Name	NEIL MCDONALD
Position	Senior Group Leader
Year joined *(Crick or founder institute)*	1994

Career History

1987 – 1991: Birkbeck College, University of London, UK - Ph.D. in Crystallography (Supervisor Professor Tom L. Blundell)
1991 -1994: Columbia University, New York, USA - Postdoctoral fellow (in lab of Professor Wayne A. Hendrickson, HHMI)

Major Awards, Honours and Prizes

1991- 1994: Lucille P. Markey Scholar (competitive fellowship award)
2006: Promoted to Full Professor at Birkbeck College
2007: Shortlisted for the Queen's Anniversary Prize, Birkbeck College
2020: Nominated for the David Cooksey Translational Award

Membership of external committees, editorial boards, review panels, SABs etc

2016 – 2019 CR-UK Science Programme Grant Ad Hoc Committee Member
2016 - present Editorial board of Biochemical Journal

Lab Name	***Signalling and Structural Biology Laboratory***

Research programme and achievements

The laboratory studies the molecular mechanisms regulating membrane-linked signalling hubs formed by the neurotrophic receptor tyrosine kinases and the PKC superfamily of serine/threonine kinases. Members of both protein kinase classes act to control fundamental biological processes from cell patterning and polarisation to neuronal physiology, connectivity and survival. Their aberrant activities directly contribute to neurodegeneration and neuropsychiatric disease, heart disease and cancer.

We are addressing two major questions: (1) How do neurotrophic factor receptors assemble and signal to control neuronal function and how are they subverted by oncogenic deregulation in non-neuronal contexts? (2) How do multi-component aPKCi/PKN kinase complexes establish cell membrane polarity and cell shape through a network of activating and inhibitory interactions? Our long-term ambition is to leverage our understanding of these kinase control mechanisms to explain their emergent biological properties and manipulate their functional states using chemical and biological tools.
Our structure-driven programme uses crystallography, cryo-EM and cell biology on reconstituted membrane-linked kinase assemblies, integrated with *in vivo* models through

collaborations within the Crick Institute. We have exploited opportunities to develop chemical and biological tools against our molecular targets to contribute to drug-discovery programmes.

The main achievements of the laboratory since 2015 are as follows:

1: We have determined cryo-EM structures of GDNF ligand-bound RET receptor complexes. Insights from these studies have allowed us to pursue a translational project supported by Crick i2i funding with the aim of producing bispecific DARPin anti-RET antagonists. We are probing bispecific DARPin properties on AML cell lines dependent on RET function, both in laboratories at the Crick (Bonnet lab) and in Heidelberg (Scholl lab).

2: We have explored mechanisms of RET tyrosine kinase oncogenic deregulation by missense mutation and identified novel modes of RET chemical inhibition through P-loop engagement. Importantly, this new mode of RET inhibition is largely insensitive to oncogenic gatekeeper mutation.

3: We have reconstituted a key polarity complex containing aPKCi and determined its structure using cryo-electron microscopy. We have combined this analysis with collaborations within the Crick to examine the impact of targeted disrupting mutations using polarised cells and externally using an *in vivo* model organism.

4. We initiated a successful aPKC iota drug design programme with CRT, together with Peter Parker. This programme directly led to potent, selective inhibitors against aPKC used by Barry Thompson, Kathy Niakan and Nate Goehring for their research programmes and publications. Pre-clinical candidates and a backup series are currently being assessed in a topical formulation to target aberrant Hedgehog signalling in basal cell carcinoma.

4: Structural studies on RPEL motif-containing complexes with G-actin in collaboration with the Treisman Lab have given insights into surprising stoichiometries and mechanisms
of G-actin function. These studies show how G-actin acts to control RPEL motif containing protein function, impacting on either subcellular localisation (MRTFs) or enzymatic activity (Phactrs and RhoGAPs) by steric exclusion mechanisms.

5. Our cryo-EM structures of the DNA repair endonuclease complex XPF-ERCC1 identified an auto-inhibited XPF-ERCC1 complex and a form bound to a ss/dsDNA substrate. We have exploited our structural findings to propose a collaboration with Astra-Zeneca to identify chemical inhibitors in a drug discovery programme.

Future work of the laboratory will focus on the molecular roles of both neuronal tyrosine and serine/threonine kinases in neuronal connectivity, survival and polarisation. This will include: (1) Investigation of how neuronal receptors assemble in membrane and trans-synaptic contexts visualised using a combination of cryo-EM and cryo-ET (2) The development of biological agonists and antagonists against neuronal receptors in relevant disease contexts (3) Defining the interplay between reconstituted polarity complexes that we have solved by cryo-EM and their respective membrane recruitment mechanisms (4) Visualising and manipulating distinct activity states of polarity complexes and their subcellular locations using nanobody tools.

Research outputs

Jones M, Beuron F, Borg A, Nans A, Earl CP, Briggs DC, Snijders AP, Bowles M, Morris EP, Linch M, McDonald NQ. (2020) ***Cryo-EM structures of the XPF-ERCC1 endonuclease reveal how DNA-junction engagement disrupts an auto-inhibited conformation*****. Nat Commun 11(1):1120. DOI: 10.1038/s41467-020-14856-2**
This first cryo-EM structure from the lab determined the first full length XPF family endonuclease structure revealing an auto-inhibited conformer and the initial steps in DNA-junction recognition opening up the endonuclease structure.

Soriano EV, Ivanova ME, Fletcher G, Riou P, Knowles PP, Barnouin K, Purkiss A, Kostelecky B, Saiu P, Linch M, Elbediwy A, Kjær S, O'Reilly N, Snijders AP, Parker PJ, Thompson BJ, McDonald NQ. (2016) ***aPKC Inhibition by Par3 CR3 Flanking Regions Controls Substrate Access and Underpins Apical-Junctional Polarization*****. Dev Cell 38(4):384-98. DOI: 10.1016/j.devcel.2016.07.018**
This three-way collaboration within the Crick combined a molecular and cellular analysis of Par3 interaction with aPKCiota, together with an *in vivo* examination of the polarised follicular epithelium in *Drosophila*. The study suggests a dual role for Par3 as both substrate and competitive inhibitor of aPKCiota function.

Diring J, Mouilleron S, McDonald NQ, Treisman R. (2019) ***RPEL-family rhoGAPs link Rac/Cdc42 GTP loading to G-actin availability*****. Nat Cell Biol 21(7):845-855. DOI: 10.1038/s41556-019-0337-y**
This collaborative publication continued our efforts to apply X-ray crystallography to define the interaction mode and stoichiometry of G-actin binding to three known classes of RPEL motifs: MRTF, Phactrs and RhoGAPs. The study concludes that a subset of RhoGAPs are regulated by G-actin binding a single RPEL-motif, blocking access to the rho-GTPase binding site.

Plaza-Menacho I, Barnouin K, Barry R, Borg A, Orme M, Chauhan R, Mouilleron S, Martínez-Torres RJ, Meier P, McDonald NQ. (2016) ***RET Functions as a Dual-Specificity Kinase that Requires Allosteric Inputs from Juxtamembrane Elements*****. Cell Reports 17(12):3319-3332. DOI: 10.1016/j.celrep.2016.11.061**
This study unexpectedly revealed a serine auto-phosphorylation site within the RET kinase domain required for activation both *in vitro* and *in vivo*. These findings are consistent with a dual-specificity kinase function for this receptor tyrosine kinase.

Adams SE, Purkiss AG, Knowles PP, Nans A, Briggs DC, Borg A, Earl CP, Goodman KM, Narowtek A, Borg A, McIntosh PB, Houghton FM, Kjær S, McDonald NQ. (2021) ***A two-site flexible clamp mechanism for RET^{ECD}-GDNF-GFRα1 assembly reveals both conformational adaptation and strict geometric spacing.*** **Structure 29, 1-15. DOI: 10.1016/j.str.2020.12.012**
RET is an unusual receptor tyrosine kinase that recognizes a set of five GDNF family ligand-co-receptors. Our paper reveals the basis for ligand-co-receptor recognition by RET. By comparing crystallographic and cryo-EM structures of unliganded and ligand-co-receptor-bound RET, we explain how RET acts as a two-site molecular clamp. It has a flexible arm that engages different co-receptors through conformational adaptation while a rigid arm recognizes the dimeric nature and molecular dimensions of each GDNF family ligand to drive receptor activation. Our findings have implications for the design of RET receptor modulators relevant to both Parkinson's disease and RET-driven cancers.