i-GPCRnet
International Research Network (IRN) on GPCRs

Welcome to the

4th IRN i-GPCRnet Annual Meeting 2024

#iGPCRnet2024

ABOUT

Welcome to the 4th IRN i-GPCRnet annual meeting which will take place at the University of Nottingham (United Kingdom) on July 8th – 10th 2024.

Organizing Committee:

Stephen Hill (University of Nottingham)
Ralf Jockers (Institut Cochin, Paris)
Meritxell Canals (University of Nottingham)
Robert Lane (University of Nottingham)
Jeanette Woolard (University of Nottingham)
Stephen Briddon (University of Nottingham)
Julie Sanchez (University of Nottingham)
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Simon Platt (University of Nottingham)
Laura Kilpatrick (University of Nottingham)
Chloe Peach (University of Nottingham)
Clare Harwood (University of Nottingham)
Eddy Wragg (University of Nottingham)
Xavier Iturrioz (INSERM CEA, Gif-sur-Yvette)

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School of Life Sciences
University of Nottingham

CONFERENCE TIMETABLE

Day 1 – Monday 8th July, 2024

12:00 **Opening and registration** — ESLC Foyer

14:15 **Introduction** — Coates Road Auditorium

Session 1:

Chaired by Natasha Dale & Steve Hill

14:30 **Carsten Hoffmann** (Institute for Molecular Cell Biology, CMB – Center for Molecular Biomedicine, University Clinic Jena, Jena, Germany):
"Novel insights into GPCR regulation by using GRK-KO cells"

15:15 **Maria Tindara Ignazzitto** (Barcelona, Spain):
"Photoswitching azobenzenes to reversibly control β-adrenergic receptors with light"

15:30 **Yair Ben-Chaim** (Ra'anana, Israel):
"Voltage dependence of G protein coupled receptors"

15:45 **Laura Klement** (Jena, Germany):
"Targeting cellular kinases to prevent Influenza A virus infection utilizing encapsulated kinase inhibitors"

16:00 ***Coffee break*** — ESLC Foyer

Chaired by Hannah Lockington & Rob Lane — Coates Road Auditorium

16:30 **Darren Heywood** (Promega, United Kingdom):
"Tools for Studying GPCR Biology"

16:45 **Cyril Goudet** (Montpellier, France):
"Optical control of adenosine A2A receptor using istradefylline photosensitivity"

17:00 **Amod Godbole** (Jena, Germany):
"The molecular basis for GRK/arrestin-mediated regulation of adenosine A1 receptor"

17:15 **Ana Novak** (Orléans, France):
"Identification and characterization of nanobodies targeting serotonin 5-HT7 receptor for application in GPCR Drug Discovery"

17:30 **Chris Tate** (MRC Laboratory of Molecular Biology, Cambridge, United Kingdom):
"GPCR dimerisation: insights from cryo-EM structures of the yeast pheromone receptor Ste2"

18:15 ***Poster session and dinner*** — ESLC Foyer

Day 2 – Tuesday 9th July, 2024

Session 2:

Chaired by Simon Platt & Chloe Peach — Coates Road Auditorium

09:00 **Andrea Kliewer** (Institute for Pharmakology und Toxikology, University Clinic Jena, Jena, Germany):
"Cortico-Subcortical Dysconnectivity Following Opioid Administration Correlates with Analgesia in the Awake Mouse Brain"

09:45 **Frederic Bihel** and **Frederic Simonin** (Strasbourg, France):
"Characterization of a novel orally bioavailable NPFF receptor antagonist for the treatment of opioid-induced hyperalgesia and analgesic tolerance"

10:00 **Lauren Brown** (Nottingham, United Kingdom):
"Does receptor reserve underpin the improved therapeutic windows of opioid partial agonists?"

10:15 **Dominique Massotte** (Strasbourg, France):
"Mu-delta opioid heteromers and neuropathic pain"

10:30 ***Coffee break*** — ESLC Foyer

Chaired by Eline Koers & Raphael Haider — Coates Road Auditorium

11:00 **Isabel Alves** (Bordeaux, France):
"Impact of membrane lipid polyunsaturation on dopamine D2 receptor activation, signaling and internalisation"

11:15 **Marjorie Damian** (Montpellier, France):
"Gq protein peptidomimetics as allosteric modulators of the ghrelin receptor"

11:30 **Farhad Dehkhoda** (Brisbane, Australia):
"*Constitutive activity of the ghrelin receptor provides a dominant second- messenger switch to re-code dopamine D2 receptor signal output"*

11:45 **Katia Befort** (Strasbourg, France):
"Regulation of the endocannabinoid system in binge eating disorder and obesity"

12:00 **Martyna Szpakowska** (Department of Infection and Immunity, Luxembourg Institute of Health, Esch-sur-Alzette, Luxembourg):
"Novel insights into the function and modulation of GPR182/ACKR5"

12:30 ***Lunch break and poster session*** — ESLC Foyer

Session 3:

Chaired by Eddy Wragg & Laura Kilpatrick — Coates Road Auditorium

14:20 ***Group photo***

14:30 **Françoise Bachelerie** (Université Paris-Saclay, INSERM, Inflammation, Microbiome and Immunosurveillance, Orsay, France):
"The CXCL12 chemokine receptors CXCR4/ACKR3 in skin biology and interplay with papillomavirus"

15:15 **Omolade Otun** (Montpellier, France):
"Linking conformational landscape of Atypical Chemokine Receptor 3 (ACKR3) to ligand efficacy using Hydrogen/Deuterium Exchange Mass Spectrometry"

15:30 **Christopher Schafer** (Amsterdam, The Netherlands):
"Specific phosphorylation barcodes attenuate CCL25-mediated G protein coupling by CCR9"

15:45 **Revvity** (sponsor talk)

16:00 ***Coffee break*** — ESLC Foyer

Chaired by Clare Harwood & Steve Briddon — Coates Road Auditorium

16:30 **Rym Ben Boubaker** (Montpellier, France):
"Activation dynamics of mGluR5 and Novel Positive Allosteric Modulator: A Computational Approach and Mechanistic Insights"

16:45 **Luke Pattison** (Cambridge, United Kingdom):
"A role for the proton-sensing GPCR, GPR65 in inflammatory joint pain"

17:00 **Juliette Gourdon** (Nouzilly, France):
"Spatio-temporal organization of LHCGR signalling is essential to regulate gonadal activity"

17:15 **Martine Smit** (Amsterdam Institute for Molecular and Life Sciences, Vrije Universiteit Amsterdam, Amsterdam, The Netherlands):
"Nanobody-based modulation of oncogenic chemokine receptors"

18:00 ***General assembly***

18:30 ***Networking and dinner*** — ESLC Foyer

Day 3 – Wednesday 10th July, 2024

Session 4:

Chaired by Joelle Goulding & Jeanette Woolard — Coates Road Auditorium

09:00 **Maria Marti-Solano** (Department of Pharmacology, University of Cambridge, Cambridge, United Kingdom):
"A multi-dimensional view of receptor – G protein coupling: learning from rare disease variants"

09:45 **Isabella Maiellaro** (Nottingham, United Kingdom):
"Leveraging Drosophila and AI for High-Throughput Studies of Inflammation-Induced Pain"

10:00 **Shuguang Yuan** (Basel, Switzerland):
"Advancing GPCR drug discovery via computational methods in an ultimately efficient way"

10:15 ***Coffee break***

Chaired by Sam Cooper & Ralf Jockers

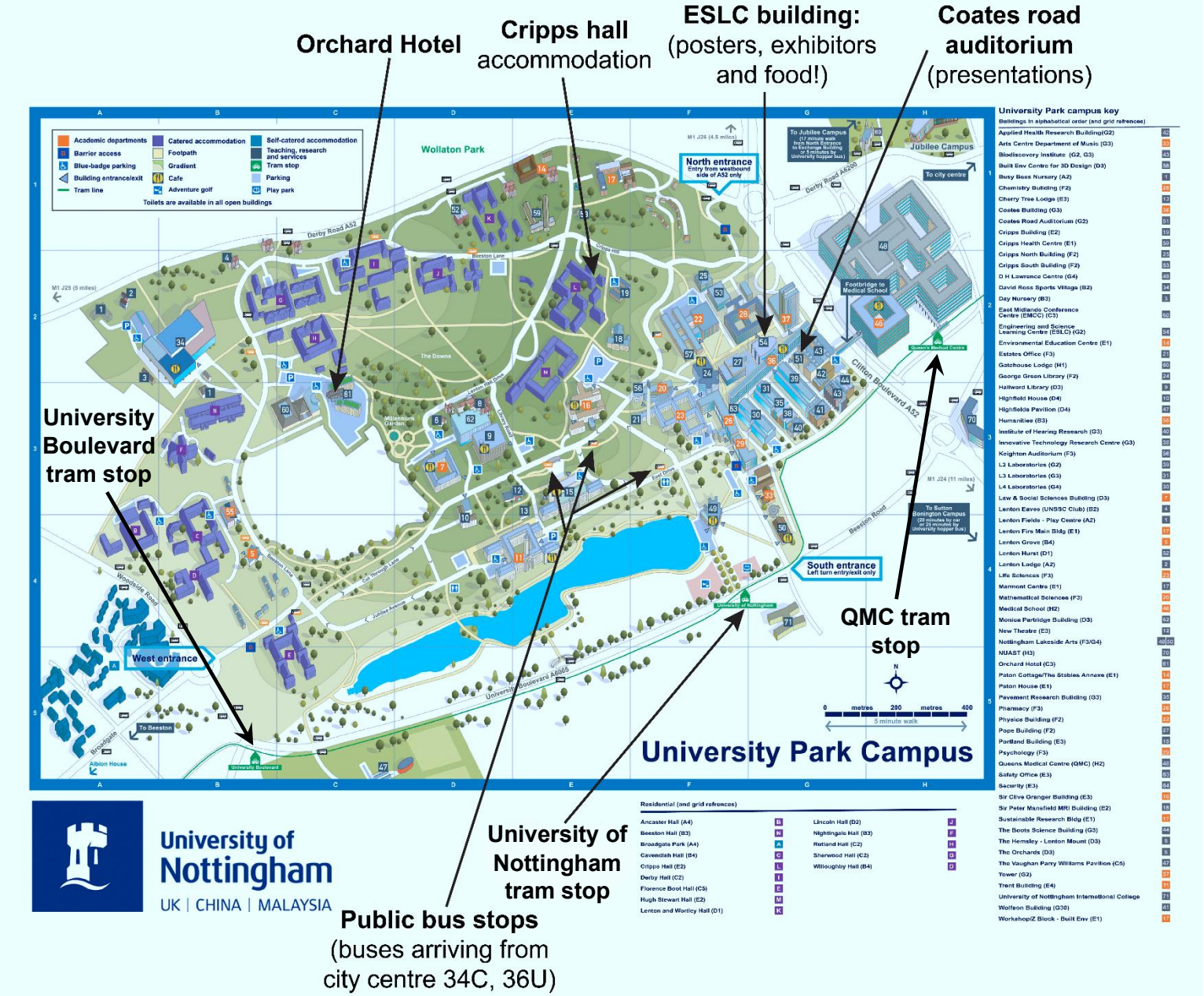
10:45 **Catherine Wark** (BMG LABTECH, United Kingdom):
"Plate based GPCR studies – How to get the best from your microplate reader!"

11:00 **Davide Calebiro** (Institute of Metabolism and Systems Research, College of Medical and Dental Sciences, Centre of Membrane Proteins and Receptors, Birmingham, United Kingdom):
"Untangling the GPCR puzzle with light: from arrestin to intracrine signalling"

11:45 ***Conclusion and Awards***

12:00 ***End of meeting***

CAMPUS MAP


Orchard Hotel
Cripps hall
accommodation
ESLC building:
(posters, exhibitors
and food!)
Coates road
auditorium
(presentations)
University
Boulevard
tram stop
QMC tram
stop
University of
Nottingham
tram stop
Public bus stops
(buses arriving from
city centre 34C, 36U)
University Park Campus
University of
Nottingham
UK | CHINA | MALAYSIA
North entrance
South entrance
West entrance
Wollaton Park
Jubilee Campus
University Park campus key

PRACTICAL INFORMATION

How to get to Nottingham:

By train (international)

Eurostar to London St. Pancras

Eurostar trains arrive in London St. Pancras. Direct trains to Nottingham leave from this station.

By air

East Midlands Airport

Directly connected to Nottingham, *via* skylink bus 1 hour, leaves every 20 minutes. Alternatively, 25-minute drive by car/taxi.

Birmingham Airport

Connected to Nottingham *via* train, connecting in Birmingham New Street. Journey time 2 hours.

London Luton Airport

Directly connected to Nottingham *via* train, 1 hour 40 minutes.

Other London airports

When flying into other London airports such as Stansted, Gatwick, or Heathrow, change trains in London. Trains to Nottingham leave from London St. Pancras. Total journey time 3-4 hours.

Getting around Nottingham:

Both buses and trams can be paid for using contactless debit/credit cards.

Trams

The tram is the most efficient option to travel from Nottingham Train Station to the University.

Toton Lane tram line runs from the centre of Nottingham and Nottingham Train Station to the University of Nottingham tram stop which is the closest to the conference venue and the Cripps hall (University Park campus), the Queen's Medical Centre (QMC) tram stop which the closest to the workshop venue, and the University Boulevard tram stop which is the closest to the Orchard hotel (via the West entrance).

Buses

NCT bus routes 34, 35, and 36 run the centre of Nottingham to Queen's Medical Centre and University Park.

Hotels:

The Orchard, University of Nottingham Campus

Located on the University Park campus (15-minute walk to the conference venue and 15-20 walk to the University Boulevard tram stop)

The Jubilee Hotel & Conferences

Located in the University of Nottingham Jubilee Campus (20-minute walk to the conference venue)

Jury's Inn/Leonardo Hotel, Nottingham

Located next to Nottingham Train Station. Short walk from city centre.

Connected to the University of Nottingham and QMC via Toton Lane tram line.

Ibis Hotel, Nottingham City Centre

Located at Lace Market tram stop, city centre. Tram and bus connections to the University.

Checking into Cripps Hall:

For attendees who requested accommodation, it will be provided in single study rooms with shared bathroom facilities in the Cripps Hall. Check-in will take place at the welcome point North and will be open from 12 pm (noon) on Monday 8th of July 2024. Breakfast is included and will be served in the Cripps dining hall. Check-out time is until 10am.

Places to eat:

All include some vegan and gluten free options.

https://www.baribericotapas.com

https://theangelmicrobrewery.co.uk

https://handandheartnottingham.co.uk

https://www.kayalrestaurant.com

https://www.castlerockbrewery.co.uk/pubs/canalhouse/ (space for large groups and outdoor space)

https://vichotelbeeston.co.uk

Places to network:

https://www.31knotts.com

https://thekilpin.co.uk

https://www.pitcherandpiano.com/bars/nottingham (space for large groups)

https://wollatonhall.org.uk

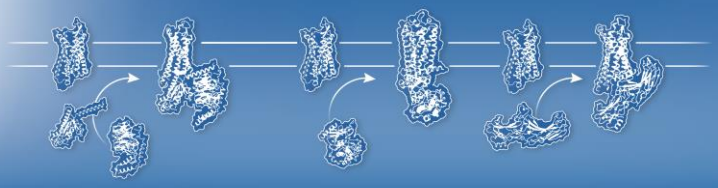
https://www.nottinghamcity.gov.uk/HighfieldsPark

POSTER PRESENTATIONS

Posters will be available to view for the duration of the conference. Poster presentations will be Monday 8th July (18:15) and Tuesday 9th July (12:30-14:20). Presentation prizes will be awarded on Wednesday 10th July at the end of Session 4.

1	Pauline Raynaud	A single domain intrabody specifically targeting the follicle-stimulating hormone receptor (FSHR) affects receptor signalling and trafficking
2	Juliette Gourdon	Spatio-temporal organization of LHCGR signalling is essential to regulate gonadal activity
3	Katia Befort	Regulation of the endocannabinoid system in binge eating disorder and obesity
5	Rong Fu	Development of phosphosite-specific antibodies for an orphan GPCR
6	Shuangyu Lian	Development of novel positive allosteric modulators of GLP-1R – in vitro screening
7	Romane Guisiano	Photoswitchable Ligands for Targeting Melatonin Receptors Activation Using Light
8	Anne-Cécile Van-Baelen	
10	Julie Dam	Functional impact of human GLP1R variants associated with impaired glucose control and increased adiposity: from experimental determination towards AI-based prediction
11	Farhad Dehkhoda	Constitutive activity of the ghrelin receptor provides a dominant second- messenger switch to re-code dopamine D2 receptor signal output
12	Amelia Chorfa	Positive allosteric modulator identification from Pistacia vera extract on purified immobilized Melatonin receptor 2
13	Nedjma Labani	Positive allosteric modulator identification from Pistacia vera extract on purified immobilized Melatonin receptor 2
15	Chloe Bonef	
16	Erika Cecon	Non-canonical ERK signaling activation through Gi/Gq obligatory cooperation in class A GPCR dimers
17	Laura Klement	Targeting cellular kinases to prevent Influenza A virus infection utilizing encapsulated kinase inhibitors
18	Andrew Brown	The state-of-the-art in secondary pharmacology and its impact on the safety of new medicines
19	Ludivine Houzé	β adrenergic receptors activation induces tau pathological transformation linked to Alzheimer's disease
20	Fanny Malhaire-Ferreux	Azoglurax, a photoswitchable allosteric agonist of the metabotropic glutamate receptor mGlu5
21	Cyril Goudet	Optical control of adenosine A2A receptor using istradefylline photosensitivity
22	Maria Ignazzitto	Photoswitching azobenzenes to reversibly control β-adrenergic receptors with light
23	Mona Reichel	The distinct roles of GRK5 and 6 for the Endothelin-1 A receptor (ETAR) in ovarian cancer
24	Isabella Maiellaro	Leveraging Drosophila and AI for High-Throughput Studies of Inflammation-Induced Pain

25	Friederike Wunsch	Dynamics-based assessment of subtle changes in GPCR ligand recognition and function
26	Marjorie Damian	Gq protein peptidomimetics as allosteric modulators of the ghrelin receptor
27	Julie Kniazeff	Constitutive activity of the ghrelin receptor (GHSR): a biased- or a general phenomenon?
28	Xavier Gómez-Santacana	Development of photoswitchable allosteric modulators for mGlu receptors to improve selectivity
29	Amod Godbole	The molecular basis for GRK/arrestin-mediated regulation of adenosine A1 receptor
30	Iona Truong	Exploring the mGluR5 interactome in an amyloid-β context
31	Jack Moton	Arenavirus-based infection as a targeted non-lytic anti-cancer virotherapy
32	Morgan Dennis	The Design, Synthesis and Pharmacological Characterisation of Fluorescent Ligands for the Human Beta-3 Adrenoceptor (β3AR).
33	Lucy Adam	Sensing Extracellular pH Changes: Overlooked Abilities of Diverse G Protein-Coupled Receptors in Pain and Inflammation
34	Sasha-Gaye Richards	CXCR2 is expressed in lymphangioleiomyomatosis (LAM) and may contribute to parenchymal damage.
35	David Reiner-Link	Molecular functions of single-nucleotide variants exclusive for the long isoform of the dopamine D2 receptor
36	Jenny Filor	The intracellular loop 3 of muscarinic acetylcholine receptor M2 and M4 determines the molecular acrobatics of β-arrestin2
38	Meg Sambrook	Modelling the difference in activity between pre- and post-synaptic mu opioid receptors
39	Yu Melanie Tao	Investigations on voltage sensitive behaviour in 5HT2A receptors
40	Sandra Lecat	The molecular mechanism of analgesic tolerance to agonist targeting the delta opioid receptor is dissociated from degradation of the receptor
41	Michaela Ulrich	Pharmacological characterization of the human prostanoid EP1-receptor.
42	Ludovic Berto	Regulation of the ghrelin receptor conformation and signalling by G protein-coupled receptor kinases (GRKs)
43	Farhaan Napier-Khwaja	NanoBRET: Ligand-Binding At The Parathyroid Hormone 1 Receptor, A Prototypical Family B1 GPCR.
44	Thomas Lamme	Specific phosphorylation barcodes attenuate CCL25-mediated G protein coupling by CCR9
45	Aruba Farooq	Regulation of macrophage polarisation in the acidic pancreatic tumour microenvironment.
46	Amaia Nunez-Del-Moral	CCR2 homo- and CCR2/CCR5 hetero-dimers characterization
47	Eline Koers	The use of ThermoBRET for ligand-engagement screening on marginally stable G protein coupled receptors.
48	Julie Delaroche	Split-NanoBiT-BRET-based Biosensors as tools to Query Mitochondrial GPCRs Localization
49	Florence Gbahou	MT2 fluorescent tracers : from technological tools to ligands deciphering structural basis of MT2 selectivity



50	Tamara Miljus	Dynamic characterisation of β-arrestin activation mechanisms by single-molecule FRET
51	Rebecca Murray	Investigating Activation of the Adenosine A2A Receptor Encapsulated in Lipid Particles Generated by SMA-Like Polymers.
52	Abdulrasheed Abdulrahman	Bioluminescence Resonance Energy Transfer-based Biosensors for the Precise Detection of Mitochondrial Protein Localization
53	Foteini Patera	Fluorescent ligands as a tool to investigate adenosine receptors' profile on immune cells: a study of A2A and A2B expression levels on human primary cells
54	Mae Sherrin	Elucidating the downstream signalling partners of intracellular FFAR4 & their role in regulating lipolysis
55	George Farmer	Novel Fluorescent Antagonists for the Nociceptin Opioid Receptor (NOR) to Facilitate the Development of Improved Opioid Painkillers.
56	Dominic Huxley	Elucidating the Signalling Mechanisms of Natriuretic Peptide Receptor (NPR)-C to Facilitate Drug Design & Development for Ischaemic Heart Disease and Heart Failure
57	Andy Chevigné	NanoLuX: a network-wide Nanoluciferase-based platform to monitor activation of classical and atypical chemokine receptors
58	Carmela Perri	The absence of TSHR-GPER heteromers is a potential marker of thyroid cancer
59	Lara Baschieri	Evaluation of the role of DENND1A in the pathogenesis of PCOS
60	Clément Peyron	The nucleotide-free conformation of the endogenous Gi protein illuminates the activity of G protein-coupled receptors

ABSTRACTS FOR SELECTED TALKS

Session 1:

Photoswitching azobenzenes to reversibly control β-adrenergic receptors with light

Maria Tindara Ignazzitto, Xavier Gómez-Santacana, Amadeu Llebaria, Xavier Rovira

MCS, Institute for Advanced Chemistry of Catalonia (IQAC-CSIC), Barcelona, Spain

β-adrenoceptors (β-ARs) are class A G Protein-Coupled Receptors (GPCRs), which regulate several physiological responses and represent the main pharmacological target for the treatment of cardiac and respiratory diseases. However, their ubiquitous expression in the human body and the poor β1-/β2-ARs selectivity of current ligands involve side effects at both respiratory and cardiac level. Moreover, many aspects on β-ARs function and organization are not yet fully understood. This includes mechanisms of activation, receptor dynamics and signaling. To overcome these issues, we focused our research on the development of photopharmacological approaches. These strategies make use of ligands with light-dependent properties that allow the control with light of the drug action site and the time course of drug effect as well as receptor activation/inactivation processes. To design light-regulated molecules targeting β-ARs, we introduced a photochromic moiety (i.e. an azobenzene) within the molecular scaffold of β-ARs agonists. Thus, a series of six photoswitchable molecules have been synthetized. Using suitable light wavelengths, the azobenzene moiety can be reversibly photoisomerized from trans to cis configurations and back. This involves large changes in the ligand geometry, polarity and electron-density, which are expected to alter their binding properties (affinity) or functional properties (activity). After testing the azocompound pharmacological activity on β-ARs, none of the ligands showed significant affinity in dark for both β1- and β2-ARs. However, upon illumination, two compounds showed the highest affinity for both β1- and β2-ARs and potent β1-AR agonism, enabling reversible receptor activation/inactivation by using light. Overall, we expect that these azo-compounds will provide a spatiotemporal control of β-ARs which could be of major interest for both research and therapeutic applications.

Voltage dependence of G protein coupled receptors

Merav Tauber and Yair Ben Chaim
Department of Natural Sciences, The Open University of Israel, Ra'anana, Israel

G-protein coupled receptors (GPCRs) mediate the vast majority of signal transduction processes in the body. Although GPCRs span the cell membrane, they traditionally were not considered to be voltage dependent. In recent years, it was found that the affinity and activity of several GPCRs are regulated by membrane potential. The most studied voltage sensitive GPCRs are the muscarinic receptors. We found that the affinity of these receptors toward acetylcholine is voltage sensitive; the affinity is lower at resting potential than under depolarization for the M1R and higher at resting potential than under depolarization for the M2R. We further showed that this voltage sensitivity is an intrinsic property of the receptor as it was shown that depolarization induces charge movements in these receptors, analogous to “gating currents” that were measured in voltage-gated channels. More recent studies from our laboratory, as well as from others, demonstrated that voltage-dependence may be a general property common to many GPCRs. The molecular mechanism that underlies the voltage dependence of GPCRs is not fully understood and several recent studies suggest that a novel mechanism is responsible for this property of GPCRs.

Targeting cellular kinases to prevent Influenza A virus infection utilizing encapsulated kinase inhibitors

Laura Klement (1), Josefine Schroeder (2), Jana Ismail (3), Amod Godbole (1), Stephanie Schubert (3), Christina Ehrhardt (2), Carsten Hoffmann (1)

1) Institute of Molecular Cell Biology, Center for Molecular Biomedicine, University Hospital Jena, Hans-Knöll-Str. 2, 07745 Jena, Germany
2) Section of Experimental Virology, Institute of Medical Microbiology, University Hospital Jena, Hans-Knöll-Str. 2, 07745 Jena, Germany
3) Laboratory of Organic and Macromolecular Chemistry, Friedrich Schiller University Jena, Humboldtstr. 10, 07743 Jena, Germany

Worldwide circulating and emerging viruses represent a global burden for public health as known for the annual influenza A viruses (IAVs). Treating severe IAV cases is an unresolved challenge as specific antiviral treatment strategies are rare and viruses can rapidly develop resistances. Since viruses depend on the host cell machinery for successful infection, host factors indispensable for the viral life cycle represent promising targets for therapeutic interventions. Recently, G protein-coupled receptor kinases (GRKs) or protein kinases C (PKCs) have emerged as potential players that support viral infection. However, the limited bioavailability and cytotoxic effects of some kinase inhibitors necessitate innovative approaches such as their incorporation into nanoparticles (NPs).

This project aims to adopt existing kinase inhibitors into NPs for tailored treatment of host cell proteins essential for the IAV life cycle.

The impact of various GRK and PKC inhibitors on viral titers and protein expression was analyzed in human lung epithelial cells infected with IAV. Subsequently, the most effective compounds were encapsulated into NPs based on different polymer compositions. The cellular uptake mechanism of the particles was examined via confocal microscopy.

Unlike proposed in literature, the GRK inhibitors paroxetine and CMPD101 did not decrease viral replication. However, we identified two PKC inhibitors, which significantly inhibit IAV multiplication in cell culture without affecting cell viability. The PKC inhibitor 1 was successfully incorporated into NPs, which were effectively internalized by the cells and significantly decreased viral propagation without displaying cytotoxic effects. Currently, we are further investigating the mechanism of action of PKC inhibitor 1 during IAV infection and the effect on virus-mediated regulation of cellular signaling mechanisms.

These findings demonstrate the potential of targeting host kinases via encapsulated inhibitors as a novel treatment strategy to counter infections by respiratory viruses in future epidemics and pandemics.

Optical control of adenosine A2A receptor using istradefylline photosensitivity

Anaëlle Dumazer[1,2], Xavier Gómez-Santacana[2], Fanny Malhaire[1], Chris Jopling[1], Damien Maurel[1], Guillaume Lebon[1], Amadeu Llebaria[2*], Cyril Goudet[1*]

[1] IGF, Université de Montpellier, CNRS, INSERM, 34094 Montpellier, France
[2] MCS, Laboratory of Medicinal Chemistry and Synthesis, Institute of Advanced Chemistry of Catalonia (IQAC-CSIC), Barcelona, Spain

In recent years, there has been growing interest in the potential therapeutic use of inhibitors of adenosine A2A receptors (A2AR) for the treatment of neurodegenerative diseases and cancer. Nevertheless, the widespread expression of A2AR throughout the body emphasizes the importance of temporally and spatially selective ligands. Photopharmacology is an emerging strategy that utilises photosensitive ligands to attain high spatiotemporal precision and regulate the function of biomolecules using light. In this study, we combined photochemistry, cellular and in vivo photopharmacology to investigate the light sensitivity of the FDA-approved antagonist istradefylline and its potential use as an A2AR photopharmacological tool. Our findings reveal that istradefylline exhibits rapid trans to cis isomerization under near UV light, and prolonged exposure results in the formation of photocycloaddition products. We demonstrate that exposure to UV light triggers a time-dependent decrease in the antagonistic activity of istradefylline in A2AR-expressing cells and enables real-time optical control of A2AR signaling in living cells and zebrafish. Together, these data demonstrate that istradefylline is a photoinactivatable A2AR antagonist and that this property can be utilized to perform photopharmacological experiments in living cells and animals.

The molecular basis for GRK/arrestin-mediated regulation of adenosine A1 receptor

Amod Godbole*, Alina P. Handreg*, Carolin Grosse*, Julia Drube*, Carsten Hoffmann*
*(University Hospital Jena, Germany)

Introduction:
The ubiquitously expressed Gi-coupled A1 adenosine receptor and its endogenous agonist adenosine play a crucial role in homeostasis. During inflammation and hypoxia, the increased levels of adenosine and A1 signaling contribute to sepsis and tumor development. Interactions of GPCRs with GRK/arrestins combined with receptor internalization (to decrease surface expression) generally aid in regulating GPCR signaling.

Objective:
Investigation of GRK/arrestin-mediated regulation of the A1 receptor on a molecular basis has however been complicated due to lack of sensitive readouts.

Results:
Using pharmacological inhibitors combined with fluorescence imaging, we show that the A1 receptor, upon adenosine stimulation, internalizes via a caveolae-, dynamin-dependent route. With the help of barr1/2 KO cells combined with microscopy and bystander BRET, we show that this internalization is mediated only via barr2 and that only the A1 internalizes into early endosomes while the barr2 persists on the membrane as distinct clusters. Using GRK KO cells and intermolecular BRET we also show that only the cytosolic GRK2 and membrane-anchored GRK6 are able to initiate this transient A1/barr2 interaction. Substituting serines with alanine, our data indicates that this interaction is dependent on the serines in the ICL3 of A1. Using the recently published PIP2-non-binding mutant of barr2, our data indicate that the barr2/PIP2 interaction supports the low amount of phosphorylable serines and this interaction is indispensable for translocation and formation of barr2 membrane clusters and A1 internalization. Ultimately, with the help of cAMP-based FRET sensors, we also show that GRK/arrestin-mediated regulation influence the rate of cAMP regulation.

Conclusion: We propose, that in addition to GRK2/6-specific phosphorylation, barr2 initially depends on the PIP2 around the activated A1 to translocate to and interact with the phosphorylated receptor and initiate receptor internalization. Using FRET-based ERK1/2 sensors, we are now investigating the potential impact of this transient receptor/arrestin interaction on ERK activation.

Identification and characterization of nanobodies targeting serotonin 5-HT7 receptor for application in GPCR Drug Discovery

Ana Novak[1], Chayma El Khamlichi[1], Nadège Hervouet-Coste[1], Enora Pigeon[1], Cyril Guimpied[1], Vincent Aucagne[1], Gilles Bruneau[2], Eric Reiter[2] and Séverine Morisset-Lopez[1]

[1] Centre de Biophysique Moléculaire, group « Neurobiology of receptors and therapeutic innovations », CNRS UPR 4301, rue Charles Sadron, 45071 ORLEANS Cedex 01
[2] INRAE, UMR 7247, Tours, France

In our laboratory we are focused on the last discovered member of the serotonin receptor family, the 5-HT7 receptor. This receptor belongs to the G protein-coupled receptor superfamily (GPCRs), one of the most successful therapeutic target families. In the last decade this receptor has become a promising target for the treatment of neuro-psychiatric disorders, sleep and circadian rhythm disorders and pathological pain (El Khamlichi et al., 2022). Owned to some limitations of synthetic molecules (lack of bioavailability, selectivity) development of new pharmacological tools based on antibodies and nanobodies (antibody fragments) have emerged (Heukers, R. et al,2019.). Nanobodies are small proteins (~15 kDa) derived from the variable domain (VHH) of naturally occurring heavy-chain antibodies with high affinity and selectivity (Muyldermans, 2013). According to their convex shape they are particularly interesting for targeting buried epitopes such as those found in ligand-binding pockets of receptors (De Genst et al, 2006; Muyldermans et al, 2009). To identify nanobodies to human 5-HT7R, we used different antigen format for in vivo immunization of Llama. Here we report the successful selection of a few potential candidates, after performing phage display screening, that showed specific binding to targeted receptor 5-HT7 (ELISA, HTRF and flow cytometry). Further we did characterization from molecular (In-vitro) to behavioral (In-vivo) level of selected candidates /nanobodies targeting 5HT7 in different functional assays. We conducted epitope research by integrating computational predictions with molecular biology techniques, enabling precise identification and characterization of epitopes. With the most promising VHH candidate we demonstrated its therapeutic potential in preclinical models in mice. The new developed tools (nanobodies) may provide insights into the biology of 5HT7 receptor and thus enable us to better understand the role of the receptor in various physiopathological conditions.

Session 2:

Characterization of a novel orally bioavailable NPFF receptor antagonist for the treatment of opioid-induced hyperalgesia and analgesic tolerance

Frédéric Bihel[1] and Frédéric Simonin[2]

Affiliations
1. Laboratoire d'Innovation Thérapeuthique, UMR7200 University of Strasbourg-CNRS, Illkirch, FRANCE
2. Biotechnologie et Signalisation Cellulaire, UMR7242 CNRS-University of Strasbourg, Illkirch, FRANCE

Chronic pain is a major public health problem in humans for which current treatments often prove to be ineffective and display adverse side effects. In the United States, the massive use of opioids for the treatment of chronic pain led to more than 500 000 deaths by overdose over the last fifteen years highlighting the urgent need to develop new therapeutic strategies for pain relief. Neuropeptide FF receptors (NPFF1R and NPFF2R) and their endogenous ligand neuropeptide FF have been shown previously to display anti-opioid properties and to play a critical role in the adverse effects associated with chronic administrations of opiates. We describe here the design and optimization of a drug-like orally active NPFFR ligand called RF1359 that display nanomolar affinity and potent antagonist activity for NPFF1R subtype. It displays promising properties for future drug development in terms of metabolic stability, pharmacokinetic profile, solubility, selectivity and safety profile. In naïve mice, the pharmacological blockade of NPFF receptors with RF1359 efficiently prevents the development of morphine-induced hyperalgesia while preserving its analgesic effect over chronic administration. Moreover, when evaluated in two different clinically relevant models of pain, RF1359 significantly improved morphine-induced analgesia allowing to reduce morphine dose and thus potential adverse side effects associated with chronic morphine. Altogether, our data indicate that RF1359 represents a promising lead compound for the development of therapeutic agents that could improve opiates efficacy upon chronic administration and limit adverse side effects.

Does receptor reserve underpin the improved therapeutic windows of opioid partial agonists?

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In the last two decades, functional selectivity at the MOR towards G-protein signalling and away from beta-arrestin2 recruitment has been suggested to explain the improved therapeutic window of novel opioid agonists for analgesia over respiratory depression. However, a recent analysis proposes that reduced intrinsic efficacy rather than G-protein bias may explain this. If this hypothesis is correct, it would be expected that differences in receptor reserve in areas of the CNS attributable to distinct opioid behaviours underlie the different effects of partial agonists, with the tissues mediating antinociception exhibiting the greatest MOR reserve compared to those involved in the adverse effects. This proposal requires full validation. This study was designed to investigate the impact of receptor reserve on opioid-related behaviours of antinociception, respiration and constipation using irreversible antagonism of the MOR. Behaviours were measured in awake freely moving male C57BL/6J mice using the hot-plate, whole-body plethysmography and faecal boli accumulation assays respectively. Animals were pre-treated with methacinnamox (MCAM) (0.1-10 mg.kg-1, i.p.), then injected with a single dose of a MOR agonist (morphine (15 mg.kg-1, i.p.), 7-OH mitragynine (3 mg.kg-1, s.c.) or tianeptine (30 mg.kg-1, i.p.)). All MOR agonists induced significant antinociception, respiratory depression and constipation. Treatment with MCAM alone did not affect antinociception or respiration but induced significantly increased baseline faecal boli production at the highest dose. MCAM also blocked ligand-induced antinociception, respiratory depression and constipation in a dose-dependent manner. The dose of MCAM which significantly blocked each opioid-induced behaviour was both behaviour- and ligand- specific. These data suggest that differences in receptor reserve across areas of the CNS responsible for distinct opioid-related behaviours alone cannot explain the enhanced therapeutic window of low intrinsic efficacy agonists and highlight potential limitations of using MCAM to block opioid-induced effects in different tissues.

Mu-delta opioid heteromers and neuropathic pain

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Neuropathic pain represents an unmet medical challenge which requires to develop novel therapeutic strategies. Mu and delta opioid receptors are widely expressed in pain related areas and can functionally interact to form heteromers. Activation of mu-delta heteromers reduces thermal and mechanical nociception in Neuropathic pain represents an unmet medical challenge which requires to develop novel therapeutic strategies. Mu and delta opioid receptors are widely expressed in pain related areas and can functionally interact to form heteromers. Activation of mu-delta heteromers reduces thermal and mechanical nociception in naïve animals but their antinociceptive efficacy remains poorly explored in neuropathic conditions. We therefore examine changes in mu and delta opioid receptor distribution in two models of neuropathic pain. We determined nociceptive thresholds in naïve and neuropathic male and female mice following acute injection of CYM51010 or MP135, two ligands targeting mu-delta heteromers and compared them to those following mu agonist morphine administration. We also investigated the ability of the compounds to activate G protein and beta arrestin dependent signaling cascades. In parallel, we assessed changes in mu-delta neuronal co-expression in neuropathic conditions.

Impact of membrane lipid polyunsaturation on dopamine D2 receptor activation, signaling and internalisation

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The heterogenous and dynamic constitution of the membrane fine-tunes signal transduction. In particular, the polyunsaturated fatty acid (PUFA) tails of phospholipids influence the biophysical properties of the membrane, production of second messengers, or membrane partitioning. Few evidence mostly originating from studies of rhodopsin suggest that PUFAs directly modulate the conformational dynamic of transmembrane proteins. However, whether such properties translate to other G protein-coupled receptors remains unclear. The brain is highly enriched in PUFAs and their deficiency has been associated with several neuropsychiatric disorders. We focused on the dopamine D2 receptor (D2R), a class A GPCR, that is consistently impacted in these disorders and represents the main target of most antipsychotics. Membrane enrichment in n-3, but not n-6, PUFAs potentiates ligand binding. Molecular dynamics simulations show that the D2R preferentially interacts with n-3 over n-6 PUFAs. PUFA cell enrichment strongly impairs agonist-induced endocytosis of D2R in HEK293 cells, without affecting clathrin-mediated endocytosis or β2 adrenergic receptor endocytosis. Using live cell TIRF imaging, we show that D2R clustering is not affected, but that recruitment of β-arrestin2, that occurs prior to receptor internalization, is strongly impaired and endocytic vesicle formation is slowed down. Furthermore, in vivo n-3 PUFA deficiency blunts the effects of D2R ligands. These results suggest that n-3 PUFAs act as allosteric modulators of the D2R and provide a putative mechanism for their potentiating effect on antipsychotic efficacy.

Jobin et al, Mol. Psy. (2023) 28:2171.
Baccouch et al, BioRxiv; doi: https://doi.org/10.1101/2023.12.14.571632

Gq protein peptidomimetics as allosteric modulators of the ghrelin receptor

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Our understanding of G protein-mediated GPCR signaling, a major event in intercellular communication, is far from complete, as it is a dynamic process involving a variety of intermediates in addition to those described in three-dimensional structures. In this context, we used the ghrelin receptor and original G protein peptidomimetics to provide a description of the GPCR:G protein interplay. We show that coupling to the G protein mimetic not only affects the conformational features of the cytoplasmic regions of the receptor where the G protein binds, but also has an allosteric effect on the extracellular ligand binding pocket. In addition, our data clearly indicate that the interaction of the receptor with the Gαq C-terminal helix, which is the major belt drive between the receptor and the nucleotide-binding pocket of the Gα subunit, is different whether the pre-assembled complex with the GDP-loaded G protein or the active assembly with nucleotide-free Gq are considered. This illuminates the GPCR:G protein coupling process with different states on the way to G protein activation.

Constitutive activity of the ghrelin receptor provides a dominant second- messenger switch to re-code dopamine D2 receptor signal output

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The dopamine D2 (DRD2) and ghrelin (GHSR) receptors are co-expressed in preganglionic neurons of the spinal defecation centre. Pharmacological stimulation of either receptor results in colonic pressure waves, however ghrelin is absent from the central nervous system. In most neurons, activation of DRD2 results in neuronal inhibition via Gαi/o and subsequent G protein-gated inwardly rectifying potassium channel activation. Contrastingly, DRD2 activation in defecation centre neurons results in depolarisation. This cellular output switch has also been reported in hypothalamic neurons co-expressing GHSR and DRD2 and was previously attributed to heterodimerisation of these receptors. In spinal defecation neurons, those exhibiting an excitatory response to dopamine also exhibited an excitatory response to GHSR agonists. Inverse agonists of GHSR blocked DRD2 dependent excitatory response, which was also dependent on intracellular calcium stores. This switch in DRD2 coupling to calcium mobilisation was also observed in recombinant cells, however this does not appear to be the result of a switch in the G protein preference of DRD2. Moreover, we were unable to detect allostery between these receptors using ligand binding, nor did we observe them to be close enough to indicate dimerisation using super-resolution microscopy. Instead, we observed a GHSR dependent priming of phospholipase C beta (PLC-b), that was dependent on GHSR's constitutive activity. In cells co-expressing a GHSR polymorph that lacks constitutive activity, coupling of DRD2 to calcium was restored by priming with a low concentration of ghrelin. The requirement for PLC-b was also seen in spinal defecation neurons, where PLC-b inhibition reversed the response of these cells to dopamine from excitatory to inhibitory. Together, this data indicates that dopamine mediated excitation is dependent on GHSR constitutive activity via a dominant second-messenger switch. This work has broad implications for determining metabotropic neurotransmitter responses via modulation through other G protein-coupled receptors.

Regulation of the endocannabinoid system in binge eating disorder and obesity

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Binge eating disorder (BED) and obesity are complex conditions characterized by maladaptive eating behaviors, often directed towards highly palatable foods. The mesocorticolimbic dopamine system, known as the reward system, plays a central role in the hedonic food intake and is known to be dysregulated in both eating disorders and obesity. The endocannabinoid system (ECS), which is expressed in the reward system, exerts significant influence over feeding behavior, but the extent of its involvement in binge eating and obesity remains incompletely understood. Recent research has highlighted its pivotal role in regulating feeding behaviors, particularly in the context of addictive-like eating patterns. We propose that the ECS undergoes similar regulatory changes in reward-related brain regions in both BED and obesity.

To test this hypothesis, we developed models of BED and of obesity in adult male rats. The rats were subjected to either a continuous (obesity) or intermittent (BED) diet over a period of 6 weeks, with free access to high-fat food and a 10% sucrose solution. Binge-eating behavior was evaluated by measuring significantly higher intake of sucrose/fat during the first hour of access in the intermittent, compared to the continuous, access group. Following completion of the protocol, we assessed the effects of diet exposure on ECS gene expression using qPCR, and on endocannabinoid levels using mass spectrometry, in several brain regions. Our findings reveal distinct modulation of the ECS based on the pattern of access to palatable foods, suggesting that BED and obesity exert varying effects on the ECS within the reward system. Intriguingly, correlation analysis indicates that the macronutrient composition of palatable foods may have a specific influence on ECS gene expression. Our findings broaden the comprehension of ECS molecular adaptation in obesity and BED models. This insight could facilitate the discovery of novel ECS targets for therapeutic intervention.

Linking conformational landscape of Atypical Chemokine Receptor 3 (ACKR3) to ligand efficacy using Hydrogen/Deuterium Exchange Mass Spectrometry

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Chemokine receptors are class A GPCRs known for their ability to mediate migration of immune cells in response to a chemical trigger, the phenomenon known as chemotaxis. Whilst the majority of chemokine receptors signal in a G-protein mediated manner, a small subset of atypical chemokine receptors exhibit an apparent inability to activate G-proteins. Atypical Chemokine Receptor 3 (ACKR3) is one such receptor and has been shown to signal predominantly through β-arrestin mediated pathways in a constitutive manner. To further probe this functional divergence, small ligands have been developed that act as agonists or inverse agonists for β-arrestin recruitment at ACKR3.

Hydrogen-Deuterium exchange-mass spectrometry (HDX-MS) monitors the rate of deuterium incorporation to follow the dynamics within a protein. This can be used to compare the conformation of a protein in different states to determine ligand-induced effects. We have used HDX-MS to identify structural changes that occur at ACKR3 upon ligand binding, aiming to correlate specific conformational fingerprints to the signalling effect of small ligands. This allowed us to obtain distinct HDX profiles for agonists and inverse agonists at ACKR3. HDX experiments performed in the presence of β-arrestin 1 allowed us to propose a binding mechanism for arrestin at ACKR3. In parallel, we performed MD simulations and mutagenesis studies to support changes observed by HDX highlighting differences in binding modes between agonists and inverse agonists. Combining these approaches, we describe an activation mechanism for ACKR3 and propose structural changes that may correspond to ACKR3's bias towards the β-arrestin pathway.

Session 3:

Specific phosphorylation barcodes attenuate CCL25-mediated G protein coupling by CCR9

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CCR9 mediates immune cell migration between the thymus and the gut through activation by its chemokine ligand CCL25. As a G protein-coupled receptor (GPCR), CCR9 activates Gi and Gq pathways to drive cell motility. Dysregulation of CCR9 or CCL25 lead to inflammatory diseases and overexpression correlates with malignant tumor metastasis. Although robust downstream G protein responses have been reported, we observe that G protein activity, determined by live-cell BRET assays, by CCR9 is severely and rapidly attenuated. What is leading to this low level of CCR9 activity is unclear. We proposed that rapid desensitization by arrestins or phosphorylation of the receptor is suppressing the G protein response in our system. Here, we test this hypothesis by observing G protein activation and signaling in cells lacking arrestins or GPCR kinases (GRKs) due to CRISPR knockout. Canonically, arrestins physically interfere with GPCR signaling, however, the absence of arrestins had no impact on CCR9 activation. Instead, a 6-fold increase in activation was observed in the absence of GRKs. Site-directed mutagenesis identified that the proximal phosphorylation sites were responsible for the attenuation. The modulation was not due to CCR9 re-localization or internalization, but rather phosphorylation appeared to interfere with G protein binding. This suggests that phosphorylation directly regulates the G protein-dependent activity of CCR9 and may have implications for targeting the receptor therapeutically and other GPCRs.

Activation dynamics of mGluR5 and Novel Positive Allosteric Modulator: A Computational Approach and Mechanistic Insights

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Metabotropic glutamate receptors (mGluRs) are critical Class C G-protein coupled receptors (GPCRs) involved in various neural processes, with their dysfunction linked to numerous neurological disorders.

mGluRs operates as dimers, with each monomer consisting of a large extracellular ligand-binding domain, a cysteine-rich domain, and a seven-transmembrane domain containing an allosteric site.

Allosteric modulators are of particular interest as they enhance receptor response to the endogenous agonist (L-glutamate), offering greater mGluR subtype-specificity and potentially, more refined pharmacological outcome and fewer side effects compared to orthosteric ligands. However, the mechanism of mGluR allosteric modulation is unclear, hampering the design of allosteric modulators. Indeed, mGluR activation is a complex, step-wise process which is only partially understood [1,2].

Here, we focus on mGluR5 and its positive allosteric modulators (PAMs), which have shown promise in the therapeutic intervention of schizophrenia.

To elucidate the activation mechanism and allosteric modulation of mGluR5, we performed extensive molecular dynamics (MD) simulations, using a state-of-the-art enhanced-sampling technique, Reservoir-Replica-Exchange MD (R-REMD). This approach captured detailed conformational changes and activation dynamics of the receptor.

Further MD simulations with this PAM demonstrated its role in facilitating mGluR5 activation by accelerating the TM6-TM6 contact between dimers, promoting the receptor's transition to an active state. The findings enabled in silico screening and the discovery of a novel PAM, which shows promising therapeutic effects in preclinical mouse models of schizophrenia.

In conclusion, our computational approach has successfully identified a novel mGluR5 PAM and provided detailed insights into its activation mechanism. These findings contribute to the understanding of mGluR activation dynamics, offering a foundation for future drug discovery efforts targeting this important GPCR family.

Pin J-P, Bettler B. 2016. Organization and functions of mGlu and GABAB receptor complexes.Nature. 60–68

Krishna Kumar K, Wang H, et al. 2024. Stepwise activation of ametabotropic glutamate receptor.Nature. 1–6

A role for the proton-sensing GPCR, GPR65 in inflammatory joint pain.

Luke A. Pattison[1], Rebecca H. Rickman[1], Helen Hilton[1], Susanne Wijesinghe[2], Graham Ladds[1], Li Yang[3], Simon Jones[2] and Ewan St. J. Smith[1]

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Therapeutic options for treating inflammatory pain are limited and plagued by side-effects. Localised acidosis is characteristic of inflammation and many receptors expressed by pain-sensing neurons (nociceptors) are sensitive to extracellular protons, which makes them potential targets for treating inflammatory pain. While proton-sensitive ion channels have more established roles in nociception, less is known about the roles of proton-sensing G protein-coupled receptors (PS-GPCRs). In several inflammatory pain models increased expression of PS-GPCRs has been reported. In particular, GPR65 (also known as TDAG8) expression often increases following inflammatory insult, and thus we sought to further explore the role of GPR65 in inflammatory pain. First, in a uniform cellular background the signalling responses elicited following GPR65 activation by protons and two other reported agonists, BTB09089 and psychosine, were compared. BTB09089 best recapitulated proton-induced signalling, highlighting its use as a more-selective tool to probe GPR65 function. Injection of BTB09089 into the knee joint of mice caused inflammation and pain-related behaviours. Additionally, knee-innervating sensory neurons supplying the BTB09089 injected knee were hyperexcitable. However, this could not be re-created by directly stimulating sensory neurons isolated from naïve mice with BTB09089, thus suggesting the involvement of another cell type. Fibroblast-like synoviocytes (FLS) resident in the joint also express GPR65. Stimulation of FLS with BTB09089 increased cAMP accumulation and induced secretion of inflammatory mediators capable of sensitising naïve sensory neurons. The specificity of these results was confirmed through studies with GPR65-/- mice. Similar effects of BTB09089 on FLS isolated from arthritic human patients were also observed. We thus postulate that GPR65 is a key mediator of cell-cell interactions responsible for the manifestation of inflammatory pain in both humans and mice, and thus of interest in the development of more targeted therapeutics for treating inflammatory diseases.

Spatio-temporal organization of LHCGR signalling is essential to regulate gonadal activity

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Luteinizing hormone (LH) is essential to control reproductive functions through binding its G protein-coupled receptor (GPCR), the LHCGR, expressed by gonadal cells. Activation of LHCGR by its ligand engenders a complex signalling network that ultimately controls steroidogenesis and ovulation. A long-known key regulator of this signalling network is the Gs/cAMP/PKA signalling pathway, and studies have recently highlighted the importance of receptor endocytosis in the control of cAMP dynamics. Upon hormone binding, LHCGR internalization occurs preferentially through a subset of signalling endosomes called Very Early Endosomes (VEE), distinct from the classical Rab5-positive Early Endosomes (EE). So far, it is unknown whether LHCGR can signal from multiple endosomes and whether cAMP compartmentation is physiologically relevant. GPCR signalling compartmentation is a growing field, but in the case of LHCGR, it remains unclear why and how gonadal cells spatially and timely organize cAMP signalling to control and regulate specific biological responses. Using a panel of approaches aiming at studying receptor post-endocytic signalling, our data support a crucial role of LHCGR endocytosis to regulate cAMP response organization and gene transcription. In addition, in both Leydig cell line mLTC-1 and mouse primary Leydig cells, we demonstrate that endosomal cAMP signalling is essential to steroidogenesis.

Session 4:

Leveraging Drosophila and AI for High-Throughput Studies of Inflammation-Induced Pain

Isabella Maiellaro[1], Zsuzsa Markus[1], Meritxell Canals[1], Kevin Webb[2]

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Sensory neurons are responsible for sensing and adapting to noxious stimulation. Following noxious stimulation, sensory neurons normally return to a basal/inactive level. Conditions like inflammation impair this ability to restore equilibrium, leading to hypersensitivity that, if persistent, can result in chronic pain.

Acute nociception and hypersensitivity reflect two functional states of the nociceptive circuit. The former aims to protect the injured body, while the latter, when persistent, becomes chronic and debilitating rather than serving as an acute warning.

Elucidating the differences between these two states is important, as it will likely shed light on chronic pain mechanisms and lead to the discovery of specific factors underlying inflammatory pain.

Our aim is to establish an invertebrate alternative to the more challenging rodent and simian models for studying the mechanisms underlying inflammation-induced sensitization.

Drosophila larvae respond to noxious stimuli (heat, mechanical, or chemical) with a stereotyped behavioural response called nocifensive escape locomotion (NEL), where the larvae rotate around their body axis in a corkscrew-like motion. Genetic screens have identified the genes and cells responsible for such behaviour. Importantly, Drosophila are ideally suited for genetic and pharmacological screening, as a large number of mutants and treatments can be created in a short time.

However, current behavioural assays to detect nociceptive responses are severely limited by their low throughput, requiring manual stimulation with a mechanical or chemical probe and categorization by direct observation, one larva at a time.

We have established a platform to monitor nocifensive escape locomotion and its underlying signalling at the single-neuron level, allowing the study of acute nociception and hypersensitivity. We further optimise the behavioural assays by harnessing optogenetics and AI classification to trigger and codify behavioural events in real time, thereby increasing the throughput from single to multiple larvae, making it suitable for genetic screening purposes.

Advancing GPCR drug discovery via computational methods in an ultimately efficient way

Shuguang Yuan
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Modern drug discovery is a long and tedious process which costs at least 10 years and 2 billion USD in average. How to speed up this expensive process has become one of the most essential topics in pharmaceutical industry. With the progresses in both artificial intelligence and computational biology, advancing modern drug discovery via computational pharmacy plays more and more important roles. In this talk, Dr. Yuan will illustrate applying computational biology and artificial intelligence to answer fundamental questions in life science, especially in the area of G protein-coupled receptors (GPCRs). He will also discuss how to speed up modern drug discovery in an ultimate efficient way. Finally, Dr. Yuan will also talk about his successfully story on how to develop “first-in-class” drug molecules into clinical trials.

WORKSHOP (SEPARATE REGISTRATION)

Day 4 – Thursday 11th July, 2024

4th IRN i-GPCRnet Workshop

Pharmacology in Drug Discovery

Dear PhD students and postdoctoral researchers of the IRN i-GPCRnet and attendees of the meeting,

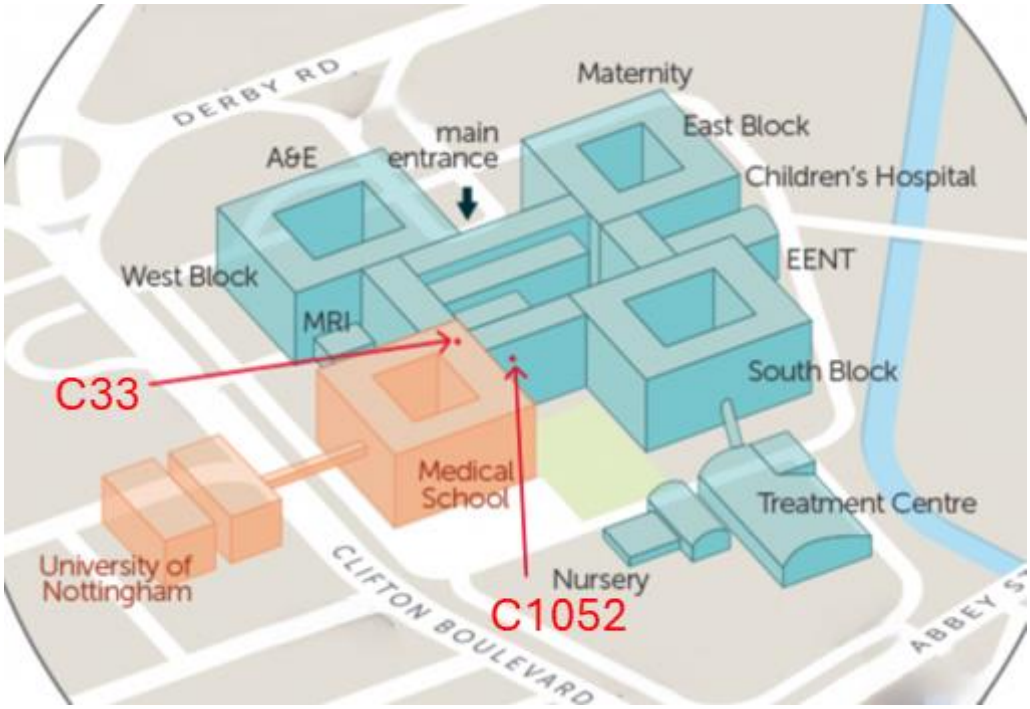
Commencing on the day after the 4th annual i-GPCRnet meeting (Thursday 11th July, 2024), the workshop in Nottingham will be based on a well-established due diligence exercise where small teams of 4-5 will work through data provided by three companies on a promising drug candidate. The ultimate aim is for each group to make a recommendation to senior management for in-licencing of one selected candidate drug. The exercise will be preceded by two lectures giving the background to analysis of ligand-binding and functional responses of GPCRs.

PROGRAMME

09:00 - 09:15:	**Dr. Laura Kilpatrick** *An introduction to Pharmacology in Drug Discovery*	Medical School Building, C1052
09:15 - 10:15:	**Prof. Steve Hill** *Lecture 1: An Introduction to Ligand Binding*	
10:15 - 10:45:	***Coffee break***	Medical School Building, C33
10:45 - 11:45:	**Prof. Steve Hill** *Lecture 2: Measurement of Functional Responses*	Medical School Building, C1052
11:45 - 12:00:	Introduction to the Workshop	
12:00 - 13:00:	***Lunch***	Medical School Building, C33
13:00 - 15:00:	**Workshop:** Due Diligence Data Exercise and Hands-On Cellular Assays Provided by BMG Labtech and Promega	
15:00 - 15:30:	Workshop Feedback	

Access and locations:

The workshop will take place in the Medical School Building, within the Queen's Medical Centre (hospital). The closest tram stop to the Medical School is the Queen's Medical Centre (QMC) stop on the Toton Lane. The introduction and lectures will be held in C1052 (C Floor of the South block of the QMC) and the breaks and workshop will be held in C33 (C Floor of the Medical School, red core). There will be volunteers (in red T-shirts wearing "HELPER" badges) ready to guide you from the footbridge (linking University Park to the Medical School), the Coates Road auditorium, or the QMC tram stop to the lecture theatre.



Security and emergency contact numbers

The university's Security service provides a 24-hour uniformed presence on all campuses. One of Security's main purposes is to respond to calls for service, provision of routine patrols, checking, locking and unlocking buildings. If you need to contact Security, please use the first number below:

Phone: +44 115 951 3013 for 24 Hour Security Control
Phone: +44 115 951 8888 for Emergencies ONLY (fire, medical emergency)