

PRECR® REPAIR MIX RESTORATION OF PARTIAL STR PROFILES RESULTING FROM DNA DAMAGING AGENTS

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Forensic evidence obtained from crime scenes, in best cases, presents challenges to STR profile analysis. These challenges may be in the form of limited quantity, sample type, age, storage conditions, collection methods, etc. Extraction method and STR analysis, to include mini-STRs, must be carefully considered in order to obtain results that can be presented in court or assist in the investigation. Such consideration is further challenged when a perpetrator attempts to clean the crime scene through the introduction of chemicals, such as bleach and hydrogen peroxide. These cleaning agents cause oxidative damage and hydrolysis of DNA, altering DNA structure. Other methods of DNA damage may be environmental and include exposure to: UVA, extreme environmental conditions, ionizing radiation (gamma irradiation), etc. The resultant DNA damage includes: abasic sites, single strand breaks (SSBs), base mismatches, modified bases, modified 3' ends, 8-oxo-dG formation, thymidine dimers, and double strand breaks. Though the amount and type of DNA damage varies based upon exposure time, concentration, and type of damaging agent, when enough damage has occurred STR analysis becomes difficult. Issues encountered include allele drop out, peak imbalance, partial profile generation, and the reduction in amplifiable loci or generation of the "ski-slope" effect.

The New England Biolabs (NEB) PreCR® Repair Mix contains a number of enzymes that function to correct damaged DNA regardless of the damaging method used. The kit includes a specialized polymerase, a ligase, endonucleases, and glycosylases to correct the aforementioned DNA damage. The objective of this research was to evaluate the NEB PreCR Repair Mix for repair of DNA lesions induced by bleach, hydrogen peroxide, gamma irradiation, and extreme environmental conditions. The success of the repair mix is dependent upon the extent and type of DNA damage. In this work, the repair mix protocol was optimized using various DNA sources to include: HL-60, 9947A, blood, semen, and saliva. Each DNA source was damaged to produce lesions that resulted in partial, "ski-slope", or no profiles using a variety of damaging methods. The optimized repair protocols used were a modification of both the manufacturer's suggested protocol as well as a method published by the ATF and AFDIL. The success of the repair was evaluated using (either individually or in combination): AmpF[®]STR[®] Identifiler[®] PCR Amplification Kit, AmpF[®]STR[®] MiniFiler[™] PCR Amplification Kit, the PowerPlex[®] S5 System, and the PowerPlex[®] ESX System. Depending upon fluid type and damaging method, the PreCR[®] Repair Mix improves STR profiles through restoration or enhancement of alleles. ☘