Background and Aims of Programme

The Wellcome Four Year PhD Programme in Integrative Cell Mechanisms (iCM) is closely associated with the Wellcome Centre for Cell Biology and trains the next generation of cell and molecular biologists in the application of quantitative methods to understand the inner workings of distinct cell types in different settings.

A detailed understanding of normal cellular function is required to investigate the molecular cause of disease and design future treatments. However, data generated by biological research requires increasingly complex analysis with technological advances in sequencing, mass spectrometry/proteomics, super-resolution microscopy, synthetic and structural biology generating increasingly large, complex datasets. In addition, innovations in computer sciences and informatics are transforming data acquisition and analysis and breakthroughs in physics, chemistry and engineering allow the development of devices, molecules and instruments that drive the biological data revolution. Exploiting technological advances to transform our understanding of cellular mechanisms will require scientists who have been trained across the distinct disciplines of natural sciences, engineering, informatics and mathematics.

To address this training need, iCM PhD projects are cross-disciplinary involving two primary supervisors with complementary expertise. Supervisor partnerships pair quantitative scientists with cell biologists ensuring that students develop pioneering cross-disciplinary collaborative projects to uncover cellular mechanisms relevant to health and disease.

We aim to recruit students with a variety of backgrounds across the biological and physical sciences, including Biochemistry, Biomedical Science, Cell Biology, Chemistry, Computational Data Sciences, Engineering, Genetics, Mathematics, Molecular Biology and Physics. Students are trained to adapt, broaden and apply their skill set to the understanding of cellular mechanisms of biomedical importance.

The next deadline for applications will be early December 2021 and will be posted at: https://www.wcb.ed.ac.uk/how-apply
The expansion of research in cell biology was planned in 1992 as a result of the vision of Professor Sir Kenneth Murray, who was at the time Biogen Professor at the Institute of Cell and Molecular Biology. A seed contribution of £2.5 million from the Darwin Trust was followed by financial commitments from The Wolfson Foundation, the University and the Wellcome Trust, allowing construction of the Michael Swann Building. The majority of research space was earmarked for Wellcome Trust-funded research. Recruitment, based on research excellence at all levels in the area of cell biology, began in earnest in 1993, mostly but not exclusively, through the award of Research Fellowships from the Wellcome Trust. The Swann Building was first occupied by new arrivals in January 1996 and became “The Wellcome Trust Centre for Cell Biology” from October 2001. Core funding for the Centre from the Wellcome Trust was renewed in 2006, 2011, 2016 and 2021.
Director’s Report

The Wellcome Centre for Cell Biology (WCB) is one of fifteen UK-based, Wellcome-funded research centres. The WCB research groups, and our associated research facilities, occupy the Michael Swann Building on the King’s Buildings Campus of the University of Edinburgh. Constructed in the mid-1990s, the Michael Swann Building was purpose built as a centre for research in molecular cell biology.

WCB has benefitted from Wellcome Centre status since 2001, and this has allowed us to bring together outstanding scientists in a dynamic, supportive environment, where their work is enhanced by access to a set of world-class core facilities. Such a situation is usually found only in research institutes, but WCB also benefits from being embedded in a major university. This setting offers unrivalled opportunities for interaction with researchers across the range of disciplines in science, medicine and social science, as well as the arts.

This is my last report as Centre Director; after 10 years in the post, I will be passing on the role to Prof. Adèle Marston, who is currently Deputy Director. A key event for the Centre in the past year was the extension of our Core Grant. Adèle successfully led this bid and will take over as Director at the start of the new funding period – in December 2021.

It has, of course, been an unusual and difficult year. For most of us, life has changed substantially since the time I sat down to write the previous report, in March 2020. Then, the term of lockdown was strange and new, but still seemed to be something that happened to people elsewhere – although getting ominously closer. During the past year, I have been hugely impressed by the fortitude and resilience shown by the members of WCB. I must also express my deep gratitude to all of the support staff in Swann and elsewhere in King’s Buildings, many of whom have continued to work through all of the lockdowns.

This brochure presents a very brief overview of our research and highlights some of the exciting results reported during the past year. I am delighted that, despite the ongoing challenges posed by lockdown, which almost halted our research, and continuing greatly reduced building occupancy, WCB groups have again made excellent progress, reporting innovative, world-class research, as reflected in our many notable publications.

In addition, I would like to congratulate individual WCB members on grant funding successes during the year. Bill Earnshaw renewed his Wellcome Principal Research Fellowship while, in addition to leading the centre renewal, Adèle Marston, who is currently Deputy Director. A key event for the Centre in the past year was the extension of our Core Grant. Adèle successfully led this bid and will take over as Director at the start of the new funding period – in December 2021.

Centre staff have also managed to deliver an excellent programme of public engagement, despite the necessity of switching to an entirely online delivery. Particular thanks to Sarah-Jane Judge, who took on the role of Public Engagement Manager just as we went into the first lockdown; making for a challenging start. Thanks to everyone who gave up their time and brought their enthusiasm to share our work with the broader community in Edinburgh. Let me also welcome Karen May in her new role as Centre Manager, which she stepped into at the start of this year.

In the absence of a normal seminar programme, we have established a weekly “Renowned Seminar Speaker Programme”. This is proceeding well, so thanks to Robin Allshire for getting the ball rolling on this. The first cohort has joined our Wellcome 4-year PhD programme in “Integrative Cell Mechanisms (ICM)”, again under difficult circumstances. Thanks to everyone who participated; particularly Vasso Makrantoni who organised the start-up of the programme, before leaving to start her own independent career. This exciting, cross-disciplinary partnership brings cell biologists together with physical and data science experts across Edinburgh.

Centre members were also successful in obtaining prestigious posts elsewhere and deserve our warmest congratulations, even though we are very sorry to see them leave. Tony Ly moved to a new position in Dundee University, Vasso Makrantoni (Marston lab) was appointed as a lecturer at the Zhejiang University - University of Edinburgh Institute, Clémentine Delan-Forino (Tollervey lab) obtained a prestigious CNRS post in the Institut de Microbiologie de la Méditerranée in Marseille, France. Marie-Luise Winz (Tollervey lab) was appointed as a Junior Professor, in the Johannes Gutenberg University in Mainz, Germany. Our best wishes to them all in their future careers. The continuing success of Centre alumni is a great source of pride.

Prof. Jean Beggs, who was a founding member of WCB, took retirement last year, after a hugely successful and influential research career. She is greatly missed as a colleague in WCB and throughout the RNA community. Happily, however, the WCB group leaders unanimously voted to create the position of Emeritus Centre Member, with Jean as first recipient of this honour. Jean has therefore contributed an entry in the newly-established Emeritus Centre Member section of the brochure. It is a great pleasure to see her contributions resume.

Let me end, as usual, by reiterating my sincere thanks to all of our talented and dedicated researchers and support staff, and congratulate them on their excellent work over the year. Your efforts underpin all of the continuing successes of the WCB.

In addition, I would like to congratulate individual WCB members on grant funding successes during the year. Bill Earnshaw renewed his Wellcome Principal Research Fellowship while, in addition to leading the centre renewal, Adèle Marston received a Wellcome Investigator Award, and Julie Welburn was the recipient of a COVID Reboot grant. Notable awards were given as marks of esteem in recognition of outstanding work by WCB PIs: Donal O’Carroll was awarded the lifetime honour of EMBO (European Molecular Biology Organisation) Membership. Robin Allshire was elected as a Fellow of the Academy of Medical Sciences. Adrian Bird was announced as the joint winner of the Brain Prize – the most valuable research career. She is greatly missed as a colleague in WCB and throughout the RNA community. Happily, however, the WCB group leaders unanimously voted to create the position of Emeritus Centre Member, with Jean as first recipient of this honour. Jean has therefore contributed an entry in the newly-established Emeritus Centre Member section of the brochure. It is a great pleasure to see her contributions resume.

Let me end, as usual, by reiterating my sincere thanks to all of our talented and dedicated researchers and support staff, and congratulate them on their excellent work over the year. Your efforts underpin all of the continuing successes of the WCB.

In addition, I would like to congratulate individual WCB members on grant funding successes during the year. Bill Earnshaw renewed his Wellcome Principal Research Fellowship while, in addition to leading the centre renewal, Adèle Marston received a Wellcome Investigator Award, and Julie Welburn was the recipient of a COVID Reboot grant. Notable awards were given as marks of esteem in recognition of outstanding work by WCB PIs: Donal O’Carroll was awarded the lifetime honour of EMBO (European Molecular Biology Organisation) Membership. Robin Allshire was elected as a Fellow of the Academy of Medical Sciences. Adrian Bird was announced as the joint winner of the Brain Prize – the most valuable research career. She is greatly missed as a colleague in WCB and throughout the RNA community. Happily, however, the WCB group leaders unanimously voted to create the position of Emeritus Centre Member, with Jean as first recipient of this honour. Jean has therefore contributed an entry in the newly-established Emeritus Centre Member section of the brochure. It is a great pleasure to see her contributions resume.

Let me end, as usual, by reiterating my sincere thanks to all of our talented and dedicated researchers and support staff, and congratulate them on their excellent work over the year. Your efforts underpin all of the continuing successes of the WCB.
Chromosomal DNA wraps around nucleosomes containing core histones (H3/H4/H2A/H2B). At centromeres however, a specific histone H3 variant, CENP-A, replaces histone H3 to form specialized CENP-A nucleosomes. CENP-A chromatin is critical for chromosome segregation machinery assembly (kinetochores) and specific chromosomal locations. Kinetochores are flanked by histone H3 lysine 9 methylation (H3K9me)-dependent heterochromatin. Our goal is to decipher conserved mechanisms that establish, maintain and regulate heterochromatin and CENP-A chromatin domain assembly. Heterochromatin is required for CENP-A chromatin establishment on centromere DNA. One objective is to provide further insight into mechanisms that promote heterochromatin formation on pericentromeric repeats. Heterochromatin might also silence genes throughout the genome: we therefore also investigate how heterochromatin formation is regulated and whether such mechanisms influence phenotype. We endeavour to determine how heterochromatin, spatial nuclear organisation and non-coding RNA transcription combine to mediate CENP-A incorporation at centromeres.

Our main questions are: 1. How do chromatin architecture and subnuclear compartmentalization affect specialized chromatin domains? 2. How does chromatin architecture alter nucleosome chromatin loop size relative to flanking euchromatic host chromatin (Figure D). Thus, altered loop size likely contributes to the distinct appearance of heterochromatin (Figure E).

It is not known why heterochromatin has a distinct architecture on mitotic chromosomes. We find that large regions of fission yeast chromatin which adopt a mitotic organisation distinct from surrounding host chromosome regions (Fitz-James et al. 2020). Heterochromatin assembled on this inserted fission yeast DNA alters chromatin loop size relative to flanking euchromatic host chromatin (Figure D). Thus, altered loop size likely contributes to the distinct appearance of heterochromatin (Figure E).

Selected Publications

Robin Allshire
Co-workers: Tatiana Aucynnikava, Roberta Carloni, Tadhg Devlin, Andreita Fellas, Elisabeth Gaberdel, Dominik Hoelper, Marcel Latos, Nile Tse London, Sunil Naha, Alisson Pidoux, Sevena Pociunaitė, Desislava Staneva, Manu Shukla, Silo Torres-Garcia, Sharon White, Welfang Wu, Imtiazy Yaseen, Rebecca Yeooba

Epigenetic inheritance: establishment and transmission of specialized chromatin domains

A. Unstable caffeine resistant epimutants UR-1 and UR-2 exhibit novel islands of H3K9me-dependent heterochromatin compared to wild-type cells (wt).

B. UR-1 and UR-2 caffeine resistant (CAF) epimutants are cross-resistant to clinical (CLT, Clotrimazole, FLC, Fluconazole) and agricultural (TEB, Tebuconazole) fungicides used to treat human and crop-plant fungal infections, respectively.

C. Model. Resistant isolates arise after lethal insult exposure. Resistance can be mediated by changes in DNA (resistant mutants) or reversible, heterochromatin-based epimutations (resistant epimutants). Upon withdrawal of insult epimutants lose ectopic heterochromatin islands, resistance, reverting to wild-type (sensitive phenotype). In contrast genetic mutants continue to exhibit mutant resistance phenotype.

D. Large inserts of fission yeast DNA in mammalian chromosomes exhibit an unusual mitotic chromosome structure associated with H3K9me-dependent heterochromatin formation. Hi-C analyses demonstrate more frequent contacts per kb over the fission yeast chromosome insert compare to flanking mouse chromatin.

E. A higher ratio of condensin to chromatin over fission yeast DNA assembled in heterochromatin results in more contacts and less chromatin per unit length and consequently, reduced mitotic chromosome width.

F. Models. Heterochromatin formation triggers condensin overloading of chromatin, reducing mitotic chromosome width.


Cell division is an essential biological process that ensures genome integrity by equally and identically distributing chromosomes to the daughter cells. Errors in cell division often result in daughter cells with inappropriate chromosome numbers, a condition associated with cancers and birth defects. Key events that determine the accuracy of cell division include centromere specification, kinetochore assembly, physical attachment of kinetochores to spindle microtubules and successful completion of cytokinesis. These cellular events are regulated by a number of mitotic molecular assemblies (including the Chromosomal Passenger Complex (CPC), KMN (Knl1-Mis12-Ndc80) network, the Ska complex, Spindle Assembly Checkpoint and the Anaphase Promoting Complex) involving an extensive network of protein-protein interactions.

Although much is known about the basic mechanisms of cell division, structural level mechanistic details of pathways regulating error-free chromosome segregation are still emerging. In particular, a high-resolution understanding of centromere inheritance and how kinetochores employ dynamic protein interaction to harness the forces generated by spindle microtubules to drive chromosome segregation is yet to be obtained. To address these important questions requires an approach that integrates structural and functional methods capable of dissecting and probing individual roles of protein interactions mediated at varying timescales. We use molecular biology and biochemical approaches to characterize protein interactions in vitro, X-ray crystallography, Cross-linking/Mass spectrometry, Small Angle X-ray Scattering (SAXS) and Electron Microscopy for structural analysis and a combination of in vitro and cell-based in vivo functional assays using structure-guided mutations for functional characterization.

The specific questions that we aim to address currently are: i) What is the molecular basis for the establishment and maintenance of CENP-A nucleosomes at centromeres? ii) How do the outer kinetochore microtubule binding components cooperate to facilitate spindle-driven chromosome segregation? and iii) How CPC, a key player required for eliminating incorrect kinetochore-microtubule attachment is targeted to the kinetochore? We address these questions by characterizing protein complexes involved in centromere maintenance (Mis18 and Mis18-associated), physically coupling chromosomes to kinetochores (the Ska complexes and outer kinetochore microtubule binding factors) and error-correction (CPC and its centromere/kinetochore receptors). The structural and functional insights from these studies will also provide new avenues for targeting specific protein-interactions to fight mitosis-related human health disorders.

Our recent crystal structures of Cal1 bound to CENP-A/H4 heterodimer and CENP-C provided a mechanistic understanding of how Cal1 targets CENP-A/H4 to centromeres to maintain centromere identity. Our work revealed that Cal1 combines functions of human Mis18 complex and HJURP, through evolutionarily conserved and adaptive interactions, to target CENP-A/H4 to centromeres in a self-sufficient manner. (Medina-Pritchard et al., 2020).

**Selected Publications**


Regulation of gene expression in eukaryotes is complex, but probably ultimately depends on proteins that recognise specific DNA sequences. Our recent work emphasises this relationship by showing an unexpected dependence of gene function on proteins that recognise short, frequent base sequence motifs (Figure 1). This points to a broad-brush category of control whereby genes can be up- or down-regulated based on the DNA sequence properties of their genomic location.

Our studies of this phenomenon have centred on two proteins that are implicated in human disease: MeCP2 and SALL4. MeCP2 contains a DNA binding domain that requires DNA methylation in either a CG or a CAC context. This protein is highly expressed in mature neurons and its deficiency causes the profound neurological disorder Rett syndrome. We showed previously that MeCP2 restrains gene expression by recruiting a corepressor to the genome. Recently, we questioned the importance of the unusual methyl-CAC target site, which is abundant only in neurons. To do this, we replaced the DNA binding domain of MeCP2 with one that can bind mCG but not mCAC. Mice dependent on this protein had Rett syndrome-like phenotypes, indicating that mCAC binding is essential for the function of MeCP2. These experiments also allowed us to identify a subset of de-regulated genes that are mis-expressed in other neurological disorders and may be implicated in Rett syndrome. Future work will explore this possibility further.

The second protein under study is SALL4, a multi-zinc-finger protein that plays an important role in development and disease. For example, SALL4 is highly expressed in many cancers with poor prognosis. We speculated that it might interpret DNA base composition by recognising AT-rich DNA and, in agreement with this notion, found that a zinc finger cluster specifically targets short A/T-rich motifs in the genome and recruits a partner corepressor. In embryonic stem cells, this represses a set of AT-rich differentiation genes, thereby prolonging the pluripotent state. Prevention of AT-binding activates these genes, leading to precocious differentiation. It has been recognised for decades that the mammalian genome is organised in long domains with distinct, evolutionarily-conserved base compositions. Our SALL4 study provides the first evidence that base composition can be read as a biological signal to regulate gene expression.

Despite major differences between MeCP2 and SALL4 in protein sequence, pattern of expression and the physiological consequences of mutation, they share striking similarities. Both recognise short DNA sequence motifs that are frequent in the genome and both recruit corepressor complexes leading to widespread modulation of gene expression. Future work will seek a comprehensive mechanistic picture of the ways in which mechanisms of this kind help to define and stabilise cell states, and how defects in this system lead to disease.

Selected Publications


Understanding proteins that interpret the genome to stabilise cell states

Similarities and differences between SALL4 and MeCP2 function. Both mediate the effects of short, frequent DNA sequence motifs on gene expression by recruiting corepressors containing histone deacetylases (HDACs). However, they recognise different base sequences (methylcytosine-containing versus AT-rich) and recruit different corepressor complexes (NCoR versus NuRD) associated with distinct HDACs (HDAC3 versus HDAC1/2).
The central nervous system is a complex network of neurons and supporting cells that form the information relaying unit of an organism. During neural development, pioneer neurons extend axons in response to guidance cues from other neurons and non-neuronal cells to establish the framework that build the neural circuits. The assembly of this circuit is a highly orchestrated event that involves neurite outgrowth, fasciculation (axon bundling) and synapse formation to generate a functional nervous system. How these organizational features emerge during development is poorly understood.

Microtubules are critical for neuron formation and function. As neurons develop, microtubules are organized and sculpted by the cell machinery to form the axons, dendrites and the neural network. Several human neurodevelopmental disorders are linked to mutations in microtubule cytoskeleton-related proteins. Despite the central role of the microtubule, little is known about how the microtubule cytoskeleton contributes to the assembly of the neural circuit. We aim to understand how the microtubule cytoskeleton uses distinct molecular machinery to build and regenerate 3 dimensional neuronal circuits using the simple multicellular organism C. elegans as a model.

During my post-doc, I discovered an unexpected role for kinetochore, the chromosome segregation machinery, in developing neurons of C. elegans. Our work showed that the evolutionarily conserved 10 subunit KMN (Knl1-Mis12-Ndc80) network, the microtubule coupler within the kinetochore, acts post-mitotically in developing neurons. A similar function for kinetochore proteins has also been described in Drosophila and rat hippocampal cultures. KMN proteins are enriched in the dendritic and axonal outgrowth during neurodevelopment. Removal of KMN components post-mitotically from developing neurons resulted in a disorganized nerve ring, a network of 181 axons and synapses, considered as the “brain” of C. elegans. We hypothesize that the kinetochore proteins facilitate nerve ring assembly by promoting the proper formation of axon bundles. Starting from this unique angle, we aim to understand how the microtubule cytoskeleton integrates distinct molecular machinery to build and regenerate 3 dimensional neuronal circuits in C. elegans. Our goal is to define the function of the kinetochore proteins in building the nerve ring; 2) build a functional map of microtubule cytoskeleton during nerve ring assembly by addressing the function of non-kinetochore microtubule factors; 3) investigate how kinetochore proteins build and maintain neuronal network by addressing its role in dendritic branching and regeneration.

Role of microtubule cytoskeleton in building and regenerating the neural connectome

Nerve ring assembly in C.elegans
A. The C.elegans head nervous system in L1 larvae (PH marks the membrane and histone the cell body). The axon bundle in the nerve ring is between white arrowheads (scale 10 μm).
B. Schematic of KMN network: Mis-12 interface (red) with the centromere, Ndc80 (purple) binds the microtubule and Knl1 (blue) functions as a scaffold.
C. Structure of nerve ring

Selected Publications
The expression of individual genes is controlled at the levels of mRNA transcription and also post-transcriptionally, by processes such as splicing, localization, modification or editing, and degradation. To gain a mechanistic understanding of these processes it is important to understand the interactions between the individual players, including both protein and nucleic acid components, at the molecular level. We have used structural approaches to tackle mechanistic questions about how protein-RNA interactions can control RNA maturation and RNA editing and how transcriptional repressors are recruited to methylated DNA. By combining structural studies with biochemical, biophysical and cell-based functional assays we can gain powerful insights into these molecular processes.

We have contributed to the understanding of the molecular basis for the neurological disease known as Rett syndrome, which is caused by mutations in the DNA binding protein MeCP2. In collaboration with Adrian Bird’s lab, we showed that a cluster of Rett syndrome associated missense mutations on MeCP2 are part of a motif that binds to a component of a nuclear co-repressor complex. This interaction supports a model where MeCP2 acts a bridge between methylated DNA and complexes that drive gene repression.

Recently, we solved a crystal structure of a yeast RNA binding protein, Ssd1, that is important in cell wall biogenesis. It is thought that Ssd1 functions by repressing translation of cognate transcripts. Using CRAC, we found that Ssd1 binds to specific sequences in the 5’UTRs of a small set of transcripts, several of which encode proteins required for cell wall biogenesis. The structure of Ssd1 shows that it has a classical fold of an RNase II family nucl ease. However, RNA degradation activity has been lost by two mechanisms. First, the catalytic residues have been altered during evolution. Second, a channel that, in active enzymes, allows RNA substrates to funnel into the active site has been blocked. We propose that Ssd1 has evolved a new RNA interacting surface.

A domain overview of Ssd1 is shown along with the crystal structure with domains marked in blue, cyan, green and pink. The Ssd1-specific insert is shown in the domain overview and structure in orange. The yellow lollipops are phosphorylation sites. Two segments of the Ssd1 structure are shown in black – these block the active site funnel, as can be seen by comparison with the structure of DIS3L2 (left), where RNA is bound.

**Selected Publications**


Structural biology of macromolecular complexes in RNA metabolism and transcriptional silencing

![Structural diagram](image)
In 2020/21, our research focused on structural transformations during mitotic chromosome formation, epigenetic regulation of kinetochore assembly/stability and the role of the chromosomal passenger complex (CPC) in regulating cell division. We spent most of 2020/21 at home analysing data, working on manuscripts, chewing fingernails and attending Zoom Journal Club and Lab Meetings (other brands are available, but we don’t use them). The Zoom format may persist after restrictions ease, as we have become quite comfortable with it.

Bill’s PRF renewal application was submitted 1 week before Scotland went into lockdown. Painstaking practice for written responses to the grants panel and subsequent negotiations with Wellcome about the final award were ultimately rewarded with success, providing a glimmer of light in this dark period. Bill and Elisa also spent many hours preparing for our return to the lab in July. For the foreseeable future we are limited to 5 people in the main lab at any one time. Thus, progress is impeded, but not completely blocked.

Underpinning most of our studies is Kumiko’s chemical-genetic system for obtaining highly synchronous entry of cultured cells into mitosis. We can now do biochemical analysis on processes that could only previously be studied by live-cell microscopy.

• We are writing up a comprehensive study of protein-DNA transactions during mitotic entry. This paper focuses on disassembly of the nucleus during very early prophase. This turned out to be unexpectedly interesting, in part because dramatic changes occur in the nucleolus long before any condensation of the DNA can be seen (see Figure).

• We are also preparing to publish our multidisciplinary study of interactions between major chromosome scaffold proteins during mitotic chromosome formation, a collaboration with the groups of Job Dekker, Leonid Mirny and Anton Goloborodko. We do the genetics, cell biology and imaging. They do Hi-C and polymer modelling, respectively.

• We continue to study the enigmatic and perplexing protein Ki-67 and the RNA/protein-rich mitotic chromosome periphery compartment (MCPC). Preliminary studies reveal that Ki-67 still has surprises in store for us and indicate that components of the mysterious MCPC may be required for accurate chromosome segregation.

• Our work on mitotic chromosome segregation focuses on the formation of human artificial chromosomes (HACs). We discovered that major genome scrambling similar to chromothripsis often occurs at an early stage in HAC formation. Ongoing experiments aim to understand these early steps in HAC formation, and hopefully minimize the rearrangements. This is critical for planned efforts to construct synthetic human chromosomes which would be futile if chromosomes assembled at great effort and cost were scrambled when introduced into human cells.

Our work is supported by a Wellcome Principal Research Fellowship and by the Centre for Mammalian Synthetic Biology.

Selected Publications


J. CELL SCI. 133: JCS242610: PMID: 32576667; PMC7390644; DOI: 10.1242/jcs.242610.


PMID: 32889546; PMCID7754191; DOI: 10.1021/acssynbio.0c00326.

The role of non-histone proteins in chromosome structure and function during mitosis

A. tSNE map showing protein clusters that move away from DNA at various times during early mitosis.

B. The order of movement of the 6 clusters shown here. The left-most two clusters are highly enriched for proteins involved in pre-RNA processing and leave the DNA before chromatin condensation is visible. Many of these proteins end up on the mitotic chromosome periphery. Nuclear pore proteins and proteins of the inner nuclear envelope are also early to move away from DNA.
Our lab is interested in the organisation, establishment and maintenance of specialised chromatin states. Epigenetic transmission of centromere identity through many cell generations is required for proper centromere function and when perturbed can lead to genome instability and disease. We use Drosophila and human tissue culture cells as model organisms to address the following questions:

**What is the role of transcription at the centromere?**

Loading of CENP-A at the centromere occurs outside of S-phase and requires the removal of H3 "placeholder" nucleosomes. Transcription at centromeres has been linked to the deposition of new CENP-A, although the molecular mechanism is not understood. Using fast acting transcriptional inhibitors we demonstrate that centromeric transcription is required for loading of new dCENP-A and promoting dCENP-A transition from chromatin association to nucleosome incorporation (Bobkov et al., 2018). Unlike placeholder nucleosomes, previously deposited CENP-A is specifically retained by Spt6 both in human and Drosophila cells, identifying Spt6 as a CENP-A maintenance factor that ensures the stability of epigenetic centromere identity (Figure 1). We are currently investigating the molecular mechanism how some histones like CENP-A are maintained while others like H3.3 placeholders are evicted to preserve epigenetic centromere identity.

**How is the centromeric chromatin fiber organised?**

To map centromere proteins on the linear centromeric chromatin fiber, we have recently developed a novel approach where proteins-of-interest fused to ascorbate peroxidase (APEX) or Turbo ID leave a "footprint" on the underlying nucleosome through proximity-biotinylation. With this methodology we have described novel localization patterns of a subset of centromere proteins at human centromeres (Kyriacou and Heun, 2018). We are extending this approach to proteins localising to all layers of the centromere, including the inner centromere, CENP-A loading factors and the outer kinetochore.

**How does Su(var)2-10/PIAS contribute to heterochromatin organisation?**

The Su(var)2-10 gene has been originally identified in position-effect-variegation (PEV) assays designed to uncover proteins involved in heterochromatin formation. Cloning of the gene revealed its homology to the protein family SUMO E3-ligase PIAS (Protein Inhibitor of activated STAT), but how sumoylation promotes heterochromatin formation remains unknown. While PIAS does not localise to pericentric heterochromatin in somatic cells, it is enriched next to centromeres in early fly embryogenesis, suggesting a role in heterochromatin establishment. Specific depletion of PIAS as this point of development will shed light on the link between PIAS' SUMO targets and chromatin organisation.

---

**Selected Publications**


Specialization of chromosome segregation mechanisms in meiosis

Meiosis generates gametes with half the parental genome through two consecutive chromosome segregation events, meiosis I and meiosis II. Meiotic errors are prevalent in humans, accounting for frequent miscarriages, birth defects and infertility. Our vision is to elucidate the molecular basis of the adaptations that sort chromosomes into gametes during meiosis. We use budding and fission yeast as general discovery tools, and Xenopus and mouse oocytes to uncover meiotic mechanisms in vertebrates. Using patient-donated oocytes and ovarian tissue, we address the relevance of our findings for human fertility.

Structural and functional organisation of pericentromeres

During meiosis, chromosomes undergo remarkable remodelling for transmission into gametes. Chromosomes are broken and reciprocally exchanged in prophase, specifically cohered at centromeres during meiosis I and permanently separated at meiosis II. Chromosome morphogenesis begins before S phase, when cohesion establishment links sister chromatids coupled to DNA replication. The cohesin complex is a major definer of chromosome structure, establishing intra and inter-sister chromatid linkages and providing the context for spatial control of homolog interactions. Our group revealed how chromosomes are structured for their segregation during meiosis. We established the region around the centromere, called the pericentromere, as a paradigm for chromosomal domain organisation. We discovered that cohesin is loaded at the centromere and that the borders of pericentromeres are marked by convergent genes that trap cohesin. This folds pericentromeres into a looped structure that is important for accurate chromosome segregation. Our current work is aimed at understanding how pericentromere structure is adapted during meiosis to suppress meiotic recombination and to accommodate the co-segregation of sister chromatids during meiosis I.

Specialization of meiotic kinetochores

Kinetochores link centromeric nucleosomes to microtubules for chromosome segregation. Our goal is to understand how the kinetochore is adapted to perform its meiosis-specific functions in suppression of meiotic recombination, directing the co-segregation of sister chromatids during meiosis I, and maintaining linkages between sister chromatids until meiosis II. Recently, we defined the proteomic landscape of yeast kinetochores and centromeric chromatin during meiosis, revealing extensive remodelling during prophase and meiosis I. We are now addressing the mechanism of kinetochore remodelling, as well as its functional importance. In many organisms, sister kinetochores are fused in meiosis I, while a lack of fusion in human oocytes may account for susceptibility to segregation errors and fertility problems. Ongoing work in Xenopus, mouse and human oocytes aims to test this hypothesis.

Selected Publications


Regulation of gene expression by non-coding RNA and RNA modification

Genomic DNA methylation is erased (reprogramming) and reset (de novo methylation) during germ cell development. Our current model of MIWI2-piRNA directed TE methylation based on functionally defined interactions is presented.
Accurate segregation of chromosomal DNA is essential for life. An error in this process could result in cell death or aneuploidy. Furthermore, chromosome segregation in oocytes is error-prone in humans, and mis-segregation is a major cause of infertility, miscarriages and birth defects. Chromosome segregation in oocytes shares many similarities with those in somatic divisions, but also has notable differences. Distinct features of oocytes potentially hinder accurate chromosome segregation. They include (1) lack of centrosomes, the major microtubule nucleation centres in mitosis, (2) exceptionally large cell volume, and (3) cell cycle arrests at two stages. Oocytes are likely to have specific molecular mechanisms which mitigate negative impacts of these features, but little is known about how oocytes set up the chromosome segregation machinery. Defining the oocyte-specific mechanisms would be crucial to understand error-prone chromosome segregation in human oocytes. Furthermore, it may provide an insight into whether and how cancer cells might gain resistance to anti-mitotic drugs by activating these pathways.

To understand the molecular pathways which set up the chromosome segregation machinery in oocytes, we take advantage of Drosophila oocytes as a ‘discovery platform’ because of their similarity to mammalian oocytes and suitability for a genetics-led mechanistic analysis. In Drosophila oocytes, as in human oocytes, meiotic chromosomes form a compact cluster called the karyosome within the nucleus. Later, meiotic chromosomes assemble a bipolar spindle without centrosomes in the large volume of the cytoplasm, and establish bipolar attachment. We have identified genes/proteins and regulations specifically important for chromosome organisation and/or spindle formation in oocytes.

The synaptonemal complex assemblies during meiotic prophase I and assists faithful exchanges between homologous chromosomes, but how its assembly/disassembly is regulated remains to be understood. We showed how two major post-translational modifications, phosphorylation and ubiquitination, cooperate to promote synaptonemal complex assembly. We found that the ubiquitin ligase complex SCF is important for assembly and maintenance of the synaptonemal complex in female meiosis. This function of SCF is mediated by two substrate-recognizing F-box proteins, Slimb/Trcp and Fbxo42. SCF-Fbxo42 down-regulates the phosphatase subunit PP2A-B56, which is important for synaptonemal complex assembly and maintenance.

Selected Publications

Hiro Ohkura
Co-workers: Fiona Cullen, Jule Niekens, Charlotte Repton, Emiliya Taskova, Dan Toddle-Moore, Gera Pavlova, Xiang Wan

The meiotic spindle and chromosomes in oocytes

SCF-Fbxo42 promotes synaptonemal complex assembly by downregulating PP2A-B56.
A. Two substrate-recognition subunits of SCF, Slimb/Trcp and Fbxo42, are required for synaptonemal complex assembly
B. Fbxo42 co-immunoprecipitates with PP2A and 14-3-3 in addition to SCF subunits
C. Fbxo42 downregulates the PP2A-B56 subunit Wrd
D. A schematic model showing that SCF-Fbxo42 down-regulates PP2A-B56 to tip the balance of phosphorylation towards synaptonemal complex assembly. Barbosa et al (2021)
Selected Publications


Structural biology has made amazing advances on providing us models of proteins alone or in complexes by technologies such as crystallography, electron microscopy or nuclear magnetic resonance spectroscopy. These methods continue to advance yet fail mostly on requiring proteins to be extracted from their native environments, a process that many proteins do not honour with keeping their native structure. We are one of the pioneers of developing crosslinking mass spectrometry as an alternative approach. This sees proteins crosslinked in their native environment and the sites of crosslinking then being determined by mass spectrometry and data analysis. We are optimising many steps of this process: choice of crosslinking reagents, protein-to-reagent ratio, digestion conditions, sample preparation, mass spectrometric acquisition, peak picking, data base construction. We are also further developing our search software xiSEARCH and integrated data visualisation tool xiVIEW. We fully embrace ideas of open sharing our insights and tools by making use of preprint repositories, providing our code via GitHub, submitting our data to public repositories and establishing field standards in close collaboration with key stakeholders such as HUPO PSI and EBI. The continuous progress on our tools has allowed us now to move into (simple) cells.

Structural biology performed inside cells can capture molecular machines in action within their native context. We developed an integrative in-cell structural approach using the genome-reduced human pathogen Mycoplasma pneumoniae. We combined whole-cell crosslinking mass spectrometry, cellular cryo-electron tomography (in collaboration with Julia Mahamid at the EMBL Heidelberg), and integrative modeling to determine an in-cell architecture of a transcribing and translating expressome at sub-nanometer resolution. The expressome comprises RNA polymerase (RNAP), the ribosome, and the transcription elongation factors NusG and NusA. We pinpointed NusA at the interface between a NusG-bound elongating RNAP and the ribosome, and propose it can mediate transcription-translation coupling. Translation inhibition dissociated the expressome, whereas transcription inhibition stalled and rearranged it. Thus, the active expressome architecture requires both translation and transcription elongation within the cell. This is in stark contrast to structures obtained by single particle cryoEM using reconstituted transcription-translation in E. coli, which see NusG and not NusA at the nexus of RNAP and ribosome. Future work will have to clarify whether this difference can be accounted for by working with different species or that working in cells captures a different state then working in a reconstituted system.
We are interested in two general areas related to cellular organisation: 1) regulation of cell polarity, under both normal and stress conditions, and 2) the molecular mechanisms underlying microtubule nucleation. In both areas we use fission yeast Schizosaccharomyces pombe as a model single-celled eukaryote. We combine classical and molecular genetic analysis with live-cell fluorescence microscopy, biochemistry, proteomics/phosphoproteomics, and structural biology methods.

Cell polarity in fission yeast is regulated by multiple internal cues that cooperate and compete with each other. The Rho-family GTPase Cdc42 and its associated regulators and effectors control the actin cytoskeleton and exocytosis. Microtubules provide an additional level of control, through the microtubule-associated protein Tea1 and its interactors. We have shown how the Tea1/microtubule system coordinates polarity regulation by a conventional Cdc42 guanine-nucleotide exchange factor, Scd1, with regulation by an unconventional exchange factor, Gef1. Our work has also led to the discovery of new cell-polarity regulators outside of the Cdc42- and Tea1/microtubule-based systems, and a new understanding of how the conserved NDR kinase Orb6 regulates cell polarity. A major current focus is on how the stress-activated kinase Sty1 (homolog of human p38 MAP kinase) regulates cell polarity; we are addressing this through large-scale phosphoproteomics and genetics approaches.

Microtubule nucleation depends on the γ-tubulin complex, a large multi-protein complex enriched at microtubule organising centres such as the centrosome. Many aspects of γ-tubulin complex regulation remain a mystery. We discovered the fission yeast proteins Mto1 and Mto2, which form an oligomeric “Mto1/2 complex”. The Mto1/2 complex targets the γ-tubulin ring complex to different sites in the cell and also activates γ-tubulin complex. Mutations in the human homolog of Mto1 lead to the brain disease microcephaly. Our current work involves understanding the mechanism of γ-tubulin complex activation by the Mto1/2 complex, using yeast genetics, microscopy, and biochemistry approaches. In recent work we have reconstituted multi-protein complex-dependent microtubule nucleation in vitro using purified proteins, and we have characterised elements of functional nucleation complexes through cross-linking mass spectrometry as well as X-ray crystallography. We have also developed new methods to interrogate protein-protein interactions in complex “solid-phase” subcellular structures in vivo, and we have used these to investigate how Mto1/2 complex is localised to nuclear pores.

In all of our work we adopt and develop new tools and techniques as necessary to address the biological questions of interest.

Selected Publications


Cell polarity and cytoskeletal organisation

A. Global phosphoproteomics after inhibition of NDR kinase Orb6 in vivo. Many phosphorylation sites with decreased phosphorylation after Orb6 inhibition match the NDR consensus. Phosphosite mutation of Sec3 (component of exocyst complex) impairs exocytosis and cell separation after cytokinesis.

B. Model for docking Mto1/2 microtubule nucleation complex at the nuclear pore. Mto1 mimics a nuclear export cargo but uses this for docking at the pore, not for nuclear export.

C. Negative-stain electron microscopy of reconstituted fission yeast γ-tubulin ring complex.

Co-workers: Tanyu Dudnakova, Ye Dee Tay, James Le Cornu, Ankita Gupta, Adam Kovac, Ana Rodriguez, Monique Scott

Kenneth E. Sawin
RNA-binding proteins (RBPs) have important functions at all steps in gene expression, including transcription, RNA processing and mRNA translation. Defects in RBPs underpin many genetic diseases, while responses to environmental stress are frequently mediated by altered RNA-protein interactions.

We examined global RBP dynamics in Saccharomyces cerevisiae in response to stress, using our recently developed technique of total RNA-associated protein purification (TRAPP). Stresses induced very rapid remodeling of the RNA-protein interactome, without corresponding changes in RBP abundance (Bresson et al., 2020). A set of "scanning" translation initiation factors (eIF4A, eIF4B, and Ded1), were remarkably rapidly evicted from mRNAs (<30 sec after glucose withdrawal) driving translation shutdown (Fig). Selective mRNA 5'-degradation by the exonuclease Xrn1 was seen following heat shock, particularly for translation-related factors, reinforcing translational inhibition. Notably, these responses are distinct from previously characterized pathways for stress-induced translation inhibition.

An outstanding question in nuclear RNA quality control is how "defective" RNAs are identified and targeted for degradation. During surveillance, the RNA exosome functions together with the TRAMP complexes (Delan-Forino et al., 2020). These include the DEAD-box RNA helicase Mtr4 together with an RBP (Air1 or Air2) and a poly(A) polymerase (Trf4 or Trf5). TRAMP acts to make substrates more susceptible to degradation, by addition of a single-stranded "tail," and also targets them to the exosome. Combining biochemistry, genetics and genomics, we identified three distinct TRAMP complexes formed in yeast, which preferentially assemble on different classes of transcripts. Surprisingly, the poly(A) polymerases Trf4 and Trf5 emerged as crucial in TRAMP targeting and recruitment.

Transcription elongation rates are important for many aspects of RNA processing, defining the "window of opportunity" for interaction and folding. However, sequence-specific regulation of elongation rates is poorly understood. Notably, elongation by RNA polymerases is only moderately processive, being based on a "Brownian Ratchet" rather than an energy-driven mechanism. To analyse RNA polymerase I elongation in S. cerevisiae (Turowski et al., 2020) we combined in vivo RNAPI profiling, in vitro biochemical analyses and a quantitative, mechanistic model of transcription elongation. Unexpectedly, these revealed that folding of the nascent pre-rRNA close to the transcribing polymerase has a major effect on the elongation rate, with a modest contribution from the stability of the RNA-DNA duplex in the active site. RNAPI from S. pombe was similarly sensitive to transcript folding, as were S. cerevisiae RNAPII and RNAPIII. For RNAPII, unstructured RNA, which favours slowed elongation, was associated with faster cotranscriptional splicing and proximal splice site usage, indicating regulatory significance for transcript folding.

**Selected Publications**

The overarching goal of research in our lab is to elucidate how histone modifications regulate gene expression. We are keen to understand how different histone modifiers and readers interact to establish complex regulatory systems that control development and cause disease if misregulated. We are taking a multidisciplinary approach to tackle these questions, combining biochemistry with proteomic, genomic, cell-biological, imaging-based, and systems biology-inspired techniques.

We aim to clarify how Polycomb and trithorax group proteins regulate expression of developmental genes in embryonic stem cells (ESCs). Bivalent domains harbour a distinctive histone modification signature featuring both the active histone H3 lysine 4 dimethylation (H3K4me3) mark and the repressive H3K27me3 mark. They have been suggested to maintain developmental genes in a poised state, allowing for timely expression upon differentiation while maintaining repression in ESCs. Bivalent nucleosomes adopt a previously unknown asymmetric conformation, carrying the active and repressive mark on opposite copies of histone H3. However, it has remained unclear how bivalent domains function to poise genes for expression in ESCs and whether they are essential for proper ESC differentiation and embryonic development.

To address these questions mechanistically, we performed pulldown experiments with recombinant asymmetric, bivalent nucleosomes and ESC nuclear extract (Figure 1A). We found that bivalent nucleosomes recruit repressive H3K27me3 binders but not activating H3K4me3 binders, despite the presence of H3K4me3 (Figure 1B), both in vitro and in ESCs. Moreover, we have discovered readers that specifically recognise the bivalent histone acetyltransferase complex KAT6B (MORF) and the histone variant H2A.Z chaperone complex SRCAP (Figure 1C). To determine the role of these novel bivalent binders in regulation of developmental genes, we performed knockout of KAT6B. Loss of KAT6B diminishes neuronal differentiation, indicating that bivalency-specific readers are critical for proper ESC differentiation.

Our findings thus suggest a model where bivalent nucleosomes drive poising by setting up a repressed but plastic state at developmental promoters through recruitment of repressive and bivalent binders and exclusion of activators (Figure 1D). We are now testing this model by examining how repressive and bivalent readers such as PRC2 and KAT6B cooperate to maintain a repressed but plastic state in ESCs while allowing for activation of developmentally regulated genes upon differentiation.

**Selected Publications**


**Molecular Mechanisms of Epigenetic Gene Regulation**

A. Outline of the nucleosome pulldown approach used to identify binding proteins of asymmetric bivalent nucleosomes.

B. H3K27me3 but not H3K4me3 readers are enriched with asymmetric bivalent nucleosomes. Additionally, novel, bivalency-specific binders are recruited (highlighted in C).

C. Model illustrating how bivalent nucleosomes support establishment of a poised state.
Research
To maintain their genomic integrity, eukaryotic cells must replicate their DNA faithfully and distribute it equally to the daughter cells. Mitotic defects lead to aneuploidy and cancer. This indicates that the mitotic mechanisms that are in place to allow faithful division have been compromised. The segregation of chromosomes is mediated by polarized and highly dynamic filaments, termed microtubules. Microtubules depend on motor proteins to assemble into a spindle and segregate chromosomes. These motors play key roles in cytoskeletal organization during cell division but also in cell migration, polarity, and axonal and cytoplasmic transport. However, the reductionist approach to studying these motors in isolation is not sufficient to understand their function in the cellular context. It remains unclear how the activities of individual motors and their interacting regulatory networks cooperate to generate physiological cellular function such as chromosome segregation. We aim to define how kinesin motors are modulated by their cargos to provide a specific output, and how the coordinated activities of kinesin motors are greater than the sum of their individual activities in vitro and in human cells.

Kinetochore and motors
CENP-E is a huge motor (730 kDa), recruited to unattached kinetochores. CENP-E moves kinetochores along microtubules to facilitate chromosome alignment. How CENP-E associates with the kinetochore, how human CENP-E is activated to walk on microtubules and how CENP-E motor ensembles are currently not known. Using a non-biased approach, we recently found the C-terminal kinetochore-targeting region of CENP-E interacts with BubR1, amongst other proteins in nocodazole-arrested mitotic cell extracts. We have defined the molecular determinants that specify the interaction between BubR1 and CENP-E. The basic C-terminal helix of BubR1 is necessary but not sufficient for CENP-E interaction, while a minimal key acidic patch on the kinetochore-targeting domain of CENP-E, is also essential. We then demonstrated that BubR1 is required for the recruitment of CENP-E to kinetochores to facilitate chromosome alignment. In collaboration with the Gruneberg lab, University of Oxford, we showed this BubR1-CENP-E axis is critical to align chromosomes that have failed to congress through other pathways and recapitulates the major known function of CENP-E. Overall, our current studies define the molecular basis and the function for CENP-E recruitment to BubR1 at kinetochores during mammalian mitosis. Our future work now focuses on what is the basis and function for the 2 distinct recruitment pathways of CENP-E to kinetochores and defining the activation mechanism of human CENP-E to ensure faithful mitosis.

Mitotic motors and microtubule dynamics
Our lab has made new discoveries on the mechanism of mitotic microtubule dynein-polymerizations for two families recently published: the Kinesin-8 and the Kinesin-13 motors. Our current work now addresses how MCAK and Kinesin-8 motors cooperate to control microtubule length in mitosis.

Selected Publications

Microtubule motor cooperation in cell division

A. Top, SEC analysis and elution profile for CENP-E

B. Thermodynamics of BubR1

C. CENP-E interaction determined by isothermal titration calorimetry. The y-axis indicates lmc/mol of injectant. The dissociation constant (Kd) between BubR1 and CENP-E was determined to be 3.18 ± 0.90 nM.

D. Sequence alignment of the C-terminus of human BubR1 with mouse and Xenopus BubR1, and human Bub1. Boxed red and blue are the conserved and similar amino acids across all 4 proteins, respectively. Amino acids in red are those with conserved properties in at least 3 sequences. The sequence necessary for BubR1 binding on CENP-E is highlighted in orange.

E. Representative immunofluorescence images of HeLa cells treated with BubR1 siRNA and induced to express GFP-BubR1 WT and GFP-BubR1, stained with CENP-E, CENP-C and Hoechst after treatment with MG132 for 2.5 hrs. Scale bar: 10 μm.

F. Scatter plot showing CENP-E intensity relative to GFP-BubR1 at individual kinetochores plotted as grey circles, with mean and standard deviation represented by black lines.

Graph showing percentage of cells with at least 1 misaligned chromosome for BubR1-depleted cells induced to express GFP-BubR1, or without induction. Error bars represent standard deviation. *** Indicating P < 0.0001.
We are interested in understanding how epigenetic marks are placed, read and interpreted on chromatin. Chromatin becomes decorated with a variety of chemical tags or epigenetic marks to control the myriad of DNA-related processes in the cell. Epigenetic modifications are initially deposited by writer enzymes. These are then read and interpreted in a co-operative manner by effector proteins. Epigenetic marks can also be removed by eraser proteins resetting the system (Figure 1A). We look at this process in the test tube by creating modified chromatin using chemical biology and biochemical methods. We then use our defined modified chromatin to study individual nucleosome-chromatin protein complexes using single-particle cryo-electron microscopy (cryo-EM), Biochemical, Biophysical and Cell Biology approaches. We are particularly interested in understanding how DNA damage repair and DNA methylation pathways are orchestrated by epigenetically-modified nucleosomes.

1. How is DNA Methylation guided by chromatin?
DNA methylation is a common epigenetic mark that is often associated with turning off genes and compacting DNA. Other epigenetic marks have the power to regulate DNA methylation, controlling when and where DNA methylation is placed on DNA, but we do not understand how this works. We are rebuilding the DNA methylation machinery within chromatin to help us answer this question.

DNA methylation is a highly regulated process, so by looking at the structure of the methylation machinery and the modified nucleosomes we hope to understand how methylation is targeted at specific times and to specific sites on DNA, hopefully helping us to understand how this process can become faulty leading to disease.

2. How do post-translational modifications foster DNA repair?
DNA is under constant attack, which can cause unwanted genetic mutations and cancer. Luckily our cells have a host of DNA repair proteins, which help to fix most of the damage. These highly efficient repair proteins are recruited to sites of damage by recognition of DNA damage-specific marks on chromatin. We are hoping to understand how DNA damage is signalled on chromatin and how this leads to correct repair.

Our recent work has shown that multiple DNA repair proteins interact multivalently with the nucleosome, commonly interacting with a conserved region called the acidic patch (Figure 1B). Through our in vitro studies we showed that the negatively charged surface of the nucleosome acidic patch is essential for binding of the listed DNA damage proteins. Mapping of the interaction regions and mutation has shown these are mediated by electrostatic interactions, typically through a highly conserved arginine anchor.

Selected Publications
There is no such thing as a standard year in Public Engagement, but 2020 will surely stand out as a time of particular challenge, but also new opportunities. With the pandemic putting a stop to public festivals and in-person events, it was time to get even more innovative with our approaches. From online to outdoors, post-out packs to comic strips, 2020 has required creativity to engage with our partners, new and old alike.

Having hosted the Edinburgh Care for Carers for a centre visit in February (figure 1), we were delighted to continue collaborating with this group throughout 2020. With the help of clip-on microscopes for phones, we created a post-out version of our popular workshop ‘Life Through Lens’. Whilst the booklet was self-supporting, our researchers enjoyed meeting the group online and viewing the microscopic images they had taken around their homes (figure 2).

In 2020 we were also introduced to a Young Carers group in Edinburgh and worked with them to develop a self-led, outdoor session titled ‘The Cell-ebrating nature scavenger hunt’. This involves participants photographing and collecting interesting biological items during walks (leaves, sticks, etc.) which they can then send back to the centre for feedback and microscopic analysis. We hope to roll this out with more youth groups in 2021.

Virtual gatherings such as Explorathon and the Midlothian Science Festival allowed our researchers to practice their online engagement skills (figure 3) including an exciting new international session ‘Research Around the World’. School and public audiences met Nepalese research colleagues, along with our own WCB researchers, to discuss how research is a global endeavour but varies from country to country.

Social distancing didn’t stop us from encouraging our audiences to get hands on! Community groups got artistic at home with our cell themed colouring books, science crafts and bake-along stained glass cell biscuit Zoom calls (figure 4). The Haddington Girl Guides made a splash with water and household items to learn about the scientific discovery process in our new ‘Researching research’ session, which has now been requested by various other units in East Lothian.

In 2020 we were delighted to support the Marston lab in winning a ScotPen Wellcome Engagement Award which enables us to link researchers and IVF patients through the production of glass art works. Public Engagement is becoming more integrated across the whole centre with PE Champions being recruited from each lab and our first whole centre project in the form of our WCB comic book. Artist in residence, Neil Bratchpiece, will be working with each lab to make an engaging comic strip about their research (figure 5).

2021 is set to be a busy year for us with high demand for our new socially distanced offerings but also plan afoot for in-person activities as soon as restrictions are lifted.
List of Groups

Robin Altshire
Wellcome Principal Research Fellow

Tatsiana Auczyńska
Wellcome Research Associate

Roberta Carlini
MRC Research Associate

Tadhg Davin
BBSRC EASTBIO Graduate Student

Andreas Fellas
ICM Graduate Student

Elisabeth Gabardiel
Wellcome Research Associate

Dominik Hoelper
EMBO Long Term Fellow

Marcel Latos
Wellcome Research Associate

Nitinie London
University of Grenoble CNRS

Sunit Nahta
Visiting Student

Alison Pidoux
Wellcome Research Associate

Severina Pociunaite
Utrecht University Erasmus+ MSc Student

Desislava Stanoeva
MRC Research Associate

Manu Shukla
Darwin Trust Graduate Student

Imliyaz Yaseen
Darwin Trust Graduate Student

A. Jeyaprakash Arulanandam
Wellcome Senior Research Fellow

Maria Alba Abad
Wellcome Research Associate

Fernandez
Darwin Trust Graduate Student

Amaa Al-Murtadha
ICM PhD student

Lana Buzuk
Wellcome Research Associate

Carla Chiodi
Wellcome Research Associate

Thomas Davies*
ICM Graduate Student

Kashyap Chhatbar
ERC Bioinformatician

Bethan Medina-Pritchard
Wellcome Research Assistant

Paula Sotelo Parrilla
Principal's Career Development Graduate student

Raphael Pantier
ERC Research Associate

Dhanya Cheerambathur
Sir Henry Dale Fellow

Beatrice
Wellcome Research Assistant

Malte Green
Wellcome Research Technician

Cameron Finlayson
Darwin Trust Ph.D. Student

Matthieu Flagebedzi
Wellcome ICM PhD Student

Charlotte de Cuencin
Visiting Masters Student

Atlanta Cook
Wellcome Senior Research Fellow

Laura Croenen
Cunningham Trust Graduate Student

James Le Cornu
ICM Graduate Student

Azzurra Codeino
Wellcome Graduate Student

Atika Al Haisani
Darwin Trust Graduate Student

Uma Jayachandran
Wellcome Research Associate

Pragya Srivatsava
Darwin Trust Graduate Student

Reshma Thakur
SEBR Overseas Postdoctoral Fellow

* joint ICM PhD student with Prof. Kevin Hardwick

Adrian Bird
Wellcome Investigator in Science

Kashyap Chhatbar
SRC Research Associate

Sarah Giuliani
ERC Visiting student

Jacek Guy
Wellcome Research Associate

Mathew Lyst
SRC Research Associate

Baisakh Mondal
ERC Research Associate

Verdiana Steccanella
ERC Research Associate/Lab Manager

Katie Paton
Personal Assistant

Kati SokePortal
Primary’s Career Development Graduate student

Anna Haidar
Darwin Trust Graduate Student

Theresa Schöpp
Wellcome Graduate Student

Ansgar Zoch
DFG Postdoctoral Research Associate

Bill Earnshaw
Wellcome Principal Research Fellow

Mar Carmina
Wellcome Research Associate

Fernanda Cianeros
Wellcome Research Associate

Loisone Di Pompeo
ERC Ph.D. Student

Moonmoon Deb
Royal Society Newton Fellow

Natalia Kucharova
Wellcome Research Associate

Emma Peat
Lab Manager/Wellcome Research Associate

Bram Prevo
Sir Henry Wellcome Postdoctoral Fellow

Caillín Reid
Wellcome Research Associate

Lucy Remnant
Wellcome Research Associate

Kumiko Samejima
Wellcome Research Associate

Itaru Samejima
Wellcome Research Associate

Patrick Heun
Wellcome Senior Research Fellow

Mathilde Fabe
Darwin Trust Graduate Student

Meghan Frederick
Research Assistant

Emoke Geroz
Research Assistant

Meena Krishnan
Darwin Trust Graduate Student

Heiwei Ling Tan
EastBIO Graduate Student

Alessandro Striga
Wellcome Research Associate

George Yankson
Darwin Trust Graduate Student

Adele Marston
Wellcome Investigator in Science

Weronika Borek
Wellcome Postdoc

Chuanli Huang
Darwin Trust PhD student

Diana Kocakaplan
Wellcome Postdoc

Lori Koch
Carmegie Trust PhD student

Melanie Lim
Wellcome Postdoc

Vasso Makrantonis
BBSCR Postdoc

Lucia Massari
Wellcome Postdoc

Bettina Mihalas
Darwin Trust PhD student

Anuradha Mukherjee
EastBIO BBSCR PhD student

Meg Peyton-Jones
Wellcome Postdoc

Gerard Piper
Darwin Trust PhD student

Ola Pompa
Wellcome Postdoc

Xue (Bessie) Su
Darwin Trust PhD student

Aparna Vinod
Wellcome Postdoc

Mengli (Lily) Wang
Wellcome Postdoc

Dónal O‘Carroll
Wellcome Investigator in Science

Azurra Codino
Wellcome Graduate student

Azuru DePeace
Wellcome Graduate student

Hanna Fiedler
Wellcome Graduate student

Madeleine Heep
Wellcome Graduate student

Yuka Kabayama
Wellcome Postdoc

Gabriela Korzeci
Principal’s Career Development Fund

Louie Van De Lagemaat
Graduate student

Christopher Mapparly
Visiting Scientist

Pedro Moreira
Wellcome Trust Tissue Repair PhD Programme

Theresa Schöpp
Laboratory Manager

Ansgar Zoch
Wellcome Graduate student

41
List of Groups

Hiro Ohkura
Wellcome Investigator in Science
Fiona Cullen
Wellcome Research Associate
Julie Nieman
Darwin Trust Graduate Student
Charlotte Repton
BBiRC EASTBIO Graduate Student
Emiliya Taskova
BBiRC Research Technician
Dan Toddle-Moore
Wellcome Research Associate
Gera Pavlova
BBiRC Research Associate
Xiang Wan
Darwin Trust Graduate Student

Juri Rappasliber
Wellcome Senior Research Fellow
Colin Combe
Wellcome Research Associate
Georg Kustatscher
Wellcome Research Associate
Christos Spanos
Wellcome Research Associate
Juan Zhou
Wellcome Proteomics Data Analysis Manager

Kenneth E Sawin
Wellcome Investigator in Science
Tanya Dudnakova
Wellcome Research Associate
Ye Dee Tay
Wellcome Research Associate
James Le Cornu
ICM PhD student, joint with Cook Lab
Ankita Gupta
Darwin Trust PhD student
Adam Kovac
EASTBIO PhD student
Alia Rodriguez
Darwin Trust PhD student
Monique Scott
Darwin Trust PhD student

David Tollervey
Director, Wellcome Principal Research Fellow
Stefan Bresson
Wellcome Trust Research Associate
Aziz Shihab
Wellcome Trust Research Associate
Alessandra Felis
Wellcome Trust Research Associate
Laura Milligan
EASTBIO PhD student
Elisabet Piatalski
Wellcome Trust Research Associate
Nic Robertson
Wellcome Trust Research Associate
Emanuela Sarti
ECAT Fellow
Adam Stobie
SNF Research Fellow
Tomasz Turowski
Polish Academy Research Fellow

Philipp Volgt
Sir Henry Dale Fellow
Giulia Bartolomucci
ERC Research Technician
Rachael Burrows
Wellcome Research Assistant
Ethan Hills
Darwin Trust PhD Student
Amaka Ilgo
Wellcome Research Assistant
Simone Lenzi
ERC Research Technician
Reshma Nair
ERC Postdoctoral Research Associate
Thomas Sheahan
ERC Graduate Student
Deviree Valsakumar
Darwin Trust PhD Student
Marie Warburton
ERC Graduate Student

Julie Wellburn
Wellcome Senior Research Fellow
Thomas Attard
MRC Precision Medicine Student
Ben Craik
BBiRC DTP Graduate Student
Agata Glinka
Wellcome Postdoctoral researcher
Thibault Legal
Wellcome researcher technician/part-time
Haonan (Leo) Liu
Darwin Trust Graduate Student
Toni McHugh
Wellcome Trust Postdoctoral Researcher

Marcus D Wilson
Sir Henry Dale Fellow
Hayden Burdett
MRC Postdoctoral Researcher
Gillian Clifford
Research Assistant
Dhananjay Kumar
Darwin Trust Graduate Student
Maarten Tuylj
Cryo-EM Technologist
Hannah Wapnnaar
Wellcome Postdoctoral Researcher
James Watson
Wellcome ICM PhD student

Administration/Support Staff
Greg Anderson
Centre Laboratory Manager
Elizabeth Blackburn
EPPF Research Associate
Hywel Dunn-Davies
Bioinformatics Developer
Maria Fanourgikli
Science Communicator
Carolyn Fleming
Centre Administrative Assistant
Sarah Jane Judge
Centre Public Engagement Manager (temporary)
Sarah Kees-Keer
Centre Public Engagement Manager
David Kelly
Centre Optical Instrumentation Laboratory Manager
Toni McHugh
Centre Imaging Facility Assistant
Colin McLaren
Computing Support
Karen May
Centre Manager/Wellcome 4yr PhD
Programme Administrator (temporary)
Daniel Robertson
Bioinformatics Support Officer
Christine Struthers
PA to Adrian Bird/Centre Administrator
Karin Traill
Centre Manager/Wellcome 4yr PhD
Programme Administrator
Maarten Tuylj
Cryo-EM Technologist
Shaun Webb
Centre Bioinformatics Core Facility Manager

Technical support
Scott Macrae
Wash-up Media
Denise Atteck
Andrew Kerr
Margaret Martin
Donna Pratt

1190.5x595.3


International Scientific Advisory Board

Margaret Fuller
Department of Developmental Biology and Department of Genetics
Stanford University School of Medicine
291 Campus Drive, Li Ka Shing Building
Stanford, CA 94305-5101
USA

Iain Mattaj
Fondazione Human Technopole
Palazzo Italia
Via Cristina Belgioioso, 171
20157 Milan, Italy

Michael Rout
The Rockefeller University
1230 York Avenue
New York, NY 10021
USA

Elena Conti
Max-Planck-Institut für Biochemie
Structural Cell Biology Department
Am Klopferspitz 1B
D-82152 Martinsried
Germany