PhD Program in Physics and Nanoscience

Student: Chantal Usai
Cycle: XXXVI
Tutor(s): Alberto Diaspro, Paolo Bianchini, Isotta Cainero

ANNUAL REPORT

1. Research Activity

For my PhD project, I am working at the Nanoscopy & NIC@IIT group of the Istituto Italiano di Tecnologia (IIT) using advanced optical microscopy techniques[1] to investigate human chromatin architecture and its modification correlated with genetic diseases. Specifically, I use Expansion Microscopy (ExM)[2], a versatile microscopy method that allows super-resolution imaging using conventional microscopes. This technique exploits the embedding of a biological sample in a swellable polymer network and its subsequent expansion in water to achieve nanoscale resolution.

During the 2nd year of my PhD, I started working on Hutchinson–Gilford Progeria Syndrome[3] (HGPS). HGPS is a genetic disease that causes the early onset of age-related symptoms and leads to premature death. HGPS is associated with a single nucleotide mutation in the LMNA gene, resulting in a truncated form of the protein lamin A termed ΔLA50 or progerin which causes distinctive morphological changes in the nuclear lamina. These modifications in the lamina result in abnormalities in the nuclear and DNA structure and function. During this year, I started using ExM to study modifications in chromatin compaction in a cellular model of HPGS based on human embryonic kidney cells (HEK). I investigate the spatial relationship between chromatin and lamin at the nuclear periphery. Using epigenetic modifications as a marker, I am studying the interactions between heterochromatin and nuclear lamina and the organizational differences in the peripheral chromatin both in the healthy and in the diseased cell line. The preliminary image analysis on the HGPS cell model shows a significant variation in chromatin compaction that seems to disrupt the physical association with lamin, leading to spatial separation between lamin and peripheral heterochromatin. In order to validate these results, I am now performing super-resolution imaging (STED, SIM) on non-expanded cells.

During my 3rd year, I plan to optimize the image analysis technique to evaluate the chromatin-lamin distance on 3D STED images. The next step will be to perform Correlative Light Electron Microscopy (CLEM) on non-expanded cells, exploiting GFP photooxidation to increase image contrast at the interplay between chromatin and nuclear lamin.

2. Courses and Schools

- Biologia Cellulare (Tiziana Bachetti, 6 CFU LM Biologia Molecolare e Sanitaria);
- 6th Nikon School NIC@IIT, Practical Workshop on Advanced Microscopy, Istituto Italiano di Tecnologia, Genova, November 30th to December 3rd 2021;
- International school of Physics "Enrico Fermi", course 210 "Multimodal and Nanoscale Optical Microscopy", Varenna, Italy 10-15 July 2022.

3. Exams

- Biologia Cellulare

4. Conferences

- Biophysical Society 66th Annual Meeting, San Francisco, California, February 19-23, 2022
- Focus on Microscopy, held virtually, April 10-13, 2022
- Società Italiana di Biofisica Pura e Applicata XXVI Congresso Nazionale, San Miniato (PI), September 11-14

5. Posters

- C. Usai, I. Cainero, L. Cuneo, F. Baldini, M. Mariangeli, I. Nepita, P. Bianchini, A. Diaspro, "*Investigating the role of chromatin compaction at the nanoscale in Hutchinson–Gilford Progeria Syndrome using Expansion Microscopy*", Biophysical Society 66th Annual Meeting, San Francisco, California, February 19-23, 2022

6. Oral presentations

- "*Expansion Microscopy: a tool to explore the role of chromatin compaction in Hutchinson-Gilford Progeria Syndrome*", Focus on Microscopy, held virtually, April 11, 2022
- "*Investigating the spatial relationship between Chromatin and Lamin at the nuclear periphery in Hutchinson-Gilford Progeria Syndrome using Expansion Microscopy*", XXVI Congresso Nazionale SIBPA, San Miniato (PI), September 13, 2022

7. Publications

- F. Baldini, I. Cainero, L. Cuneo, M. Oneto, E. Gatta, C. Usai, P. Bianchini, A. Pagano, L. Vergani, A. Diaspro, "Investigating nanoscale chromatin alterations involved in neuroblastoma transformation by optical nanoscopy", Il Nuovo Cimento, 45C (2022) 191;
- F. Baldini, I. Cainero, C. Usai, P. Bianchini, A. Diaspro, "Optical Microscopy to shed light on the chromatin landscapes", Biophysical Reports, in preparation ;

References

1. Diaspro A., and P. Bianchini. 2020. Optical nanoscopy. *La Rivista Del Nuovo Cimento*. 43:385–455.
2. Bianchini, P., L. Pesce, and A. Diaspro. 2020. Expansion microscopy at the nanoscale: The nuclear pore complex as a fiducial landmark. *Methods Cell Biol*. 161:275–295.
3. Vidak S., Folsner R., Molecular insights into the premature aging disease progeria, Histochem Cell Biol 145:401–417, 2016