmi Parihar: It is really good to see such a positive attitude that you have towards everything. As you already mentioned, you hold your mentor in high regard and appreciate him a lot.

Is there any specific story or memory that left a deep imprint on your career path, either from him or from any other mentor?

Professor Thelma: In terms of mentorship, during my school days—coming from a small town in Coorg—I didn't really have any specific mentor actively guiding and telling me, "do this" or "do that." It was during my Master's that I got some exposure to research, but we weren't deeply involved in lab work but limited to largely classroom learning. It was only when I came to Delhi University for my Ph. D with Professor Rao that I had real mentorship. Every day in the lab was a great learning experience. He instilled in us that how you do your work is more important than what you do. The lab culture was also unique—we used to joke that "anytime is teatime," which really meant engaging discussions were always happening, often pushing actual lab work into later hours. Every time the gardener caught a mole rat and brought to the lab was super exciting as the animal colony would get bigger and more importantly, we wouldn't know if we land up with yet another polymorphic X or Y chromosome discovery.

As far as any other specific incident, one is vivid- I was carrying a microscope—a small one—to the culture room, and it slipped from my hands and fell. I came out and told Professor Rao, expecting to be blasted. But he just said, "Don't worry, these things happen." That moment left a lasting impression on me. Most people would've shouted, "What have you done?" The other wonderful thing was- obviously I didn't have any publishable/presentable work in the first year of Ph. D. Yet, Professor Rao insisted that I attend the Cell Biology meeting, which was held at Banaras Hindu University (BHU) that year. He always said, "Paper or no paper, you have to attend these annual meetings," because much learning happens from listening and interacting with others. I preached that to my mentees too. Mentors back then were different, and so was science. It wasn't as competitive or fast paced as it is today. It was still good quality exciting science, but with a gentler pace. Getting journal reprints, for instance, involved writing to the authors and receiving them by surface mail. Doing a Ph. D back then—nearly 50 years ago—was a completely different experience, a different world, really. If we were complaining about the experiments not working or other difficulties, Professor Rao would say- Don't tell the world about the labour pains, show them the baby. You sure know what that means. His support wasn't limited to the lab and that made a huge difference to students like me who moved to Delhi from the other end of the country.

Reflections on Scientific research and Collaboration

Deepashree: Your research journey spans from foundational studies to translational genomics. Was this shift planned, or did it evolve over time?

Professor Thelma: Looking back, it was indeed a very natural growth. I was always interested in medical genetics or anything to do with human genomics—though the term "genomics" didn't really exist back then. When I was working on the mole rat, we identified fragile sites on the X and Y chromosomes. These fragile sites were located in the heterochromatic regions and resulted in specific deletions, and we later found that they were folate sensitive

My colleague, Dr. Rita (now a Professor at Nottingham university), confirmed this in lymphocyte cultures from this mole rat. Around that time, studies in humans had already revealed that a small proportion of inherited X-linked intellectual disability was associated with such folate-sensitive fragile sites. We were excited to find similar properties in our animal model and they had to be characterised.

Around then I moved on for my postdoctoral work at the Children's Hospital in Basel, Switzerland, where I shifted to human genetics and worked on several projects. One was on human male infertility where we dealt with unexplained cases and used in vitro fertilization (IVF) models with Chinese hamster eggs to study human sperm chromosome abnormalities, if any, which could be the contributing factor to infertility.

There was another project on familial cancers that I was loosely involved in. These early experiences marked my first steps towards translational research. When I returned to India, I had no job lined up but was determined to continue my research. I rejoined Professor Rao's lab as a pool officer and got back to working on mole rats. I was fortunate to get a Lecturer's post in the Department of Genetics, South Campus, Delhi University, soon after. My first two Ph. D students worked on characterizing the fragile sites in the mole rat. By then, molecular biology had started to gain ground in India. I must mention that a few months after my return as a post-doc, I attended the first molecular biology workshop at BHU. Faculty came from UK and included eminent scientists like Dr. Ken Jones, Dr. Lalji Singh, Dr. Ed Southern. We learned the basics of recombinant DNA techniques there, including Southern blotting for the first time.

The real turning point for me came with the discovery of the FMR1 gene in 1991 by the Dutch group led by Dr. Ben Oostra. They showed that Fragile X Syndrome in humans was caused by trinucleotide repeat expansions in this gene. We immediately wrote to Dr Oostra for the probe, and he happily shared it. That along with Southern blotting expertise in the lab allowed us to begin work on Fragile X screening in India. My first clinical collaborator, Professor Meena Gupta from G.B. Pant Hospital, joined me on this journey. We would go to schools for special children during parent-teacher meetings to educate families about Fragile X Syndrome. We used pictures of affected Western children to explain the features, as there were no known genetically diagnosed cases of fragile X from India at the time. The first child we suspected and tested for the expansion mutation turned out to be positive, and there was no looking back since. We received one of the earliest DBT grants in 1993 for Fragile X diagnosis, screening and counseling. Samples poured in from across the country. It had taken nearly 20 years to complete that cycle from starting my Ph. D working on a rodent system to establishing human Fragile X testing, and making a direct societal impact.

Rashmi Parihar: Since you've worked closely with many patients and families over the years, is there any specific incident, discovery, or story that really surprised you or left a strong impact?

Professor Thelma: In terms of Fragile-X, there wasn't anything very different from what was already reported—we knew that the mutation had to be present, and that the premutation would pass through the carrier mother and sometimes become a full mutation in the next generation. But what really was disturbing were the social situations we encountered. There were parents who knew their son had Fragile-X Syndrome, was intellectually challenged and yet they insisted on getting him married—because, in their view, the family line must continue. These were not unusual cases in our country. Despite having a diagnosis and offering genetic counseling, families often stuck to deeply rooted cultural expectations. We had many cases where couples underwent prenatal diagnosis for Fragile-X mutation. Unfortunately, some had repeated pregnancies with a fetus carrying the full mutation

While a negative result brought relief, these recurring cases were emotionally difficult. Even more complicated were the situations involving carrier daughters. Mothers would come and ask, “Should we tell the family she’s marrying into that she’s a carrier?” This was the early 1990s, but even today, the awareness and social comfort around disclosing such information remain limited. One example that stands out involved a family with four boys affected by a syndrome. The second son was a little more functional than the others, so the family got him married. The girl he married was an orphan, likely seen by her guardians as someone they could “give away.” We had already counseled them, but families have their autonomy to decide.

There have been exciting scientific moments too. The first one that we were really thrilled with was the identification of a gene causing inherited intellectual disability.

Professor Madhulika Kabra from AIIMS and I were collaborating, and she referred a family from Chandigarh with multiple affected boys. They showed Fragile X-like features but tested negative—no trinucleotide expansion, no point mutation. This was ideal for discovery genomics. Professor Kabra along with my Ph. D student Thenral, flew to Chandigarh to examine affected individuals and collect samples. This was much before next-generation sequencing (NGS) era. Using microsatellite markers, we mapped the linkage region to the X chromosome—about a 50 Mb span with 400 candidate genes. We couldn’t have possibly screened all 400 genes with conventional PCR-Sanger sequencing, so while we were prioritizing, fortunately for us, NGS technologies became available. Although targeted sequencing was extremely expensive (about ₹1.2 lakhs per sample, only available in the U.S.), we carefully selected informative family members and got them sequenced for the targeted region and identified the disease-causing gene variant. This is probably one of the earliest examples of NGS for discovery genomics in India. Our submission of this discovery to the OMIM (Online Mendelian Inheritance in Man) database in 2014 earned MID2 a new entry—our very first OMIM-recognized gene. A few more such discoveries stemmed from our Fragile X screening/testing program.

Deepashree: Your work extends beyond Fragile X to complex disorders like Parkinson’s and schizophrenia. What drives your interest in these conditions?

Professor Thelma: Thank you for asking such thoughtful questions. Our work on schizophrenia actually began quite organically. When we received the DBT grant for Fragile X—which involved diagnostics, screening, and counseling—the DBT officer suggested I collaborate with a trained counselor, since we weren't formally licensed to counsel. He recommended Dr. Smita Deshpande, a psychiatrist at Ram Manohar Lohia Hospital. When I approached her, she kindly agreed to counsel our families but she also had a request—she asked if I would collaborate with her and team on the genetics of schizophrenia. They had many families with schizophrenia and were looking for a geneticist. Of course, I agreed or rather jumped. In a university setting, we try to avoid overlapping projects among Ph. D students to prevent unhealthy competition. So, while we spread ourselves thin, taking on multiple disorders, we also gained breadth. This was a golden opportunity to transition from studying neurodevelopmental disorders to neuropsychiatric ones. We received our first schizophrenia grant through an Indo-US Fogarty collaboration. We had a committed team, including a trained psychologist Dr. Triptish Bhatia who helped recruit families and collect data. She would return from the field with extensively filled diagnostic (and family) interviews for genetic studies and blood samples. Soon, our lab scaled from handling a few dozen samples to hundreds. This growth required dedicated staff for tasks like DNA extraction and storage. Initially, we relied on candidate gene association approach.

Since dopamine is central to schizophrenia, we focused on genes involved in its synthesis, transport, metabolism, and reuptake. On the other hand, we expected families with schizophrenia to follow Mendelian inheritance, but those turned out to be rare—less than 1%. Most were oligogenic, with multiple heterozygous variants contributing to small effects. But these findings added notably to genetics of schizophrenia. Dopamine is also central to Parkinson's disease, but in an opposite trend, and so we moved on with these two phenotypes. Using SNP data from the Human Genome Project, which was publicly available just around that time, and with a timely, generous funding support from DBT, we conducted modest-sized association studies, keeping with the time and trend.

We were fortunate to have the appropriate samples when next-generation sequencing technologies became available a few years after. That's why I often say: timing, being prepared, and luck all matter. Having well-characterized samples and families allowed us to use these new techniques immediately. This led to discoveries across a spectrum of brain disorders—from intellectual disability to schizophrenia and Parkinson's. So, while I say I've spread myself thin, I also had the opportunity to study a wide range of diseases which spanned neurodevelopmental, neuropsychiatric, and neurodegenerative domains—completing the full arc of brain-based genetics research.

Rashmi Parihar: You have championed your research in neurodevelopmental to neuropsychiatric and then neurodegenerative disorders. Not only this, you've also integrated genomics with pharmacogenetics and Ayurgenomics. So how do you personally bring these things seemingly diverse sets together? Is there a framework you follow or do insights emerge more organically?

Professor Thelma: When working with schizophrenia, a small proportion of patients on antipsychotic medication developed tardive dyskinesia—an iatrogenic disorder resembling Parkinsonism. We wondered if the issue was in dopamine metabolism or CYP family genes and whether pharmacogenetics could help predict responders and non-responders. Based on the individual's metabolizer status (rapid, intermediate, slow), a drug dose may be too low or too high. We partnered with an Israeli group through Indo-Israel funding to study tardive dyskinesia and began SNP-based candidate gene association studies. This was before exome sequencing era. Soon after, during a DBT brainstorming meeting, a well-known rheumatologist Professor Ashok Kumar from AIIMS raised a clinical problem to be addressed: only 40–50% of rheumatoid arthritis patients respond to methotrexate, the first line of treatment and it takes six months to determine non-responder status. This led us to investigate pharmacogenetics of methotrexate. As we moved on, we transitioned from candidate gene studies to genome-wide association studies (GWASs). In 2008, we proposed a center of excellence project for GWAS across five complex diseases. The committee limited us to work on two diseases, and we focused on rheumatoid arthritis and ulcerative colitis, both inflammatory conditions. These first GWASs yielded India-specific gene signatures, which encouraged us to carry out functional genomics, identify druggable targets, etc. However, despite technological advancements, large-scale association studies and more, predictive and preventive medicine in complex disorders remains a continuing challenge and a distant aim. Addressing phenotypic and genetic heterogeneity in this group of disorders seemed (and seems) beyond the reach of available tools and technologies. Association studies were at their peak, but with poor replication across studies.

Around this time, I met an excellent Ayurveda physician and learned about the concept of 'Prakriti'—the Ayurvedic method of deep phenotyping and subgrouping of individuals-healthy and with disease, in ways modern medicine doesn't.

Coining the term 'Ayurgenomics' in 2008, the plan was to combine the prakriti concept or deep phenotyping principles of ayurveda with genomics to get better insights into genetics of complex traits. We received AYUSH funding in 2008 to explore rheumatoid arthritis in different Prakriti types (Vata, Pitta, Kapha). SNP association studies showed that inflammatory markers dominated in Vata, oxidative stress markers in Pitta, and a mix in Kapha. This matched Ayurveda's description, and we published this in 2012. I continue to work on Ayurgenomics and am convinced that this is a promising paradigm for actionable genetic leads for complex disorders.

Deepashree: We understand from your recount of your scientific journey that one project led to another to another, but what has been the most satisfying to you or made you feel that you have realised your dream?

Professor Thelma: Thank you very much, I honestly feel that the answer to this one question would summarise the pursuit of my passion and the satisfaction that I have been able to do my little bit for science for society. My very first project in human genetics on the fragile-X syndrome diagnosis and the service we provided to so many individuals and families for the first time in the country was the foremost fulfilment of my desire to contribute to predictive and preventive medicine. Then came disease causal gene discoveries and more through the years adding to the disease genetic testing panels, etc. The most fulfilling/satisfying was however my debut with newborn screening (NBS) for inborn errors of metabolism. With the magnanimous vision of Professor C N R Rao and unstinted support from Dr T Ramasami, I could with Dr Seema Kapoor and several clinical collaborators carry out a prospective study on 200,000 newborns across Delhi state hospitals. We not only demonstrated the feasibility of NBS but generated the first ever epidemiological data for over 40 inborn metabolic disorders all with just a drop of blood from heel prick of the newborn. NBS is indeed truly a translational effort for the people and the best example of predictive and preventive medicine, globally ongoing but barely started in India. Every newborn deserves a healthy life and benefits of science must reach every citizen. Therefore, this data to policy research effort is indeed one which I am really happy about and hope that sooner or later pan India data would be generated and NBS becomes mandatory in the country.

Vision as ISCB President

Deepashree: We've talked at length about your scientific interests, but due to time constraints, we'd now like to hear more about your leadership roles. We understand that you've had a long association with the Indian Society of Cell Biology. Could you share a bit about your early involvement?

Professor Thelma: Absolutely. The first meeting I attended was way back in 1977. Except for times when I was abroad, I continued to actively participate in the annual meetings. At the time, as I mentioned before, I was working on mammalian cytogenetics. In fact, most of the founding members came from genetics and cytogenetics background. Broadly speaking, there were four main groups in the country then—Calcutta (largely focused on Drosophila), Banaras (doing both mammalian and Drosophila work), Mysore (Drosophila), and Delhi (primarily mammalian cytogenetics). For me, cell biology meetings were not just academic events. Every annual meeting felt like an extended family reunion. We all knew each other—every mentor, every student, every lab, and the environment was incredibly collegial. It wasn't just about academic learning; it fostered strong interpersonal relationships and resource sharing. I valued these meetings deeply. As my work shifted towards human genetics, I naturally became more involved with the Indian Society of Human Genetics. However, that happened much later, because ISHG was then largely dominated by the medical community, and it was more challenging for basic researchers to enter. Still, my connection with the cell biology community remained strong and this is sure evident now.

The camaraderie and academic enrichment from those early cell biology meetings left a lasting impression. That's why I always encourage my students to attend these meetings as these are good opportunities to meet like-minded individuals, build collaborations, and learn immensely. You never know which interaction will open new doors. Today, subject boundaries are blurring—cell biology, human genetics, and other fields are integrating rapidly. You can't talk only about microscopy or one niche area anymore. We must embrace interdisciplinary approaches to tackle complex questions in biology. Cell biology, too, is evolving to meet that challenge. I see this integration as essential, as the future lies in breaking silos and collaborating across disciplines. That's how we'll decode the complexities of human and organismal biology.

Rashmi Parihar: We could feel from your expressions how deeply associated you are with the Society of Cell Biology. As the current president of ISCB, what is your larger vision for taking the society to its next phase?

Professor Thelma: I believe that regardless of the society we belong to or affiliated with, scientists will continue their work—research, experiments, discoveries. But as a society, we have a broader role. We must enable interdisciplinary research, translatable to some extent, increase public outreach, and, most importantly, spark interest in science among young minds. Many young people today don't realize the daily excitement and thrill and the opportunities that scientific research offers. So, one of my priorities would be to share that enthusiasm widely—outside our labs and conferences. I'd also like to initiate teacher training programs as a part of the outreach activities.

In today's context, scientific networking plays a pivotal role. While societies may not directly orchestrate collaborations, they can foster platforms where people meet, learn, and exchange ideas. ISCB should remain vibrant by spotlighting exciting research and making sure that exposure is for a diverse audience. These aren't entirely new ideas—some of them are already happening. But we need to do more and amplify the message of the beauty of doing science.

Motivation and Balance

Deepashree: After all these years, what keeps you motivated to continue in academia and research?

Professor Thelma: Every single day in the lab feels new. If I were born again, I would still choose to be a scientist. The intellectual satisfaction, the unknowns we face and solve—are endlessly rewarding. We'll never solve all the mysteries of biology in our lifetimes, and that itself is a motivation. Enthusiasm and integrity in our approach will carry us through challenges. If you have a goal and stay focused, you'll find an appropriate and ethical way to get there or if I may say the path unfolds.

Rashmi Parihar: With so many responsibilities—as a researcher, leader, mentor, and administrator—how do you maintain balance and find time for yourself?

Professor Thelma: Oh, my early mornings — about two to three hours—are only mine. That's sacred time for yoga and meditation. No phones, no disturbance. Occasionally, I attend meditation retreats to recharge. Early morning over, the rest of the day is work time or time for others. There's no fixed 9-to-5 in our way of life—it could be a 20-hour day sometimes. It's about fulfilling responsibilities with sincerity, whether that's mentoring students, serving on scientific committees, or performing other roles. To do justice, you must give your 200%—and that takes time.

Balance: from chakras to chromosomess.

Deepashree: Is there a particular mentor or philosophy that has shaped your leadership or mentorship style?

Professor Thelma: I live by two principles: faith and surrender. Faith that whatever is best for you will happen, and surrender to that process, but giving your best all through for whatever you have to do.

Advice to Young Scientists

Rashmi Parihar: You've already shared so many insights, but as a closing note, what advice would you give to young Indian students or early-career scientists aspiring to work in genetics or cell biology?

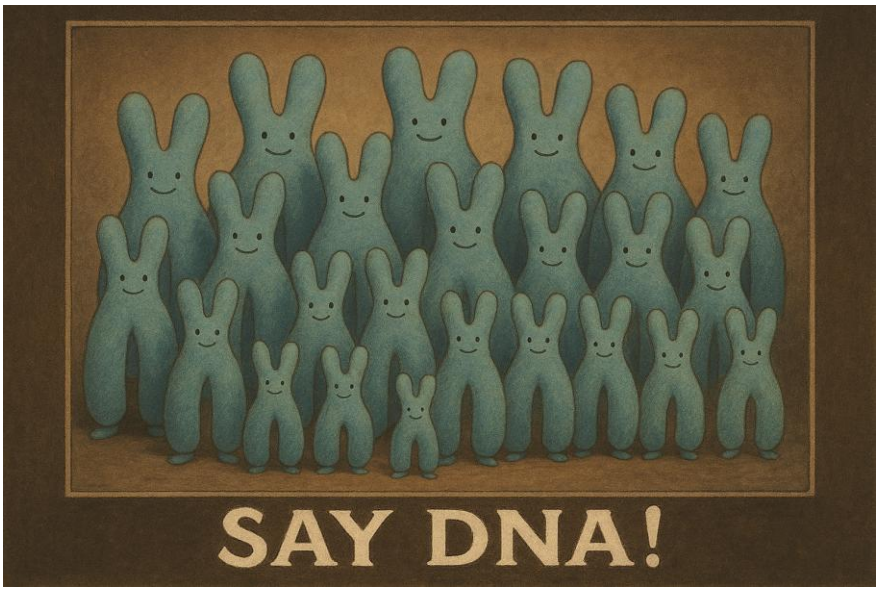
Professor Thelma: If you feel a calling to give something back to the society, science is just one way of doing it—especially for health and well-being. Before choosing a field, take your time. Meet people in that domain and discuss, read deeply, and assess whether you have the commitment, patience and passion to pursue the aims (and, in a lighter vein if it would fill your pocket the way you wish it to). That introspection is critical. Most health conditions, including lifestyle-related ones, have a genetic underpinning and you can't change that, and diseases will occur. For aspiring health scientists, in the context of your question, I see there are two core domains: prediction and prevention, which are genetics-driven, and treatment and cure, which rely heavily on understanding the biology. The latter is where cell biology plays a huge role. To sum it up, if you're passionate, patient, and committed, science will be rewarding—intellectually and through its societal impact. Choice is yours.

Deepashree and Rashmi Parihar: Thank you very much, Professor Thelma. This has been such an engaging and enriching conversation.

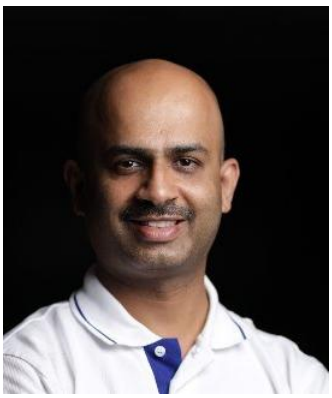

Rashmi Parihar and Deepashree Sheshadri serve as Research Establishment Officers at IIT Kanpur.

Fun time

Family Photograph! (22+XY)



Stop codon recoding: Physiological significance and potential therapeutic applications



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The genetic code was deciphered in the 1960s through the pioneering work of scientists such as Har Gobind Khorana, Marshall W. Nirenberg, and J. Heinrich Matthaei. It consists of 61 codons that specify the 20 standard amino acids, along with three stop codons (UAG, UGA, and UAA) that signal the termination of translation. Genetic code is nearly universal, with most studied organisms adhering to the same codon assignments [1]. However, deviations from the standard code, known as codon reassignments, have been well documented in specific organisms and contexts. These reassignments often involve stop codons being interpreted as sense codons. For instance, UGA can encode the amino acid selenocysteine in mRNAs that contain a selenocysteine insertion sequence (SECIS) in their 3' untranslated regions (UTRs). Additionally, under certain conditions, stop codons can be recognized as sense codons by the translational machinery, allowing translation to continue until the next available stop codon. This phenomenon is referred to as stop codon readthrough (SCR), a form of translational recoding (Fig. 1) [2].

The process of SCR has been observed in all domains of life, from prokaryotes to humans, even in viruses. Our laboratory is interested in identification of novel SCR events in mammals, and the characterization of the mechanism and the physiological significance of these events. In the past ten years, we have experimentally investigated SCR in the following mammalian mRNAs – AGO1, MTCH2, NNAT and FEM1B [2-6]. Using computational tools to detect ribosome footprints after the canonical stop codons, we have identified 144 mRNAs that exhibit SCR in Arabidopsis thaliana [7]. This was the first report to identify SCR in plants. SCR results in a longer protein isoform with a unique C-terminus, which can confer unique properties to the isoform – different localization or function or stability compared to the canonical isoform. Thus, SCR serves as a mechanism of regulation of function as well as expression of a gene [4].

SCR plays a regulatory role in a variety of cellular processes. Our study on AGO1 mRNA demonstrated that SCR generates an isoform termed Ago1x, which functions as a competitive inhibitor of the microRNA pathway [3]. Similarly, MTCH2, which encodes a mitochondrial membrane protein, undergoes SCR to produce an unstable isoform.

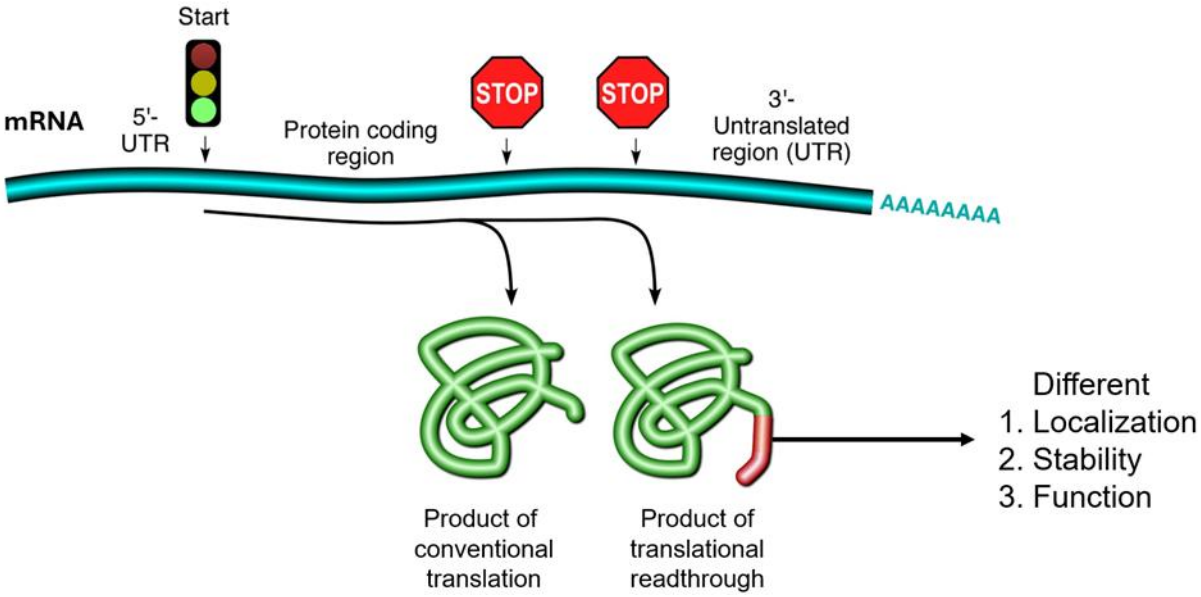

Figure 1. Stop codon readthrough results in longer isoforms with extended C-terminus. These isoforms can have different functions, or localization, or stability depending on the sequence, compared to their canonical isoforms generated from conventional translation.

Loss of this SCR event leads to diminished mitochondrial membrane potential and reduced ATP production [4]. In the case of NNAT, which encodes neuronatin, SCR results in an alternative isoform NNATx that fails to bind the Sarco/Endoplasmic Reticulum Calcium ATPase (SERCA2) calcium pump, unlike the canonical NNAT isoform, leading to impaired calcium-mediated neuronal differentiation [5]. Another example is FEM1B, where SCR gives rise to the unstable isoform FEM1Bx. When SCR is absent, cells accumulate more FEM1B, a known negative regulator of the cell cycle. Thus, SCR at FEM1B promotes cell cycle progression by lowering levels of the canonical FEM1B protein [6]. Additional studies have implicated SCR in regulating processes such as angiogenesis and myelination [8, 9]. Collectively, these findings highlight SCR as a key post-transcriptional mechanism modulating diverse cellular functions, including cell cycle progression, mitochondrial activity, neuronal differentiation, microRNA function, angiogenesis, and myelination. As research progresses, it is likely

Why do only certain mRNAs undergo SCR? This remains a fundamental question in the field. While a complete answer is still elusive, several observations point toward a general underlying mechanism. In the gag-pol region of murine leukemia virus (MLV), SCR, which is crucial for viral replication, is driven by a downstream RNA pseudoknot located five nucleotides beyond the stop codon [10]. In another example, the RNA-binding protein HNRNPA2B1 binds nine nucleotides downstream of the stop codon in VEGFA mRNA and promotes SCR, contributing to an antiangiogenic effect [8]. Similarly, the microRNA let-7a binds nine nucleotides downstream of the stop codon in AGO1 mRNA and enhances its SCR [3]. These findings point to a common theme: transient molecular obstacles to ribosome movement, such as RNA-binding proteins or microRNAs can promote stop codon recoding and facilitate SCR. Our laboratory is currently testing this model in additional mRNAs to evaluate its broader applicability.

SCR has significant clinical relevance, particularly for genetic disorders caused by nonsense mutations that introduce premature stop codons into mRNAs. Conditions such as thalassemias, Duchenne muscular dystrophy, and cystic fibrosis fall into this category. In such cases, inducing SCR across the premature stop codon could restore full-length protein production and offer therapeutic benefit. Several efforts have been made to exploit this mechanism therapeutically [4]. Compounds such as aminoglycosides and ataluren have been shown to promote SCR; however, their clinical application has been limited due to issues like toxicity and insufficient efficacy in vivo. These drugs act by targeting ribosomes non-specifically, raising concerns about widespread disruption of normal translation. This underscores the urgent need for mRNA-specific approaches that can selectively induce SCR at disease-relevant sites while minimizing off-target effects.

To address the need for mRNA-specific induction of SCR, we have developed two targeted strategies based on the principle that transient molecular roadblocks can promote SCR. The first approach involves the use of antisense oligonucleotides (ASOs) designed to bind immediately downstream of premature stop codons caused by nonsense mutations. This binding creates a localized barrier to ribosome progression, thereby promoting SCR. We have successfully tested this strategy in patient-derived cells from individuals with β-thalassemia and Duchenne muscular dystrophy, and are currently working to enhance its efficiency [11].

Our second approach leverages a CRISPR-Cas13–-based system, in which a catalytically inactive Cas13 (dCas13) is directed to bind the region just downstream of the stop codon. This patented method has been shown to induce readthrough of both natural and disease-associated premature stop codons (Fig. 2). We are now developing a lentivirus-based delivery system to enable in vivo application of the CRISPR-dCas13 platform [12]. Unlike conventional readthrough-inducing agents such as aminoglycosides, our ASO- and CRISPR-dCas13–-based strategies are transcript-specific, offering a key advantage in minimizing off-target effects and adverse outcomes. However, because these approaches must be tailored to the precise location of the premature stop codon, they require patient-specific customization.

Overall, our work in the field of SCR exemplifies how insights into fundamental genetic mechanisms can be translated into innovative therapeutic strategies. It underscores the critical role of both basic and applied research in advancing precision medicine.

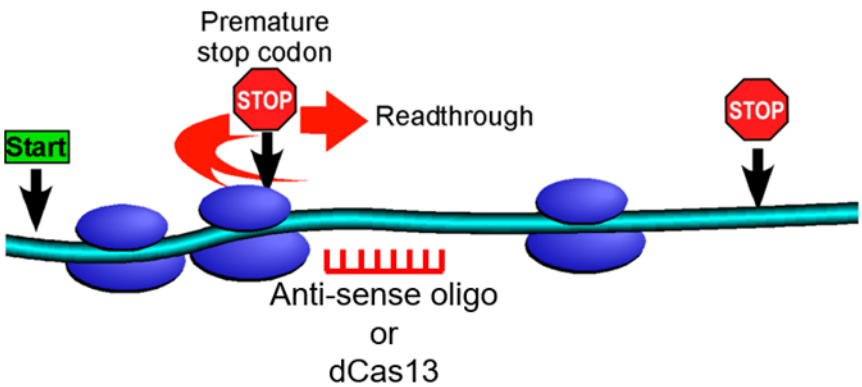

Figure 2. Antisense oligonucleotides and CRISPR-dCas13 system can be used to induce therapeutic readthrough across disease-causing premature stop codons.

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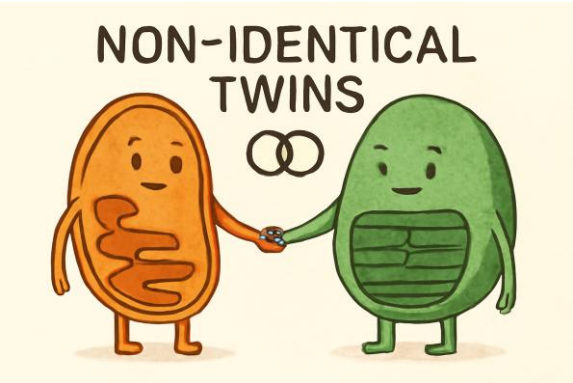
Fun time!



Fruit flies as a model for rare and undiagnosed genetic disorders

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Roadblocks of rare and undiagnosed genetic disorders

Rare and undiagnosed disorders are medical challenges that pose a significant emotional, social, and economic burden. Despite their individual rarity, more than 300 million people worldwide are collectively affected by rare diseases [1]. At least 5–7% of the population in India are currently living with a rare disease [2,3]. A major fraction, i.e. more than 75% of rare diseases arise from diverse genetic and genomic abnormalities [1,4]. Some of the common underlying genetic causes include: i) point mutations, where a single nucleotide base pair is altered in a gene; ii) copy number variations, which involve deletions or duplications of DNA segments; iii) chromosomal abnormalities that includes structural changes (such as translocations or inversions) or numerical abnormalities (such as an extra chromosome 21 in Down syndrome) in chromosomes. These genetic anomalies cause abnormal development, metabolic imbalances, organ malfunctions, etc., leading to a disease condition. A major challenge associated with the rare/undiagnosed diseases includes a lack of diagnosis. Highly variable symptoms are observed not only between different diseases but even among individuals with the same condition. Because many rare diseases share common symptoms, they are frequently misdiagnosed, leading to delays in receiving appropriate treatment. In addition, the extremely low prevalence of individual rare diseases presents significant challenges for conducting large-scale studies to reach a diagnostic conclusion. One study suggests that only ~1,200 rare diseases have more than five documented cases reported [1]. This scarcity of data contributes to long diagnostic delays.

After 2010, technological advancements such as genome and exome sequencing have significantly enhanced our ability to identify genetic variants [5]. This led to faster and more accurate diagnoses of many rare and previously undiagnosed diseases. However, at least 40% of such cases still remain unsolved even when next-generation sequencing is combined with bioinformatic analysis [2]. This happens especially when a gene is not linked to any known disease, or it is a variant of unknown significance. The lack of functional data makes it challenging to determine the pathogenicity of the identified variants.

Non-mammalian model organisms provide an in vivo platform for rapid and effective functional assays to test the pathological nature of gene variants. In addition, they facilitate the investigation of disease mechanisms and the screening of drugs in a controlled and scalable manner. For example, Drosophila melanogaster, commonly known as the fruit fly, helped discovering the causal involvement of over 50 genes in rare or undiagnosed diseases in recent years [6]. In some cases, it has also provided insights into disease mechanisms and potential therapeutic strategies for disease management. So, what makes the fruit fly such a successful model for studying rare diseases?

Fruit flies provide a platform for variant functional assessment

Over the past 100 years, fruit flies have been extensively used in biomedical research. Notably, more than 70% of human disease-related genes have functional orthologs in fruit flies [6,7]. It allows researchers to investigate the biological consequences of human disease-associated genetic variants in a living organism. In many cases, flies carry a single gene where humans may have multiple related genes, known as co-orthologs. This occurs as a result of gene duplication events during evolution [5]. Despite this divergence, these human co-orthologs often share functional conservation with the ancestral fly gene, which enables researchers to study putative pathogenicity of a candidate gene variant using fruit flies. In addition, its short life cycle (approximately 10 days) and low maintenance cost make it ideal for large-scale genetic studies and quick functional assessment of putative disease-causing variants. Additionally, fruit flies offer robust and scorable phenotypic assays. Importantly, the well-annotated genome and sophisticated genetic toolkit make fruit flies a preferred model for variant functional analysis.

A few core strategies (Fig. 1) are commonly used to analyse the functional nature (whether a variant is loss-of-function, or gain-of-function, or dominant-negative) of a variant. include ectopic expression of a human reference gene and the suspected pathogenic variants in different tissue of fruit flies in a wild-type genetic background. The Gal4/UAS binary expression system enables this precise temporal and spatial control of gene expression using transgenic lines [8].

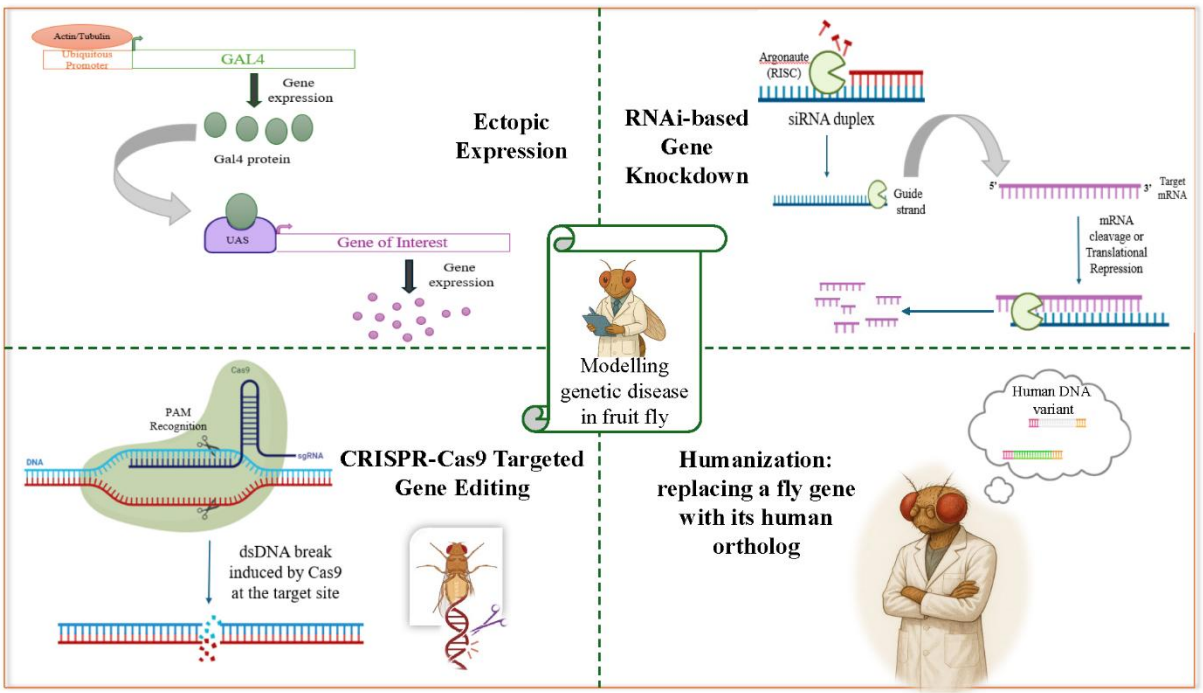

Figure 1. Tools and strategies for variant functional assessment using fruit flies. The image was created using BioRender, ChatGPT, and Microsoft PowerPoint.

In this system, the driver line contains the Gal4 transcription factor-encoding gene under the control of a tissue- or cell-specific enhancer, thereby restricting Gal4 expression to defined set of cells. The responder line carries the gene of interest (GOI) downstream of Upstream Activating Sequences (UAS), which serve as binding sites for Gal4. When both fly lines are crossed, Gal4 binds to the UAS sequences in the progeny, activating transcription of the GOI specifically in the targeted tissues. It enables temporal and spatial control of gene expression, resulting in scorable phenotypes such as lethality, or tissue-specific effects in the wings, eyes or other organs, or neurological phenotypes such as locomotory defects, etc. The differences in the observed phenotypes help understand the functional character of the variant. For example, Marcogliese et. al used this strategy to determine the functional nature of variants in IRF2BPL, a gene involved in a severe paediatric neurodevelopmental disorder called NEDAMSS (neurodevelopmental disorder with regression, abnormal movements, loss of speech, and seizures) [9]. They identified seven individuals and all of them had heterozygous variant in this gene. The patients had neurological symptoms including seizures, developmental delay, ataxia, hypotonia, speech issues, etc. For functional assessment of the disease-causing variants, they used the ectopic expression strategy. When IRF2BPL reference transgene was ubiquitously expressed, it caused lethality, indicating that the fully functional IRF2BPL shows a cellular toxic effect. On the other hand, majority of the patient-derived variants did not cause lethality in this assay, implicating that the variants are loss-of-function in nature [9].

This ectopic expression strategy offers a straightforward way to assess the nature of a putative variant. However, if the ectopic expression of either the reference or the variant(s) do not cause a scorable phenotype, functional assessment becomes difficult. In this situation, some other strategies, such as RNAi-mediated knockdown, are adopted to draw a parallel between fly and patient phenotypes and to test the nature of variants. For example, bi-allelic mutations in DNMBP have been reported to cause cataract and visual impairment in infants [10]. DNMBP interacts with E-cadherin and helps maintaining proper architecture of the cellular junctions in epithelial cells in humans. When an RNAi-mediated knockdown of the corresponding fly gene, sif, was carried out, it led to the mislocalization of E-cadherin, impaired cellular junction formation, and a significantly reduced phototransduction ability in flies [10]. These parallels helped establishing that loss of DNMBP indeed causes this infantile visual disorder in humans. This approach provides supportive evidence to prove the pathogenic mechanisms caused by the loss of a gene; however, it does not directly test the nature of the patient-derived variants. This limitation is overcome by using a ‘humanization’ strategy.

During fruit fly humanization, the reference human transgene or suspected variants are expressed in a genetic background where the orthologous fly gene is mutated. If the expression of human transgene rescues the mutant fly phenotype, it establishes functional conservation between the fly and human proteins. Subsequently, the variants are expressed in the mutant background, and their ability to alter the phenotype in flies are scored. Recently, by using this strategy, Paul et. al. identified that deleterious variants in PPFIA3 cause a neurodevelopmental disorder [11]. The patients presented with developmental delay, dysmorphisms, hypotonia, etc. Additionally, they showed neurological symptoms such intellectual disability, autism-related indications, and micro or macrocephaly. Strong loss-of-function mutations in the fly ortholog, Liprin-α, cause lethality at different developmental stages of fruit flies. Ubiquitous expression of the human PPFIA3 partially rescued the lethality in fly mutants, whereas the patient-derived variants showed a significantly reduced ability to rescue the lethality [11].

This key assay helped establishing the damaging nature of the patient-derived variants. Although the human reference and variant transgenes are expressed in a mutant background, this strategy offers limited information about the cell type-specific origin of a disease. For example, if ubiquitous expression of the human ortholog rescues a lethal phenotype in fly mutants, the specific cellular origin of the phenotype (whether glial, neuronal, or another cell type) remains unclear. To overcome this, CRISPR-Cas9 is used to insert a Gal4-expressing cassette (such as T2A-Gal4 or Kozak-Gal4) either into the coding intron or to replace the coding region of the orthologous fly gene [12-14]. This creates a loss-of-function/null allele, and the inserted Gal4 can further be used to drive expression of various UAS-transgenes under the native expression pattern of the fly gene. When crossed with a UAS-RFP/mCherry, this Gal4 allele helps to examine the tissue and cell type-specific gene expression. In addition, it ensures that human reference or variants are expressed only in those cells where the endogenous fly gene is expressed. This strategy was used to solve multiple undiagnosed disease cases including the one related to TOMM70, which encodes for a protein that helps in mitochondrial protein import. Two patients were reported with de novo variants in TOMM70. Both patients exhibited neurological symptoms such as dystonia, ataxia, hyperreflexia, and white matter abnormalities in the brain. However, other symptoms such as microcephaly, ptosis, and dysarthria were present in either one of the two patients [15]. As the symptoms were puzzling and TOMM70 was not reported earlier to cause a genetic disease, functional assessment was required to test the pathogenicity of these variants. For that, a null allele of fruit fly Tom70 was created by replacing the Tom70 coding region with a GAL4 transgene via CRISPR-Cas9 technology. The allele helped determining the expression pattern of this gene at different stages and cell types in fruit flies. It was also noted that the complete loss of Tom70 leads to pupal lethality, which could be rescued by expressing human TOMM70 reference transgene. However, expression of the patient-derived TOMM70 variants only partially rescued this lethality suggesting they are loss-of-function alleles [15]. This approach enables a direct assessment of how a disease-causing variant affects gene function, exerts a cell type-specific effect, and contributes directly to disease mechanisms.

Future Directions

Despite their low individual prevalence, rare disorders collectively impose a significant burden on global health. Unlike cancer or other common diseases, rare and undiagnosed conditions are often deprioritized by pharmaceutical companies in the drug development pipeline due to their low prevalence. Additionally, the genetic heterogeneity and clinical variability of these disorders make diagnosis particularly challenging. However, these very challenges make rare diseases uniquely valuable for research. In many cases, genes implicated in rare or undiagnosed diseases are not well studied in the context of human genetic disorders. Therefore, studying these diseases leads to the discovery of novel gene functions. For example, before 2018, IRF2BPL/EAP1 was primarily known for its role in regulating the onset of female puberty in mammals [16]. The discovery of NEDAMSS disease using fruit flies first revealed a role for IRF2BPL in the nervous system. Subsequent studies demonstrated that IRF2BPL regulates Wnt signalling and supports neuronal maintenance [17]. As the field of rare disease research continues to evolve, fruit flies will remain a powerful tool for gene discovery and mechanistic investigation. Because of their high evolutionary conservation, fruit flies help bridge the gap between genomic data and biological understanding. Research using fly models has produced transformative insights into gene function, phenotypic expansion, and disease-modifying pathways across multiple rare disorders.

The continued integration of fruit fly genetics with human genomics, high-throughput screening, and personalized modelling will be critical for advancing diagnostics and developing targeted therapies for individuals affected by rare/undiagnosed genetic diseases.

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Antibacterial and anti-biofilm potential of green-synthesized silver nanoparticles and antibiotic combinations against multiple-antibiotic resistant ESKAPE bacteria

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Antimicrobial resistance (AMR) has become a global health challenge of unprecedented proportions. The emergence of Multiple Antibiotic Resistance (MAR) in ESKAPE (Enterococcus faecium, Staphylococcus aureus, Klebsiella pneumoniae, Acinetobacter baumannii, Pseudomonas aeruginosa, and Enterobacter spp.) pathogens is of great concern because conventional antibiotics have become increasingly ineffective in controlling infections caused by these microorganisms. Therefore, there is an urgent need for alternative strategies to tackle MAR ESKAPE microorganisms. The inherent antimicrobial properties of silver nanoparticles (AgNPs) make them promising candidates for combating antibiotic-resistant pathogens. This study reports potential new antimicrobial agents, prepared by combining patented green-synthesized AgNPs with antibiotics (SACs), which are effective against MAR ESKAPE microorganisms [1]. Mechanistic insights into their anti-biofilm activity have also been provided, suggesting that these biofilm-eradicating SACs probably result in the loss of bacterial cell membrane integrity, leading to leakage of cytoplasmic content and eventual cell death.

AgNPs were synthesized using a commonly available but hitherto unused microorganism, Streptococcus pneumoniae ATCC 49619, as per the method illustrated in Figure 1 (Process Patent No. 458867). Transmission Electron Microscopy analysis of the synthesized AgNPs confirmed spherical nanoparticles with an average diameter of 7.37 ± 4.55 nm [1].

To test the antibacterial activity of the AgNPs, MAR ESKAPE food bacterial isolates (previously isolated in the AMR laboratory; [2, 3]) were used. These isolates were highly resistant to multiple antibiotics. The AgNPs showed antibacterial activity against all selected MAR ESKAPE isolates, demonstrating their wide-spectrum antimicrobial activity.

Subsequently, the antibacterial effectiveness of AgNPs in combination with individual antibiotics (silver nanoparticle-antibiotic combinations: SACs) was tested against the selected isolates by determining the Fractional Inhibitory Concentration (FIC) using the 2D checkerboard method. The antibiotics to which the MAR ESKAPE isolates showed resistance were chosen for the experiments.

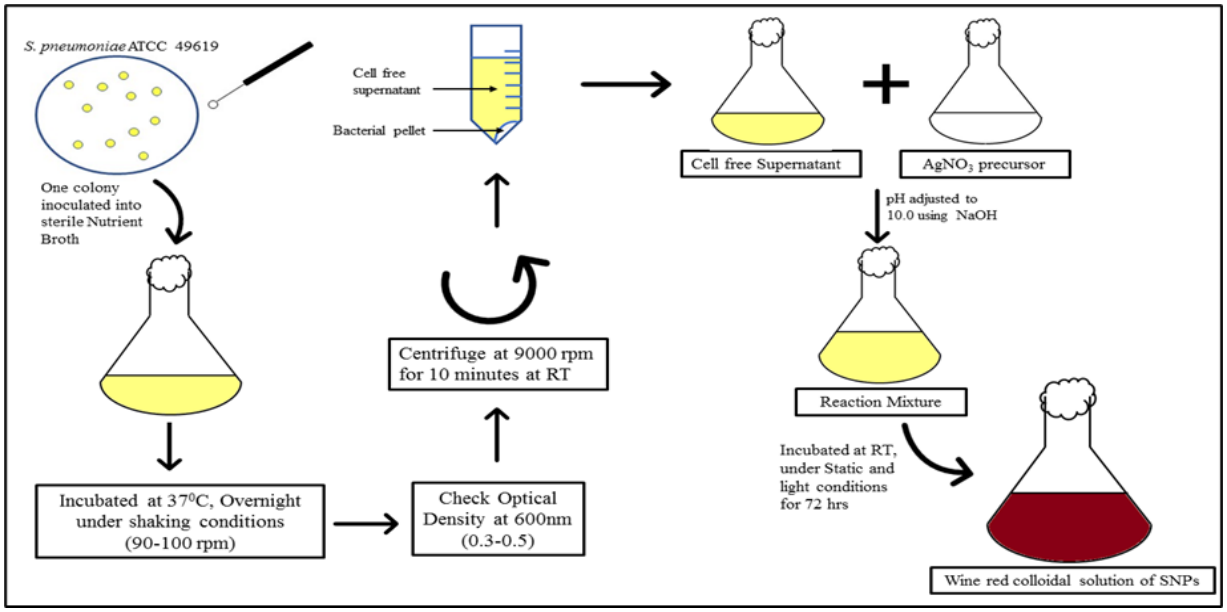

Figure 1. Schematic representation of the synthesis process of green silver nanoparticles (AgNPs) using S. pneumoniae 49619. A single colony was inoculated into sterile nutrient broth and incubated overnight. The cell-free supernatant, which was then used for the synthesis of AgNPs. A change in colour from pale yellow to wine red gave visual confirmation of the synthesis of AgNPs [1].

SACs were prepared by adding AgNPs and the respective antibiotics at their combination minimum inhibitory concentrations (MICs) just prior to the experiments. The nature of the interaction between the AgNPs and antibiotics (synergistic, additive, neutral, or antagonistic) was determined by calculating the FIC index (FICI) of the SACs. The lower the FICI value of the SAC, the higher the synergistic interaction between AgNPs and antibiotics. Synergistic and additive interactions were observed for all SACs, with FICIs ranging from 0.035 to 1.14. Moreover, the effective MICs of the antibiotics were reduced by 6.4 to 32-fold in the SACs compared to their MICs when used alone. Such a high-fold reduction in the effective concentrations of antibiotics against MAR ESKAPE isolates when used in combination with AgNPs has not been reported previously. The antibacterial activity of both antibiotics and AgNPs is enhanced in the SACs, thus highlighting the importance of using the AgNPs in combination with antibiotics. The high antimicrobial activity of the SACs against a battery of MAR ESKAPE microorganisms in the

the synthesized AgNPs and their potential as effective antimicrobial agents [1]..

One of the challenges of AMR is the presence of highly resistant persister cells in bacterial biofilms, which help bacteria survive various stressors, such as antibiotic treatment [4]. Moreover, bacteria in biofilms are difficult to eradicate due to their complex structure. Therefore, in the present study, the effectiveness of the SACs against the biofilms of selected MAR isolates was assessed. Both eradication of pre-formed biofilms and inhibition of biofilm formation, in the presence of SACs, were tested. All the tested SACs caused significant eradication of pre-formed biofilms (Fig. 2) and inhibition of biofilm formation [1] of the MAR ESKAPE bacterial isolates.

Amongst the various mechanisms proposed for the antibacterial action of AgNPs is the destabilization of cell membrane structure by formation of pits in the cell wall of bacteria, leading to increased membrane permeability [5]. Moreover, bacterial plasma membranes consist

of a lot of sulphur-containing proteins, which may represent sites suitable for the attachment of AgNPs [6]. The binding of AgNPs to membrane proteins may result in the loss of membrane integrity leading to the leakage of intracellular content, eventually resulting in cell death. To assess whether the SACs in the present study acted via a similar mechanism, membrane leakage assays were performed. Bacterial cells were exposed to SACs for 3 hours, and samples were checked hourly for leakage of cellular contents. Sugar and protein leakage was observed within 1 hour of treatment with the SACs (Fig. 3). This leakage of cellular components suggests that exposure to SACs may have induced a loss of membrane integrity. AgNPs have been reported to damage cell membrane integrity, leading to the leakage of cytoplasmic contents such as K+ ions, carbohydrates, proteins, and nucleic acids [5-7]. In most cases, treatment with SAC caused more pronounced membrane damage in all the tested isolates than treatment with AgNPs or antibiotics alone. Thus, the membrane leakage assays suggest that the AgNPs in the SACs might be inducing loss of the bacterial cell membrane integrity, allowing easy entry of the antibiotics into the bacterial cell, thus enabling them to act more efficiently even at low concentrations [8, 9]. The subsequent antibiotic stress may be causing further damage to the integrity of the cell, leading to increased leakage of cytoplasmic contents. The increased membrane leakage is probably one of the mechanisms of the synergistic antibacterial activity of the SACs against MAR ESKAPE microorganisms [1].

In conclusion, the green-synthesized AgNPs displayed promising antibacterial activity against diverse MAR ESKAPE bacteria, and this activity was further enhanced upon physical combination of AgNPs with antibiotics as SACs. The ability of AgNPs to show synergistic activity with antibiotics belonging to various classes further suggests their potential for multiple clinical applications. Such SACs can be potent alternatives to conventional treatment strategies against antimicrobial resistance. Detailed in-vitro cytotoxicity and biocompatibility studies with AgNPs and SACs are underway.

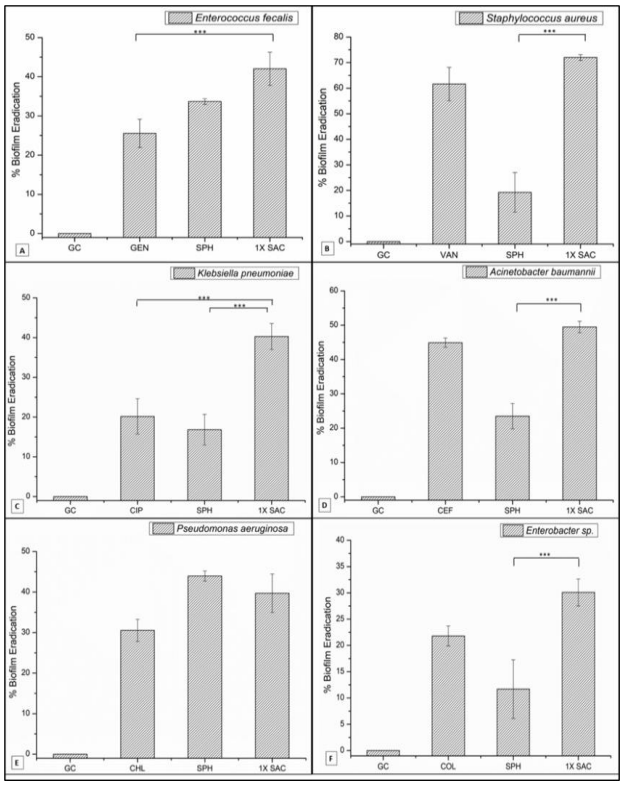

Figure 2. Eradication of biofilms formed by MAR isolates in the presence of SACs: A) E. fecalis, B) S. aureus, C) K. pneumoniae, D) A. baumannii, E) P. aeruginosa, and F) Enterobacter spp. CEF-cefotaxime, CHL-chloramphenicol, CIP-ciprofloxacin, GEN-gentamicin, VAN-vancomycin, COL-colistin, SPH-AgNPs under study, and GC-growth control. Error bars represent the standard deviation of triplicate sets of experiments. The data was statistically significant compared to the control at $P<0.05$. *$P<0.05$, **$P<0.01$, ***$P<0.001$ [1].

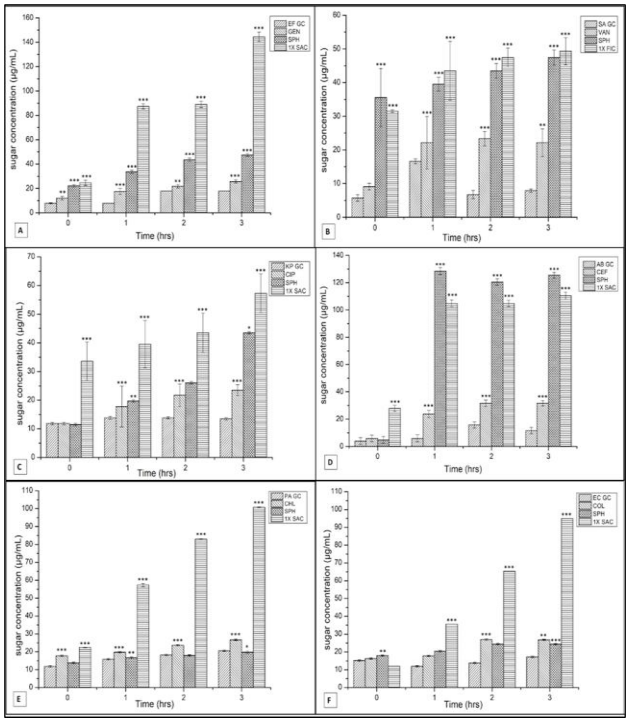

Figure 3. Leakage of carbohydrates from SAC-treated isolates: A) E. fecalis, B) S. aureus, C) K. pneumoniae, D) A. baumannii, E) P. aeruginosa, and F) Enterobacter spp. CEF-cefotaxime, CHL-chloramphenicol, CIP-ciprofloxacin, GEN-gentamicin, VAN-vancomycin, COL-colistin, SPH-AgNPs under study, and GC-growth control. Error bars represent the standard deviation of triplicate sets of experiments. The data was statistically significant compared to the control at $P<0.05$. *P <0.05, **P <0.01, ***P <0.001 [1]

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Outsmarting cancer by engineered nanoparticle: A synergistic therapy with doxorubicin and quercetin



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Background

One of the foremost challenges in modern oncology is cancer's adaptive resistance to chemotherapeutic agents [1]. Among the key contributors to this resistance is the P-glycoprotein (P-gp) efflux pump, which actively expels drugs from tumor cells, thereby reducing intracellular drug retention and diminishing therapeutic outcomes [2]. To address this, our research team led by Prof. Sri Sivakumar at IIT Kanpur has developed an innovative core–shell porous nanoparticle designed to co-deliver doxorubicin, a standard chemotherapeutic [3], and quercetin, a natural polyphenol [4] with chemosensitizing properties. This dual-agent system utilizes a tumor-responsive, sequential release mechanism to enhance treatment efficacy while mitigating systemic toxicity [5,6].

Conceptual innovation: Stimuli responsive, targeted and sequential drug delivery paradigm

Central to this nanoplatform is a polydimethylsiloxane (PDMS) polymer core, chosen for its biocompatibility and capacity to house hydrophobic agents like quercetin. This core is enveloped by a shell of polyethyleneimine and hyaluronic acid (HA), providing aqueous stability and active targeting of CD44 receptors extensively overexpressed in cancer cells. Doxorubicin, owing to its slight hydrophilic nature, is sequestered within this HA layer (Fig. 1).

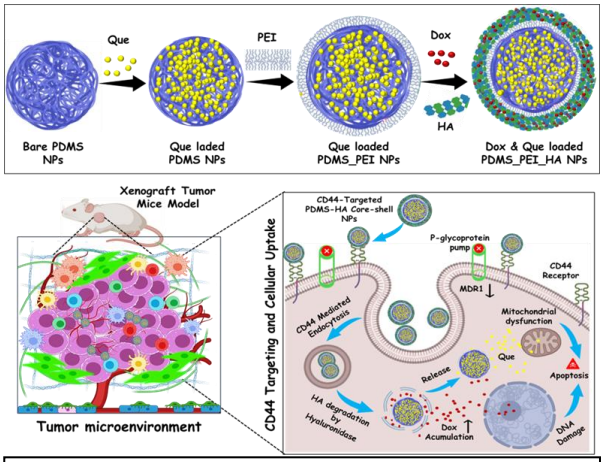

Figure 1. Graphical Abstract: A dual drug nanocarrier composed of polydimethylsiloxane (PDMS) and hyaluronic acid (HA) enables sequential release of doxorubicin and quercetin for enhanced tumor targeting and reversal of multidrug resistance. HA targets CD44+ tumor cells. This nanoplatform significantly improves drug accumulation, apoptosis, and tumor regression, highlighting a promising strategy for spatiotemporally controlled, low-toxicity combination chemotherapy.

Upon reaching the tumor microenvironment where hyaluronidases are prevalent, the outer shell is enzymatically degraded, triggering the initial release of doxorubicin and inducing apoptotic cell death. Subsequently, quercetin is released from the core to inhibit P-gp activity, thereby sustaining intracellular drug levels and reinforcing therapeutic potency. This orchestrated release strategy exemplifies a precision-guided, timed therapy rather than mere drug administration. Hence, the scheme offers programmable and targeted release kinetics while maintaining biocompatibility and enabling therapeutic synergy.

Efficacy across biological models: In vitro and in vivo

In vitro assessments using MCF-7 breast cancer cells demonstrated a significant enhancement in drug sensitivity. The co-loaded nanoparticles reduced IC_{50} values for both agents by nearly tenfold compared to their free-drug counterparts. Apoptosis analysis revealed that, at optimal doses, nearly 97% of cancer cells underwent programmed cell death outperforming all monotherapies and even free drug combinations. Additional investigations confirmed substantial G2/M phase cell-cycle arrest, increased caspase-3/7 activity, and downregulation of efflux-related P-gp, highlighting the mechanistic soundness of the platform. In vivo studies using a mouse xenograft model further reinforced these findings, showing a ~65% reduction in tumor volume, achieved with lower overall drug dosages (Fig. 2A). Histopathological (Hematoxylin & Eosin) and TUNEL analyses (Figs. 2B and C) showed extensive tumor necrosis with minimal adverse impact on healthy tissues attesting to the system's targeted delivery and safety profile.

Why this matters: Scientific and clinical implications

The co-delivery of a chemotherapy drug with a plant-derived chemosensitizer in a programmed nanocarrier underscores an important translational trend: combination therapy did not mean increased number of active agents and related toxicity, but rather smarter design of their carrier and strategic delivery.

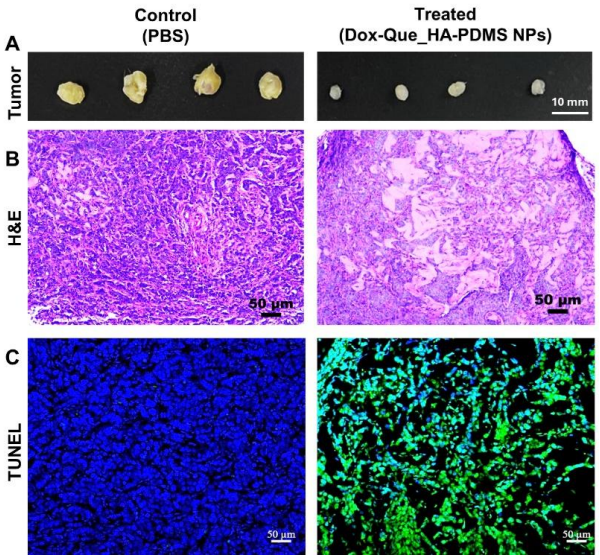

Figure 2. In vivo antitumor efficacy studies in MCF-7 tumor-bearing xenograft mice models. (A) Representative images of tumors extracted after the completion treatment which corresponds to control (PBS treated) and experimental (Dox-Que_HA-PDMS NPs) group. (B) Characteristic histological images of H&E staining of tumor sections. (C) CLSM images of TUNEL assay of tumor sections: blue (DAPI) and green (TUNEL). (Reproduced from [7])

This method redefines how we approach multidrug resistance. The increased retention of doxorubicin is not due to higher dosing but to quercetin's inhibition of P-gp, as validated by Immunofluorescence via flow cytometry. The strategy achieves pharmacodynamic synergy through both spatial and temporal separation of drug release. Moreover, it repositions bioactive plant compounds often sidelined for their poor solubility as vital therapeutic allies when delivered via well-engineered nanocarriers.

Considerations for future translation

Despite promising outcomes, additional research is essential. The long-term biocompatibility and clearance of PDMS-based nanocarriers must be thoroughly evaluated to satisfy regulatory requirements. Further exploration into the stability and kinetics of drug release under varied physiological conditions will help ensure translational robustness. Expanding the platform to encompass other CD44+ tumor types could widen its therapeutic scope. Long-term safety data, immune response profiling, and scalability assessments will be crucial for eventual clinical application.

A human-centered ethos

Beyond the molecular mechanisms and statistical readouts lies a more human aspiration designing a chemotherapy experience that is not only potent, but patient centered. This work exemplifies how interdisciplinary collaboration in chemistry, engineering, and medicine can produce technologies that respect both science and individual. Our novel approach provides not just efficacy but also confronts resistance not with brute-force doses but with elegant strategy; outwitting cancer at the molecular level rather than overwhelming it, seeking to restore quality of life alongside therapeutic success.

Final reflection

This study offers more than a novel formulation—it proposes a new framework for the future of cancer therapy. Through the precise pairing of a conventional cytotoxic agent and a natural resistance modulator, encapsulated in a thoughtfully engineered nanocarrier, this work lays the groundwork for combination regimens that are both scientifically rigorous and deeply humane. The PDMS–HA delivery system represents a strong step forward in the evolution of smarter, safer, and more effective cancer treatments.

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Neuromuscular junction dysfunction in glycogen storage diseases: Insights from Lafora disease

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Neuromuscular junctions (NMJs) serve as critical synaptic interfaces enabling communication between motor neurons and skeletal muscles. These highly specialized, tripartite structures comprising presynaptic motor neuron terminals, the postsynaptic muscle endplate, and the synaptic cleft are essential for voluntary motor control [1]. The integrity of this interface ensures precise muscle contraction, and its impairment has been implicated in a variety of neuromuscular and neurodegenerative disorders. Classical NMJ disorders such as Myasthenia Gravis and Lambert-Eaton Myasthenic Syndrome arise due to defects in the post and pre-synaptic elements, respectively, leading to muscle weakness and fatigability. More broadly, NMJ dysfunction is a key pathological feature in conditions such as Amyotrophic Lateral Sclerosis, Spinal Muscular Atrophy , and Charcot-Marie-Tooth disease, contributing to progressive motor impairment [2].

Recent findings highlight that metabolic dysregulation, particularly in glycogen storage disorders (GSDs), is an underappreciated contributor to NMJ vulnerability. A subset of GSDs, including Pompe (GSD II), Cori (GSD III), Andersen (GSD IV), Tarui (GSD VII), and GSD IX, have shown direct involvement of NMJ dysfunction [3, 4]. Although Lafora disease (LD) is classified as a GSD and is primarily recognized as a form of progressive myoclonus epilepsy, the contribution of NMJ dysfunction to its pathophysiology has remained understudied. This commentary first examines the emerging evidence of NMJ dysfunction across various GSDs, highlighting shared mechanisms of synaptic vulnerability. Building on this foundation, it further expands upon recent findings published in Disease Models & Mechanisms, which provide the first detailed analysis of NMJ pathology in LD, and introduce the concept of "metabolic synaptopathy" as a unifying framework.

Neuromuscular junction vulnerability in Glycogen Storage Disorders

GSD are a diverse group of inherited metabolic conditions that impair key enzymatic pathways involved in the synthesis or breakdown of glycogen. While they have long been characterized by dysfunction in the liver, heart, or skeletal muscles, a growing body of research suggests that their pathological effects extend further, specifically to the NMJ, a site

increasingly recognized for its sensitivity to metabolic disturbances. A compelling example is Pompe disease (GSD II), where deficiency of the lysosomal enzyme acid alpha-glucosidase not only leads to profound skeletal and cardiac muscle involvement, but also induces structural and functional abnormalities at the NMJ [5]. These include reduced synaptic vesicle density and evidence of denervation, particularly affecting respiratory and postural muscles, thus highlighting the NMJ as an important and vulnerable target in the disease's neuromuscular manifestations [6]. Similarly, Cori disease (GSD III), caused by a deficiency in the glycogen debranching enzyme, is associated with electrophysiological abnormalities, including altered electromyography patterns and nerve conduction delays. These disturbances, along with severe vacuolar myopathy, have been linked to the presence of malformed glycogen and polyglucosan inclusions in muscle fibers and peripheral nerves [7]. Additionally, in Andersen disease (GSD IV), NMJ dysfunction appears to be driven in part by the accumulation of polyglucosan bodies within glial and neuronal compartments, contributing to both peripheral neuropathy and muscle pathology [8]. This highlights the broader neuro-muscular impact of aberrant glycogen processing. A late-onset variant of this condition, known as adult polyglucosan body disease, further underscores the neurological impact, presenting with gait instability, bladder dysfunction, and progressive cognitive decline due to compromised glycogen branching enzyme activity [9]. Extending this theme of NMJ susceptibility, McArdle disease (GSD V), resulting from mutations in the gene encoding muscle glycogen phosphorylase, leads to impaired energy production during physical exertion [10]. The resulting symptoms such as early muscle fatigue, cramping, and myalgia may not solely arise from intrinsic muscle deficits but could also be influenced by compromised synaptic transmission at the NMJ, where high metabolic demands render the synapse particularly susceptible to energetic stress. Collectively, these disorders reveal a common thread that NMJs are particularly sensitive to disruptions in cellular energy and glycogen level. This cross-cutting vulnerability strengthens the case for investigating NMJ impairment as a central mechanism in other metabolic disorders, including LD, and supports the broader conceptual framework of "metabolic synaptopathy.".

Lafora disease: Expanding the neurocentric paradigm to neuromuscular synaptopathy

LD is a rare, autosomal recessive neurodegenerative disorder classified among GSDs. It typically manifests during early adulthood and is characterized by the progressive accumulation of aberrant, insoluble glycogen-like inclusions known as Lafora bodies (LBs) in neurons and other tissues [11]. Clinically, LD presents as a severe form of progressive myoclonus epilepsy, with hallmark symptoms including myoclonus, generalized seizures, ataxia, cognitive decline, and eventual loss of motor function [12]. While the disease has long been studied primarily for its profound effects on the central nervous system (CNS), emerging data and patient reports point to significant involvement of peripheral tissues, including skeletal muscle. Over the last two decades, research on LD pathophysiology has largely centered on two proteins: laforin and malin, encoded by the EPM2A and NHLRC1 genes, respectively. These proteins form a functional complex that regulates glycogen architecture and cellular homeostasis. In the absence of either, glycogen metabolism becomes disrupted, leading to poorly branched, hyperphosphorylated glycogen accumulation [13,14]. In addition to regulating glycogen metabolism, laforin and malin play key roles in glucose uptake and autophagic clearance of misfolded proteins. Their interactions with protein targeting to glycogen, glycogen synthase, and the autophagy adaptor p62 underscore their multifunctionality. Disruption of this regulatory network in knockout mouse models leads to LB formation, impaired

autophagy, neuroinflammation, and increased seizure susceptibility [15-17]. Therapeutic efforts have so far focused on strategies to reduce glycogen synthesis or enhance glycogen clearance within the brain. However, these interventions are primarily CNS-targeted, despite growing recognition that LD patients also suffer from muscle atrophy, motor coordination deficits, and reduced muscle strength, features that may stem from peripheral synaptic dysfunction.

Until recently, the mechanisms underlying these peripheral manifestations remained poorly understood. Our recent study, published in Disease Models & Mechanisms, aimed to fill this gap by providing the first in-depth characterization of NMJ pathology in LD mouse models [18]. We demonstrated that NMJ transmission deficits begin as early as five months of age in LD mouse models (Fig. 1). Repetitive nerve stimulation revealed a progressive decline in compound muscle action potential amplitudes, indicative of impaired synaptic transmission. Immunohistochemical and ultrastructural analyses revealed fragmented motor endplates, reduced acetylcholine receptor density, and decreased pre and post-synaptic arrangement. These structural defects were further compounded by downregulation of synapse-stabilizing genes such as Agrn, Musk, and Lrp4. A particularly notable observation was the presence of collateral sprouting, where surviving motor neurons attempted to re-innervate the denervated endplates, suggesting an early compensatory response to ongoing degeneration. In parallel, we identified alpha-motor neuron loss, reactive gliosis in the spinal cord, and hypermyelination of sciatic nerves, collectively supporting a complex neuro-myopathic axis in LD.

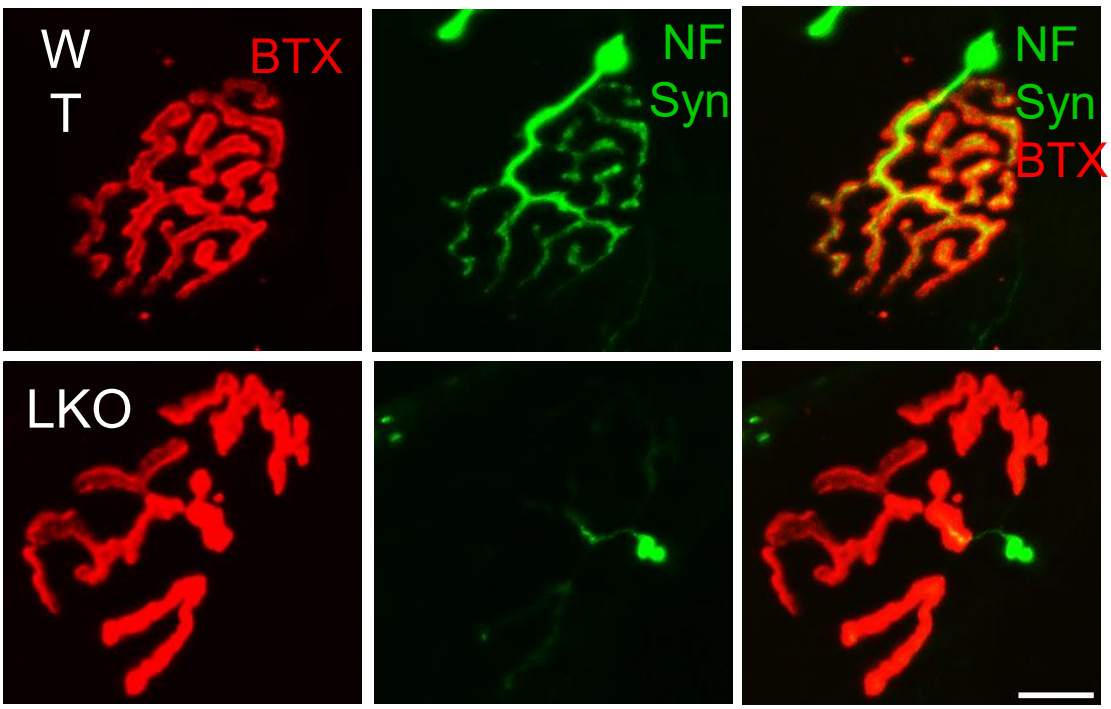

Figure 1. Immunofluorescence images depicting the neuromuscular junction (NMJ) morphology in 10-month-old wild-type (WT) and laforin-deficient (LKO) mice: Here, postsynaptic acetylcholine receptors (AChRs) are stained in red with Alexa-594 conjugated α-Bungarotoxin (BTX), presynaptic nerves and terminals are labeled in green with an anti-neurofilament (NF)* antibody, and synapsin-1 antibodies respectively. WT mice display a compact, pretzel-shaped NMJ structure, while LKO mice show a more dispersed and less organized NMJ morphology. Scale bar = 10 µm (data adapted from Shukla et al., 2024 [18]

These findings challenge the notion of LD as a purely CNS-centric disorder and instead support a more comprehensive perspective that recognizes the involvement of peripheral neuromuscular components in disease progression. The accumulation of LBs within muscle fibers and spinal cord neurons likely creates a metabolically stressful environment at the NMJ, disrupting both structural integrity and synaptic function. Taken together, our findings support redefining LD as a systemic GSD, one that impacts not only central neuronal circuits but also peripheral neuromuscular communication. By uncovering early and progressive defects at the NMJ, our study introduces the concept of metabolic synaptopathy, defined as synaptic dysfunction driven by underlying metabolic stress. This framework may extend to other GSDs, many of which exhibit comparable synaptic vulnerabilities. Crucially, our work highlights the NMJ as a promising therapeutic target in LD and related GSDs, offering new opportunities for intervention through strategies such as glycogen-modulating small molecules or gene therapies aimed at preserving synaptic structure and function.

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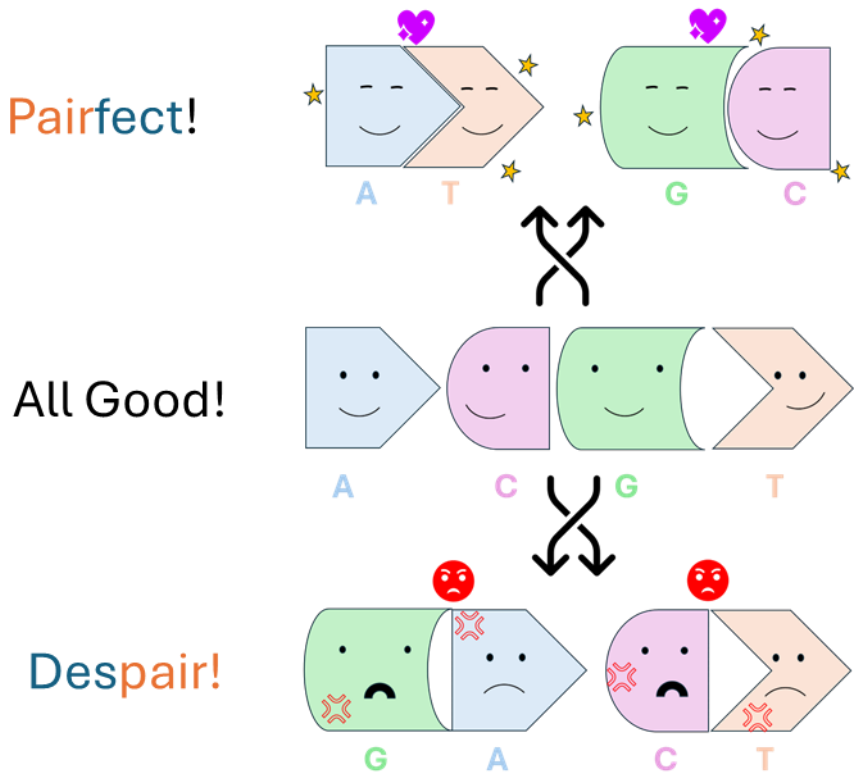
Fun time!



Roller coaster ride of autophagy in cancer: Understanding in distinct phases of tumorigenesis

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The cellular homeostasis mechanism, autophagy, recycles cytoplasmic components via the lysosomal machinery [1]. In autophagy, misfolded proteins, damaged organelles, and other cytoplasmic components are sequestered into double-membrane vesicles called autophagosomes, which fuse with lysosomes for degradation and recycling. Autophagy is activated in response to various stressors, including nutrient starvation, oxidative stress, and accumulation of damaged or misfolded cellular components. Autophagy is tightly regulated by nutrient-sensing pathways, including mTOR and AMPK, and is orchestrated through a series of autophagy-related (ATG) proteins [2]. Various autophagy-related genes are involved in the regulation of autophagy in different stages of autophagy, starting from autophagy initiation to cargo degradation. In the context of cancer, autophagy plays a paradoxical role, acting as a tumor suppressor and a tumor promoter depending on the stage and cellular environment. Autophagy behaves as a “double-edged sword” in oral cancer. The functional role of autophagy in cancer depends on tumor type, stage, genetic context, and tumor microenvironment [3, 4]. The autophagy status during tumor progression varies from the tumor initiation to invasion and metastasis. Autophagy levels regulate the tumor burden. The protective autophagy increases tumor burden, while the cytotoxic autophagy relieves the tumor burden via autophagic cell death (type-II cell death). In healthy cells, autophagy acts like a cellular custodian. It clears out damaged proteins and organelles, preventing the kind of genetic instability that can spark cancer. During the early stage of cancer, autophagy starts to decline as cells undergo malignant transformation [5]. Studies have shown that cells are more likely to become cancerous when key autophagy genes including BECN1, ATG5, or ATG7 are deleted or suppressed. Interestingly, partial loss of BECN1 is frequently seen in early-stage breast, ovarian, and prostate cancers [6].

However, once a tumor forms, autophagy reactivates, and the role of autophagy shifts toward protective autophagy. During this situation, autophagy provides energy and raw materials, allowing them to adapt to harsh conditions. This shift of autophagy from basal level of autophagy to tumor pro-survival autophagy to sustain cancer and metastasis in a nonlinear manner illustrates the roller coaster

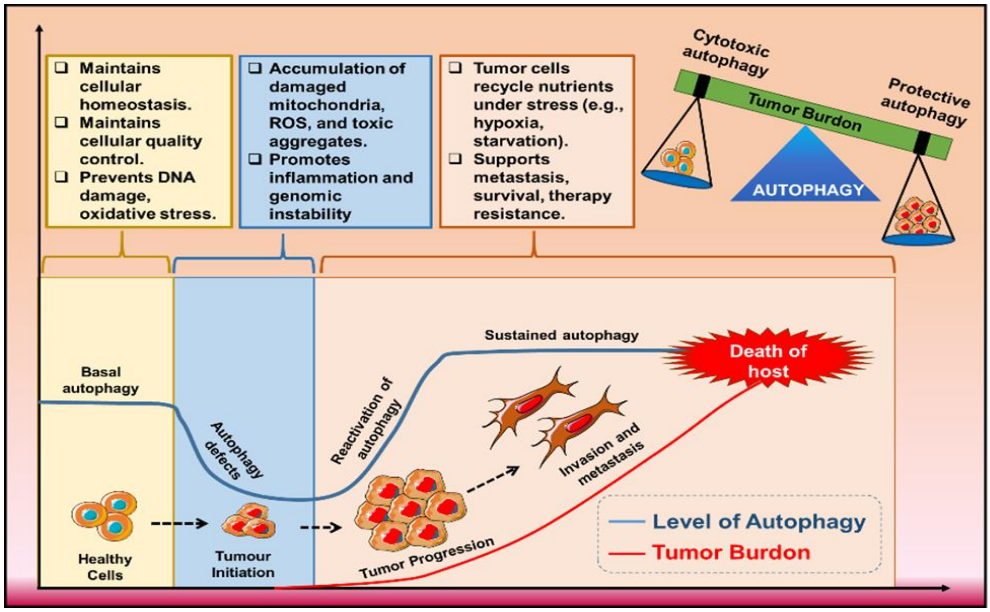

Figure 1. **Roller coaster ride of autophagy in cancer progression**. Basal autophagy is essential for maintaining cellular homeostasis, preventing healthy cells from oncogenic transformation. Later, autophagy becomes cytoprotective to the tumor cells and supports their growth, survival, and malignant transformation under an adverse microenvironment via reactivation of autophagy status.

nature of autophagy in cancer. In the later stages of cancer, autophagy remains active and plays a key role in helping cancer cells survive. It does this by preserving cancer stem cells and supporting tumor dormancy [5]. This sustained activity allows cancer cells to stay in a suspended, stem-like state, giving them the flexibility to reactivate and spread. The protective mode of autophagy supports cancer progression by maintaining cancer stem cells, sustaining tumor dormancy, preserving a paused pluripotent state, and promoting the formation of polyploid giant cancer cells (PGCCs), and helps in metastasis and invasion [7].

To better understand how autophagy contributes to cancer progression, we analyzed the expression of key autophagy-related proteins across different histological grades of tumor differentiation of oral squamous cell carcinoma (OSCC). A total of 53 oral tissue samples were examined for ATG14 expression, including 10 normal tissues and 43 tumor samples subdivided by grade: 12 Grade I (well-differentiated), 14 Grade II (moderately differentiated), and 17 Grade III (poorly differentiated) OSCC cases. Our findings reveal a grade-wise increase in ATG14 expression from normal tissue to more aggressive tumor forms. Only 10% of normal tissues showed high ATG14 expression, while 33.33% of Grade I, 57.14% of Grade II, and 82.35% of Grade III tumors exhibited high ATG14 levels. The difference in expression across tumor grades was statistically significant ($p = 0.006$), suggesting that ATG14 expression correlates with tumor aggressiveness and may serve as a biomarker for OSCC progression. In parallel, we evaluated ULK1, a kinase that regulates autophagy initiation and directly phosphorylates ATG14. In a sample set of 64 tissues, only 10% of normal cases showed elevated ULK1 levels. In contrast, 44.1% of Grade I, 47.05% of Grade II, and 69.23% of Grade III OSCC cases demonstrated high ULK1 expression ($p = 0.04$)(Figure 2, adapted from Naik et al, 2020) [8]. This study suggests that as cancer progresses, the autophagy status increases accordingly.

However, from the above finding, it is impossible to conclude that the roller coaster model of autophagy is involved in cancer progression. To study the roller coaster ride of autophagy in cancer progression, it is essential to investigate the autophagic status in each stage of cancer progression. One of the major challenges in understanding how autophagy behaves across different stages of cancer is the lack of access to stage-specific human tissue samples. In most clinical settings, cancer is only diagnosed once it has progressed beyond the early stages. Early lesions like hyperplasia or mild dysplasia often go undetected, making it nearly impossible to collect tissue from the full timeline of cancer development in patients.

To overcome this limitation, we turned to a well-established hamster buccal pouch model using 7,12-Dimethylbenz[a]anthracene (DMBA)-induced oral carcinogenesis. In our ongoing studies of how autophagy changes during cancer development, we are working on a complex, stage-dependent pattern of autophagy. Our preliminary work using the hamster buccal pouch model of oral cancer offers strong evidence of autophagy dynamic behavior. To model stage-specific oral carcinogenesis, we treated male Golden Syrian hamsters with a 0.5% solution of DMBA in liquid paraffin, three times per week, for four weeks. The experiment was completed in 16 weeks, with animals sacrificed at various time points for tissue analysis. We observed a clear, time-dependent cancer progression including hyperplasia at 4 weeks, dysplasia by 8 weeks, and neoplasia emerging by 12 and 16 weeks.

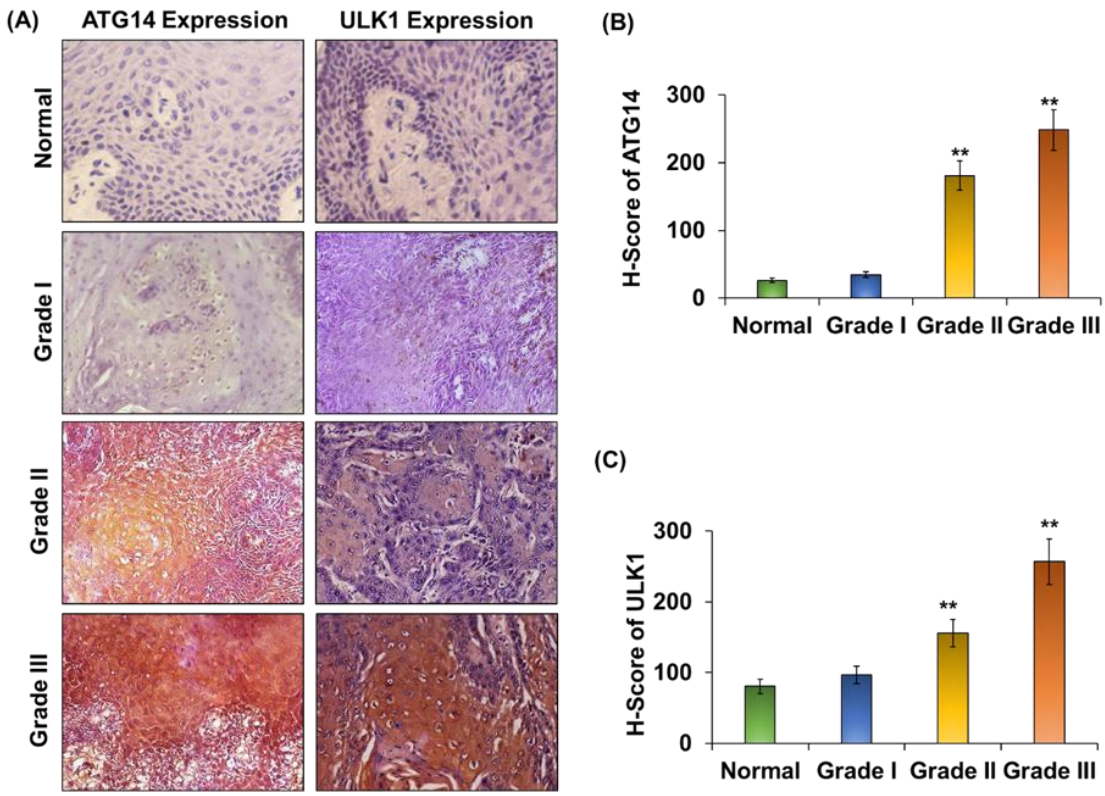

Figure 2. Expression of key autophagy-related proteins across different histological grades of tumor differentiation of oral squamous cell carcinoma using immunohistochemistry (A). The semiquantitative immunoreactive analysis for ATG14 (B) and ULK1 (C) was carried out using the histoscore method, calculated from the percentage of positive cells and staining intensity of different tissue samples. **p < 0.01 was considered significant compared to control (data adapted from Naik et al, 2020).

This confirms that DMBA reliably induces staged tumor development, allowing us to track changes in key molecular processes, including autophagy. To monitor autophagy, we focused on the protein LC3, particularly the LC3-II form, a marker for autophagosome accumulation. Western blot analysis revealed a striking pattern: LC3-II levels decreased at 4 and 8 weeks precisely during cancer initiation and then rose again at 12 and 16 weeks as tumors progressed. Similarly, other autophagy-related markers (ATG14, ULK1, and ATG5) also showed the same expression pattern (Figure 3). These findings suggest that autophagy is suppressed in the early stages, potentially allowing damaged cells to escape clearance and begin cellular transformation.

As the tumor advances, autophagy appears to reactivate, likely helping cancer cells survive under stress and fuel their continued growth. Our results provide insight into how autophagy shifts roles involved in cancer progression.

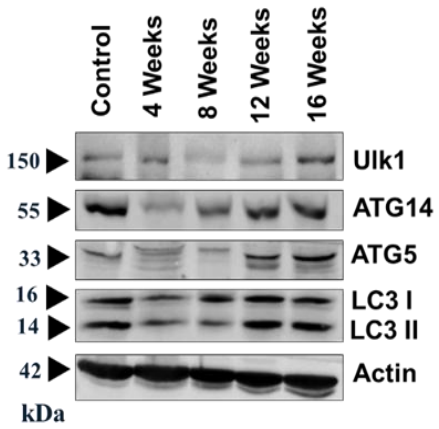

Figure 3. Expression analysis of key autophagy-related proteins during oral cancer progression. Western blot analysis was performed in hamster buccal pouch tissue lysates from control animals and those subjected to carcinogen treatment for 4, 8, 12, and 16 weeks. A biphasic expression pattern was observed: LC3-II levels decreased at 4 and 8 weeks, corresponding to early stages of tumor initiation, and increased again at 12 and 16 weeks during advanced tumor progression. Similar expression trends were observed for ATG5, ATG14, and ULK1. β-actin was used as the loading control. (Unpublished data).

However, extensive studies need to be documented on stage-specific autophagy status during cancer progression and its molecular mechanisms for better cancer therapeutics.

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DNA damage response in *Deinococcus radiodurans*: An example of logical shift in paradigm

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LexA is characterized as an auto-repressor that keeps a large number of Escherichia coli genes containing LexA box in their upstream region, repressed during the normal growth of the bacterium [1]. Upon DNA damage, the co-protease activity of RecA-ssDNA cleaves LexA and derepresses these genes [2,3]. The cellular and molecular events characterized post DNA damage and arrest of cell division have been collectively termed as DNA damage response and cell cycle regulations (SOS response) in E. coli and was established as a key mechanism by which bacteria respond to DNA damage and thus, regulate cell cycle [4–6]. More than 100 genes involved in genome integrity, energy metabolism, and growth have been identified as transcriptionally regulated in SOS regulon in response to DNA damage in E. coli . Majority of them are involved in DNA metabolism for repairing the damaged DNA, and other functions required to maintain genome integrity. Some SOS-regulated proteins, such as SulA in *E. coli* , *Cyanobacteria*, *Arabidopsis thaliana*, and YneA in *Bacilli*, function to halt cell division following DNA damage [7–9]. For instance, SulA inhibits the activity of FtsZ (the master regulator of bacterial cell division) attenuated till SulA is

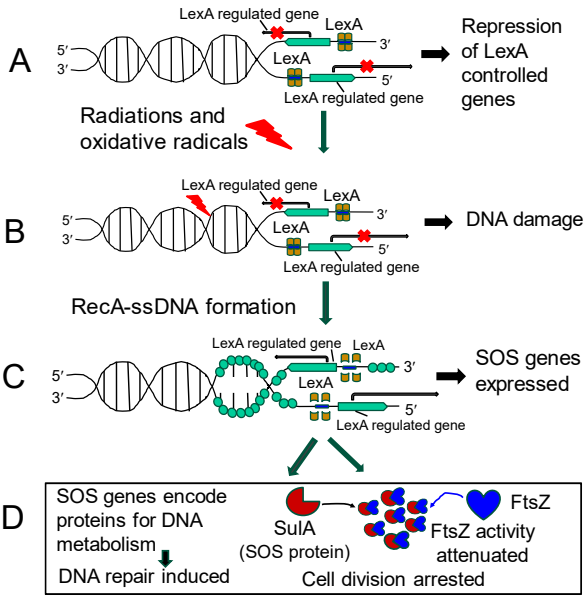

Figure 1. **Schematic representation of LexA/RecA-mediated SOS response in *Escherichia coli*.** The *E. coli* genome containing genes with LexA boxes are repressed by LexA under normal growth conditions (A). Upon DNA damage (B), the co-protease activity of RecA-ssDNA cleaves LexA (C) and depressed all SOS genes under LexA. The DNA repair SOS proteins repair damaged DNA while SulA a SOS protein binds with FtsZ and attenuate its function (D), till SulA is degraded by specific protease in *E. coli* . .

brought down by the specific protease and FtsZ resumes its functions. This concept was evaluated in many bacteria albeit mostly mesophilic ones, but held strongly in almost all the cases studied and thus LexA/RecA-mediated canonical SOS response became a paradigm of DNA damage response and cell cycle regulation in bacteria (Fig. 1).

Notably, findings from bacterial research particularly in organisms harbouring the limited copies of single circular chromosome have established several paradigms to explain the DNA metabolic processes in eukaryotes including the regulation of origin of replication in chromosome, firing of 'ori' once per cell cycle and segregation of duplicated genome before cytokinesis. The LexA/RecA-mediated DNA damage response mechanisms unilaterally drove the major research on bacterial response to DNA damage and cell cycle regulation and became a paradigm. LexA/RecA-mediated SOS response could not explain the similar processes in non-prokaryotes. Rather, ATM/ATR kinases were discovered as the sensor kinases of DNA damage in eukaryotes and the cascades of serine/threonine (Ser/Thr) phosphorylation of DNA repair proteins involved in DNA metabolisms and cell division regulation became the paradigm of DNA damage response and cell cycle regulation in eukaryotes. While the majority bacterial encodes the genome elements for LexA/RecA-mediated canonical SOS response and showed the sporadic occurrence of Ser/Thr protein kinases (STPKs) undermine the possibility of Ser/Thr phosphorylation/dephosphorylation-based regulation in bacteria. Consequently, the DNA damage response and cell cycle regulation in prokaryotes and eukaryotes were literally separated between LexA/RecA-mediated SOS response in bacteria and Ser/Thr phosphorylation /dephosphorylation by STPKs in eukaryotes. Recently, the S/T phosphorylation of DNA repair proteins by eukaryotic type STPKs have been reported and demonstrated that the function of these proteins is regulated by phosphorylation /dephosphorylation mechanism(s) in those bacteria [10].

Extreme radioresistance in *Deinococcus radiodurans*

Bacteria belonging to *Deinococci* family are characterized by their extreme resistance to DNA damaging agents. These bacteria have a highly complex cell envelopes, which include a lipid-rich layer but lack lipopolysaccharides (LPS), instead being supported by a proteinaceous S-layer. D. radiodurans R1 was the first identified member of *Deinococci* family, was isolated from the spoiled meat irradiated with a sterilizing dose of Y radiation, in 1956. An efficient double-strand breaks (DSB) repair and strong oxidative stress tolerance mechanisms have been attributed to the extreme phenotypes in this bacterium [11]. The first genome sequence of *D. radiodurans* R1 was published in 1999 [12]. Since then, a large number of *Deinococci* species have been isolated and characterized from diverse climatic conditions ranging from Antarctica to hot springs, and more than 200 genome sequences are currently at various stages of release in public databases. Unlike other bacteria, *D. radiodurans* can resists ~200 DSB and ~3000 single strand breaks, and harbours multipartite genome system comprising of 2 chromosomes and 2 mega-plasmids [13,14]. Each of these genome elements are present in 6-10 copies per cell at logarithmic growth phase. The role of ploidy and genome multiplicity in radioresistance was long speculated till recently demonstrated. While the primary chromosome is indispensable, a reduction in copy number of secondary genome elements made cells compromising wild type resistance to Y radiation [15], providing evidence for the role of secondary genome elements in extreme phenotypes in this bacterium. Although, oxidative stress tolerance and an efficient DNA repair processes contributing to extreme phenotypes explain much of its resilience, the presence of polyploid chromosomes especially those containing a typical *E. coli* -like oriC, whose regulation governs chromosome copy number, is intriguing and merits further investigation.

The RecBC pathway of homologous recombination, a well-known mechanism of DSB repair in majority bacteria, has been ruled out in *D. radiodurans* [16], instead the RecFOR pathway of homologous recombination is found to be crucial to DSB repair in this bacterium [17,18]. It is now established that RecA dependent DSB repair is fundamental to DNA repair [19] albeit RecA activity is differentially regulated by the Ser/Thr phosphorylation [20]. An efficient RecA depended DSB repair supported by the Extended Synthesis Dependent Strand Annealing (ESDSA) mechanism for high fidelity repair of shattered genome [19]. Regarding the mechanisms controlling base damage / mismatch repair, it contains both the pathways of nucleotide excision repair like UVR and UVS system, short patch base excision repair and mismatch repair [11.13.21].

DNA damage response in *Deinococcus radiodurans*

One of the remarkable features in *D. radiodurans* is a rapid and error-free reassembly of shattered genome through various DNA damaging agents. DNA damage response activated by canonical error-prone LexA/RecA-mediated recombination repair which is common in *E. coli* [22] found to be absent in this bacterium. Additionally, though *D. radiodurans* encodes RecA and three LexA orthologs, it does not possess UmuD and a translesion DNA polymerase involved in error-prone DNA synthesis during SOS repair [12]. Transcriptome analysis in cells exposed to Y radiation showed a rapid upregulation of several genes including recA in response to DNA damage. However, this increase in recA transcription was found to be independent of LexA suggesting that the classical *E. coli* -type DNA damage response and cell cycle regulation mechanisms are absent in *D. radiodurans* [23–27]. Nonetheless, this bacterium adjusts it's both proteome and transcriptome upon Y radiation exposure [28–30], arguing in favour of some mechanism that would help the bacterium to respond the Y radiation effects, and relay it to the genome functions and protein homeostasis.

The mechanisms underlying desiccation and Y radiation-induced transcriptional regulation of gene expression have been studied independently. For example, IrrE (also called PprI) was demonstrated to act as a master regulator of recA expression in this bacterium [31].

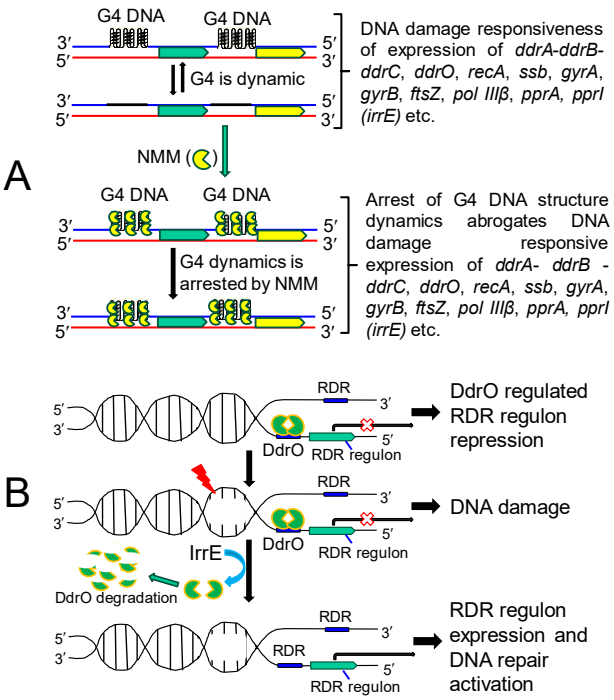

Figure 2. **Alternate probable mechanisms for regulation of gene expression in response to DNA damage in *Deinococcus radiodurans*.** (A) A large number Y radiation responsive deinococcal genes contain guanine quadruplex (G4) DNA in their upstream region. The arrest of G4 DNA structure dynamics synthetically using G4 DNA binding drug N-methyl mesoporphyrin IX (NMM), the Y radiation inducible expression of these genes is abrogated. (B) Similarly, some genes contain Radiation and Desiccation Responsive Motifs (RDRM) in their upstream region. DdrO binds with RDRM and keep these genes repressed. In response to DNA damage IrrE is induced and replace DdrO from RDRM. Free DdrO is degraded and repression is relieved. .

PprI a metalloprotease, cleaves DdrO and results into relaxation of DdrO repressed genes containing Radiation Desiccation Response Motifs (RDRM) in their upstream regions [32,22]. The other mechanism that could explain DNA damage responsive expression of gene was through the Guanine quadruplex (G4) DNA structure dynamics [34]. It was shown that the arrest of G4 DNA structure dynamics results to loss of radioresistance [35], and an attenuation of DNA damage responsive gene expression in response to Y radiation [36]. It may be noted that all Y radiation responsive genes are not under the control of G4 DNA structure dynamics. Therefore, the regulation of Y radiation responsive gene expression using either RDRM mediated PprI/DdrO regulation, and /or through G4 DNA structure dynamics provided strong evidence of some alternate mechanism of DNA damage responsive gene expression (Fig 2). These examples were limited to regulating a few genes in the *D. radiodurans* genome, and the mechanisms that would regulate the synthesis or activity of other SOS proteins are not clear. However, a strong possibility of their functional cross talks with other mechanism(s) cannot be ruled out.

Discovery trail of new mechanism of DNA damage response and cell division regulation

Pyrroloquinoline quinone (PQQ) was known as a coenzyme for certain redox enzymes including gluconate dehydrogenase (GDH) and methanol dehydrogenase [37]. PQQ synthesis in bacteria requires at least 5 enzymes (PqqA, PqqB, PqqC, PqqD, and PqqE/PqqF) and genes encoding them are organized in an operon. The PqqE is unique and said to be a regulatory enzyme of PQQ biosynthesis. In bacterial physiology, the PQQ role was known in mineral phosphate solubilization. The gluconic acid produced by direct oxidation of glucose would diffuse out from bacterial periplasm to the surrounding medium including soil, that would help in the solubilization of insoluble metal phosphates into inorganic phosphorous. The free phosphorous is then used by plants and microbes as biofertilizer. The PQQ interaction with bacterial dehydrogenases is known, through the conserved β propeller motifs called PQQ Interaction motifs (PQIM) [38]. In *D. radiodurans*, the genome contains a gene (pqqE) predicted to encode a putative PQQ synthase. Although, this enzymes do not directly involved in DNA repair and radioresistance. Interestingly, when *E. coli* was genetically engineered to produce PQQ, it exhibited high tolerance to the photodynamic effects of Rose Bengal. These cells showed an unexpected increase in resistance by approximately 8–10 log cycles to UVC radiation (254 nm) [39] (Fig. 3). Since UVC-induced cell death primarily results from DNA damage rather than oxidative stress, these findings suggested that PQQ may function beyond its known role in oxidative stress protection, prompting further investigation into its potential role in DNA damage response in *D. radiodurans*.

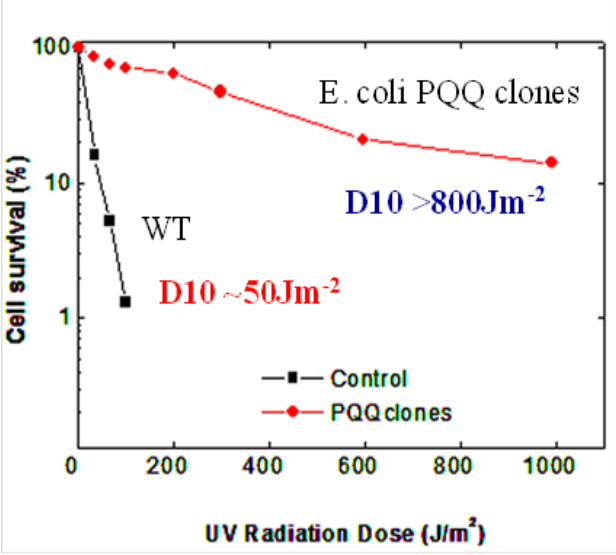

Figure 3. **UVC (254nm) response curve of *Escherichia coli* cells producing pyrroloquinoline quinone (PQQ).** The transgenic *E. coli* cells expressing pqqE synthase gene of *Deinococcus radiodurans* and making PQQ (*E. coli* PQQ clones) were exposed to different doses of UVC radiation at 254 nm, and cell survival was compared with wild type cells without PQQ (WT). (Courtesy: [39]).

In *D. radiodurans*, the *Δ* pqqE (PQQ-deficient) mutant was assessed for Y radiation response and reassembly kinetics of shattered genome by pulsed field gel electrophoresis. The findings revealed a previously unrecognized role of PQQ in DSB repair, firmly establishing its importance in radioresistance [40]. We provided mechanistic evidence that PQQ contribution in radioresistance and DSB repair was through a protein encoded by DR_2518 ORF in the genome of *D. radiodurans*. The Δ2518 cells showed ~3 log cycle drops in wild-type resistance to Y radiation and the DSB assembly in these cells were completely impaired till 6-8 hours of post irradiation recovery (PIR) [41] (Fig. 4). The expression and activity of both PQQ and DR2518 were responsive to oxidative stress and DNA strand breaks. Structurally, DR2518 is divided into N terminal STPK domain and C-terminal region containing seven tandem repeats of PQIM (beta propeller motifs).

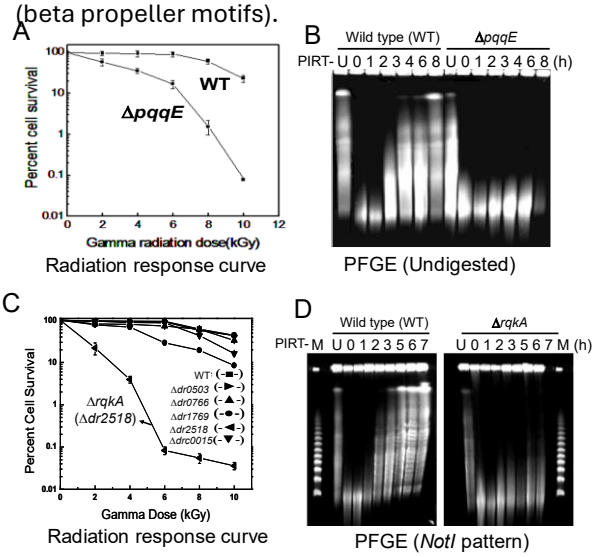

Figure 4. **Gamma radiation response curve and shattered genome assembly kinetics in *Deinococcus radiodurans*.** The ΔpqqE cells (PQQ minus) (A, B) and *Δ* dr2518 *Δ* rqkA) (C, D) were exposed to different doses of Y radiation and cell survival (A, C) and reassembly of shattered genome in cells exposed to 6.5kGy Y radiation (B, D) were monitored. Results were compared with wild type and other mutants as applicable. (Courtesy: [40,41])

Altogether, DR2518 protein was characterized as a Radiation Responsive Ser/Thr Quinoprotein kinase and named as RqkA [41]. Transcriptome analyses of Δdr2518 and ΔpqqE cells strongly indicated that PQQ and RqkA interaction affect genome functions in response to Y radiation [42]. These results suggest that both the synthesis and activity of PQQ and RqkA are oxidative stress and DNA damage responsive, and their functional interaction stimulate the autokinase activity of RqkA and global genome functions upon DNA damage.

Ser/Thr quinoprotein kinase (RqkA) mediated stress response to DNA damage

The proteome of *D. radiodurans* was searched for the conserved phosphor-S/T epitopes containing proteins. A number of proteins harbouring conserved phospho-Ser/Thr (S/T) epitopes, including RecA, PprA, DnaA, DprA, SbcC, FtsZ, FtsA and DivIVA [14] were detected. The effects of RqkA phosphorylation on the functions of DNA repair or DNA replication related proteins like RecA, PprA, and DnaA, and cell division proteins such as FtsZ, FtsA, and DivIVA have been characterized in significant details. RecA, an important DSB repair protein and PprA a pleiotropic protein involved in radiation resistance [43-44] showed the requirement of a specific S/T phosphorylation for the improved in vitro actvities, and in vivo roles in extreme radioresistance and DSB repair in this bacterium [20,45,46] (Fig. 5). Similarly, RqkA phosphorylation of FtsZ and FtsA aborts the interaction of these proteins that are required for productive cells division47. Phosphorylation of FtsZ at 235 and 335 positions leads to destabilization of FtsZ polymer and in vivo polymerization / depolymerization dynamics [48] (Fig 6). RqkA phosphorylation of DivIVA at T19 position could change the orientation of septum formation, shifting it from perpendicular to tilted leading to non-productive cytokinesis. Further, RqkA phosphorylation affect the interaction preference of DivIVA between cell division and genome segregation proteins [49].

The enhancement of DNA repair function with the concurrent arrest of cell division upon Ser/Thr phosphorylation by RqkA nearly mimics the function of sensor kinases in response to DNA damage in eukaryotes [50]. Interestingly, DprA a key player of natural transformation (NT) that plays a crucial role in switchover of ESDSA to single-strand annealing repair in *D. radiodurans*, is also identified as an RqkA phospho-substrate, may implicating DprA phosphorylation roles in DNA repair and NT. These observations point to a potential eukaryote-like role of RqkA in DNA damage signalling and cell cycle regulation in *D. radiodurans*, analogous to the functions of eukaryotic Ser/Thr kinases such as ATM and ATR, and warrant further investigation in other bacteria with unique genome architectures.

In conclusion, research on bacterial responses to bacterial response to DNA damage and cell cycle regulation often aligns with established paradigms such as the canonical LexA/RecA-mediated SOS response. However, new findings and understanding occasionally, challenge the established paradigm. When such odd results are explained with robust experimental supports and sound reasoning that leads to scientific novelty. LexA/RecA-mediated canonical SOS response is a paradigm and synonym to bacterial response to DNA damage and cell cycle regulation. However, current understanding of DNA damage response and cell cycle regulations of *D. radiodurans* does not conform to the classical SOS paradigm. Instead, it appears to use an alternative mechanism involving STPKs and phosphorylation-dephosphorylation cascades, a system more commonly associated with eukaryotic cells.

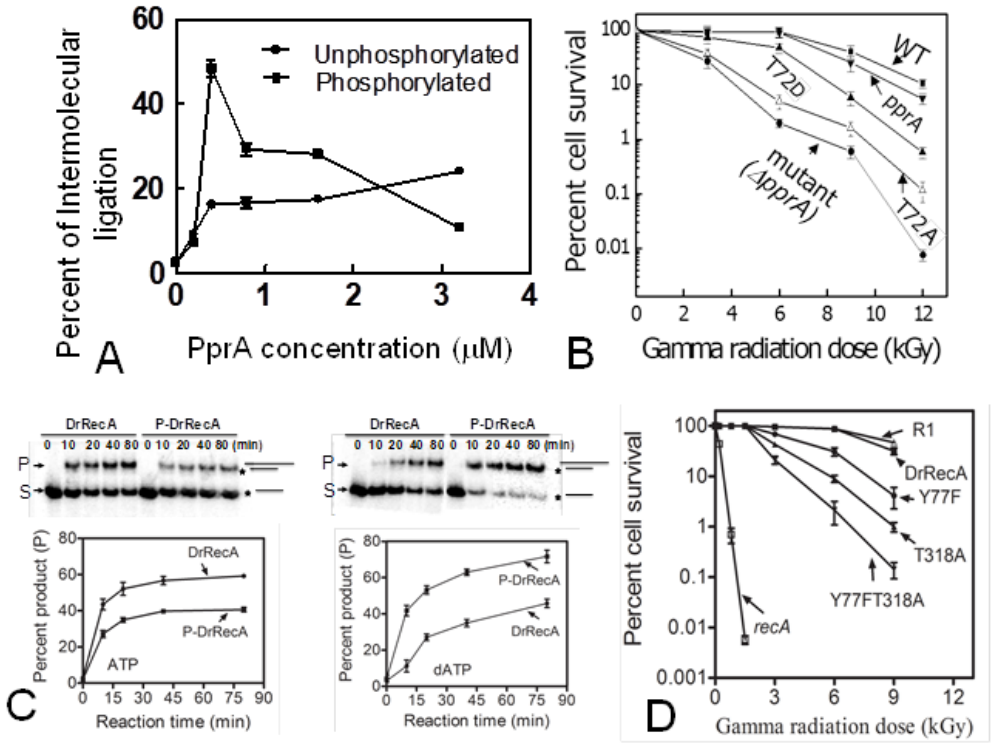

Figure 5. **Effect of RqkA phosphorylation on DNA metabolic activity of DNA repair proteins and their roles in radioresistance in *Deinococcus radiodurans***. The effect of RqkA phosphorylation on the stimulation of DNA end joining activity of T4DNA ligase by PprA (unique to *Deinococci*) (A) and functional complementation of ΔpprA mutant's loss of radioresistance by phosphomutants of PprA (B) were monitored. [Courtesy: 45]. Similarly, the effect of RqkA phosphorylation on strand exchange activity of drRecA in the presence of ATP and dATP (C) and functional complementation of Δ recA mutant's loss of radioresistance by phosphomutants of RecA (D) were monitored. [Courtesy: 20].

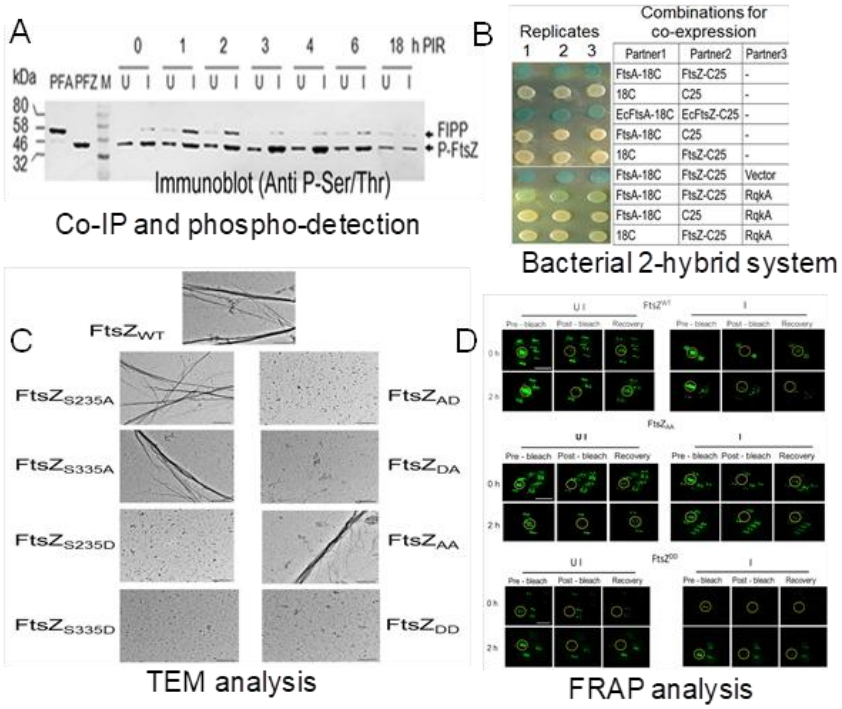

Figure 6. **Effect of RqkA phosphorylation on FtsZ in its cell division roles in Deinococcus radiodurans**. In vivo phosphorylation of FtsZ in cells collected at different time interval of post irradiation recovery period (PIR) was SHAM control of unexposed cells (A). The interaction of drFtsZ with drFtsA as a function of RqkA was monitored using bacterial 2 hybrid system (B). (Courtesy: [47]). Phosphosites S235 and S335 in drFtsZ were replaced with alanine (S235A/S335A or aspartate (S235D/S335D) and their polymerization characteristics were monitored in vitro by TEM (C). The effect of phosphorylation on polymerization/depolymerization dynamics of selected mutants were monitored by FRAP assay in host cells (D). (Courtesy: [48])

Here, we have summarised the output of a long-standing research on *D. radiodurans* that was not done to test a frozen hypothesis but with an open question on how does this bacterium withstands to a non-permissible dose of ionizing radiation. All the work carried out for last several years, when viewed collectively, reveals a new dimension on bacterial response to DNA damage and cell cycle regulations. The proposed mechanism centres around a well-established pathway in eukaryotes including yeast, which operate through STPKs and S/T phosphorylation/dephosphorylation cascades. Our findings suggest that *D. radiodurans* follows a distinct mechanism of DNA damage response and cell cycle regulation through the phosphorylation-mediated regulation of proteins functions, and RqkA seems to be a key regulator of oxidative stress and DNA damage response in this bacterium. RqkA phosphorylation of both DNA repair proteins like RecA and PprA and cell division proteins like FtsZ, FtsA, and DivIVA (studied so far), and the effect of phosphorylation on their activity regulation offer an analogous model of DNA damage response and cell cycle regulation in *D. radiodurans*, to the one that bears similarities to eukaryotes (Fig. 7) [51]. A possibility of G4 DNA structure dynamics controls of DNA damage response, and DdrO/PprI-RDRM interaction mediated regulation of DNA damage responsive gene expression, possibly operating through RqkA-mediated phosphorylation, cannot be ruled out. Further work would be required to ascertain whether S/T/Y phosphorylation-mediated DNA damage response and cell cycle regulation represents a broader, yet undiscovered, mechanism in bacteria. .

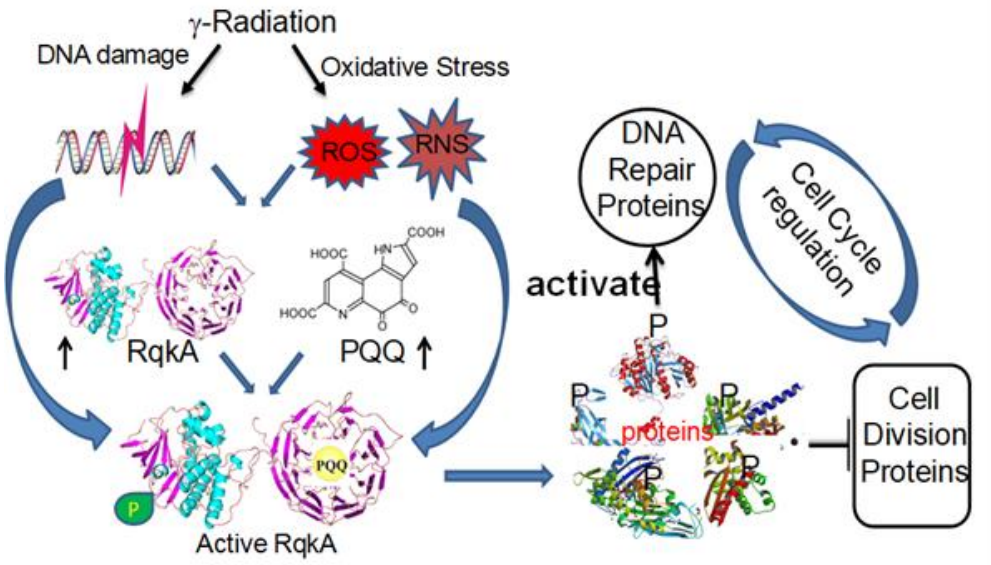

Figure 7. **Schematic representation of a Y radiation responsive Ser/Thr quinoprotein kinase (RqkA) mediated DNA damage response and cell cycle regulation in *Deinococcus radiodurans*.** The Y radiation induces the synthesis of both RqkA and pyrroloquinoline quinone (PQQ). The PQQ activates the autokinase activity of RqkA, which is predicted to have a number of putative substrates in *D. radiodurans* that include DNA repair proteins like RecA, PprA, and DnaA, and the cell division protein like FtsZ, FtsA, and DivIVA. All these proteins undergo phosphorylation in vivo which showed a kinetic change during post irradiation recovery. On one hand, the phosphorylation of RecA, PprA, and DnaA enhances their activity that would be required for their efficient function in radioresistance. On the other hand, FtsZ and FtsA phosphorylation alters their interaction, the polymerization/depolymerization dynamics of FtsZ and deregulated activity of DivIVA. (Courtesy: [51]).

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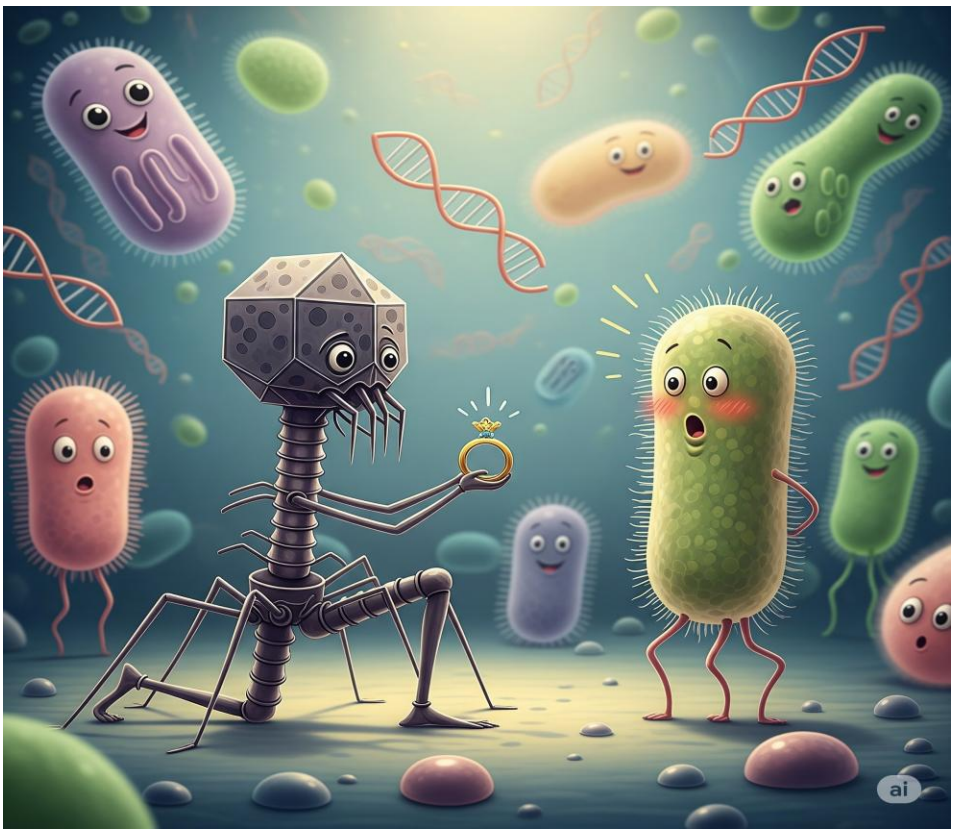
Fun time!



Textbook to petri dish: Why hands-on training in cell biology matters

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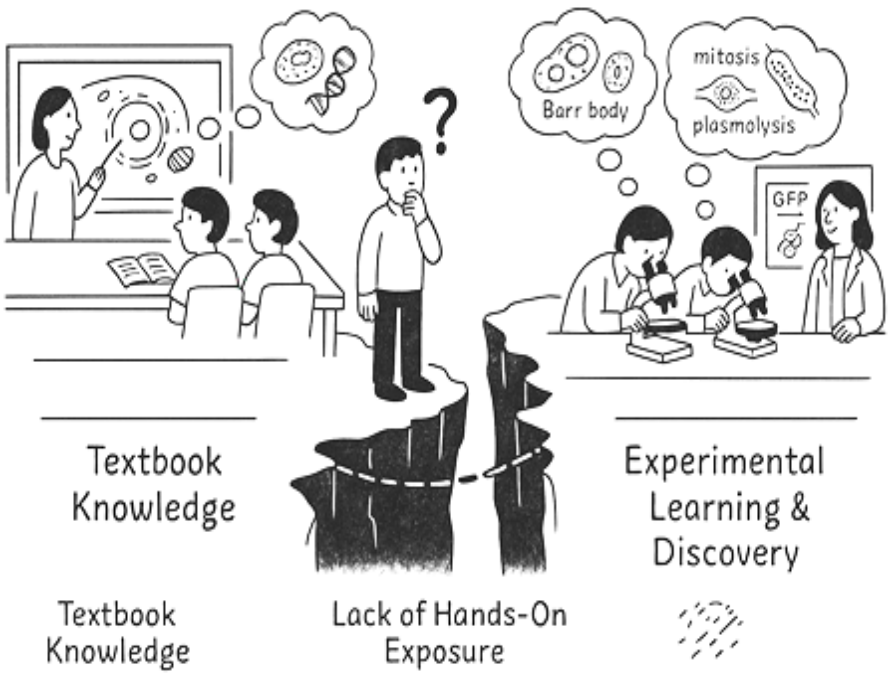
In recent years, there has been a growing recognition of laboratory courses as an essential component of science education and a crucial stepping stone for a career in academic research or industry, particularly at the undergraduate and postgraduate levels. Among various disciplines of biology taught in colleges and universities, cell biology stands out as an attractive field, as it provides foundational insights into how cells—the building blocks of life and autonomous units that decode genetic information—function, communicate, and respond to the environment. In a simplistic view, while genes provide instructions to build proteins, it is the cell that contains the machinery for gene-decoding to synthesize the molecules necessary to achieve the properties of life. While unicellular organisms are self-sustaining and independent, the cells of higher-order multicellular organisms coordinate their functions to drive the complex behavior of an organ and, ultimately, an organism, thus broadly constituting the defining features of life. Understanding cellular processes and behaviors is therefore essential for students, not only to gain insights into how cellular machinery governs the features of life, but also to comprehend more advanced concepts in genetics, physiology, biochemistry, and molecular biology. Moreover, a strong grounding in cell biology education helps students understand the cellular basis of diseases such as cancer, neurodegenerative disorders, and infections, and prepares them for careers in medicine, pharmacology, and biomedical research. Intersection of cell biology with diverse fields such as bioengineering, synthetic biology, immunology, and nanotechnology opens up opportunities for students in interdisciplinary careers and translational science with advanced specialization.

While theoretical knowledge provided in classrooms is the first and essential step in training students in cell biology, hands-on laboratory trainings are what actually spark curiosity among students, build a foundation for scientific thinking, and enable students to truly begin to understand how a cell and its components function. Peering down a microscope to visualize cells and cellular components or observing how cells divide and chromosomes separate during division can greatly transform a student's theoretical

knowledge may prepare students for college/university examinations or help them succeed in national-level competitive tests, those who have not been exposed to hands-on training will remain ill-equipped to perform basic laboratory techniques or to develop as critical thinkers—a prerequisite for future experimentalists. For a subject like cell biology, which is inherently experimental and predominantly visual, a lack of meaningful practical training can not only impede deep understanding of the subject, but may also stifle creativity and innovation. This article highlights the crucial role of hands-on training in cell biology education and explores strategies to bridge the gap between classroom teaching and experimental learning through practicals or experiments.

Challenges in cell biology lab training at undergraduate and postgraduate levels

Over the past few decades, cell biology has remarkably advanced from merely observing cell structures using light microscopy and basic staining techniques, with limited ability to manipulate the genome, to visualizing cellular details with unprecedented clarity using high-resolution imaging techniques, observing cellular processes in real-time, and precise gene editing using tools like CRISPR-Cas9. However, except for a few well-equipped and well-funded institutes and universities, the vast majority of colleges and universities in India still lack basic, let alone state-of-the-art, equipment, essential for hands-on training in cell biology. This lack of infrastructure severely limits students' exposure to hands-on training or their ability to conduct even small-scale cell biology experiments. Due to the lack of proper infrastructure and adequate reagents, undergraduate and postgraduate programs often resort to "demonstration experiments" rather than hands-on teaching. As a result, students are limited to observing a fixed slide under the microscope and are deprived of the essence of cell biology experimentation, often leaving the lab without a proper understanding of the rationale behind the protocols used in the experiment. In many cases, students are evaluated on their 'lab performance' based on solely written records as no actual experiments could be conducted. Overcrowded classrooms and under-resourced labs further exacerbate the disconnect between theoretical and practical knowledge.



In addition to resource limitations in the labs, a shortage of faculty, many of whom are distanced from active research, and trained technical staff designated for lab operations remains a barrier to fulfilling the needs of students for effective cell biology lab training. Also, urban–rural divide in our country is a major concern in cell biology education with colleges and universities in villages and smaller towns grappling with a variety of issues such as limited infrastructure, insufficient teaching staff, and unreliable electricity. Students in these settings are often deprived from real hands-on training and are instead reduced to performing dry-lab simulations, model building or diagram drawing in their so-called 'practical' modules. This disparity, faced predominantly by rural students, not only stands as an impediment for their educational growth, but also dampens their scientific aspirations. Even many educational institutes in urban or suburban areas face similar challenges. For meaningful cell biology education with a strong emphasis on practical learning, the basic infrastructural and resource needs must be fulfilled.

Small experiments, big excitement

Given the challenges encountered by many colleges and universities in providing cell biology lab training and considering the resource limitations, innovative teaching approaches must be developed to meet students' needs even with limited infrastructure. At the same time, experiments should be designed in a way that they will spark excitement among students, encouraging many of them to pursue cell biology or related research, or to choose career options where they can benefit from such training. Below are a few examples of experiments that are relatively simple and easy to execute:

- Studying mitotic cell division using squash technique: This experiment involves using root tips, fixing them in acetomethanol, staining with toluidine blue or Giemsa, and squashing the tissue on a glass slide with a cover slip. The stained tissue samples are then observed under a microscope. Students can be trained to identify different phases of mitotic cell division.
- Identifying Barr bodies in buccal epithelial cells: Barr bodies are found in the nuclei of female mammalian cells. To visualize Barr bodies, buccal smear from a female is spread on a glass slide, fixed, and stained with either methylene blue or cresyl violet. After mounting with a cover slip, the slide is observed under a microscope. Barr bodies can be visualized as a darkly stained structure on the periphery of the nucleus.
- Blood cell count using a hemocytometer: A blood sample is taken by pricking a finger with a fine needle and the sample is diluted in a solution. The dilution factor is noted, and a measured amount of the diluted sample is loaded into the hemocytometer's counting chamber using capillary action. After putting a coverslip over the counting chamber, the sample is observed under a microscope. The cells are counted and the total number of counted cells is multiplied with the dilution factor and a correction factor to obtain the cell concentration.
- Plasmolysis in plant cells: To observe the effect of osmosis in plant cells, this simple experiment can be designed. A thin layer of epidermis from the inner surface of an onion is peeled using forceps and carefully placed on a glass slide. The sample is first observed under a microscope, then treated with 5% NaCl solution for 10 minutes, and observed again. Students can visualize how the cell membrane shrinks away from the cell wall and the cytoplasm retracts due to plasmolysis.
- Visualizing cell organelles (for well-equipped labs): Cultures cells can be processed for indirect-immunofluorescence using antibodies targeting marker proteins of different organelles like mitochondria, Golgi complex, lysosomes, microtubules, etc. The cells can be counterstained for nuclei or actin using appropriate dyes.

Upon staining, different cell organelles are visualized under an epifluorescence or confocal microscope.

• Live-cell imaging to visualize organelle dynamics (for well-equipped labs): Cultured cells are transfected with GFP or RFP-tagged organelle markers, and organelle dynamics are visualized in real-time using a fluorescence microscope with a live-cell handling module.

The above experiments are just a few suggestions that can be incorporated into cell biology practical labs. Depending on available resources, many such simple yet exciting practicals or experiments can be thoughtfully designed. However, appropriate protocols must be followed and conditions should be optimized by lab instructors before the initiation of practical sessions.

Educating the educators

In order to cater to the needs of the growing number of students enrolled in life sciences programs across the country, faculty members in the first place should possess a strong foundation in cell biology, both in theoretical and practical aspects. Many biology teachers in underfunded or rural colleges or universities have not had the opportunity to gain exposure to cell biology techniques, nor are they familiar with the recent advances and discoveries in cell biology. For them, regular faculty development programs are essential. National institutes and research centers should design programs tailored for educators and organize hands-on training workshops and refresher courses. To involve faculty members located in remote areas and unable to be physically present for trainings and workshops, virtual learning platforms can be utilized. Additionally, faculty awareness should be enhanced regarding the use of open-access educational tools, resources, and journals.

To strengthen cell biology education and equip educators with up-to-date knowledge, several central universities and national research institutes or centers such as the IITs, IISERs, IISc, NCBS, NISERs, AIIMS, DBT or CSIR institutes, are serving as regional training hubs. These institutions already have the infrastructure and expertise to conduct cutting-edge cell biology experiments and host faculty development programs. Additionally, faculty members in colleges are greatly benefitting from Government initiatives such as the DBT STAR College Programme, aimed at improving science teaching across the country. However, It is important that faculty members who require training are aware of such opportunities and supported to participate in regular intervals, so they can impart updated knowledge in cell biology and foster a spirit of inquiry-driven learning among students.

Looking ahead

As India intensifies its investments in biotechnology parks, precision medicine and digital health, and aspires to become a global leader in life sciences by supporting to a mature research ecosystem, there is, and will continue to be a growing demand for life science graduates. In this rapidly evolving scientific landscape, cell biology stands at the core of many transformative breakthroughs—from understanding disease mechanisms to devising next-generation therapeutic strategies. Those equipped with a strong foundation in cell biology and skilled in cutting-edge techniques will play a pivotal role in driving this scientific progress and contributing to the country's biomedicine-driven economy. Strengthening cell biology education, especially through hands-on training and up-to-date methodologies will be the key to building a competent, future-ready scientific workforce.

Featured Article

Peering through the lens of imagination: How science learned to see

Savitha Sekhar, Science Communicator and Senior Graphic Designer at CACTUS Communications.

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Even in the dimmest corridors of human history, that is long before "science" even became a word, we can find evidence of humanity's desire to understand and communicate the mysteries of the natural world—a desire that found a voice in visual representation. To appreciate the evolution of visualization in the sciences, let's rewind to prehistoric times. And from there, let's traverse the ages and explore the contributions of some of the artists and scientists who pioneered the art of depicting the natural world.

We'll also stop and look at a few milestones—randomly chosen landmark discoveries (some pivotal, others simply intriguing)—which contributed to the development of tools and methods that allow us today to visualize everything, spanning from the subatomic to the galactic, analyzing data sets from the smallest to the largest.

In prehistoric times—and this we all know—visual representation emerged through cave paintings; like those in Chauvet and Bhimbetka, portraying animals and reptiles. As settled communities arose, abstract concepts found expression in illustrations. Case in point: a map, as in the Babylonian clay tablet. Advancements in materials and techniques led to more detailed depictions of the natural world, such as the ancient Egyptian Green Room. We can observe from such early artifacts—depicting concepts ranging from geometric figures for calculations to celestial charts and navigational maps—our growing understanding of the world and a desire to explore beyond one's immediate surroundings.

With the Middle Ages came treatises featuring meticulous illustrations, enabled by the printing press. The Renaissance witnessed a leap in quality with artists like Leonhart Fuchs and Maria Sibylla Merian producing realistic depictions of plants and insects. Notable contributions to natural history illustrations continued with John James Audubon, Elizabeth Gould, and Ernst Haeckel.

Anatomical illustrations too progressed similarly. Artists and physicians would collaborate to produce paintings, drawings, and sculptures of the human body. In some cases, such as that of Da Vinci, artists would dissect bodies themselves. Physician–artists like Vesalius, challenged the old guard, correcting wrongly held notions, with accurate illustrations.

Modern masters like Brödel and Netter built on this, while Kahn's metaphors like Man as Machine made complex science accessible to the public.

With the development of the microscope, emerged a new field of science. Robert Hooke's Micrographia revealed the microscopic world of cells. Leeuwenhoek, in 1675, using even better microscopes, captured the microbial world of bacteria, protozoa, red blood cells, and more, through detailed drawings (with the help of local illustrators). Santiago Ramón y Cajal would observe sections of the brain using microscopy and draw what he perceived (such beautiful illustrations!). This led him to postulate the neuron doctrine, which laid the foundations of modern neuroanatomy.

While, many were raptly absorbed in studying the microscopic words, others were more intrigued by the Cosmos. The Nebra sky disc is one of the oldest depictions of astronomic phenomena. Aristarchus calculated (using shadows and geometry) and illustrated the sizes of celestial bodies, and this work is believed to have influenced Copernicus who proposed the Heliocentric theory.

The Antikythera mechanism, a marvel of ancient engineering, (perhaps the earliest known predecessor to the modern computer), could calculate and display information about astronomical phenomena. The invention of the telescope broke new grounds. Galileo, who kept developing improved versions of the telescope, captured unseen landscapes of the moon and stars which he shared through printed illustrations.

Hevelius built upon this legacy with meticulous lunar-surface and star maps. Edmund Halley further showcased the power of visualization by using illustrations to predict celestial events. In 1850, John Whipple and William Bond were the first to photograph a star, Vega, (other than the sun, that is); they used a 38-cm Harvard refractor. The 20th century saw artists like Chesley Bonestell who used art to inspire the commoners and shape the public perception of space exploration; his work even influenced popular media and Hollywood.

Chemistry and physics also benefitted from visualizations. Descartes' spiral effluvia, laid the groundwork for later developments in understanding magnetism.

Michael Faraday, lacking formal mathematical training, employed visual methods to understand electromagnetism—work which allowed James Clerk Maxwell to formulate equations of electromagnetism. James Clerk Maxwell created a 3D model of thermodynamic surface using clay, to visualize Gibbs theoretical concept.

John Dalton illustrated atoms. Mendeleev created the periodic table. Moseley's experiments with X-ray spectra plotted as a graph revealed a direct link between atomic number and spectral lines, revolutionizing the periodic table's organization. Archibald Couper introduced the idea of pictorial representation for structural formulas for organic compounds.

Friedrich August Kekulé proposed the structure of benzene (with a 3D model), which allowed the prediction of then unknown isomers. His student, Jacobus van't Hoff's tetrahedral view of carbon revolutionized organic chemistry. Hofmann was the first to use 3D models of molecules in his lectures.

Pauling and his colleague Corey pioneered the concept of space-filling molecular models in molecular visualization, forming the basis for the widely used CPK models in chemistry. The development of X-ray crystallography in 1912 enabled the visualization of atomic arrangements in crystals, enhancing our understanding of molecular structures. John Kendrew's x-ray diffraction studies on myoglobin, illustrated by Irving Geis, were a landmark in structural biology. Roger Hayward, an artist and architect turned scientific illustrator, collaborated with Linus Pauling, making substantial contributions to molecular illustration in chemistry.

Watson and Crick unravelled the DNA structure through a combination of 3D model visualization (using cardboards!) and insights from Rosalind Franklin's Photo 51.

As strange as it may sound, mathematics also saw its fair share of visualizations. Euclid's Elements laid the foundation for geometry in 300 BCE. Euler's formula, in the 17th century, influenced key concepts like topology and graph theory, crucial for modern scientific visualization. Menaechmus, Archimedes, and Apollonius advanced the understanding of conic sections, vital in real-world applications. In the 17th century, Fermat and Descartes introduced analytic geometry, merging geometry with algebra. Pascal's calculator and Babbage's computers set the stage for advanced computing. These developments, while not all illustrative, played a pivotal role in shaping modern scientific visualization methods.

The concept of visualization, especially with respect to conveying data, first originated with the use of maps, charts, and graphs. The earliest evidence of this is in the tenth century, where planetary movements were represented graphically. Nicole Oresme pioneered the plotting of theoretical functions in the 14th century, and Michael Florent van Langren created the first known statistical graph in the 17th century.

Edmond Halley's plotted the first data map in 1686. Dr. John Snow charted the location of deaths from cholera in central London (1854). Florence Nightingale revolutionized healthcare data visualization in the 19th century (her Coxcomb chart was a game changer). By the mid-1800s, new forms of statistical graphics were used for economic and national data.

The 20th century saw the development of stereograms, contour plots, and computer graphics, with pioneers like Marie Tharp (seafloor maps using sonar data) and Etienne-Jules Marey (pioneered the graphic method) making significant contributions. Bertin established a framework for effective visual communication with his Semiology of Graphics (1967).

The 20th century brought a new era with computer graphics, using tools like MATLAB, Mathematica and Tableau, for dynamic visualization. The digital revolution marked a turning point in science visualization. Computer graphics and 3D modeling expanded possibilities, offering new ways to represent complex scientific concepts. Key moments in this revolution include the development of sophisticated software tools and platforms, allowing researchers to create immersive visualizations for a deeper understanding of scientific phenomena.

The journey continues, marking new developments every day. Of course, there are many who have directly and indirectly contributed to the ever-growing field of scientific visualizations. Captured in this article are only a select few of the many whose legacy lives on in the dazzling spectrum of scientific visualizations we witness today

Puzzle 1: Find and mark the words listed

(Contributed by Arveen Kaur, IIT Kanpur)

B E M T T T S I E O B E G T B A R L C E O O G L L
O A Y S U E T C Y L L C E L E G A N S M I L P T E
C E O M E E M C I U D T O G E N A S E C N M P A I
D R U Y O O I G B F A R I B O S O M E P A Y C Y L
C D H Y T O A U Y R T A O H E M A T O L O G Y A I
T Y L M M T T A D N U M U S M U S C U L U S F I O
T A T P I O A Y M N H S A E O M I Y O O L C G Y C
O H P O R I H M S H L N I E T P S E C Y R P B I U
R P N C C O M U G E I R L E G S H G E N O T Y P E
E H I A B H M B T K S E H S D E C I L U O E D U L
O M M R I S E B S T L T E E Y N H L L D T T O A I
L O A U O A D M R O E M T U S O D A E A B S M G N
Y C M I Y D H I I I Y E E A R M A O M Y I I U Y G
G O Y N R I H Y M S C O R C N R S O S M O U D O L
O X G D T A Y G N I T T O L B O N U M M I Y I O L
L Y D A E O I A U B L R Z A U H U A O I N Y C O M
O L A O M N T C I L E D Y U P X E L P I F D A I N
I A L I O E Y I B C N O G A H D T P L E O I C G O
B C A R R M C G L C C S O A Y N A H Y T R C I E E
L O M O T A H E U I U D U L L S I A Y Y M A E G O
L C L N C T F I I M M S S E O O R F G C A O L M H
E Y D O E O T S Y H O P T L G S S O L O T N C L U
C L B M P D Y H E T S A L R E N T L U O I I U D C
E G U L S E T Y B I U G C Y N O L D L N C M N L E
Y C O O S I T C A B O E O S Y D X Y I P S A R L A

DROSOPHILA
HETEROZYGOUS
RIBOSOME
NEMATODE
GLYCOCALYX
C ELEGANS
MICROTUBULE
MUS MUSCULUS
AMYGDALA
CARBOHYDRATE
AMINO ACID
BIOINFORMATICS
OOCYTE
SPECTROMETRY
PHYLOGENY
CYTOCHEMISTRY
HEMATOLOGY
CELL BIOLOGY
GENOTYPE
IMMUNOBLOTTING
AICBC
ALPHA FOLD
KINASES
SDS PAGE
THYMUS
LIGATION
HORMONES
NUCLEIC ACID

Puzzle 2: Find and mark the words listed

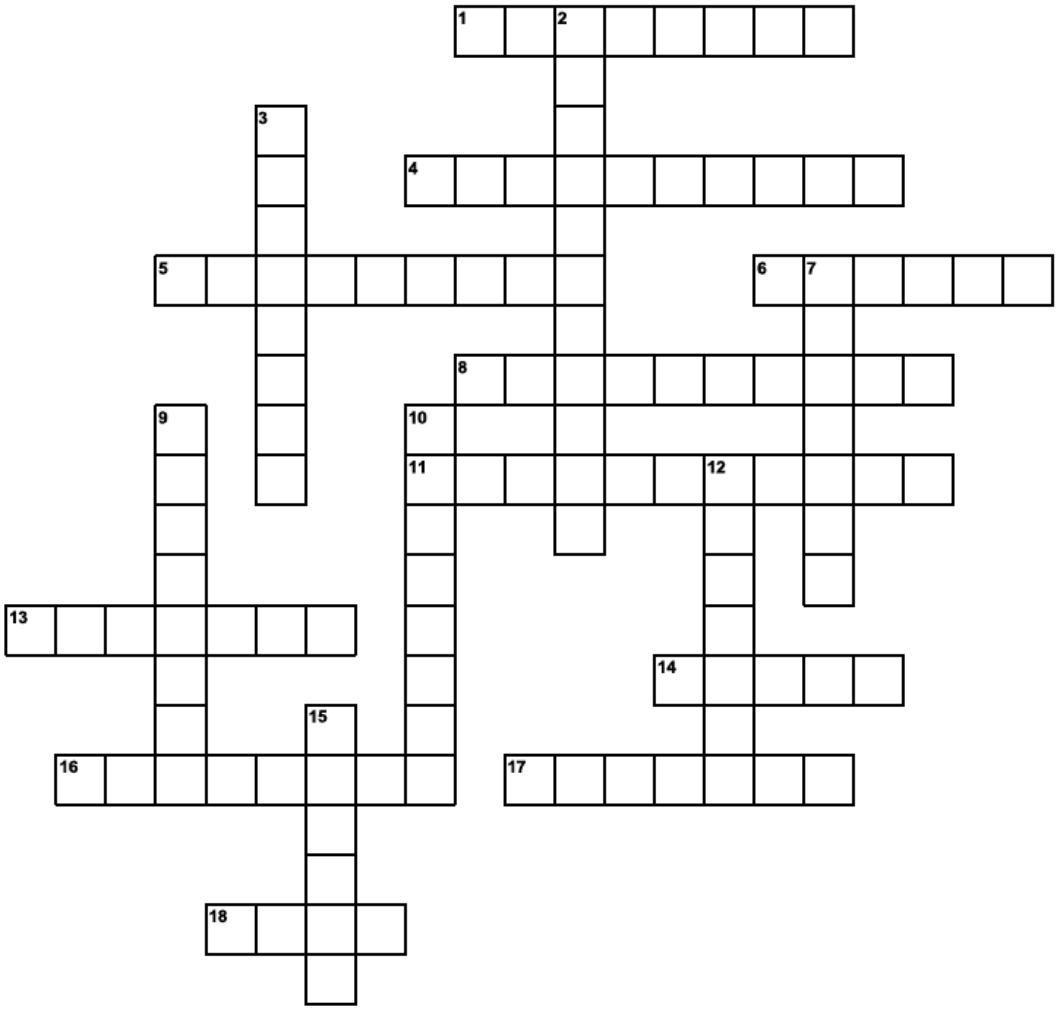
(contributed by Arveen Kaur, IIT Kanpur)

L E N I G T S I S E R O H P O R T C E L E T E C N
M I S R N T R F O S E N O M R O H E O H N L C T E
A L I A I O D L L N E N O I T A N I T U L G G A O
S I L E L O R T E R O N I T E I O P O R H T Y R E
E A Q C A M O U F L A G E B I O M A R K E R S L F
M Y P P E L H S E I R A L L I P A C R T C O C R S
T A P I N O C R I S T A E M I T O P H A G Y I N E
Z E U S N N A N T K O A I A A R I S S O I S R O A
S D I C A Y T T A F C I O U R L C E L U M S T H O
E N U C L E O L I L I N F N L A I E P S T I A R F
E R H S N O I O R S N B E E R T G G U I E P I L U
A G O E P S E S F C A A R I T C U P N L I E R E A
Q N T R M I A I T R I S R I S H G G R A E F E N A
U F C S I O E A E O M A O O N R I G A H N I G T M
A M A R H S P L I U N R P F T O L I P E N C L I B
P P I H A N A H L O S A T T P M G A P F L S Y V A
O E Q T I P D A I E E Y O S I O Y E L I P T Z I E
R I H B L S E I U L N H S U R S L A N D S E O R M
I S I S O T A R E K I P I S P O M U A A U I P U F
N E A H C N Y S Y F M A S I A M O R P P N G A S M
M A D I C U L L E P A N O Z A E P O T O S I I E I
R N R E P R I S T A P L A S R N R R N D H U I S T
C C K N Z E Y O S F O N O O T N I H P O R T S Y D
C S V S C S R H I R D M N Z E O O M K F Y E A S T
A N A C H E A P T Z E B R A F I S H S I E T H D H

LENTIVIRUSES
BIOMARKERS
AQUAPORIN
INFLAMMASOME
YEAST
ELECTROPHORESIS
ERYTHROPOIETIN
FIBRINOGEN
CRISTAE
DOPAMINE
MALIGNANCY
HEMOPHILIA
NUCLEOLI
DYSTROPHIN
CHROMOSOME
MITOPHAGY
FATTY ACIDS
GERIATRICS
HORMONES
ZEBRAFISH
CAPILLARIES
FERROPTOSIS
ZONA PELLUCIDA
ANNEALING
AGGLUTINATION
ISOTOPE
KERATOSIS
CAMOUFLAGE

Word Puzzle 1: Find the solutions

(Contributed by Arveen Kaur, IIT Kanpur)



Across

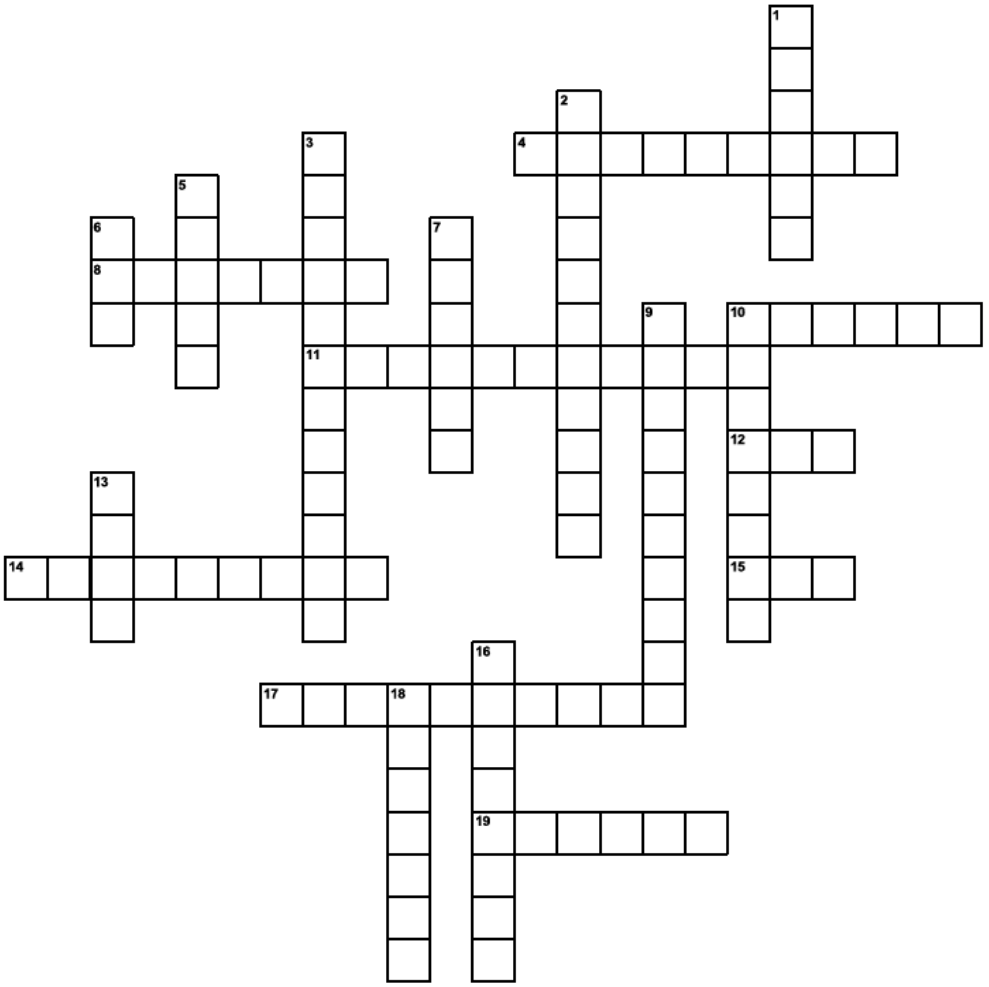
[1] Cancer-causing gene
[4] Flattened membranous sacs in a chloroplast
[5] Programmed cell death
[6] The young of any organism in an early stage of development
[8] Any deviation from the normal diploid number of chromosomes
[11] The division of the cytoplasm following mitosis to generate two daughter cells
[13] Lack of adequate oxygen at the tissue level
[14] A spasmodic, usually painful contraction of a muscle [16] Process extending from the cell body of a neuron [17] The middle part of the small intestine
[18] Biological unit of heredity

Down

[2] Classified as a lipid, the most abundant steroid in animal tissues
[3] An RNA molecule with catalytic activity
[7] a special type of cell division that occurs during maturation of germ cells
[9] A disease-producing microbe
[10] A lysosome-like organelle in the head of a sperm cell containing enzymes
[12] The functional unit of the kidney
[15] A chemical substance that binds to a specific receptor

Word Puzzle 2:

(Contributed by Arveen Kaur, IIT Kanpur)



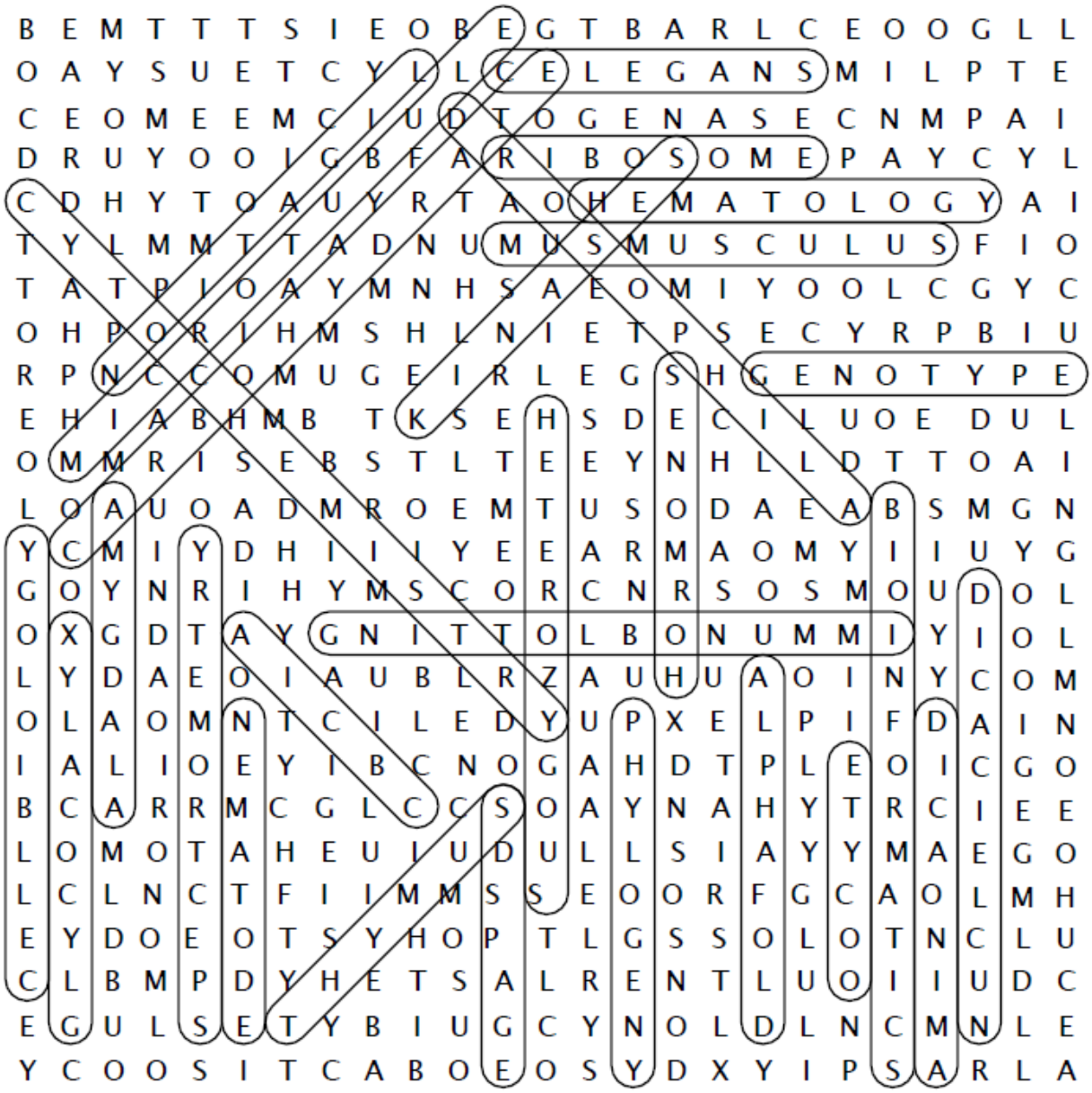
Across

[4] complete set of chromosomes in an individual
[8] aqueous phase of the cytoplasm excluding organelles, membranes, and insoluble cytoskeletal components
[10] protein that catalyzes a particular chemical reaction
[11] interacting effectively with water
[12] region of sequenced DNA that is not interrupted by stop codons
[14] tags proteins for degradation by the proteasome
[15] first nucleic acid
[17] spread of cancer cells from their site of origin
[19] outer proteinaceous coat of a virus

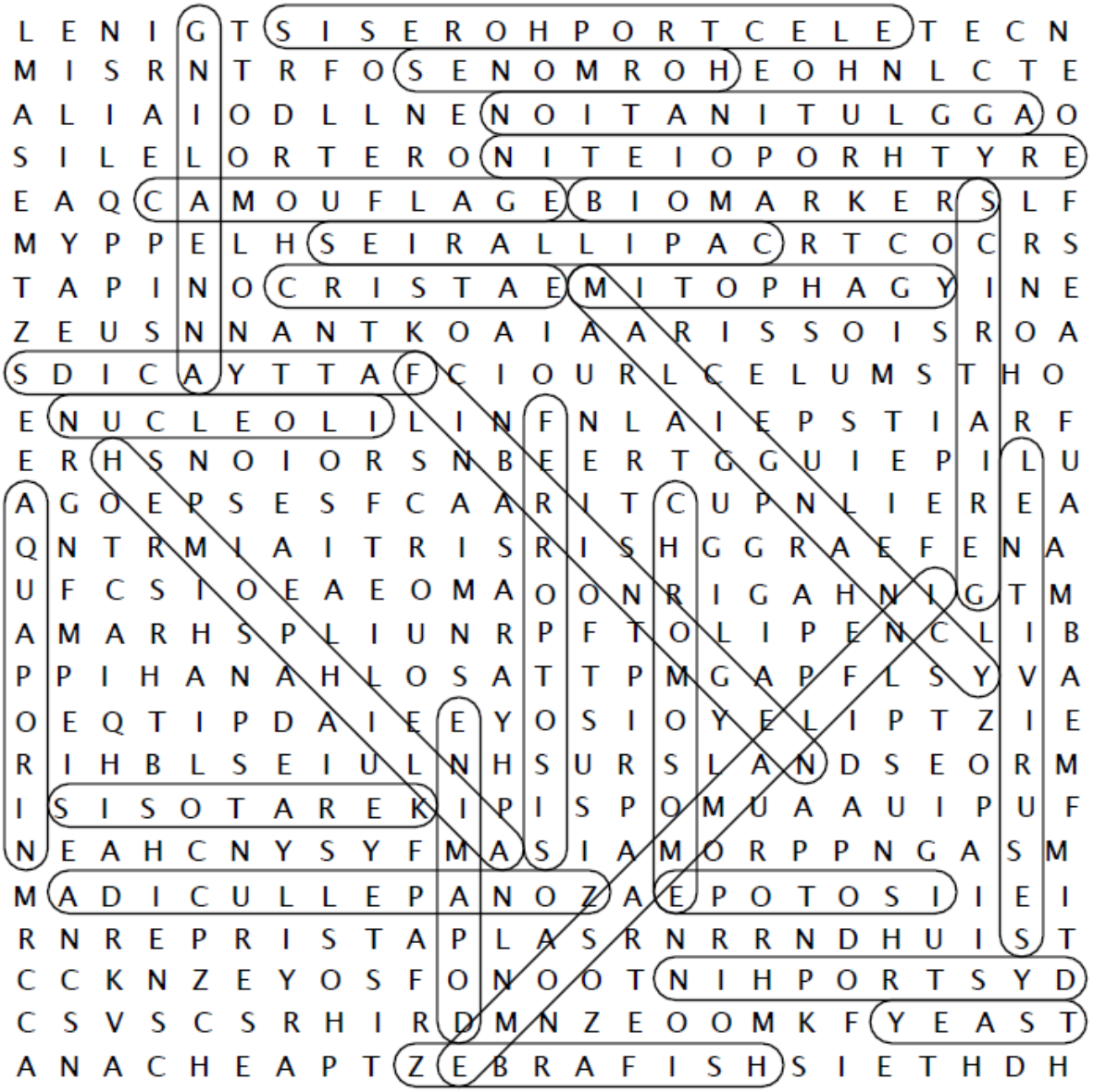
Down

[1] metazoan egg cell
[2] phagocytic leukocytes
[3] cell organelle with it's own DNA
[5] structural protein in eukaryotic cells
[6] technique for amplifying specific DNA segment [7] very long, branched polysaccharide, composed exclusively of glucose units
[9] metabolic pathway in which sugars are degraded [10] outermost of the three primary cell layers in embryo [13] supporting cells of nervous tissue
[16] self-renewing cell
[18] material that elicits an immune response

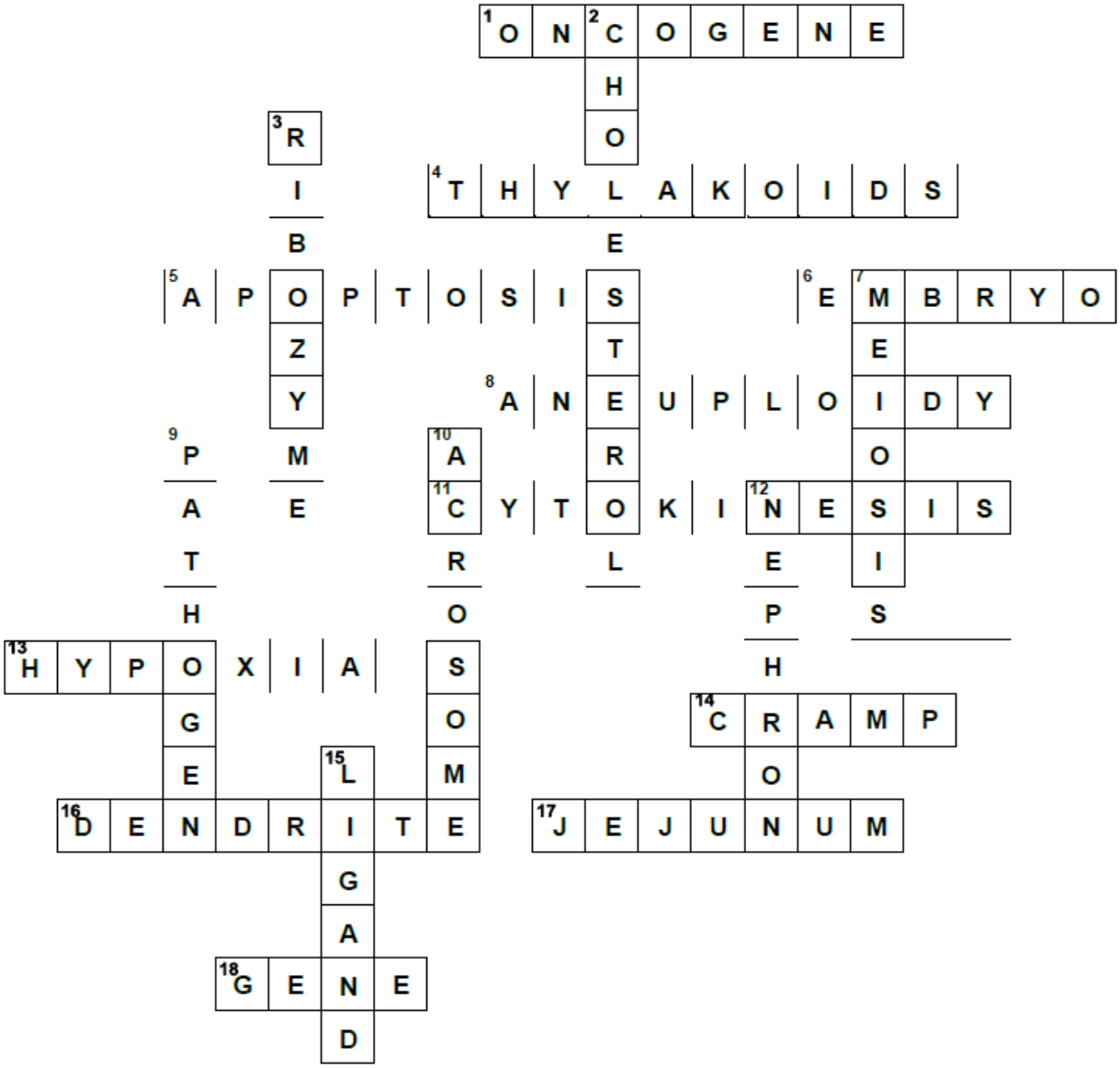
Solutions (Puzzle 1)



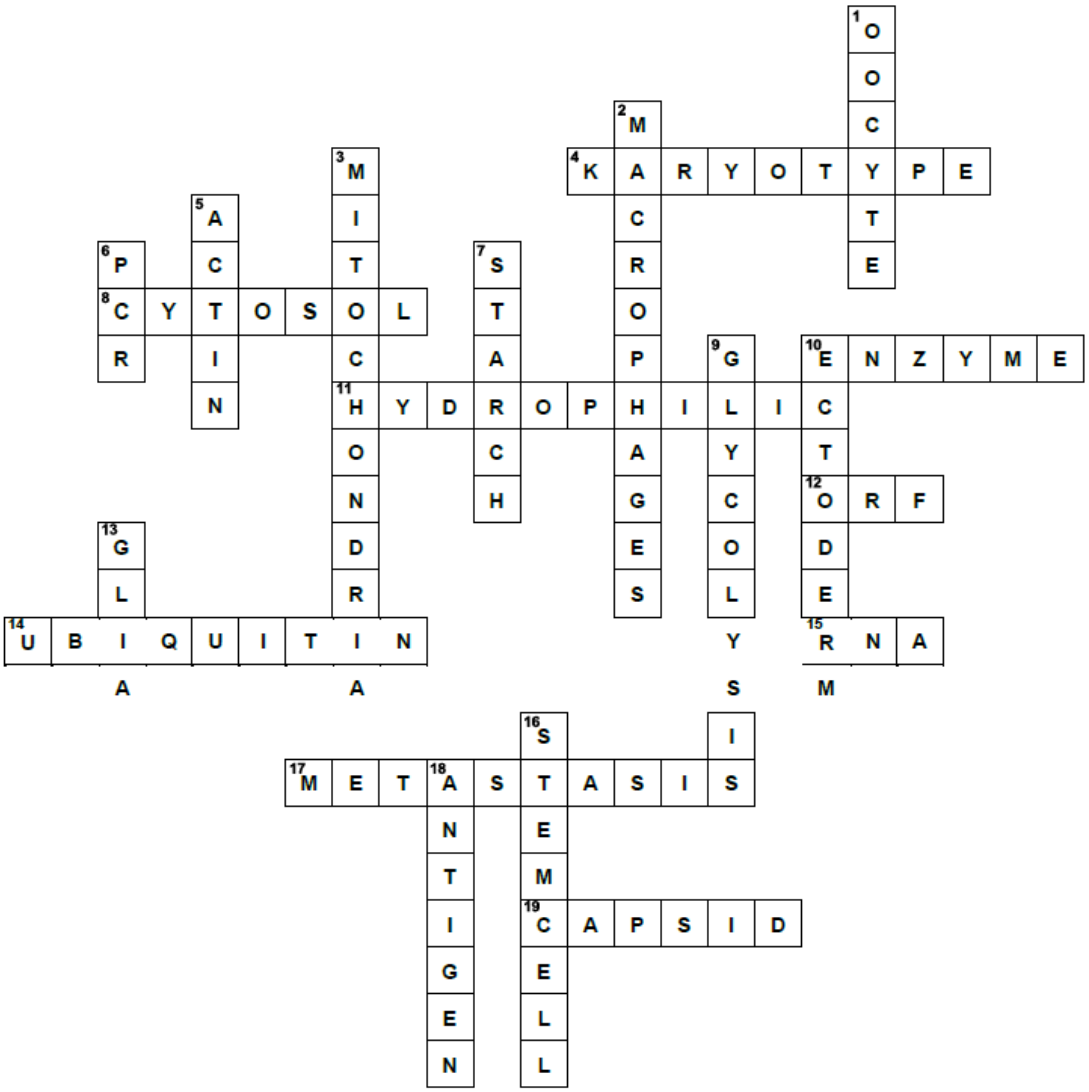
Solutions (Puzzle 2)



Solutions (Word Puzzle 1)



Solutions (Word Puzzle 2)



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Disclosure: Some of the images (cartoons) featured in this newsletter were generated using AI tools.

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