Abstract

Burkholderia thailandensis is a soil bacterium that frequently duplicates a 208.6 kb region of DNA containing 157 genes. This region is duplicated through RecA-dependent recombination between homologous IS elements flanking the region. Duplications are more abundant in bacteria grown as biofilms, suggesting that there is an advantage to having two or more copies of one or more genes within this region during biofilm growth. In order to find the gene(s) conferring this advantage when duplicated, a fragmented reporter system was used to observe formation and resolution of duplications. These fragmented gene pairs contain >500 bp of homology, but are non-functional until after recombination duplicates the intervening sequence, forming a complete gene at the duplication junction. A fragmented kanamycin-encoding gene reporter system which confers kanamycin resistance to only cells with a duplication is commonly used to quantify frequency of duplication.

In order to identify advantageous genes, that when duplicated, provide a fitness advantage in biofilm growth, two strains with fragmented kanamycin reporter sequences were made, capable of duplicating either the 5' or the 3' portion of the duplicating region. Each strain was grown as a biofilm to select for duplication of the region. Samples from the biofilms were resuspended in PBS, serially diluted, and plated on plates with and without kanamycin. The duplication frequency was measured by comparing growth on plates with antibiotics to those without. Neither half duplicated as much as the WT control nor as little as the negative control suggesting there are potentially advantageous genes in both halves.