Representation of Verogen Technology in Forensic Genomics Literature

ForenSeq™ library preparation and analysis and MiSeq FGx® sequencing are the technologies of choice for modern forensic laboratories.

**Highlights**

- **Preferred sequencing technology for forensic genomics**
  The versatility and superior data quality of the MiSeq FGx System make it the platform of choice for 70% of peer-reviewed publications in 2019.

- **Rapid acceptance of the Verogen portfolio**
  Over 314 peer-reviewed studies have employed Verogen platforms and applications to date—a doubling of net new papers every year.

**Preferred Sequencing Technology**

The launch of the Verogen MiSeq FGx Sequencing System in 2015 introduced the industry to next-generation sequencing (NGS), also called massively parallel sequencing (MPS). Since then, Illumina sequencing-by-synthesis (SBS) technology has been the preeminent force driving Verogen NGS technology in the forensic sequencing space. The Illumina SBS technology that underpins the MiSeq FGx System now powers 90% of the sequencing data generated worldwide. Nearly 70% of publications describing commercial library prep methods use technologies specifically designed for sequencing on a MiSeq FGx System, with the ForenSeq approach far outpacing other chemistries (Figure 1). This technical note summarizes the pertinent publications and highlights key papers.

**Community Support across Applications**

Year after year, the scientific community has embraced the Verogen portfolio, publishing more papers each year than the previous year and building academic and professional support for ForenSeq library prep kits, the MiSeq FGx System, and the ForenSeq Universal Analysis Software (UAS). As of March 2020, the community has released 314 peer-reviewed publications endorsing Verogen NGS technology across a broad range of forensic and human identification applications, including validation studies, forensic genetic genealogy (FGG), analysis of degraded and low-quantity DNA samples, missing persons and disaster victim identification (DVI), sequence allele implications, population analysis, and microhaplotypes (Figure 2).

**Complete Sample-to-Answer Solution**

The ForenSeq workflow provides the first fully validated sample-to-answer sequencing solution developed for forensic applications. With the fully kitted and simple assays of ForenSeq library prep kits, the high resolution and unmatched accuracy of the MiSeq FGx System, and the user-centric library management, data analysis, and visualization of ForenSeq UAS, Verogen NGS technology transforms the most fragile, degraded, or mixed samples into powerful results.

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Key Papers

Learn about the research behind the Verogen portfolio with a selection of peer-reviewed papers that highlight how NGS technology is powering modern forensic laboratories and furthering human identification. Verogen offers genomic solutions for a diverse range of forensic applications, including age estimation, body fluid identification, FGG, and more.

Age Estimation


Summary: Statistical modeling approach to analyze NGS data for age-correlated CpGs, with accuracy retained down to 2 ng input DNA.


Summary: Pilot study showing the potential of blood DNA methylation (DNAm) markers for age estimation to evaluate tissues other than blood.


Summary: Review that traces developments in age estimation methods, contextualizing them with forensic genomics goals and requirements while highlighting a path forward.


Summary: Study assessing DNA methylation profiles from 1156 individuals to create an accurate model for chronological age estimation from whole blood data, combining NGS and machine learning.

Ancestry-Informative Marker Set


Summary: Outline of past human population structure and how it might have influenced the distribution of contemporary human diversity, guiding the selection of ancestry-informative marker sets (AIMs) and level of geographic resolution.

Body Fluid Identification


Summary: Description of a prototype NGS mRNA profiling assay that can detect RNA mixtures from different tissues and identify the tissue source of origin.


Summary: Collaborative exercise to analyze dried body fluid stains with two panels: an mRNA panel for body fluid and tissue identification, and a coding region SNP (cSNP) panel for assigning body fluids and tissues to donors.

Forensic Genetic Genealogy


Summary: Report demonstrating the success of sequencing forensic samples to create SNP genotypes for searches in genealogical databases (i.e., GEDmatch) to generate leads to identify missing or unknown individuals.

Nomenclature


Summary: Presentation of STRidER, a free, curated forensic autosomal STR database that enables reliable frequency estimates from high-quality data and offers quality control for autosomal STR data.


Summary: Report summarizing topics discussed at the STR sequence nomenclature meeting the STRAND Working Group, a forum for presenting and discussing ideas.

Summary: Proposal to use the longest uninterrupted stretch (LUS) in allele designations as a straightforward method to represent sequence variation in STR repeat regions and facilitate probabilistic interpretation of NGS typing results.


Summary: Comprehensive revision to the sequence template file. The update expands the forensic STR list, adds annotations, and makes the file available as an FTP download with dynamic revisions and a date-stamped change log.

Population Data


Summary: Study using the ForenSeq DNA Signature Prep Kit to obtain sequences from unrelated French individuals, helping forensic laboratories increase discrimination power for human identification, kinship analysis, and mixture interpretation.


Summary: Concordance assessment of autosomal STRs and population variability using commercial STR kits, capillary electrophoresis (CE), and Verogen NGS technology to type 400 samples. Results demonstrate high concordance.


Summary: Characterization using the MiSeq FGx System and other NGS tools. The resulting population data illustrate the genetic variation in common STR markers for the selected population samples and provide allele frequencies for statistical calculations related to STR profiling.

Review

Bleka, Øyvind, Mayra Eduardoff, Carla Santos, et al., **“Open source software EuroForMix can be used to analyse complex SNP mixtures,”** Forensic Science International: Genetics 31 (November 2017): 105–110, https://doi.org/10.1016/j.fsigen.2017.08.001.

Summary: Quantitative likelihood ratio mixture calculation of a SNP panel demonstrating that uncertainty about the number of contributors to a mixture has little effect on the likelihood ratio, removing a barrier to widespread adoption of SNP crime-stain analysis.


Summary: Appeal for wider adoption of NGS in forensic genomic applications with a focus on issues essential to successful implementation in the laboratory and in court.


Summary: Methods to prepare libraries with the ForenSeq DNA Signature Prep Kit, sequence with the MiSeq FGx System, and analyze data with ForenSeq Universal Analysis Software. Includes an assessment cementing these products as reliable and robust.

Sequence Characterization


Summary: Summary of the STR Sequencing Project (STRSeq), a collaborative, international effort to facilitate the description of sequence-based alleles at the STR loci targeted in human identification assays.


Summary: A detailed population study of sequence variation with ramifications for coordinating the compilation of sequence variation on a much larger scale than was required before forensic laboratories started adopting NGS.

Validation


Summary: Methods for sequencing using Verogen NGS technology. Highlights include sequencing many forensic loci in one multiplex reaction with semi-automated genotyping, successful SWGDAM developmental validation, and generating more actionable information.


Summary: Inter-laboratory validation assessing concordance, reproducibility, sensitivity, and mixtures with a statistical comparison of inter-locus balance between CE and NGS.

**Summary:** Development and validation of a fully automated workflow with the ForenSeq DNA Signature Prep Kit and NGS STARlet. Automation improves reproducibility and reduces hands-on time, facilitating NGS adoption in forensic laboratories.


**Summary:** Inter-laboratory study evaluating forensically relevant parameters in the framework of the SeqForSTRs project. Eight laboratories sequenced a shared ForenSeq DNA Signature Prep library on MiSeq FGx Systems. All obtained quality metrics, spotlighting NGS as a promising tool for human identification.

**Y-STR Analysis**


**Summary:** Study sequencing samples from 250 unrelated Korean males. Results indicated that the MiSeq platform-based analysis system used in the study can facilitate forensic laboratories’ investigation into the sequences of the 23 Y-STRs.

**Learn More**

To learn more about Verogen applications, visit: verogen.com/applications/.