Title of Project:	State of the art sequencing and analytical approaches to unravel centromere organisation
Cell Mechanism Supervisor Name	Patrick Heun
Quantitative Supervisor Name	Chris Ponting

Summary of project

The centromere is a unique chromatin domain important for proper segregation of chromosomes during mitosis. In most organisms, the position of the centromere is determined epigenetically by the centromere-specific incorporation of the H3-variant CENP-A. Transcription at centromeres has been linked to the deposition of new CENP-A, although it is unclear whether transcription or the produced RNA is most important. We and others have recently found that several components of the transcriptional machinery including RNA Polymerase II (RNAPII) and centromere-associated RNA transcripts temporally coincide with CENP-A loading in mitosis to G1. While we have evidence that the transcriptional process itself is required for CENP-A deposition at the centromere, this does not rule out a role for the centromeric transcripts themselves. To start, we will purify the non-coding nascent centromeric RNA transcripts. With ultra-long read sequencing (ONT and/or PacBio) we will map these highly repetitive transcripts to their underlying DNA templates. We will then seek to dissect the roles of transcription and RNA transcripts by identifying transcriptional start sites, promoters, RNA Pol II binding patterns, transcription factor binding sites and RNA binding factors. Specifically targeting transcriptional repressors or RNases to centromeric loci will help to investigate the role of transcription in centromere inheritance and function. This work will be performed using Drosophila tissue culture cells as our model system.

What quantitative skills will the student acquire or develop during their PhD project?

Statistical tests; DNA/RNA read assembly; read mapping; differential expression. Python, R scripting. Data visualisation and analysis. Error handling. Data management. High performance computing.