

TrueAllele[®] Interpretation of Low-level DNA Data: Examining Cartridge DNA Information

People can touch an item and leave small amounts of DNA behind at a crime scene. Once collected as evidence, this touch DNA is often present in small amounts, producing low DNA data peaks. DNA labs often struggle to interpret this kind of data, claiming that it is uninformative. Their interpretation software may be limited as well. However, Cybergenetics TrueAllele[®] Casework probabilistic genotyping technology can get all of the information from touch DNA data when other interpretation methods cannot.

Cartridge DNA evidence

Cartridge casings are the empty shells left behind after a gun was fired. The casings are made from different metals and can be different calibers (sizes) depending on the type of firearm. Nearly 200,000 cartridge cases are recovered annually at United States crime scenes¹, including homicides, aggravated assaults, robberies, and gang-related crimes. This cartridge touch DNA evidence can contain vital DNA information of who is involved in the crime. But the DNA data generated from fired cartridge casings may be low-level, limiting interpretation for some DNA labs or potentially keep the cartridge evidence from being collected during an investigation.

Low-level DNA data

When DNA laboratories test items like cartridge casings, they take the samples through multiple steps (e.g., extraction, amplification). This process ends with generating electronic DNA data that contain all the DNA information. When there is a large amount of DNA in a sample, the DNA data show high data peaks. However, when there is a small amount of DNA in the sample, the DNA data have low data peaks. All DNA data peaks are informative, regardless of how high or low the data peaks are. Lower-level data convey to the analyst that there is a smaller amount of contributor DNA in the sample.

Touch DNA samples and those from cartridges sometimes result in low-level DNA data. These samples can be degraded or damaged due to environmental factors. DNA labs are typically unable to interpret low-level data, stating that it is below their artificial interpretation threshold. The lab's DNA report may state that the data are "uninterpretable" or "inconclusive" even for their limited interpretation software.



TrueAllele Casework can interpret all DNA data, including low-level cartridge samples. Cybergenetics has analyzed cartridge evidence data in over 90 cases. The technology has been tested on low-level data in validation studies. Cybergenetics has also recently studied TrueAllele interpretation of cartridge DNA data, examining the match information obtained from these samples.

Examining cartridge DNA information

The cartridge study was designed to determine how much single-source DNA information can be recovered from firearm cartridge evidence. The design included multiple cartridge types and DNA collection techniques. However, when reviewing the data, the TrueAllele analyst noted that the data, which was supposed to be from one person, indicated mixtures of multiple people. TrueAllele had no problem resolving this low-level mixture data.

Cartridge samples

The study examined DNA data from seven different casing materials for a total of 910 cartridge samples (Table 1). The cartridge samples were then collected using five different DNA collection methods (Table 2). Once the DNA was collected, the cartridge data were interpreted both manually and using TrueAllele.

Table 1: Cartridge study samples

Material	Total
45 Fired	90
45 Unfired	90
Aluminum Unfired	150
Brass Fired	130
Brass Unfired	150
Nickel Unfired	150
Steel Unfired	150

Table 2: Collection methods

Collection	Description
	Swab cartridge with a wet cotton swab
Wet:Wet	followed by another wet cotton swab
	Swab cartridge with a wet cotton swab
Wet:Dry	followed by a dry cotton swab
	Place cartridge into a solution to soak and
Soak and Sonicate	then in a sonicator
	Rotate cartridge over sticky portion of
Tape Lift	tape dot stuck to microscope slide
	Scrape material off of the cartridge with a
Scraping	sterile razor blade

The cartridge DNA data included 202 low-level samples and 431 mixtures (47% of the samples). The mixtures contained 2 to 5 contributors.

Page 2 of 6



Manual interpretation

Once the single source cartridge samples were created, a university forensic lab manually interpreted the DNA data. They used a peak-height threshold and an allele counting method. The laboratory counted how many alleles matched the reference that were expected to be present in the sample. The laboratory found the reference sample was present in 205 of the cartridge samples. However, the lab's manual review couldn't handle more contributors and sub-threshold peaks.

TrueAllele interpretation

TrueAllele found more DNA information than human review. When comparing to the known reference, the study results showed that the computer found the known reference in 351 cartridge samples (Table 3), compared to 251 samples using manual interpretation. The DNA match information for this known reference was assessed across all cartridge types and shown to be informative with match statistics ranging from the hundreds (less contributor DNA) to the sextillions (1 followed by 21 zeros, more contributor DNA).

	Collection					
Material	Wet:Wet	Wet:Dry	Soak and Sonicate	Tape Lift	Scraping	
45 Fired	1	0	9	N/A	N/A	
45 Unfired	12	19	6	N/A	N/A	
Aluminum Unfired	26	18	9	29	11	
Brass Fired	5	13	1	3	3	
Brass Unfired	8	7	1	22	0	
Nickel Unfired	15	24	0	18	3	
Steel Unfired	18	22	17	17	14	

Table 3: TrueAllele reference inclusions

By using all the data, the computer examined the lower-level data and mixtures and found a previously unidentified contributor. This unknown profile was informative, with an expected genotype match statistic of a nonillion (1 followed by 30 zeros). This unknown contributor was found in 138 of the samples, across all seven cartridge types

Page 3 of 6



(Table 4). Manual interpretation did not assess this unknown person, since that method focused solely on the reference's allele pair, limiting data interpretation.

	Collection					
Material	Wet:Wet	Wet:Dry	Soak and Sonicate	Tape Lift	Scraping	
45 Fired	1	2	2	N/A	N/A	
45 Unfired	10	2	8	N/A	N/A	
Aluminum Unfired	4	4	0	9	0	
Brass Fired	14	3	1	5	3	
Brass Unfired	9	1	0	10	1	
Nickel Unfired	9	3	1	10	0	
Steel Unfired	6	1	3	13	3	

Table 4: TrueAllele unknown contributor inclusions

Study conclusions

Examining the cartridge study results, TrueAllele can develop informative data from cartridge evidence, even without a reference for comparison. The computer uses all the data and does not discard anything (unlike a DNA lab). The reference or unknown cartridge match information can be used to move investigations and prosecutions forward. Additionally, TrueAllele computer interpretation was better than manual review at extracting DNA information from cartridge casings.

TrueAllele Casework Applications

More information from less DNA

Some threshold-based interpretation methods discard valuable DNA information. Using all of the data, TrueAllele Casework extracts more identification information, even on samples with low amounts of DNA. The computer can get more DNA information from less DNA data. Thus, TrueAllele can interpret the data and provide information for challenging evidence items, such as cartridges.



Cartridge database

Once TrueAllele produces the DNA information from cartridge evidence, the next step is to link the informative separated genotypes back to references (to identify suspects) or to other evidence (to find links between cases). TrueAllele Database (TADB) is an automated genotype database that can do just that. All DNA data and information can be uploaded to the TADB, including data from low-level cartridge evidence or known references. By using informative genotypes, TADB provides more distinct evidence to reference matches than a more limited allele database would produce. Additionally, TADB can link cases through evidence to evidence comparisons that help to find serial criminals.

Case example

In the fall of 2017, in separate incidents, an assailant shot two men to death in Baton Rouge, Louisiana. The perpetrator also fired shots through the door of another family's home. The random shootings appeared to be racially motivated. Detectives recovered cartridge casings from one of the crime scenes. The state crime generated DNA data from the evidence but stated that there was insufficient DNA data to perform a manual interpretation using thresholds. The evidence data were submitted to Cybergenetics for TrueAllele analysis. On the same DNA data, TrueAllele connected the suspect to the cartridge casings with a match statistic of 3.28 million. On April 23, 2021, a TrueAllele analyst testified at the East Baton Rouge Parish trial in Baton Rouge about the computer DNA match statistic. On April 26, 2021, the jury found the defendant guilty of first-degree murder, and a judge later sentenced him to life in prison.

Conclusion

Using limited interpretation methods, DNA laboratories have struggled to resolve touch DNA data, including that from cartridge cases. However, TrueAllele Casework does not leave any DNA information behind. The technology readily interprets both low-level single-source and mixture sample data, providing useful match information. The computer can also upload all DNA data into a genotype database, helping to link this evidence to suspects solving violent crimes. Overall, TrueAllele reliably interprets low-level evidence DNA data, including data from cartridge cases.



References

1. "Shelling out Evidence: NIST Ballistic Standard Helps Tie Guns to Criminals." *NIST*, 23 Jan. 2023, www.nist.gov/news-events/news/2012/08/shelling-out-evidence-nist-ballistic-standard-helps-tie-guns-criminals.

2. Prasad, Elisha, et al. "Touch DNA recovery from unfired and fired cartridges: Comparison of swabbing, tape lifting and soaking." *Forensic Science International*, vol. 330, Jan. 2022, p. 111101, https://doi.org/10.1016/j.forsciint.2021.111101.

Page 6 of 6