AMI
Africa Microscopy Initiative
Imaging Centre

Visiting Researcher Program – Call 1

Project Application Guidelines
March 2023

To apply for the AMI Imaging Centre Visiting Researcher Program, please submit a Project Application through the below Google Forms link. Please take note of all information included in the guide below. Applications will be closed on 29 May 2023 at 11:59 p.m. (UTC+2).

https://forms.gle/p77w1QmzipjRHWzh9

Note: A Google account is required to access the Project Application form. Answers and completed fields are automatically saved and the form can be revisited and completed over multiple sessions.

Within the Project Application form, you will be required to provide the following information. Please ensure that all relevant sections are completed.

- Applicant information
- Principle Investigator information *(if required)*
- Personnel information *(if required)*
- Collaboration information *(if required)*
- Project information
- Sample details

In addition to this information, applicants will also be required to upload a separate **Project Proposal**. This document, uploaded as part of the Project Application form, must contain information about the experiment(s) to be carried out at the AMI Imaging Centre. It has a word limit of 3,000 words (excluding references and figure captions) and can contain a maximum of 3 figures/images.

The Project Proposal needs to contain the following information to be fairly reviewed:

1. Project summary (max. 150 words)
Provide a short overview of the experiment being proposed. Use language that non-expert scientists can understand: avoid use of overly-technical phrases and concepts. Simply explain what is being studied, how it will be tested, and what the experiment might reveal. Real world or translational impact and justification for research is not required in the summary.

> **TIP**: the summary should contain succinct information on what biological sample is being studied and what type of biological phenomena will be measured using microscopy.

2. Project background

In this section, provide scientific background to the project and the specific biology being investigated. Please provide the scientific rationale behind the experiment, including previous data and images if possible. Describe the biological relevance of the system being studied and briefly summarise the potential future implications on the field of study.

> **TIP**: do not dedicate too much space to this section; keep it brief and clear. The goal for the project background is to provide context as to why the proposed microscopy experiment should be performed. Identifying the current limitations associated with previous experiments can provide context.

3. Specific aim and hypothesis

Within the broader goal of your research, detail the aim of your proposed experiment. In addition, include the corresponding hypothesis that adequately captures a likely outcome of the experiment.

> **TIP**: make sure that the aim and hypothesis describe a single experiment (or set of related experiments) and not an overarching project. Be very detailed and specific, making sure to link one experiment with the predicted outcome. Use the hypothesis to capture what type of information might answer your biological question.
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> **EXAMPLE**: *the goal of the experiment is to compare the inflammation of disease tissue to control tissue; the hypothesis is that the diseased tissue will exhibit a higher density of CD8+ cells and macrophages per mm².*
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> **NOTE**: if similar but distinct observations are being measured, try listing them separately (Aim 1, Hypothesis 1; Aim 2, Hypothesis 2; *etc.*).

4. Experimental approach and design

Give a brief outline of each step of the experiment. If applicable, clearly describe the experimental conditions being studied and how they will be controlled. Please indicate how the sample will be prepared and labelled for imaging as well as how it needs to be imaged using the microscope. Where possible, include as much relevant information as available, such as the sample characteristics (*i.e.*, live or fixed; fluorescent markers and antibodies to be used, mounting media, growth conditions) and potential imaging requirements (image resolution, sample depth, number of channels, duration, time interval, volume, magnification, *etc.*).

> **TIP**: be sure to detail what type of specimen is being studied (species or strain name, organ type, *etc.*) and provide as much relevant information as possible. Additionally, if it is useful, consider including a schematic outlining the experimental pipeline.
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> **EXAMPLE**: *samples of diseased intestine will be ethically obtained from patient biopsies. The samples will be fixed with formaldehyde and embedded in paraffin before being sectioned into 50μm thick slices. Each slice will be processed as standard and labelled with a primary goat anti-CD4 antibody followed by a secondary rabbit anti-goat IgG antibody conjugated to Cy5. Thereafter, the sample will be mounted onto coverslips using glycerol-based mounting solution containing DAPI. For acquisition, 40 slices each of diseased and control samples will be imaged in their entirety at 20× magnification using DAPI and Cy5 filter sets.*
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> **NOTE**: we recommend all applicants discuss experimental design options with AMI staff before submitting an application. Please schedule a project consultation in advance of the application deadline here.

5. Measurements and quantifiable outcomes

Define what biological feature will be measured and how it might be measured. Identify the aspect of the images produced during the experiment that will be informative of the hypothesis and that might be measured. If you have a quantitative image analysis approach already planned, include it here.

TIP: it is not necessary to know exactly how the feature will be measured, but it is important to identify what feature needs to be measured. Remember that microscopes provide information in the form of intensity data ("how much is present") and spatial data ("where it is present").

EXAMPLE: *the DAPI signal intensity will be used to segment nuclei, which will be used as the marker identifying all cells. Nuclei that are within a defined distance from thresholded Cy5 signal will be designated as CD8+ cells. Thereafter, the proportion of CD8+ cells relative to the total cell number will be calculated per square-millimetre and compared between different tissue locations and disease states.*

TIP: When combined in an image that represents the sample, intensity and spatial information can be measured in different ways to provide us with relevant information about specific aspects of the sample such as shape, direction, cell migration speed, gene expression, *etc.*

6. Justification for use of microscopy

Please provide justification for why you require use of the microscope in the AMI Imaging Centre to conduct your experiment. Explain this from both a technical requirement perspective (such as the need to visualise a live sample over time, or the need to measure structures that cannot be resolved on a normal microscope) as well as from a personal perspective (such as the limited access to confocal microscopes).

TIP: consider motivating why microscopy is required to answer the biological question and why another molecular or chemical technique is not sufficient. Think of which aspect of the experiment can only be done at the AMI Imaging Centre. Provide a figure showing previous result if it can be used to highlight the need to perform your microscopy experiment. Remember, information on "failed" experiments can be useful.

EXAMPLE: *to investigate the hypothesis of disease inflammation, a large number of samples need to be imaged to provide confident results. This is impractical without access to a high magnification, automated slide scanner.* **OR**: *to measure the surface area of interaction between cellular mitochondria and a plasmodium parasite, we require three-dimensional image data with resolution below 250nm. At our institute there is currently no microscope capable of achieving this resolution.*

7. Impact on you and your laboratory

Explain how this experiment will have immediate impact on your laboratory and your research. This does not refer to translatable or real-world implications such as discovering the cure for a disease. Focus on how your experiment will influence and inform future experiments in your lab. Where relevant, highlight the potential for results of this project to be included in a scientific paper, a thesis or dissertation, a grant application, or the establishment of a research collaboration.

TIP: highlight how the information gained by conducting this experiment will advance your existing research. Where possible, provide context of the other work that has been performed within the overarching project (using other techniques and experiments) and how the microscopy data might be used to complete a research manuscript or apply for a funding award. If the experiments will be included in a PhD or MSc thesis, please indicate.

Deadline: **29 May 2023 at 11:59 p.m.** (UTC+2 | CAT | GMT+2)

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