Workflow Efficiencies to Sexual Assault Casework Evidence

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QIAGEN would like to thank our speaker, Courtney A. Head, MFS, for her presentation.

The Beginning - Screening

- ~75% of our caseload is Sexual Assault related
- SA kit testing was request driven
- Requests were made to the lab via OLO (HPD's online offense report) thru a designated printer
- All requests assigned by case manager
- Traditional screening performed (AP, p30, sperm search)
- Entire item retained for DNA
- Final reports written in Microsoft Word and transferred into OLO for officer access

The Beginning – DNA testing

- Case manager assigns cases to each DNA analyst from the queue that was a filing cabinet of folders
- DNA analyst responsible for portioning retained evidence for extraction
- DNA analyst also does every step of lab work by hand
- DNA analyst will write DNA report in Microsoft Word and transfer them into OLO
- DNA analyst would make copy of the final report and hand written allele chart to be interoffice mailed to requesting officer

Technology

- Late 2009 a LIMS system was implemented
- Officer's utilize LIMS to make requests
- LIMS was used to track requests and also develop a priority hierarchy that could be searched
- LIMS allowed for a more uniform report writing system which consisted of matrix panels with canned reporting statements
- LIMS has the capacity to email the report in its entirety to the requesting officer

Lean Six Sigma

- Lab processes were evaluated to determine bottlenecks in the DNA process
- Technician positions were developed that focused on the lab work processes so that DNA analysts could focus on report writing
- Daily schedules were implemented so that each person knew what their responsibilities were each day
- Teams were developed that worked on opposite schedules

Robotics/Automation

- EZ1 robots validated to extract references and after 2 years were validated to extract all non-differential samples
- Tecan EVO 150/100 were validated for quant/amp set up and post amp plate set up

Texas Senate Bill 1636

- Mandated that all Sexual Assault kits collected in the state of Texas must be submitted to an accredited Crime Lab within 30 days of collection for testing

Brainstorming…..

- How can we process SA kits more efficiently?
  - Hire more analysts
  - Find a replacement for sperm searches
  - Do we need to send all contact DNA samples to analysis
  - Train more people to perform differential extractions
  - Can we automate the differential extraction process?

Outcome

- Hired on 8 of 10 contract screeners to assist with varying parts of analysis; most were placed in the screening section focused on SA kits
- Start the validation process of chelex extractions on swabs from SA Kits and develop a Y-screening process using PlexorHY to eliminate the need for sperm searches and limit the amount of contact samples that move forward for differential extraction
- Research possibilities for differential automation

Y-screening outcome

- Could use a batching system to process and complete several cases at once rather than one case at a time
- Implemented Lean Six Sigma strategies to work/ review cases using more of a team concept
- Using PlexorHY was a more sensitive and efficient way to determine which samples should move forward for DNA extraction
- Each analyst could complete about 15-20 cases per month total of 75-100 per month; receive about 90 cases per month (we could keep up with demand in screening)

Pros & Cons to Y-screening

Pros

- More timely results
- More sensitive and quantitative than sperm searches
- Limit the number of contact samples moving forward to DNA testing
- True negative samples did not move to extraction

Cons

- Only 1/8th of each swab was tested
- Still needed differential extractions and DNA analysis for case completion
- Female/female or male/male cases are not good candidates

Differential Extraction Process

- Traditional organic PCIA extraction
- Time consuming 2 day extraction
- Lots of tube manipulations
- Limited number of samples processed per person (typically no more than 10-12)
- PCIA is a hazardous chemical
- One extractor per week (1-5 cases a week)

QIAcube – a little automation goes a long way

- Automate the separation and wash steps of the differential extraction process
- 4 washes performed on the sperm pellet
- Cut down the sperm digestion time from overnight to 10 minutes
- Clean up performed on the EZ1; so no more PCIA
- More manageable amount of total DNA extracted (gone are the days 256.03ng of DNA on a quant)
- 2 day extraction has been reduced to 1 day

QIAcube process – 1st step

- Inventory and portion half of all swabs in the SA kit for extraction
- References are also portion and prepped for extraction (EZ1 Tip Dance Protocol)
- Any clothing items within the SA kit will be ALS'd and presumptively tested; if positive, cuttings will be prepped for extraction

QIAcube prcoess – 2nd step

- Add digest master mix to all samples and incubate at 56◦c
- Transfer substrate to spin basket and spin for 5 minutes
- Place digested samples in carriers in correct order in the centrifuge on the QIAcube
- Set up remainder of the deck to prepare for first run
- Start the 12A procedure which will separate the sperm and epithelial fractions and wash the sperm pellet twice

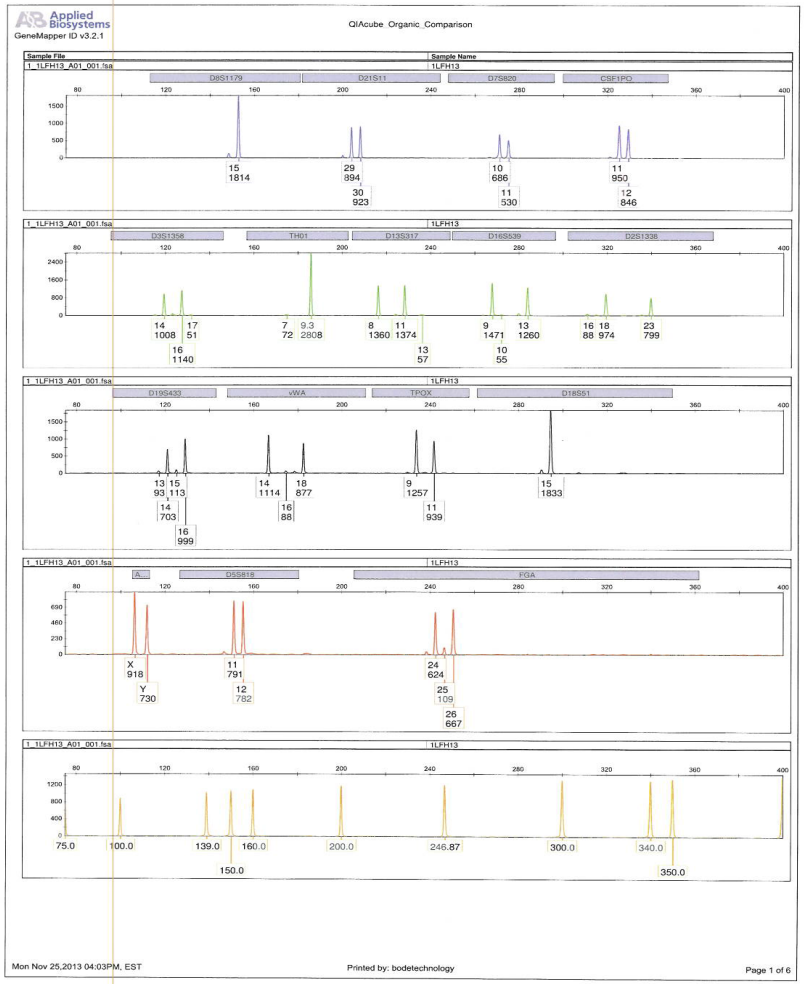
QIAcube process – 3rd step

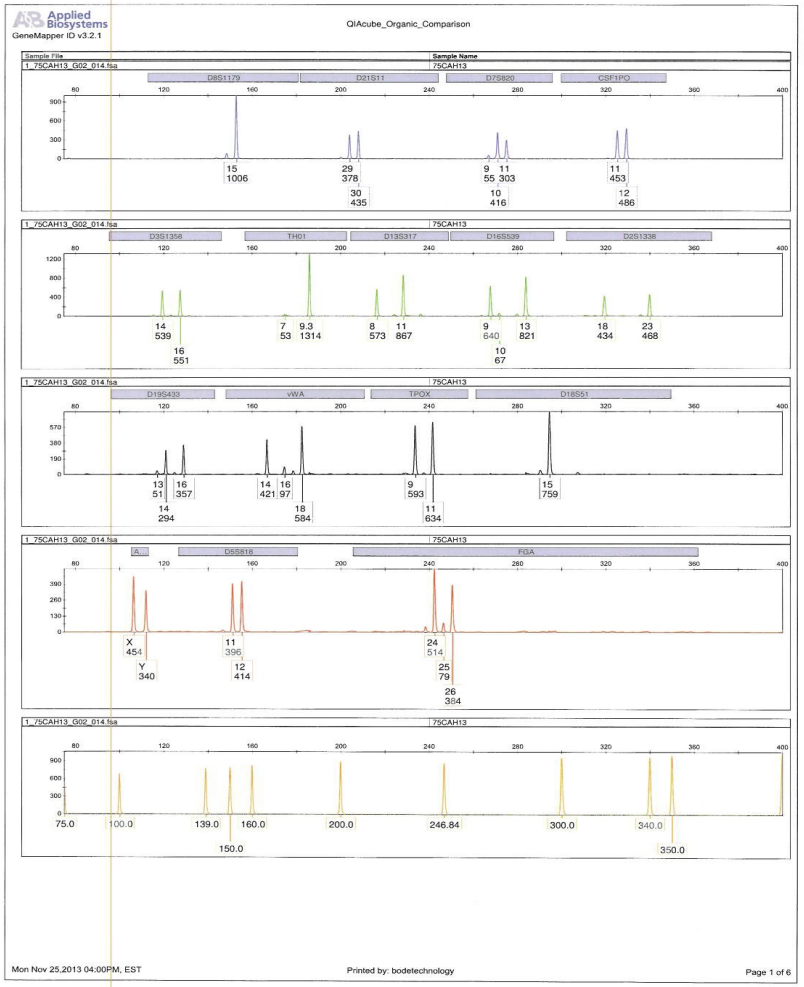
- When 12A has finished the Epithelial Fraction samples can be removed and the deck should be replenished with more tips and the sperm digestion master mix added
- Start 12B protocol which will wash the sperm pellet 2 more times and add the sperm digestion master mix
- Add MTL buffer and cRNA to the Epithelial Fraction samples and start a Large Volume Protocol run on the EZ1 (18 minutes)
- When 12B is complete remove Sperm Fraction samples and place on a heated thermomixer for 10 minutes
- After 10 minutes the Sperm Fraction samples can be started using the Trace Protocol on the EZ1 (17 minutes)

Sample Selection

- All samples Sperm and Epithelial Fractions extracted will be quanted using PlexorHY
- After quant it will be determined which samples will move forward for amplification. Example – multiple suspect or CSP cases will send all positive samples to amp and cases where there is only one suspect and no CSP we will only send the most probative sample forward
- Cases where all samples quant negative will stop and nothing will move forward to amp

Comparison of QIAcube/Organic - SF





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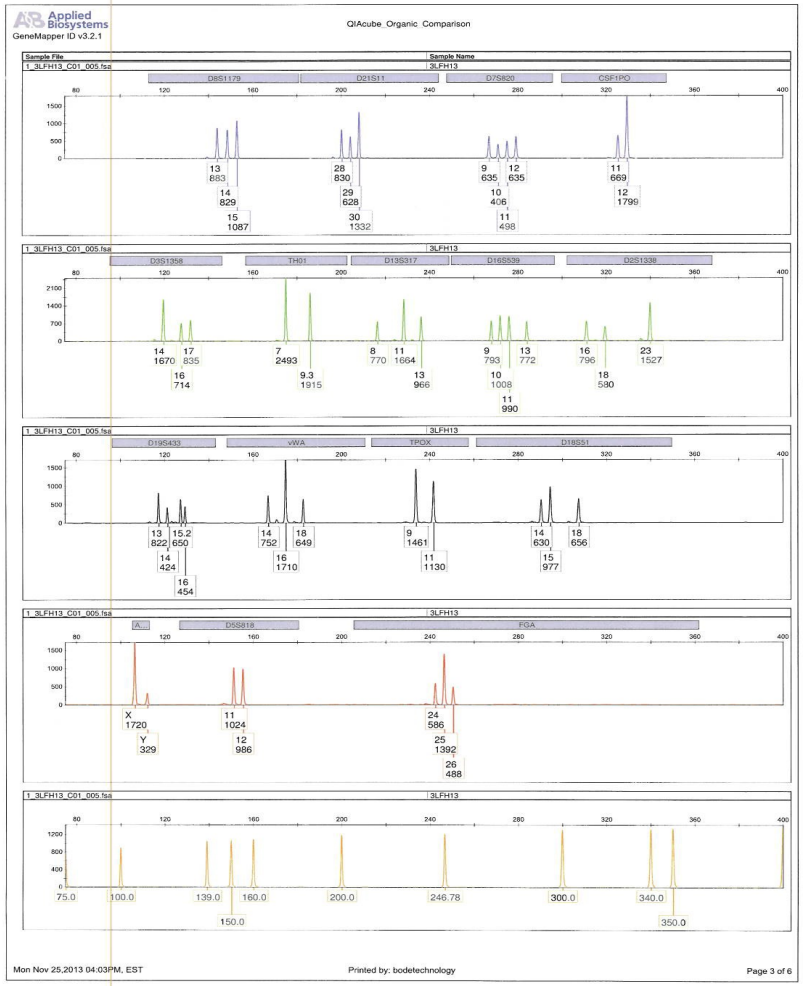

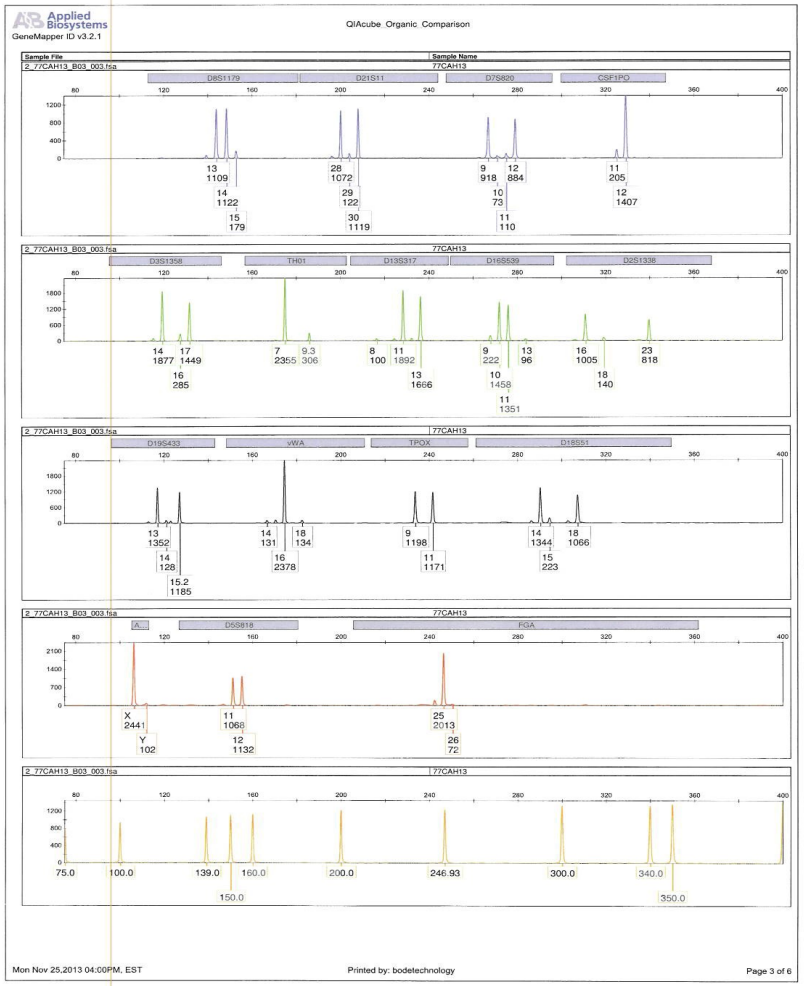

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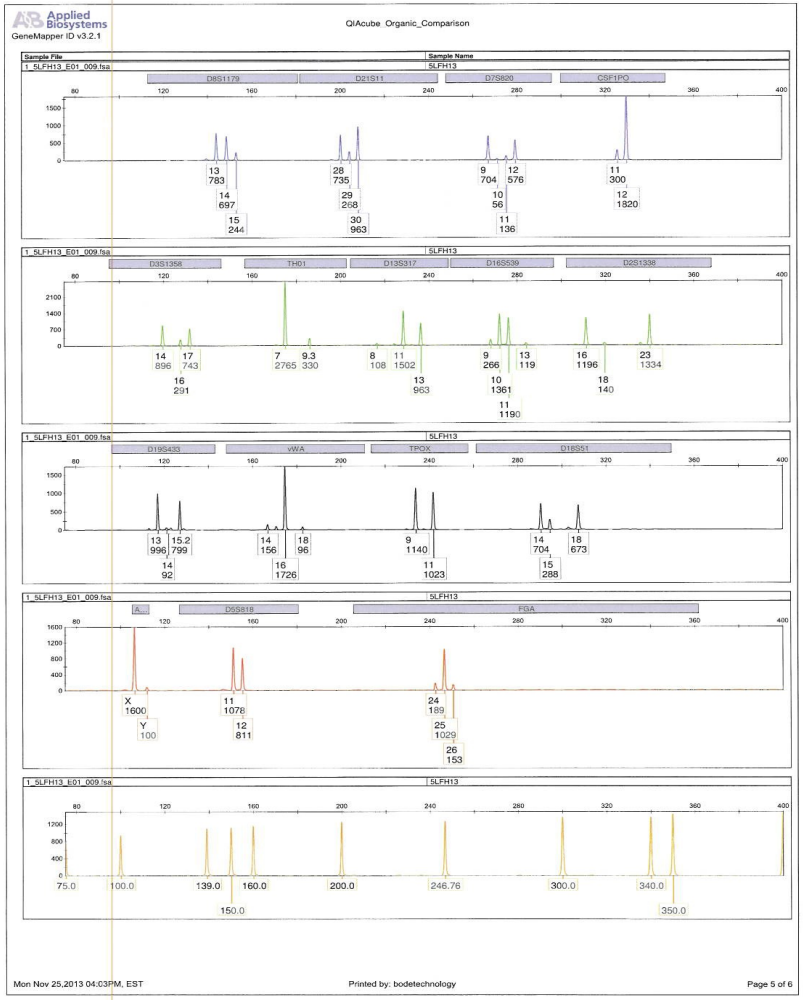
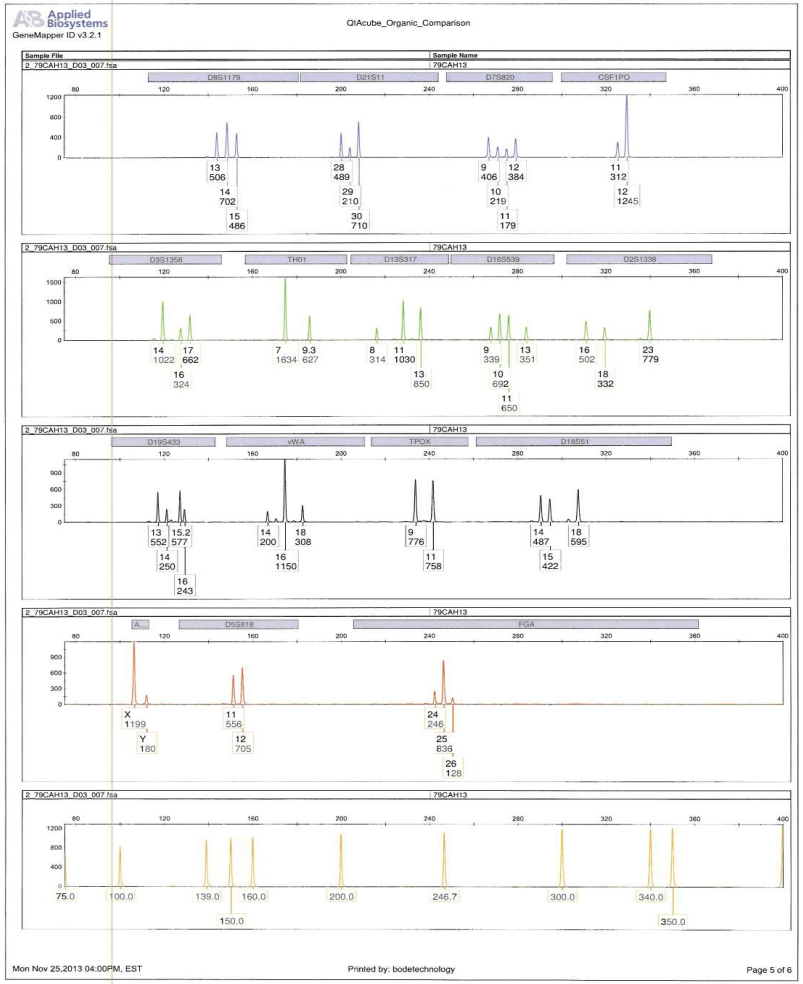
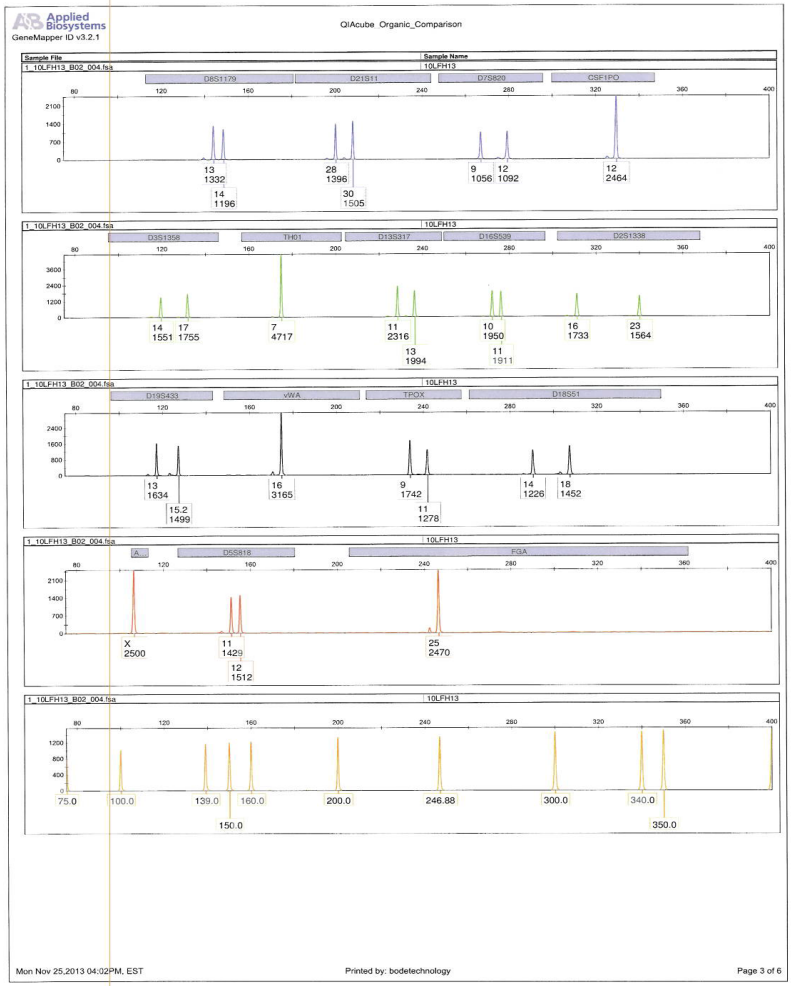
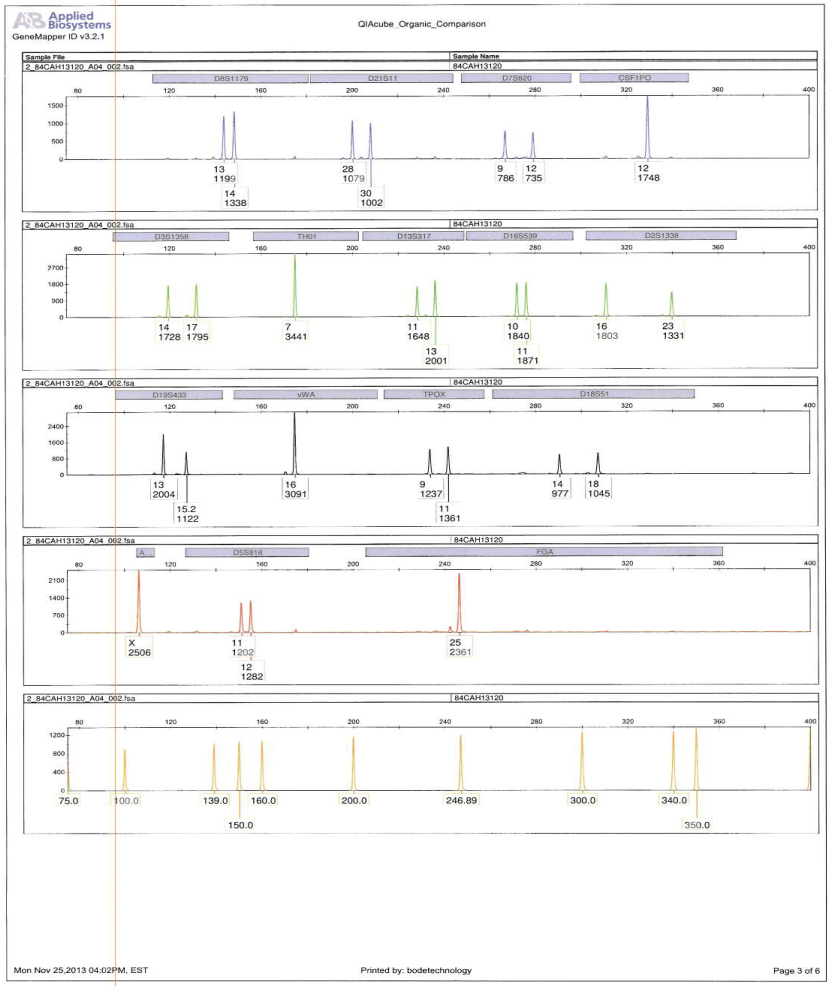




Comparison of QIAcube/Organic - EF





Two Week Processing Time Line

- Day 1 Extraction (72 samples + refs)
- Day 2 Quant (72 samples (2 quant plates) + refs)
- Day 3 Amp (1 amp plate to include paired down samples and refs)
- Day 4 3130 Load (1 plate to include paired down samples and refs)
- Day 5 Data Analysis/Rework if needed
- Day 6-10 Report writing/Tech Review/Analyst Review
- 2 week turnaround time for complete DNA testing (3-9 cases at a time worked)

Turn Around Time Improvements

- 2010 – 8 to 12 month turnaround time for screening and DNA testing on a SA kit
- 2013 – 3 to 5 month turnaround time for screening and DNA testing on a SA kit
- 2014 – 14 day turnaround time for screening and DNA testing on a SA kit

Benefits to QIAcube kit processing

- All samples in kit are extracted up front so re-work is quick and easy
- Previously, requesting officer would receive one report with the screening results and a second report with the DNA results
- New format will provide screening and DNA results all in one report

Crucial Implementations

- LIMS
- Lean Six Sigma concepts
- Qiagen EZ1 Advanced XL
- QIAcube
- Promega Plexor HY

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