

Euro-BioImaging
European Research Infrastructure for Imaging Technologies in Biological
and Biomedical Sciences

Interim Operation

**Guidelines for User Access at
Single Molecule and Super-Resolution Microscopy Nodes**

March 2016

The following guidelines are based on the principles that have been developed by the EuBI Preparatory Phase I consortium and guided successfully visiting scientists during the Proof-of-Concept Studies at participating Single Molecule and Super-Resolution Microscopy Facilities of several research institutions, such as e.g. the Bordeaux Imaging Center at Bordeaux University.

1 Guidelines for user access to a Single Molecule and Super-Resolution Microscopy Euro-Biolmaging (EuBI) Node Candidate

1.1 General outline of the project schedule

1. The user interested to conduct a project at a Single Molecule and Super-Resolution Microscopy EuBI Node Candidate can learn about the respective facilities on the EuBI web access portal. **It is strongly recommended that the user contacts informally the Node Candidate of interest** to enquire about the feasibility to conduct the project in this place, to explore the possibilities also for scientific support and to estimate together the approximate user access costs.
2. For entering the formal procedure to apply for access, the user submits a concise application via the online EuBI web access portal, including a project milestone plan to the EuBI Hub. The application can include the specific request for accessing a pre-identified Node Candidate.
3. In a first step, the user application is then scientifically evaluated by an independent board of senior scientists representing a broad scope of scientific disciplines and imaging technologies offered by EuBI.
4. After positive evaluation the user application is then forwarded to the Node Candidate for their technical evaluation and feasibility check. The Node Candidate will contact the applicant, e.g. via Skype, to clarify potentially unclear issues. If the Node Candidate approves the application in this second step, the user is invited to visit this Single Molecule and Super-Resolution Microscopy EuBI Node Candidate and conduct the project work.
5. Due to space and personnel constraints each Node Candidate defines the number of external visitors who can be accepted for user access in a given period of time. The Single Molecule and Super-Resolution Microscopy EuBI Node Candidate makes every effort to host the scientist as soon as possible after application to maintain scientific competitiveness.
6. The logistics of the visit (e.g. accommodation, travel, shipment of reagents) should be arranged with facility staff of the Node Candidate.
7. The access to Single Molecule and Super-Resolution Microscopy instrumentation is conducted in the facility of the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate supported by the scientific environment when feasible.
8. After completion, the user will be asked to provide standardized feedback on various issues of his/her stay.
9. In the post visit period the scientist will inform the EuBI Hub when the results obtained at the EuBI Node Candidate are published in scientific journal(s) with appropriate mentioning of the support provided by the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate in the acknowledgement section of the article. Such outputs will be recorded in EuBI annual reports.

1.2 Application Guidelines

The user will send the brief application for access via a standardized template on the EuBI Web Access Portal. The application includes the following items:

1. A short CV of the applicant.
2. A short scientific project description containing the following information:
 - Project title
 - Scientific background of the project
 - Description of work proposed to be conducted at the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate
 - A milestone plan of the project with clear deliverables and routes for exit if the milestones are not achieved.
 - Importance of the project for the overall research of the scientist
 - Expected results
3. Further information requested
 - Equipment/technology that is envisaged to be used
 - Consent to cover part of the access costs, which will be charged to the application's institution. This amount will be negotiated with the EuBI Node Candidate granting access, after the full technical details of the project are defined and agreed
 - Previous experience of the applicant in light microscopy techniques (in particular the one that he/she intends to use at the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate)
 - Biological hazards associated with the project
 - Agreement to acknowledge the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate in publications resulting from data obtained during the visit
 - In case the applicant is not a principal investigator: approval of the scientist's PI supporting the visit to the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate

1.3 Evaluation Guidelines

The project application will be evaluated independently by scientific experts according to the following criteria:

1. *Scientific excellence*

- Significance/importance of the project in the context of international research and standards in the field
- Relevance/contribution of the project to the scientist's overall scientific work/interests
- Progress beyond state-of-the-art
- Relevance of the project's results for inclusion in future scientific publications
- Scientific quality of the research and study concept
- Benefit for applicant (e.g. training received, results obtained, scientific networking started, being able to apply for his/her own grant)
- Impact of project on field of science, economy and society

2. Feasibility of the project

- Feasibility of the project to be successfully conducted at the EuBI Node Candidate
- Availability of required technologies and expertise at the EuBI Node Candidate
- Availability of possible required supporting laboratory or animal facilities for the project
- Technical ability of the applicant to conduct the planned experiments, or the possibility to acquire the required skills in the time frame of the proposed project
- Reasonable estimation of project duration, and availability of the EuBI Node Candidate during the proposed time frame
- Reasonable estimation of project costs and coverage by the scientist
- Necessity to conduct the research at the requested EuBI Node Candidate (or could the applicant conduct the work at another EuBI Node Candidate that would be closer to his/her home laboratory, or that would be more qualified for the specified application)

If any of the questions above are evaluated as not feasible or insufficient, the application will be rejected.

1.4 Specific guidelines for user project management at Single Molecule and Super-Resolution Microscopy EuBI Node Candidates

1.4.1 Reporting and management during the project

Project meetings will be held according to the milestone plan to discuss whether the respective milestone could be achieved (please see Appendix for exemplary milestone plan). If necessary, the timelines of the project and the milestone plan will be adapted accordingly, or the project will be terminated. Participants (also via teleconference) of these project meetings are: The scientist(s) conducting the project, the project supervisor at the home institution, and the facility staff of the EuBI Node Candidate. A brief meeting report is generated by the meeting participants for documentation of the progress of the project.

1.4.2 Reporting after project completion

After access completion, the user is asked to report on the visit, the impact the results have on his/her future work, the quality of the scientific, technical and logistic support from the Single Molecule and Super-Resolution Microscopy EuBI Node Candidate and the respective research institution. The survey will be provided online by the EuBI Hub.

- Type of instruments used
- Satisfaction concerning given advice and information on usage of most appropriate imaging instrument(s)
- Satisfaction concerning logistic support at the facility (office space, computing, libraries, accommodation)
- Satisfaction concerning technical support to make best use of the imaging instrument(s)
- Satisfaction concerning training (if received) in imaging technology
- Satisfaction concerning scientific support to set up the experiments and interpretation of results
- Rating of scientific impact of imaging infrastructure usage on the project
- Satisfaction concerning administrative support
- In the future: List of publication(s) containing project results based on using Single Molecule and Super-Resolution Microscopy EuBI Node Candidate instrument(s)

APPENDIX 1

Example for a Single Molecule and Super-Resolution Microscopy project milestone plan

Project title:

Determination of a scaffold protein impact on receptor surface trafficking properties in cultured neurons

Aims of the project:

The role of a PDZ domain containing intracellular scaffold protein in regulating the surface trafficking of a neurotransmitter receptor (Adenosin type 2 receptor) will be evaluated using over-expression of wild type and deletion mutant of the proteins as well as siRNA based knock down approaches. Surface trafficking of the receptor in the various conditions will be assayed in primary neuronal cultures using a combination of PALM and U-PAINT single molecule approaches together with FRAP and time-lapse spinning disk microscopy.

Estimated project duration:

In total the project is expected to last for 12 months. The total time spent in the facility of the Euro-Biolmaging Node Candidate is estimated to be around 4 months.

Milestone plan:

MS1 (M4): Reagents

Establishment of N-terminal EOS-tagged A2 receptor. Overexpressor cDNAs and siRNAs to the PDZ scaffold already exist.

This work will be conducted at the home institution.

Risk assessment and exit routes

If the construct fails to express properly, existing GFP and flag-tagged receptors will be used to perform anti-flag and anti-GFP based single molecule tracking. If it is delayed, following milestones will be delayed accordingly.

MS2 (M6): assay for tracking single A2 receptors

Establishment of single molecule microscopy based time-lapse imaging conditions for A2 tracking (on PALM/UPAINT microscope at the Euro-Biolmaging Node Candidate). Important feasibility controls include staining of surface EOS-A2 or GFP/flag-A2 constructs.

Risk assessment and exit routes

If the establishment fails, the project will be terminated. If it is delayed, following milestones will be delayed accordingly.

MS3 (M10): Acquisition of receptor surface trafficking properties in the different conditions by single molecule tracking. Tagged A2 receptor mobility will be assayed in condition of overexpression and knock down of the PDZ scaffold.

Risk assessment and exit routes

Once MS2 is achieved, MS3 is bound to succeed. However, delays may occur due to experimental difficulties. If MS3 is delayed, following milestones will be delayed accordingly.

MS4 (M12): Data analysis

Completion of data analysis will be performed using the specific software available at the Euro-Biolmaging Node Candidate

Risk assessment and exit routes

No risk exists in the data analysis as it is well established.