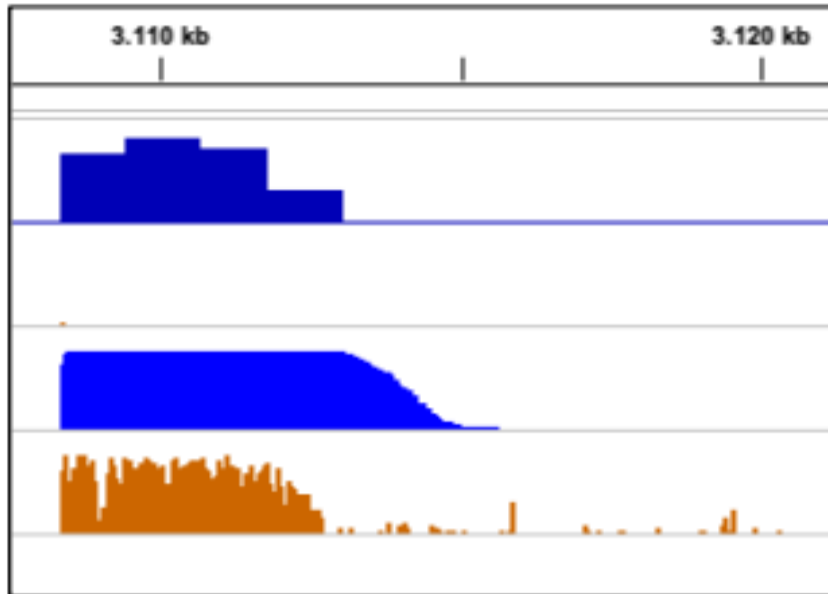
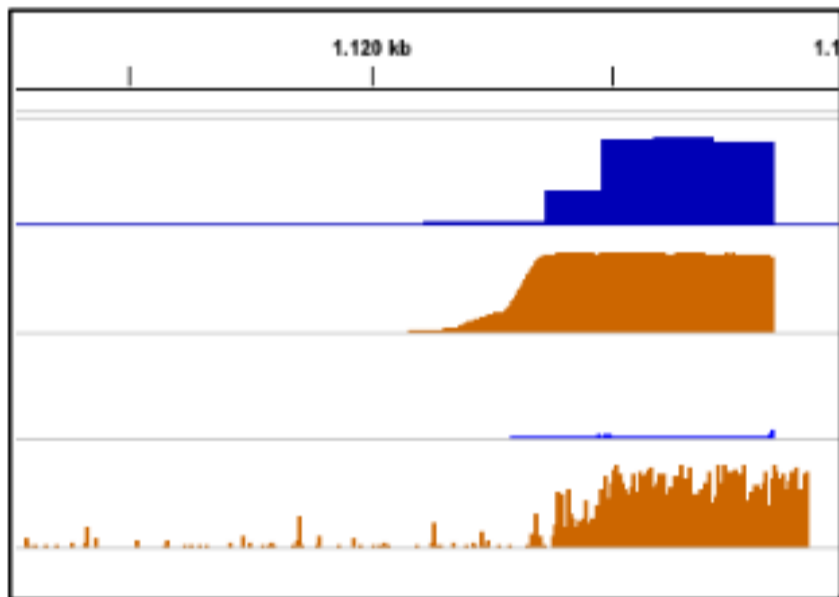


Fig. S1. Nanopore sequencing reads after base calling.

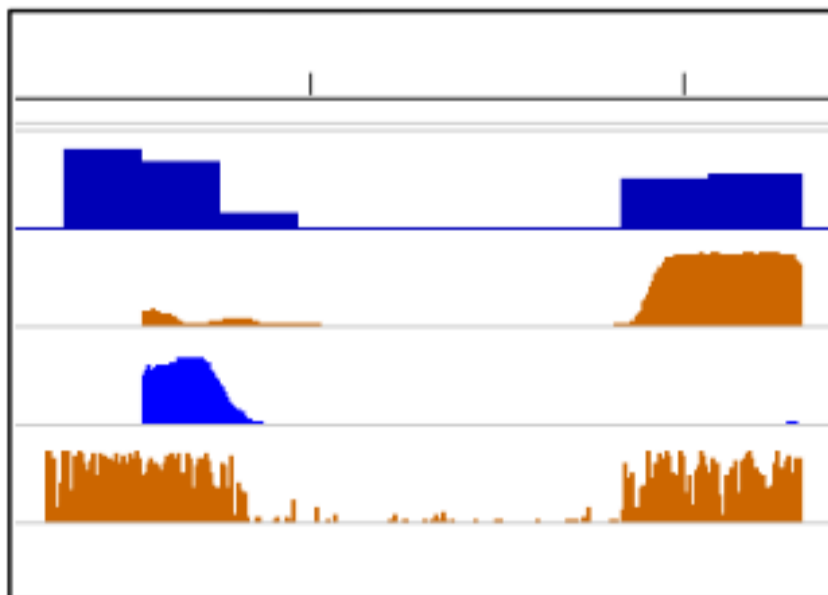
### A) Rightward moving fork



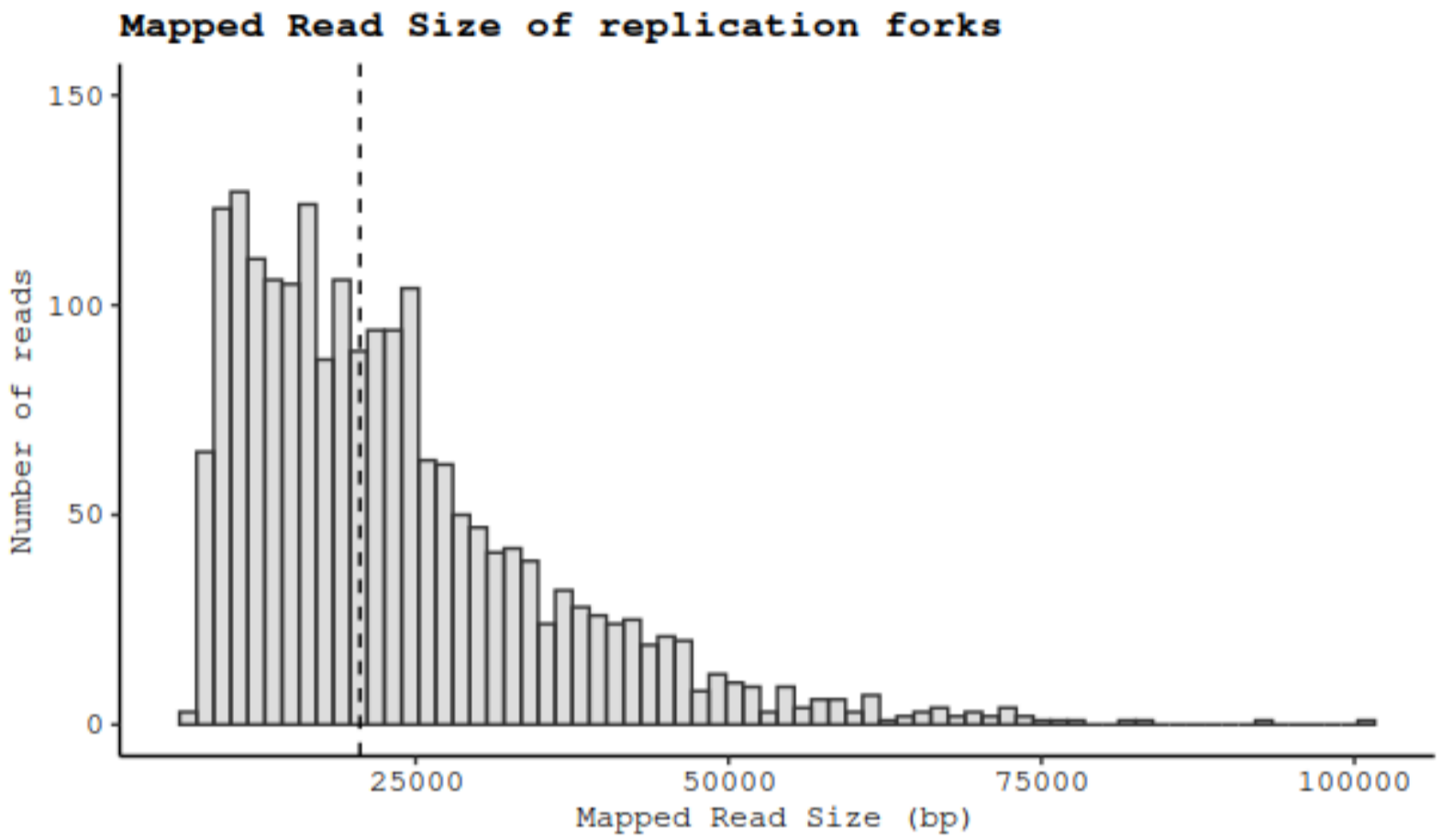
### B) Leftward moving fork



### C) Bidirectional moving forks



**Fig. S2. Detection of BrdU-incorporated nanopore sequenced molecules and movement of replication forks analyzed by DNAscent.** Bedgraphs visualized in IGV showing the probability of BrdU called at each thymidine position for three selected reads (**A-C**). Each read is represented by a group of four tracks: profile of BrdU detection (upper track), probability of a leftward moving fork (upper-middle track), probability of a rightward moving fork (lower-middle track), probability of BrdU at each thymidine position (lower track) from DNAscent forksense.



**Fig. S3. Mapped read size of replication forks.**

**Table S1. BrdU positive reads analyzed by DNAscent.** Regions containing fork direction were plotted and matched with the annotated genome to analyze replication fork and transcription directions and evaluate potential HoRT collisions.

[Click here to download Table S1](#)

**Table S2. MinION sequencing data and DNAscent analysis summary.**

Experiment	MinION runs	MinION reads	Estimated bases (Gb)	Average read length (bp)	Mapped read (%)	Positive BrdU molecules (probability $\geq 70\%$ )	%Positive reads	Input DNA per run ( $\mu\text{g}$ )
Giardia intestinalis trophozoites negative control	1	29770	0.58	41220	91.23	9	0.09	5
Giardia intestinalis trophozoites BrdU 300UMa	1	90130	1.3	38110	94.94	286	0.32	5
Giardia intestinalis trophozoites BrdU 300UMb	2	455750	8.86	30460		3377	0.74	5