Examination and Optimization of the PreCR™ DNA Repair Mix on Damaged DNA for Short Tandem Repeat and Mitochondrial DNA Analysis

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Biological specimens submitted for forensic DNA analysis are seldom pristine and unaffected by elements of their environment. The DNA contained in samples collected from crime scenes, mass disasters, mass graves and cold cases can be affected by a number of factors that cause variable forms of damage. One method of analyzing heavily damaged DNA is to amplify reduced size STR fragments using "mini-plexes." If the DNA is too degraded, however, the "mini-plexes" will suffer from the same problems as the conventional STR analysis. Being able to repair the damaged DNA *in vitro* prior to amplification could allow the analyst to perform conventional STR typing on these compromised samples. The PreCR™ Repair Mix (New England BioLabs, Ipswich, MA) contains a combination of recombinant repair enzymes designed to repair various forms of DNA damage prior to PCR amplification. The effectiveness of the PreCR™ Repair Mix on artificially damaged DNA and on non-probative casework samples was examined. Initial studies following the manufacturer's recommended protocol produced limited results with DNA samples for STR analysis, but gave good results for mitochondrial DNA (mtDNA) analysis. The standard protocol was then modified to incorporate the repair reaction components into the standard STR PCR reaction as a pre-incubation step. The modified protocol was further optimized by varying enzyme concentration and incubation times. It was found that these modifications produced a significant increase in the overall average peak heights of artificially damaged DNA samples for STR analysis. Previous research with DNA repair has shown that the formation of chimeric products can create artifacts that may interfere with interpretation. In this study, no chimeric artifacts were observed in over 150 DNA repair reactions performed on damaged DNA. This modified PreCR™ DNA Repair Mix treatment could be a simple addition to the laboratory's existing PCR parameters for obtaining STR or mtDNA profiles from damaged DNA without significantly increasing labor or time.