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Poster: Functional studies of ribosomal protein bL27 of the model bacterium *Thermus thermophilus*

For my honors project, I studied the ribosomal protein bL27 of *Thermus thermophilus*. Ribosomes synthesize proteins which is an essential part of all life. Understanding the function of ribosomes is important when studying mechanisms of antibiotic resistance, since they are a major target of many classes of drugs. Figure 1.0 shows where ribosomal protein bL27 is found in the 50s subunit. It has a tail that reaches into the peptidyl transferase active site where amino acid elongation occurs. It was my goal to understand the role that this protein has on peptide bond formation and ribosomal development and function. We deleted the *rpmA* gene in the chromosome, which codes for bL27, and replaced it with a kanamycin-resistance gene (also known as a *rpmA* knockout (KO)). Figure 3.0 shows where the bL27 gene is found within *rmpA*, and Figure 2.0 shows its actual sequence. The new kanamycin-resistant mutant strains missing bL27 (called MP) grew slowly on antibiotic agar plates and were viable. This result suggested that bL27 may only have a minor role in normal cell function. To confirm that the MP strains did not have the gene for bL27, we used a polymerase chain reaction (PCR) with the primers shown in Figure 4.0 to amplify the segment of the chromosome where *rpmA* would be. Gel electrophoresis was used to visualize this PCR product and is shown in Figure 5.0. When compared to the wild type strain, which *does* have *rpmA* (approximately 3000 base pairs in length), it appears that strains MP1 – MP8 successfully incorporated the knockout plasmid into their chromosome (which is slightly longer than wild type *rpmA*). We know this, because the longer the gene segment, the slower they will migrate through the gel. The band from wild type has migrated slightly quicker than the bands from the mutant strains. Molecular genomic sequencing of these segments would fully confirm if the gene for bL27 was removed from the MP strains. In the future, I will further classify this mutant by determining its growth rates and antibiotic resistance patterns. Experiments of this sort are so interesting, since we are able to study individual components of bacterial ribosomes and determine their role in protein synthesis and antibiotic resistance.