A panel of microsatellite and minisatellite markers for the characterisation of field isolates of Theileria parva.

Abstract

Mini- and microsatellite sequences show high levels of variation and therefore provide excellent tools for both the genotyping and population genetic analysis of parasites. Herein we describe the identification of a panel of 11 polymorphic microsatellites and 49 polymorphic minisatellites of the protozoan haemoparasite Theileria parva. The PCR products were run on high resolution Spreadex gels on which the alleles were identified and sized. The sequences of the mini- and microsatellites were distributed across the four chromosomes with 16 on chromosome 1, 12 on chromosome 2, 14 on chromosome 3 and 18 on chromosome 4. The primers from the 60 sequences were tested against all the Theileria species that co-infect cattle in East and Southern Africa and were found to be specific for T. parva. In order to demonstrate the utility of these markers, we characterised eight tissue culture isolates of T. parva isolated from cattle in widely separated regions of Eastern and Southern Africa (one from Zambia, one from Uganda, two from Zimbabwe, four from Kenya) and one Kenyan tissue culture isolate from Cape buffalo (Syncerus caffer). The numbers of alleles per locus range from three to eight indicating a high level of diversity between these geographically distinct isolates. We also analysed five isolates from cattle on a single farm at Kakuzi in the central highlands of Kenya and identified a range of one to four alleles per locus. Four of the Kakuzi isolates represented distinct multilocus genotypes while two exhibited identical multilocus genotypes. This indicates a high level of diversity in a single population of T. parva. Cluster analysis of multilocus genotypes from the 14 isolates (using a neighbour joining algorithm) revealed that genetic similarity between isolates was not obviously related to their geographical origin