

Programa Doutoral do Centro Académico de Medicina de Lisboa

Proposta de criação de Cursos Avançados

Nome do curso: 5th FLxFlow Course - Principles and Applications of Flow Cytometry
18 – 22 November 2024

Coordenação do curso: Mariana Fernandes, Flow Cytometry Unit, IMM

Área científica dominante do curso: Flow Cytometry

Preâmbulo do curso (resumo do curso a usar nos materiais de publicidade, e-mail e portal, outros)

The 5th FLxFlow (Lisbon Flow Cytometry Network) course will introduce the basic principles and applications of flow cytometry to beginners. From fluorophores and panel design, instruments operation, required controls and statistical analysis to data publication and potential applications. The course will consist of theoretical talks and theoretical practical sessions where students will learn how to set up an experiment, run a multi-color experiment, perform compensation & spectral unmixing and analyze data. There will also be practical hands-on sessions in immunophenotyping by conventional and spectral flow cytometry, cell sorting and imaging flow for selected students (selection based on course application letter).

Parte I- Objetivos e plano de estudos (se aplicável, pode/deve ser adaptado a cada curso)

1. Objetivos

This course will introduce the basic principles and applications of flow cytometry to beginners. From fluorophores and panel design, instruments operation, required controls and statistical analysis to data publication and potential applications. The course will consist of theoretical talks and theoretical practical sessions where students will learn how to set up an experiment, run a multi-color experiment, perform compensation /spectral unmixing and analyze data. There will also be practical hands-on sessions in immunophenotyping by conventional and spectral flow cytometry, cell sorting and imaging flow cytometry.

2. Destinatários

Open to CAML students and external candidates.

3. Plano curricular ou plano de curso

Unidade curricular (tema/tópico de acordo com a tipologia de curso)	Área científica	Horas de contacto	N.º de horas total	ECTS
Flow Cytometry (Basic or Advanced Module)	Flow Cytometry	13	13	0,5
Flow Cytometry (Basic + Advanced Module)	Flow Cytometry	26	26	1
Flow Cytometry (Basic or Advanced + Hands-On)	Flow Cytometry	18	18	0,5
Flow Cytometry (Basic+ Advanced + Hands-On)	Flow Cytometry	32	32	2

5th FLxFlow Course – Principles and Applications of Flow Cytometry

Basic Module			Advanced Module				
Nov 18th		Nov 19th	Nov 20th		Nov 21st	Nov 22nd	
9:00 - 9:15	Registration		Registration				
9:15 - 9:30							
9:30 - 9:45	Opening session - FlxFlow Network	Principles of multicolor panel design - Catarina Godinho	Opening session - FlxFlow Network	Imaging FC Fundamentals and Applications - José Rino	Submicron world of Cytometry - Vera Tang		
9:45 - 10:00							
10:00 - 10:15	Principles of Flow Cytometry: Fluidics, Optics and Detection - José Rino						
10:15 - 10:30							
10:30 - 10:45			SPONSOR 4				
10:45 - 11:00				SPONSOR 9	SPONSOR 10		
11:00 - 11:15	Coffee Break	Coffee Break	Coffee Break	Coffee Break	Coffee Break		
11:15 - 11:30			SPONSOR 7	Coffee Break	Coffee Break		
11:30 - 11:45	Fluorochromes - brightness, detection and resolution - Gelo Dela Cruz	Considerations for planning a flow cytometry experiment and sample preparation - Mariana Fernandes		Marine and Plant Flow Cytometry - Diana Ordóñez	Clinical Flow Cytometry - Catarina Martins		
11:45 - 12:00							
12:00 - 12:15							
12:15 - 12:30			Best Practices in publishing flow cytometry data - Vera Tang	Instrument overview	Closing Session		
12:30 - 12:45	SPONSOR 1	Proliferation, Cell Death and Apoptosis - Diana Ordóñez					
12:45 - 13:00							
13:00 - 13:15			Lunch				
13:15 - 13:30							
13:30 - 13:45	Lunch	Lunch					
13:45 - 14:00							
14:00 - 14:15							
14:15 - 14:30							
14:30 - 14:45	Spillover, compensation and spectral unmixing - Cláudia Andrade & Marta Monteiro	SPONSOR 5	Full Spectrum Flow Cytometry - Gelo Dela Cruz				
14:45 - 15:00							
15:00 - 15:15							
15:15 - 15:30			SPONSOR 8	Hands-On	Hands-On		
15:30 - 15:45	SPONSOR 2	SPONSOR 6 - Data Analysis - Basic demo, FlowJo					
15:45 - 16:00			Coffee Break				
16:00 - 16:15	Coffee Break	Coffee Break	High Speed Cell Sorting - André Mozes	Coffee Break	Coffee Break		
16:15 - 16:30							
16:30 - 16:45	SPONSOR 3 - Data Analysis - Basic demo, FCS Express	Workshop Panel Design	Data Analysis - FlowJo Plugins Serena di Cecilia (FlowJo)	Hands-On	Hands-On		
16:45 - 17:00							
17:00 - 17:15							
17:15 - 17:30			Data Analysis - FCS Express full spectrum				
17:30 - 17:45	Cytometer inside-out/How to run a flow cytometry						
17:45 - 18:00		FlxFlow network - Closing Remarks					
Welcome Drink Sponsored by Enzifarma/BD			Networking Event Sponsored by LusoPalex/Cytek				

4. Síntese programática

BASIC MODULE

DAY 1

1. Principles of Flow Cytometry: Fluidics, Optics and Detection - José Rino
 - Basic Principles
 - Flow Cytometer overview
 - Flow cytometry data representation
2. Fluorochromes - brightness, detection and resolution - Gelo Dela Cruz
 - Fluorochromes
 - Signal Resolution
 - Fluorochrome excitation & emission spectra
 - Stability, brightness and issues
3. Spillover, compensation and spectral unmixing - Cláudia Andrade & Marta Monteiro
 - Definitions of compensation and spectral unmixing
 - The Importance of compensation and spectral unmixing
 - Proper compensation/spectral unmixing
 - Rules of compensation (and spectral unmixing)
4. Data Analysis - Basic demo
 - Flow Cytometry data analysis Software
5. How to run a flow cytometry experiment
 - Walk through the cytometer
 - Setting up the Instrument

DAY 2

1. Principles of multicolor panel design - Catarina Godinho
 - Key concepts for the design of multicolor flow cytometry panels
 - Stain Index
 - Spectral Spillover
 - Instrument configurations
 - Rules for panel design
 - Panel Building Software
 - OMIPs

2. Considerations for planning a flow cytometry experiment and sample preparation - Mariana Fernandes
 - Phases of experimental planning
 - Tips & Tricks for sample preparation
 - Antibody Titration
 - Tandem Dyes
 - Viability Dyes
3. Proliferation, Cell Death and Apoptosis - Diana Ordonez
 - Principles of cell cycle and proliferation analysis
 - Measuring cell death by Flow Cytometry
 - Selecting the appropriate method for cell death and apoptosis studies
 - Recommended dyes
4. Data Analysis - Basic demo
 - Flow Cytometry data analysis Software
5. Workshop Panel Design
 - Practical Exercise

ADVANCED MODULE

DAY 3

1. Advanced experimental planning and troubleshooting - Marta Monteiro
 - Compensation misconduct
 - Antibody integrity & titration
 - Polymer dyes
 - Fluidic instability during acquisition
 - Gating strategies and pitfalls
 - Data representation
 - Intracellular staining (fixation, cytoplasm vs nucleus; stimulation for cytokine detection)
2. Best Practices in publishing flow cytometry data – Vera Tang
 - MIFlowCyt
 - Data display
 - Gating strategy
 - Materials & Methods
 - Statistics

3. Full Spectrum Flow Cytometry - Gelo Dela Cruz

- Principles of spectral flow cytometry
- Debunking the Miths
- Tools/tips

4. High Speed Cell Sorting - André Mozes

- Principles of cell sorting
- Cell sorting masks/modes
- Cell sorting controls (purity, viability...)
- Sorting Performance
- Tips on sample preparation for cell sorting

5. Data Analysis - Advanced

- Flow Cytometry data analysis Software

6. Data Analysis - Advanced

- Flow Cytometry data analysis Software

DAY 4 (MORNING)

1. Imaging Flow Cytometry - José Rino

- Fundamentals
- Applications

2. Marine and Plant Flow Cytometry – Diana Ordonez

- Fundamentals
- Applications

3. Instrument Overview

- Visit instruments at IMM Flow Cytometry Unit and all demo instruments
- Applications

DAY 5 (MORNING)

1. Submicron world of Cytometry – Vera Tang

- Refractive Index
- Challenges
- Setup the instrument, calibration
- Sample preparation

- Controls
-
2. Clinical Flow Cytometry - Catarina Martins
 - Fundamentals
 - Applications

PRACTICAL MODULE HANDS-ON SESSIONS (DAY 4 and 5 AFTERNOON)

- Session (I) - Immunophenotyping by conventional Flow Cytometry
- Session (II) - Immunophenotyping by spectral Flow Cytometry
- Session (III) – Cell Sorting
- Session (IV) – Imaging Flow Cytometry

Open sessions for data Analysis with data analysis software.

5. Corpo docente (elenco dos docentes do curso, nome, e-mail, categoria e vínculo (e.g. FMUL, IMM, HSM, outro)
Mariana Fernandes (Instituto de Medicina Molecular João lobo Antunes)
Head of Flow Cytometry Unit (mariana.fernandes@medicina.ulisboa.pt)

André Mozes (Champalimaud Foundation), Catarina Godinho (Instituto de Investigação do Medicamento), Catarina Martins (Nova Medical School), Cláudia Andrade (Nova Medical School), José Rino (Instituto de Medicina Molecular João lobo Antunes), Marta Monteiro (Instituto Gulbenkian de Ciência), Diana Ordonez (EMBL Heidelberg), Gelo Dela Cruz (Nordisk Foundation Center for Stem Cell Medicine), Vera Tang (University of Ottawa)

6. Se inserido num curso de mestrado ou doutoramento a presente proposta deve indicar qual o curso e o respetivo regulamento e ou plano e estudos.

Parte II- Organização e Funcionamento

1. Condições de acesso e seleção de candidatos
Candidatos serão selecionados pela organização (FLxFlow - Lisbon Flow Cytometry Network) após o fecho de cada uma das fases de candidatura. Os candidatos serão notificados dos resultados e os selecionados receberam informação de como devem fazer o pagamento da inscrição.
2. Inscrições e número de vagas

100 vagas. Após o fecho da primeira fase de inscrições, a organização selecionará os candidatos com base na carta de motivação. Caso a totalidade das vagas não seja preenchida, abrirá uma segunda fase.

2.1. Documentação necessária para a formalização de candidatura (e.g. CV, carta de motivação, outros)
Carta de Motivação

2.2. Período de candidaturas
1ªFase: 3 Junho a 5 de julho
2ªFase: 29 de julho a 15 de setembro

Seleção de candidaturas

1ªFase: 8 de julho a 12 de julho, os candidatos aceites devem ser notificados até 15 de julho
2ªFase: 16 de setembro a 20 de setembro, os candidatos selecionados devem ser notificados até 23 de setembro

3. Período de inscrições
1ªFase: 15 de julho a 26 de julho
2ªFase: 23 de setembro a 11 de outubro
4. Duração e horário do curso
26h
(32h for students selected for hands-on session)
9:30 to 18:00

5. Custos de inscrição e propinas
5.1. Taxa de candidatura (se aplicável)

5.2. Valor da propina e condições de pagamento (proposta de valor e data de pagamento)

CAML students: Free

Early Bird

	FLxFlow	Other Academic Institutions	Non-Academic Institutions
Basic or Advanced	75	150	250
Basic + Advanced	125	250	400
Basic or Advanced + Hands-On	200	350	500
Basic + Advanced + Hands-On	250	450	750

Late Registration

	FLxFlow	Other Academic Institutions	Non-Academic Institutions
Basic or Advanced	90	180	300
Basic + Advanced	150	300	480
Basic or Advanced + Hands-On	240	420	600
Basic + Advanced + Hands- On	300	540	900

*FLxFlow institutions:

- Instituto de Medicina Molecular João Lobo Antunes (iMM)
- Instituto Gulbenkian de Ciência (IGC)
- Champalimaud Foundation (FC)
- Nova Medical School (NMS)
- Instituto de Investigação do Medicamento (iMed.Ulisboa)