Using a Nanopore Sequencer to determine chromosome specific telomeres lengths.

Elliot Kidd, Andrew Walley, Una Fairbrother

Abstract:

Telomere length (TL) is an interesting biomarker and is associated with a wide range of disorders. This may be due to direct or indirect alteration of gene expression. Direct effects may act through Telomere Position Effect – Over Long Distance (TPE-OLD) and indirectly through production of the telomeric repeat containing RNA (TERRA).

TPE-OLD describes where telomeric ends of the chromosomes loop over and bind to gene specific regulatory elements thus repressing their expression. As telomeres shorten, this chromatin loop fails, increasing expression of certain genes. Premature shortening could lead to chronic overexpression of specific genes. As each chromosome harbours different genes, individual TLs will have different effects.

To produce data on the length of single, specific Chromosomal telomeres, we are using nanopore sequencing to generate very long reads. We aim to generate a read which contains the entire telomere, as well as enough of the unique subtelomeric region to allow us to assign the telomere to a specific chromosome.

Our lab has designed a novel technology which attaches a short stretch of DNA (oligonucleotide) to the unique telomere 3' overhang. Acting as a unique sequence tag, found nowhere else in the genome, it marks out the telomeres ends to provide a

definitive end point to the read. The first steps require validating the attachment of the tag to the 3’ overhang, which we have successfully achieved. Our methodology can potentially overcome the limitations of current methods and provide TL data in a single cell and of a single chromosome end. This will further contribute to our understanding of TL in health and disease by uncovering whether specific chromosomes with their unique gene sequences are most vulnerable to TL attrition or elongation.