High throughput single-cell genome sequencing gives insights into the generation and evolution of mosaic aneuploidy in *Leishmania donovani*

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Keywords: *Leishmania*, single-cell genomics, mosaic aneuploidy

Abstract

Aneuploidy, i.e., an imbalance in the copy number of chromosomes in a cell, is a ubiquitous feature of *Leishmania*, a protozoan parasite responsible for the group of diseases known as leishmaniasis. In these organisms, chromosome copy number (CCN) alterations represent an adaptive mechanism, modulating gene expression and possibly impacting phenotypes. Moreover, variations in CCN within single parasites in clonal populations was previously observed in a small subset of chromosomes using fluorescence hybridization methods. This phenomenon, termed mosaic aneuploidy (MA), have important evolutionary and functional implications which remains under-explored, as current methods are not capable of revealing the complete karyotype of individual *Leishmania* cells. To overcome this limitation, we applied and validated a high throughput single-cell genome sequencing method to study for the first time the extent and dynamics of whole karyotype heterogeneity in two *Leishmania* clonal populations representing different stages of MA evolution in vitro. In these two populations, we identified 117 and 208 different karyotypes co-existing among 2378 and 1516 promastigotes respectively. We observed that drastic changes in karyotypes quickly emerge in a population stemming from an almost euploid founder cell. The presence of polyploid cells at early stages suggests that these initial drastic changes may be generated by polyploidization/hybridization followed by assorted ploidy reduction, as has been observed in yeasts. During further stages of expansion, MA increases by moderate and gradual karyotypic alterations. We also observed that MA usually affected a defined subset of chromosomes, of which some display an enrichment in snoRNA genes which could represent an adaptative benefit to the amplification of these chromosomes. Our data provide the first complete characterization of MA in *Leishmania* and pave the way for further functional studies.

Acknowledgments

This study received financial support from the Flemish Ministry of Science and Innovation (SOFI Grant MADLEI) and the Flemish Fund for Scientific Research (FWO, post-doctoral grant to FVdB). A.Y. was recipient of a grant from the Agence Nationale de la Recherche (ANR) within the frame of the "Investissements d'avenir" programme (ANR 11-LABX-0024-01 "ParaFrap").