

## DEVELOPMENT OF A MICROSATELLITE MULTIPLEX PCR STRATEGY FOR DIFFERENTIATION OF *CANDIDA ALBICANS* STRAINS

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The polymorphism of five new microsatellite *loci*, located outside and inside known coding regions, in the genome of the pathogenic yeast *C. albicans*, was investigated in order to evaluate their applicability to accurately differentiate strains. The microsatellites were selected so that each was assigned to a different chromosome in order to evenly span them throughout the genome. A multiplex PCR strategy was developed allowing the simultaneous screening of the five markers, followed by GenScan analysis of the products, providing a rapid and accurate methodology for genotyping large numbers of strains. A total of 122 *C. albicans* strains, obtained from 80 patients and collected from three health institutions, were analysed using this multiplex system. Seventy-eight different genotypes were observed resulting in a discriminatory power of 0.98. When applying these microsatellites to the identification of strains isolated from recurrent vulvovaginal infections in eight patients, it was found that 13 out of 15 episodes were due to the same strain. When multiple isolates obtained from the same patient were studied the results showed that in different body sites, patients can harbour distinct clones but the infecting population at each body site is monoclonal. These new microsatellites proved to be a valuable tool to differentiate *C. albicans* strains and when compared to other molecular genotyping techniques, revealed to be simple, efficient and reproducible, being suitable for application in large scale epidemiological studies. Allele nomenclature based on the number of repeat sequences rather than fragment size is proposed for the characterization of each strain and contribute for the construction of a public database in light of what is already in use for other organisms.