Report of the Workshop on

"**Training Program in Biological Electron Microscopy**"

(August 29-September 4, 2022)

under

Synergistic Training Program Utilizing the Scientific and Technological Infrastructure (STUTI) program

(Department of Science and Technology)

Organized by

Sophisticated Analytical Instrumentation Facility

All India Institute of Medical Sciences, New Delhi

in Collaboration with **Jamia Hamdard, New Delhi**

About the venue

The Sophisticated Analytical Instrumentation Facility (SAIF)-AIIMS, New Delhi has been serving in biological electron microscopy (EM) since its inception in 1983. It is fully equipped with facilities for transmission and scanning electron microscopy of biological specimens, elemental analysis and immunogold localisation of antigens in biological tissues. It is used by university students, scientists and doctors for their front-line research and diagnosis of certain diseases in which EM acts as gold standard. Besides, users from industries and pharmaceutical agencies use this facility to test their materials for industrial applications. This facility conducts hands-on training programs for scientific investigators and summer training program in biological EM.

About the workshop and venue

A hands-on training workshop was organised at SAIF-New Delhi on "Biological Electron Microscopy" from August 29 to September 4, 2022. The aim of this workshop was to provide an outline on the use of equipment and knowledge on various methods of sample preparation for EM. Thirty participants from various parts of India participated in this training workshop.

Objectives of the training curriculum

- To teach the participants with the principles and applications of transmission and scanning electron microscopy (TEM, SEM).

- To equip them with hands on techniques for biological tissue fixation and processing for TEM and SEM.

- To deliver knowledge on section cutting (ultramicrotomy) for viewing under TEM.

- To impart knowledge on how to interpret biological cells from electron micrographs.

The training brochure

Proceedings

29-08-2022 (Day 1)

Inaugural Session

The inaugural ceremony was held on 29th August 2022 at 9:30 AM. It was attended by

Prof. Subrata Sinha, Dean (Academic) AIIMS, New Delhi,

Dr Harish Kumar, Scientist-F (DST R&D Infrastructure),

Prof. A Shariff (Head, Dept of Anatomy),

Prof. TC Nag (Co-ordinator, SAIF, AIIMS, New Delhi),

Prof. Suhel Parvez (Co-ordinator, Jamia Hamdard PMU) and

Dr Suchita Lokhande (Scientist C, DST R&D Infrastructure).

Also, all 30 participants, technical staff of SAIF-New Delhi and other invited faculty and speakers of the Institutes attended the ceremony.

Prof TC Nag welcomed the dignitaries, the invited speakers, colleagues and the participants to the program. In his speech he emphasized on the objectives of the training and the technical sessions planned for the participants to know the techniques in detail via interactions with the experts. **Dr Harish Kumar** urged the students to become scientific ambassadors not only for this Workshop but also for other DST STUTI programs as well. **Prof. Subrata Sinha**, **Prof. A. Sharif** and **Prof. Suhel Parvez** addressed the gathering and wished the participants for a successful career. The vote of thanks was proposed by **Dr Subhash Chandra Yadav, Additional Professor, Organising Secretary, AIIMS.** He thanked DST for providing the opportunity to organise such training programs and also thanked each one of them present in the program.

Inaugural ceremony: The Dean, Dr Harish kumar, the head, Department of Anatomy (upper panel), and Prof. Parvez, and Dr Suchita Lokhande (lower panel) are being welcomed with flower bouquets.

Inaugural ceremony: The Dean, Dr Harish kumar (DST), the head, Department of Anatomy, Prof. Parvez, and Dr Suchita Lokhande (DST) are lighting the lamp.

Towards the end of the inaugural function, the individual participants were asked to introduce themselves and their subject matter under study. The majority of them were in their early PhD career (20), few in MSc course (4), and there were two faculty participants, two post-doctoral fellows and one technical officer. They were from Jamia Hamdard, New Delhi, Aligarh Muslim University, HNB Garhwal University, Srinagar, Jiwaji University,

Gwalior, Central University of Rajasthan, Haryana, Institute of Home Economics (Delhi University, DPSRU (Delhi University), Central University of South Bihar, Gaya, Pondicherry University, IIT-BHU, DRDO-INMAS, New Delhi, Guru Jambheshwar University of Science And Technology, Hisar, Dr Harisingh Gaur Central University, Sagar, MP, National Institute of Immunology, New Delhi, Shaheed Rajguru College (Delhi University), Delhi Technological University, Dwarka, New Delhi, Defence Institute of Physiology and Allied Sciences-DRDO, New Delhi, Jawaharlal Nehru University and University of Delhi-South Campus. They emphasised their requirement for having such kind of training workshop periodically for update of knowledge and fulfilling their career goals, using sophisticated analytical equipment, like electron microscope.

Participants introduced themselves in the inaugural ceremony

Scientific programs

Day 1, August 29, 2022

The inaugural function was followed by the power-point presentations of two lectures: **Dr SC Yadav, Additional Professor, SAIF-New Delhi** and **Dr V Choudhary, Assistant Professor, Department of Biotechnology, AIIMS, New Delhi.** Dr SC Yadav delivered a talk on the topic '**Principles of electron microscopy'** (**lecture 1**), which was followed by **specimen preparation (primary fixation)** until 1 PM. This continued after lunch till 4 PM (on **secondary fixation of tissue samples**).

Dr V Choudhary delivered a talk on the topic '**Specimen preparation for TEM'** (**lecture 2**). Both lectures were intended for the participants to deliver basic knowledge on EM and its working principles and sample preparation method for biological TEM. In the practical sessions, participants were divided into three groups, where individual group performed **various steps in sample processing**, at shown and instructed by our technical staff.

Day 2, August 30, 2022

Lecture 3: **Ultramicrotomy** by **Dr Prabhakar Singh, SAIF-New Delhi**

Lecture 4: **Staining in EM** by **Prof. TC Nag**

Between these lectures, there were hands on practical **on dehydration for TEM samples** (morning) **and knife making and infiltration of TEM samples** (afternoon).

Physical phenomena behind staining (contrasting) and use of heavy metal salts such as uranyl acetate and lead citrate as stains were explained by Dr Nag. Also, negative staining to visualise particulate objects such as fungi, bacteria and other microbes and viruses were explained.

Day 3, August 31, 2022

Lecture 5: **Specimen preparation for SEM** by **Dr SC Yadav**

Lecture 6: **Elemental analysis by EDX** by **Dr Prabhakar Singh**

Between these lectures, there were hands on practical on **embedding for TEM/SEM processing** (morning) and **SEM processing/section cutting** (afternoon). Dr Yadav explained that biological specimens need to be processed for complete dehydration and drying before they are viewed under SEM. Accordingly, the steps of critical point drying and sputter coating were explained by him. Dr Prabhakar Singh explained **the principles of EDS** and its use for determining elemental composition in forensic medicine, geology, material science and biological research.

Three major instruments that were shown to the participants are **TALOS 200S transmission electron microscope**, **EVO 18 scanning electron microscope** and **ultramicrotome (Leica EM UC7).** Before that, the use of ancillary equipment (**glass knife maker**, **critical point dryer**, **sputter coater**, **vacuum oven** and **binocular stereomicroscope** to see small samples) were demonstrated to them. Participants were also shown cutting of thick section (400-500 nm thick) and staining with toluidine blue for light microscope observations.

TEM (TALOS 200S, Thermo Fisher Scientific)

SEM (EVO18, Zeiss)

Ultramicrotome, Leica EM UC7

Pictures of two microscopes and ultramicrotome that were demonstrated

Day 4, September 1, 2022

Lecture 7: Ultrastructure of cell by **Dr Tony G Jacob**, **Additional Professor,** Department of Anatomy, AIIMS, New Delhi. He showed the various features of cells and organelles and their identification under TEM.

After this lecture, there was a hands-on practical on **SEM processing/section cutting** until lunch.

Lecture 8: Application of TEM in regenerative medicine by **Prof. Sujata Mohanty, DBT-Centre of Excellence for Stem Cell Research, AIIMS**, New Delhi.

Dr Mohanty narrated stem cell therapy as a promising form of regenerative medicine and treating different diseases. Mesenchymal stem cells (MSCs) are the current modality of choice in regenerative medicine due to their potential of self-renewability and trans-differentiation. In the field of extracellular vesicle (EVs) biology, MSCs act via releasing EVs for cell-cell communication. Being nano-sized structures, the EVs can be visualized using EM and identified based on their morphology. Hence, there is a need of exploring more on understanding the architectural, cellular complexity and cargo of secretory vesicles.

Lecture 9: **Role of electron microscopy in understanding cilia biology and primary ciliary dyskinesia** by **Prof. Sunil Kateriya, School of Biotechnology, Jawaharlal Nehru University, New Delhi**.

Prof. Kateriya emphasised on the roles played by electron microscopy, electron tomography and Cryo-EM in our understanding of ciliary structure under physiological and pathological states. He also elaborated on new imaging techniques to improve diagnosis of ciliopathies so that it can be applied for the precise diagnosis. Cryo-EM is useful to detect ciliary motility defects in dynein arms, central apparatus and/or radial spoke mutations in human samples. Cryo-EM visualises structural defects of dynein in cilia. Cryo-electron tomography identifies nexin-dynein regulatory complex subunits which are important for ciliary motion. These tools are deployed for detection and management of human ciliopathies.

The resource faculty who delivered lectures. Top panel: Prof. Sujata Mohanty, Prof. S Kateriya, Dr Gopal Jee Jha, Prof. Asit R Mridha (extreme right), lower panel; Dr V Choudhari, Dr Tony G Jacob, Dr Prabhakar Singh, Dr Subhash C Yadav and Prof. TC Nag (extreme right).

Day 5, September 2, 2022

Lecture 10: **Cryo-ultramicrotomy** by **Prof. TC Nag**

Lecture 11: **Nanotechnology** by **Dr SC Yadav**

Lecture 12: **Application of EM in plant biology** by **Dr Gopal Jee Jha**, **National Institute of Plant Genome Research, New Delhi.**

Dr Nag discussed about the various steps in cryo-sectioning, which is useful to see the localisation of antigens in biological tissues.

After this lecture, there was practical sessions on **staining and TEM and SEM viewing** in both morning and afternoon session.

Dr Yadav spoke on Nanotechnology and its application in industries, medicine and pharmaceutical preparations to create products that are useful in everyday use. He explained about the history of nanotechnology from natural to artificial synthesis and their application in diagnostics and nanomedicines.

Dr Gopal Jee Jha highlighted on the role of electron microscopy in understanding the intricacy of plant biology. The use of EM in disease diagnosis, understanding cellular details of plant-microbe interactions, and symbiotic microorganisms was discussed.

Day 6, September 3, 2022

Lecture 13: **Biomedical application of SEM** by **Prof. Asit R Mridha, Department of Pathology, AIIMS, New Delhi.**

Under practical sessions, the participants were demonstrated **grid coating and negative staining** (morning), and **viewing of negative staining preparation** (afternoon).

In his lecture, Prof. Mridha explained how SEM is helpful to study lung injury, immune cell activation, hematopoietic cells, biofilms, cellular development and differentiation, cancer cells and drug development. SEM has wide application in biomedical science to examine the various solid tumors, thus useful for drug development and cancer treatment.

Prof. Mridha delivering his lecture topic to the participants

Materials taught in the lecture classes on the theory and practice electron microscopy were shared with the participants.

A written test was conducted, which included subjective and objective questions to assess the knowledge gained through this training. Grades in the certificates were mentioned according to their performance in the test.

In the afternoon, valedictory session and certificate distribution were arranged. This program was attended by **Prof. A Shariff, Head, Department of Anatomy, AIIMS**, New Delhi. **Dr SC Yadav, the Organising Secretary**, presented with the brief report of activities during the six days of training of the workshop. He welcomed and thanked the invited guests and concluded by wishing the participants good luck for their future endeavours. There was feedback from the participants regarding the organisation of the training, which, according to them, was found to be useful for carrying out their research in electron microscopy. They shared their experience during the workshop. They all appreciated the lectures delivered by the invited speakers, hands-on training sessions by the technicians and the hospitality and the management by the organising team. However, they all commented that a one-week training was not adequate, and suggested for an extended program, at least 10 to 12 days. This suggestion was taken care of.

The valedictory function concluded with a vote of thanks by the organising secretary. He appreciated the efforts of participants for their keen interest in the workshop and acknowledged the contribution of DST, and Jamia Hamdard, who were instrumental in making the training program successful.

Prof. Shariff then distributed the certificates to all participants.


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Sophisticated Analytical Instrumentation Facility
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A Hands-on Training Program on
Biological Electron Microscopy
Under
Synergistic Training Program Utilizing the Scientific and Technological Infrastructure (STUTI),
Department of Science and Technology, Govt. of India
(29 Aug - 4 Sept 2022)

CERTIFICATE OF PARTICIPATION

This is to certify that Ms Nida Naseem from Aligarh Muslim University successfully completed the above training organised by SAIF-New Delhi, AIIMS, with a consolidated grading OUTSTANDING (85%).

Dr. SC Yadav, Organizing Secretary, SAIF-New Delhi
Dr. Prabhakar Singh, Organizing Secretary, SAIF-New Delhi
Prof. Suhel Parvez, DST STUTI-Coordinator, Jamia Hamdard
Prof. TC Nag, Convenor, SAIF-New Delhi
Prof. A Shariff, Patron & Head, Department of Anatomy, AIIMS

Participants receiving their certificates from Prof. A Shariff.

Day 7, September 4, 2022

A Delhi tour was organised for the participants, as many of them were outside from Delhi.

Accommodation and Food

Accommodation facilities were organized and provided by Jamia Hamdard in its student guest houses and hostels. Participants commuted to the venue to and fro via taxi services. Food (lunch) was organised from AIIMS canteen by the organizing committee, breakfast and dinner were organised at the respective hostels in Jamia Hamdard under the support of Prof. Parvez.