UNSW AUSTRALIA | Medical Sciences Medicine

Department of Anatomy

ANAT 3212

MICROSCOPY IN RESEARCH

Semester 2, 2016

Course Outline

CRICOS Provider Code 00098G

ANAT3212 – MICROSCOPY IN RESEARCH

Course Introduction

Microscopy in Research (ANAT3212) is an undergraduate course for 3rd year students. It is run by the School of Medical Sciences (SoMS) in conjunction with the Mark Wainwright Analytical Centre (MWAC).

Light and Optical Microscopy has undergone rapid transformation over the last two decades and in turn it has become an vital tool in the method toolbox of researchers. This course aims to provide a theoretical and practical foundation to the suite of microscopy techniques so that they can be applied to a particular research project or hypothesis. This course is well suited for future researchers in the medical/biological sciences.

Course Administration

Course Convenors

Course Convenor: Dr. Thomas Fath
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Course Co-Convenor: Dr Renee Whan
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Dr Fath and Dr Whan share responsibility for academic and administrative matters regarding this course. Please approach them for any questions or problems concerning the course. It is best to arrange an appointment in advance by email, copying both into the email.

Ms Justine Maguire-Scarvelli is available to help with administrative problems such as enrolment or scheduling. She can be found in the Student office, G27, Ground floor of the Biosciences Building. Ph 93852426.

Email: SOMSenquiries@unsw.edu.au

If students are having difficulties of a personal nature, they should contact the School of Medical Sciences Grievance Officer, Dr Nick DiGirolamo, n.digirolamo@unsw.edu.au

Units of Credit

ANAT3212 Microscopy for Research is a 6 UoC course. It is offered in the BSc and BMedSc programs, contributing towards a major in Anatomy or a minor in Pathology in the BSc, as well as a specialisation in Anatomy or Pathology in the BMedSc. The pre-requisite for this course is the 2nd year course ANAT2241 Histology: Basic and Systematic.

Course Objectives

This is an advanced course in microscopy, which provides practical, research-oriented experience. There are four key objectives of this course;

1. Promote understanding of the principles and practice of core and advanced microscopy techniques for biomedical research

2. For students to develop skills in using microscopy to examine molecules, cells, tissue and organs to create relationships with structure and function.

3. Provide an excellent basis of core microscopy experiments for utilization in further research studies.

4. Develop oral and written communication skills, which underpin the dissemination of research findings.

Student Learning Outcomes

By the end of this course students are expected to have gained

- A demonstrable knowledge of microscopy, including specimen preparation, acquisition and image analysis.
- An understanding of the relationship between microscopy techniques and the data they produce.
- Be familiar with the application of microscopy for fields of research including but not limited to; cancer sciences, anatomy, neuroscience, physiology, immunology and biology.
- Experience and expertise in critical enquiry by designing and executing microscopy experiments to answer a scientific hypothesis.
- Work in collaborative teams to communicate concepts of microscopy use in state- of- the- art biomedical research in an oral presentation
- Work independently to communicate report and evaluate the microscopy within specialist scientific journals.
- Demonstrate reflective practice and teamwork in your development of professional skills.

Format

Teaching will include lectures, laboratory demonstrations and practical sessions, as well as tutorials. Students will gain experience in examination of microscopic specimens via a range of different methodologies.

In weeks 10 to 12, short Projects will be carried out in research laboratories on the UNSW campus. Students will be assigned to the different projects in week 5 of the course. Students' preferences for individual projects will be taken into consideration. The summaries of the projects that will be offered in 2016 are listed in page 6.

Timetable in brief:

Monday	2-3pm	WW LG02
Tuesday	10am-12pm	WWG6/7
Wednesday	3-4pm	WWLG02
Wednesday	4-6pm	WWG16/17

Attendance

In accordance with University regulations, students must attend at least 80% of all scheduled learning activities (lectures, practicals and tutorials).

There will be an attendance role taken in all classes.

Please read this manual/outline in conjunction with the following pages on the School of Medical Sciences website:

- Advice for Students
- Learning Resources

(or see "STUDENTS" tab at medicalsciences.med.unsw.edu.au)

Assessments- Summary

Assessment activity	Duration	Value	Due Details
Report- (Literature Research)- Individual	1000 words	10%	Week 9
Oral Presentation (Literature Research)- Individual	5 min	10%	Week 9
Examination Terminology & Applications of Microscopy Techniques (Format: short answers)-Individual	1 hr	35%	Week 10
Oral Presentation on Project (Presentation of project experience; should cover a description of experimental design, data analysis and interpretation)- Group	20 min	15%	Week 13
Project Research Project Report (Format: written report including Introduction, Methods, Results, Discussion, References) - Individual	2000-2500 words	25%	Week 13
Reflection Essay (Reflection on Lit Report, project report and oral presentation I and oral presentation II)- Individual	1500 words	5%	Week 13

Projects offered in 2016

PROJECT: Cellular dynamics of sub-cellular compartments in neurons

SUPERVISOR: Dr Thomas Fath

SUMMARY: The motility of cellular regions in nerve cells such as growth cones at the tips of cellular processes is dependent on the dynamics of the underlying actin cytoskeleton. The motile behaviour of a neuronal growth cone is critical to allow for establishing of complex networks between nerve cells. The aim of this project is to visualise changes in growth cone motility in response to manipulation of the actin cytoskeleton.

PROJECT: Imaging chromatin dynamics during the DNA damage response in live cells

SUPERVISOR: Dr Liz Hinde

SUMMARY: The aim of this research project is to image how chromatin organisation facilitates DNA repair factor recruitment to a DNA damage site in live cell nuclei. By application of fluorescence lifetime imaging microscopy to cells expressing fluorescently labelled histones, we will record how chromatin rearranges before and after laser induced DNA damage. This data will provide insight into how DNA repair factors can efficiently arrive at DNA damage sites located anywhere within the genome and at any point in time.

PROJECT: Synaptic Vesicle Trafficking

SUPERVISOR: Dr Vladimir Sytnyk

SUMMARY: Neurotransmission plays a key role in all brain functions. Abnormalities in neurotransmission are observed in a number of brain disorders. Neurotransmitters are stored in small organelles, which are called synaptic vesicles. These organelles fuse with the membrane in a highly regulated manner to release neurotransmitters only when required for neurotransmission. The organelles have to be quickly reformed and refilled with the neurotransmitters to be available for next rounds of neurotransmitter release. The aim of the experiment is to analyse the molecular mechanisms regulating fusion and reformation of the synaptic vesicles using vital stains of the membranes and different fluorescent reporters of the protein components of the synaptic vesicles.

PROJECT: Intravital intracellular microscopy of exocytosis of secretory granules in living mice.

SUPERVISOR: Dr Andrius Masedunskas

SUMMARY: Exocrine glands (such as pancreas, salivary glands) accomplish secretion of enzymes by delivery and fusion of large secretory granules at the apical pole of polarized epithelial cells. After membrane fusion step, actin coat is assembled around the granule that is crucial in facilitating the completion of granule exocytosis. In this project we will investigate the assembly kinetics of actin cytoskeleton machinery on the secretory granule and its role in exocytosis in live mice by using high resolution intravital microscopy. We will utilize transgenic mice and targeted drug delivery as well as image processing and quantitative analysis to achieve the aims of this project.

PROJECT: Using light-controlled molecular tools to pick apart how T cells are activated.

SUPERVISOR: Dr Greg Redpath & Dr Jeremie Rossy.

SUMMARRY: T cells are an important regulatory and effector cell type in the human immune system. T cells are activated by antigens, either foreign or from our own body, binding to the T cell receptor (TCR). Understanding how T cells work and how fundamental cellular mechanism control when and how T cells get activated is absolutely necessary for the design of novel therapeutic strategies.

Our research group is specialised in studying T cell activation using cutting edge microscopy methods. We have previously revealed a new trafficking route that controls TCR targeted recycling to and from the plasma membrane and that is an indispensable step in T cell activation. In this project, you will use a new optogenetic molecular tool that allows to control protein trafficking in precise regions of the cell, simply by using light. This project aim is to disrupt the function of proteins that are involved in TCR trafficking and determine, in real time, how this will alter T cell activation. It involves transfection of cells, imaging with a confocal microscope and data analysis.

PROJECT: Single-molecule imaging of membrane pore formation

SUPERVISOR: Dr Till Boecking

SUMMARY: Cytolytic proteins insert into the plasma membrane of a target cell to form a pore. This mechanism is exploited by cytotoxic T cells and natural killer cells to deliver proteases to cells that have been infected with viruses or bacteria. The dynamics of membrane insertion and pore formation are not understood at the molecular level. The aim of the project is to reconstitute pore formation and visualise the process using TIRF microscopy. The students will acquire movies of pore opening and analyse images using ImageJ.

PROJECT: Using single molecule FRET for the analysis of T-cell activation

SUPERVISOR: Dr Enrico Klotzsch

SUMMARY: smFRET Modern molecular biology, with its emphasis on analysis of entire genomes, has provided a 'parts list' of cellular proteins as well as an enumeration of many of their associations, including receptor- ligand interactions, cell migration and cell – matrix interaction. It appears surprising that molecular forces in this context until recently have been largely overlooked. As there are mechanical forces being involved in decision-making processes such as T-cell activation, the project aims to measure them on the pico-newton level. For that we will use sensors that change their fluorescence behaviour upon force exposure. The participants will learn how to prepare samples in the first step, measure single molecule FRET and later analyse the results with custom written Matlab code.

PROJECT: Live cell imaging of lamellipodia formation in migrating cancer cells

SUPERVISOR: Dr Nicole Bryce

SUMMARY: The aim of this project is to identify the function of newly discovered compounds that target the actin cytoskeleton in lamellipodia. We will use both fixed and live cell imaging of control and drug treated cells to look at lamellipodia structure and formation and how it impacts on cancer cell migration

PROJECT: The role of Yes Associated Protein (YAP) in skin homeostasis

SUPERVISOR: Dr Annemiek Beverdam

SUMMARY: Yes-associated protein activates epidermal stem/progenitor cell proliferation and is thought to be a key molecular player in skin cancer and eczema. This project seeks to examine protein expression of other key molecules that are associated with YAP in tissue sections from transgenic mouse models. Immunolabeling, confocal microscopy and analysis of the protein expressing

PROJECT: Imaging mechano-sensing complexes in melanoma cells

SUPERVISOR: Dr Kate Poole

SUMMARY: The migration of melanoma cells is influenced by the cell's ability to sense their mechanical surroundings. Recent data has identified a novel mechanically-gated ion channel that is required for directional migration of melanoma cells in response to extracellular matrix cues. What is not known is how the activity of this channel can influence migration. This aim of this project is to visualise focal adhesion proteins to determine if channel activity modulates focal adhesion structures in melanoma cells.