Genetic Instability and Polycystic Kidney Disease

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Given the world's estimated population, there are more than 6 million people with polycystic renal disease. We are interested in the mutagenic mechanism leading to these genetic disorders. Using the murine congenital polycystic kidney disease as a model, we identified a complex deletion at the *cpk* locus. We hypothesized that this mutation was the result of alternative DNA secondary structures such as cruciform or triplex conformation. To test this hypothesis, we cloned the murine sequence at the *cpk* locus into the chloramphenicol resistance gene in the vector pBR325. This construct could then be used to quantitate deletion by reversion back into chloramphenicol resistance. However, using host bacterial strains DH5α and Top10, we were unable to clone the full-length sequence because each isolate always contained deletion forms. Re-transformation failed to isolate the full-length tract as well. Furthermore, transformation into SURE bacterial strains, which lack the endonuclease SbcC, demonstrated tract instability, indicating that cruciform structures were not likely involved. Our current studies involve exploring melting curves of the sequence to characterize possible triplex structures, since triplex structure also appears to mediate mutations in the human *PKD1* gene that lead to autosomal dominant polycystic kidney disease. Elucidating the mutagenic mechanism leading to polycystic kidney disease may not only lead to future treatments of the disease, but may also help researchers identify the mutagenic mechanisms of other diseases that involve alternative DNA structures.