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Intended use of resource / data

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Title of resource
MinION FASTQ DNA Sequence Analysis
Authored by
When using this protocol, the following should be referenced: Samuel Armoo ¹ , Romain Derelle ² , Daniel Boakye ³ , Mike Y. Osei-Atweneboana ¹ , Charles Brockhouse ⁴ , John Colbourne ² . Adapted from Geneious Prime Software (https://www.geneious.com) user manual. ¹ Council for Scientific and Industrial Research – Water Research Institute, Accra, Ghana. ² University of Birmingham, Birmingham, UK ³ Noguchi Memorial Institute for Medical Research, Accra, Ghana ⁴ Creighton University, Omaha, NE, USA
DOI
N/A
Description
This protocol is preliminary analysis of FASTQ DNA sequence files produced by the MinION sequencer. Adapted from Geneious Prime Software (https://www.geneious.com) user manual
Intended use
Scientific research use and training purposes.
Restrictions on use
Content is not to be redistributed in the public domain (e.g. presentation, lecture, online or in publications).
Resource history

GN 26 MinION FASTQ DNA Sequence Analysis

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1.0 Introduction

The MinKNOW software of the MinION sequencer (Oxford Nanopore, UK) basecalls DNA reads in a Fast5 of Fastq format. Our analysis in this project has been based on the Fastq format. Details below.

2.0 Mapping of FASTQ Reads

Reads were mapped to the reference *Simulium sp* whole mitochondrial reads provided by Professor Charles Brockhouse, who is the coordinator of the Simulium Genomics Project. Mapping of reads were performed using the Geneious Prime Software (<https://www.geneious.com>). A graphical output of mapping showed reads peaking at the Cas9 restriction site on the mitochondrial genome (Figure 2.1).

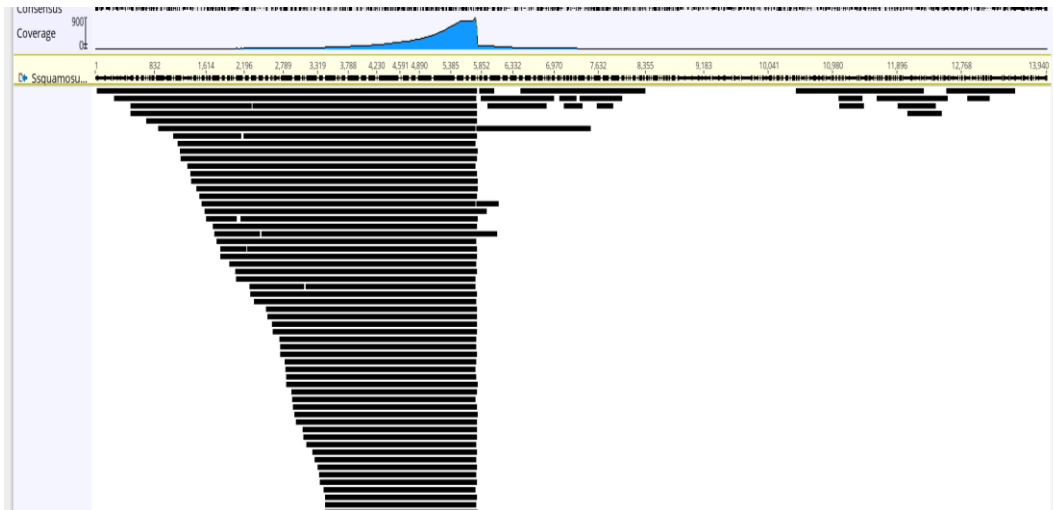


Figure 2.1: Graphical output of mapping the MinION Fastq reads to a reference *Simulium yahense* mitochondrial genome. Reads peak at the restriction site of the mitochondrial genome.

3.0 Assessing DNA Sequence Read Length

MinION sequence reads that mapped to the reference mitochondrial DNA genome, ranged from 200 to 12,400 base pairs in size. In effect, the almost the whole 15,000 bp mitochondrial genome of the blackfly was sequenced in a single sequence read.

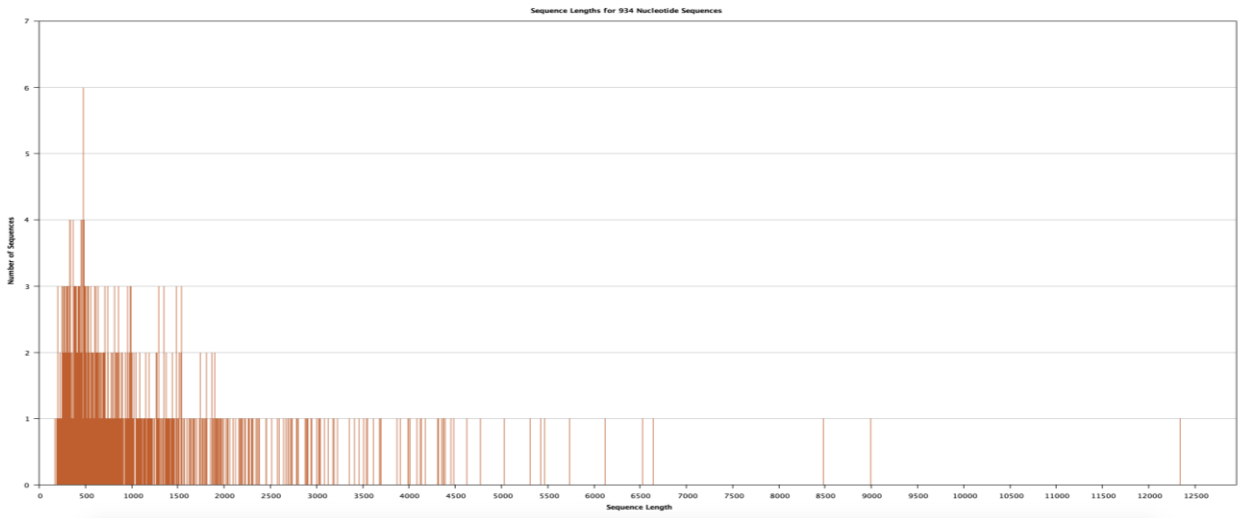


Figure 3.1: Chart showing the range of read length sizes from the MinION sequencer using the Cas9 targeted sequencing approach. The chart is produced by the Geneious Prime Software (<https://www.geneious.com>).

4.0 Phylogenomics

Mapped *Simulium sp.* mitochondrial DNA were included in a phylogenomic analysis that included several *Simulium* reference genomes from the Simulium Genomics Project, and a GeneBank search. The analysis was performed using the Geneious Prime Software (<https://www.geneious.com>), and the output showed that the MinION reads met phylogeographic as well as bioclimatic expectations (Figure 4.1).

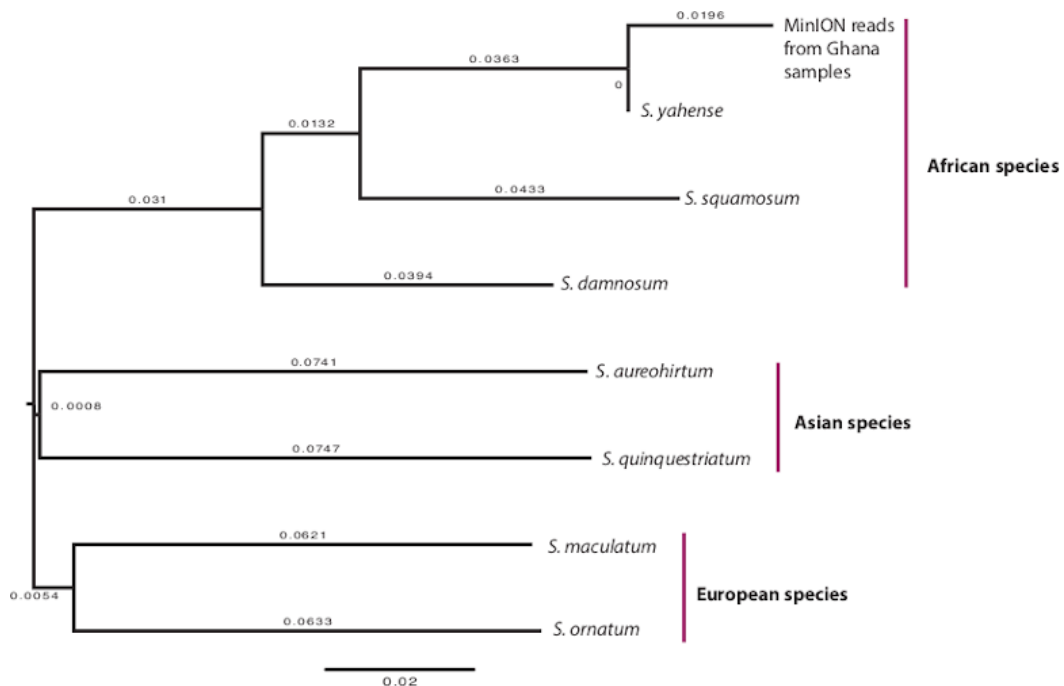


Figure 4.1: Phylogenomic analysis of MinION sequence reads from *Simulium* samples collected from Ghana together with reference whole mitochondrial genomes from the Simulium Genomics Project, and a GeneBank search showed samples from Ghana clustering together with *Simulium sp.* known to be prevalent in Ghana and Africa, whilst European and Asian species clustered differently. The analysis was performed with the Geneious Prime Software (<https://www.geneious.com>).