Ciências ULisboa

Engenharia Genética
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MULTIPLEX PCR

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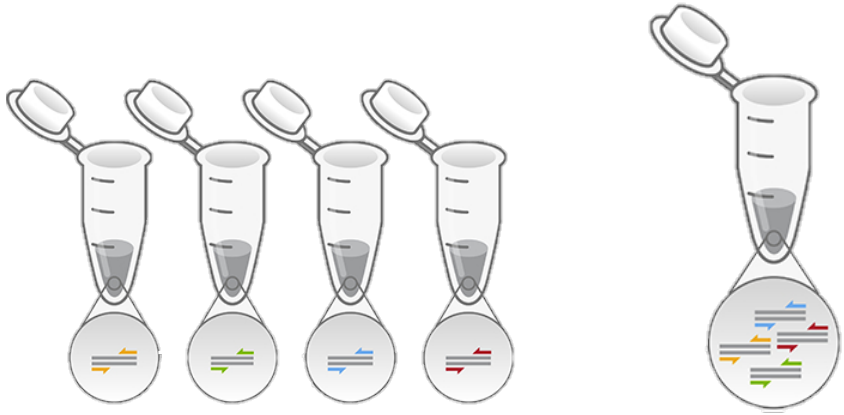
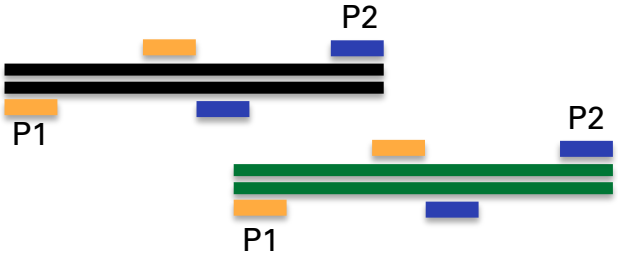


WHAT IS MULTIPLEX PCR?

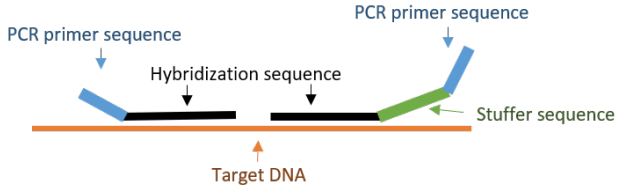
Multiple primers pairs on a single template

Multiple primers pairs on multiple templates

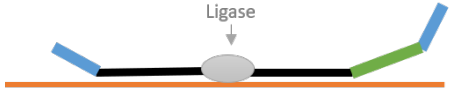


Multiplex Ligation-Dependent Probe Amplification (MLPA)

1 Denaturation; **2** Hybridization

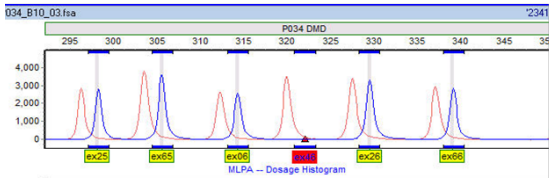


3 Ligation



4 Amplification

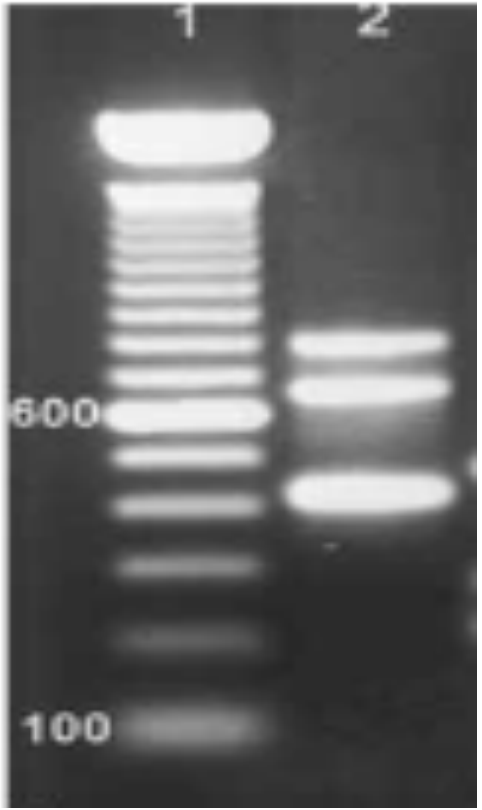
5 Fragment separation and Data analysis



ADVANTAGES	DISAVANTAGES	MLPA ADVANTAGES
Internal controls	Need for primer specificity	Very sensitive
Template quality and quantity	Differen sized or Fluorescently marked amplicons	No non-specific binding
Efficiency	Competition for resources	Easy probe differentiation (stuffer)

APPLICATIONS

- Pathogen identification;
- Gene presence analysis;
- Gene deletion analysis;
- Mutation analysis;
- Linkage analysis;
- High throughput SNP genotyping;
- RNA detection;
- Forensic studies.



Multiplex PCR for the detection of tetracycline resistant genes (Ng et al., 2001)