

## HID UNIVERSITY





## The AutoMate Express<sup>™</sup> Forensic **DNA Extraction System**

**Brad Dixon Forensic Science Applications Group** 



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## **Presentation Overview**

- Chemistry and Instrument Overview
  - PrepFiler *Express*<sup>™</sup> & PrepFiler *Express* BTA<sup>™</sup> Forensic DNA Extraction Kits
    - Protocol Overviews
    - PrepFiler LySep<sup>™</sup> Column and Cartridge Configuration
- AutoMate Express<sup>™</sup> Instrument DNA Isolation
- Developmental Validation Data





## AutoMate *Express*<sup>™</sup> Forensic DNA Extraction System

- Developed specifically to enable maximum yield of high quality genomic DNA from forensic samples using the PrepFiler *Express*<sup>™</sup> Forensic DNA Extraction Kits
  - Processes 13 samples in ~30 minutes
  - Two protocols on a single script card
    - PrepFiler<sup>™</sup> Lysis Buffer and <u>B</u>one <u>T</u>ooth <u>A</u>dhesive (BTA)











## AutoMate *Express*<sup>™</sup> DNA Extraction Workflow







## What is the PrepFiler *Express*<sup>™</sup> Kit?

- Contains all reagents needed to perform online DNA isolation in an AutoMate Express™ System specific cartridge
- A combination of uniquely structured magnetic particles and a multi-component surface chemistry optimized to provide extremely efficient DNA binding capacity and maximum DNA recovery
- No organic extraction
  No silica-based purification
  No ion exchangers
  No direct precipitation

#### Advantages of the Magnetized DNA Complex

- Efficient DNA binding capacity and maximum DNA recovery
- Stable binding during wash steps while removing PCR inhibitors
- Consistent yields of purified DNA separated from the magnetic particles at the elution step



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## PrepFiler *Express*<sup>™</sup> and *Express* BTA<sup>™</sup> Forensic DNA Extraction Kits





## **Kit Configurations**

- PrepFiler *Express*<sup>™</sup> Forensic DNA Extraction Kit\*
  - PrepFiler™ Lysis Buffer
  - PrepFiler Express <sup>TM</sup> Cartridges<sup>\*\*</sup>
  - Sample Tubes
  - Elution Tubes
  - PrepFiler LySep™ Columns
  - Tips and tip sheaths

- PrepFiler *Express* BTA<sup>™</sup> Forensic
   DNA Extraction Kit\*
  - PrepFiler BTA™ Lysis Buffer
  - Proteinase K
  - PrepFiler Express <sup>TM</sup> Cartridges<sup>\*\*</sup>
  - Sample Tubes
  - Elution Tubes
  - PrepFiler LySep™ Columns
  - PrepFiler<sup>™</sup> Lysis Tubes (for bone)
  - Tips and tip sheaths

\*1M DTT also required but not included in the kit

- \*\* Isopropanol and prepared PrepFiler™ Kit wash buffers are included in cartridge format
- \*\*\* Available as a plastics only kit

**7** 8/9/2010





## PrepFiler *Express*™/*Express* BTA™ Cartridge Configuration

Cartridges are foil sealed and pierced after protocol start







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## PrepFiler *Express*™/*Express* BTA™ Kit Off-Line Lysis





### PrepFiler *Express<sup>TM</sup>* Kit Protocol Overview







## The PrepFiler LySep<sup>™</sup> Column Simple off-line Lysis & Substrate Removal







### The PrepFiler LySep<sup>™</sup> Column Simple off-line Lysis & Substrate Removal





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## PrepFiler LySep<sup>™</sup> Filter Column

 Allows incubation of substrate with lysis buffer in assembled PrepFiler LySep<sup>™</sup> Column+Sample Tube configuration



- Filter unit plus membrane seal prevents any lysis buffer drainage during incubation
- High-speed centrifugation enables separation of lysate from substrate
  - Centrifugation force bursts membrane seal enabling lysate flow-through
- Discard PrepFiler LySep<sup>™</sup> Column containing substrate and load hingeless sample tube directly onto the AutoMate *Express<sup>™</sup>* Instrument
- Eliminates the need for a "Tip Dance" protocol





## PrepFiler *Express* BTA<sup>™</sup> Kit Protocol: Adhesives













## PrepFiler *Express* BTA<sup>™</sup> Kit Protocol: Bone Samples

- Screw Cap:
  - Needed during lysis due to bone effervescence which can pop open snap cap tubes. Also, prevents loss of volume during incubation
- 50mg Bone Powder:
  - Optimal input to recover the highest yield of DNA per mg of starting material.
  - Higher starting weight of bone may increase yield but may decrease available lysate volume
- 2 hour Incubation:
  - Minimum time for incubation but can do up to 18 hours without adverse results.
- Supernatant Processing:
  - After incubation, centrifugation required to pellet bone powder
  - Process supernatant only



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## AutoMate *Express<sup>TM</sup>* Instrument DNA Isolation





## Setup AutoMate *Express*<sup>™</sup> Instrument

Ensure Protocol Card is properly inserted



- Do Not Turn on Instrument without card inserted!
- Do Not Remove IC Card if the instrument is on!
- Power on AutoMate Express<sup>™</sup> Instrument





## AutoMate *Express*<sup>™</sup> Instrument Run

- 1. Load cartridge rack with appropriate number of reagent cartridges
- 2. Load tip and tube rack
  - Sample lysate
  - Tips and tip holders
  - Elution tube
- 3. Select appropriate PrepFiler *Express*™ Protocol
- 4. Start run



S - Sample tube T2 - Tip and tip holder T1 - (empty)

E - Elution



Select Script Press 1:PF Express 2:PF Express BTA







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## **Instrument Run and Completion**

- Both the PrepFiler Express<sup>™</sup> Standard and BTA Kits take approximately 30 minutes to complete
- When the instrument has completed the run, the instrument will beep briefly and display Finished Protocol on the display screen.
- Open the door, remove samples and store short term at 4°C or long term at -20°C
- Discard the sample tubes, cartridges, tips, and tip holders into biohazardous waste containers



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## **Developmental Validation Data**





## Sensitivity Linearity



#### N=4 for each blood sample

The blood series demonstrates a proportional increase in DNA yield as a function of blood input.





## Sensitivity STR Data: Representative Profiles







## **Correlation Study-Low Input Samples**







## **Correlation Study-High Input Samples**







## **Correlation Study-Total Allele Count**







## **Reproducibility-Low Input Samples**







## **Reproducibility-High Input Samples**







### **Case Type Samples**







## Summary

- Robust, easy-to-use system optimized for use with PrepFiler<sup>™</sup> Chemistry
- Effectively isolates high quality genomic DNA from a broad range of forensic samples
- Successfully removes inhibitors to PCR commonly found in forensic samples maximizing the performance of downstream applications.
- Extracted DNA validated for use with the Quantifiler<sup>®</sup> and AmpF{STR<sup>®</sup> kits







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