## E44 Validation of the PrepFiler® Express BTA™ Forensic DNA Extraction Kit

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**Learning Overview:** After attending this presentation, attendees will have information concerning the use of PrepFiler® Express BTA™ and the possible substrates for extraction with PrepFiler® Express BTA™, as well as the advantages and disadvantages of using PrepFiler® Express BTA™.

**Impact on the Forensic Science Community:** This presentation will impact the forensic science community, especially those in the DNA biology analysis field, by describing a method of obtaining a DNA Short Tandem Repeat (STR) profile from substrates with a limited quantity of DNA by using the PrepFiler® Express BTA™ extraction kit.

Currently, methods to extract DNA from bone, teeth, and adhesive substrates, such as cigarette butts, chewing gum, envelope flaps, and Fastape, are growing in popularity. However, there are multiple methods that give varying results when trying to extract DNA from these substrates. A possible reason for these variabilities could be the high rate of degradation/inhibition that occurs with bone, teeth, and adhesive substrates. PrepFiler® Express BTA™ DNA extraction kit was designed for those types of substrates because it is able to remove Polymerase Chain Reaction (PCR) inhibitors and helps extract more DNA from these substrates than previously used reagents when accompanied by an AutoMate Express™.

The validation for PrepFiler® Express BTA™ DNA extraction kit was conducted by the Oakland County Sheriff's Office Biology DNA Laboratory. During this validation, four different studies were performed to help determine if this extraction kit would be beneficial to implement for casework when working with these substrates. These included: (1) assess the optimal amount of sample from various types of bone/teeth, as well as determine the optimal incubation time for extraction; (2) confirm that all four of the AutoMate Express™ instruments in use in the lab give comparable results; (3) determine if PrepFiler® Express BTA™ is able to produce STR profiles from non-probative samples that are typical for case work and are suitable for comparison to known samples; and (4) determine if PrepFiler® Express BTA™ was better at removing inhibitors and extracting DNA from these challenging substrates when compared to PrepFiler® Express™. Along with these validation studies, a case study was performed on bone chips discovered in Italy that were believed to be from the 1800s. All samples were extracted on an AutoMate Express™ instrument using PrepFiler® Express BTA™, except for the samples that were extracted with PrepFiler® Express™. They were quantified using Quantifiler® Trio Quantification Kit on an Applied Biosystems® 7500 Real-Time PCR System and amplified using GlobalFiler™ PCR Amplification kit using an Applied Biosystems® 9700 GeneAmp® PCR System. Capillary electrophoresis was performed on Applied Biosystems® 3500 Genetic Analyzer.

It was determined during this validation that when using PrepFiler® Express BTA™, a complete STR profile was obtained for both femur and tooth powders from 10mg to 50mg, while rib powder was best extracted from 40mg to 50mg. However, using 50mg of powder obtained from the bone chips did not produce full profiles and produced only a partial STR profile for one of the ten samples. It was observed during this validation that when extracting DNA from multiple replicates of bone powder samples, all four of the AutoMate Express™ instruments did in fact produce similar profiles. Also, the validation illustrated that PrepFiler® Express BTA™ is able to produce profiles for non-probative evidence samples that can be used for comparison to known profiles. When comparing PrepFiler® Express BTA™ to PrepFiler® Express™, it was determined that PrepFiler® Express BTA™ gave overall better profiles with less degradation/inhibition when extracting DNA from adhesive substrate samples. However, when extracting DNA from Fastape, better quality profiles were obtained with extraction following the bone and tooth protocol instead of the adhesive substrate protocol.

Based on the results from the validation of PrepFiler® Express BTA™, it was concluded that this DNA extraction kit was able to extract DNA from bones, teeth, and adhesive substrates in a way that prevents PCR inhibition and increases the quantity of DNA extracted compared to PrepFiler® Express™. PrepFiler® Express BTA™ was able to extract DNA in a manner that produces STR profiles that can be used on forensic samples commonly processed in this laboratory.

**PrepFiler® Express BTA™, DNA Extraction, AutoMate Express™**