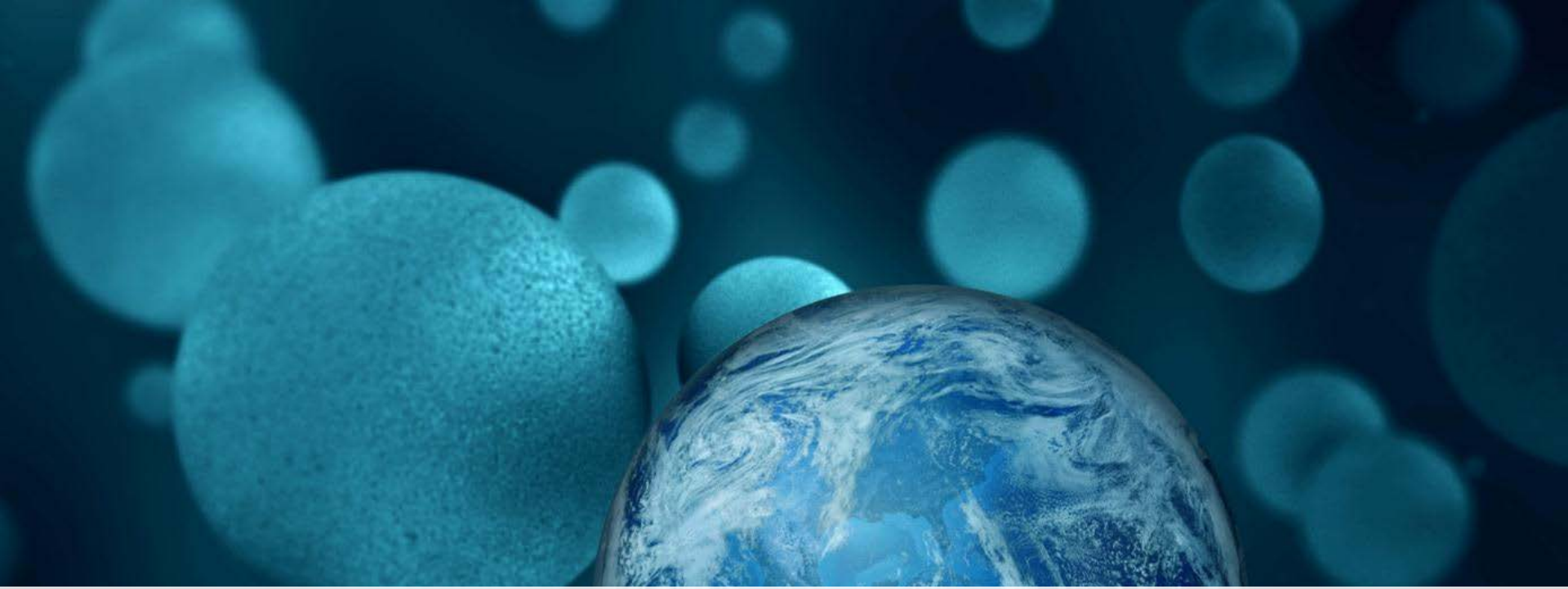


ThermoFisher SCIENTIFIC

Applications and Early Access Results on SeqStudio™ Genetic Analyzer

Steve Jackson

Just Click, That's It

Continuing our innovation in Sanger Sequencing and Fragment Analysis:

Introducing Applied Biosystems™ SeqStudio™ Genetic Analyzer for Sequencing and Fragment Analysis

- Featuring plug-and-play reagents, individualized protocols, an intuitive touchscreen and cloud connectivity

Just Click, That's It

ONE cartridge.
(capillaries, buffer, polymer, and pump incorporated)

A simple **4-capillary system**
with **4-month on-instrument cartridge stability**
4 minutes hands-on time, reduced from hours to minutes.

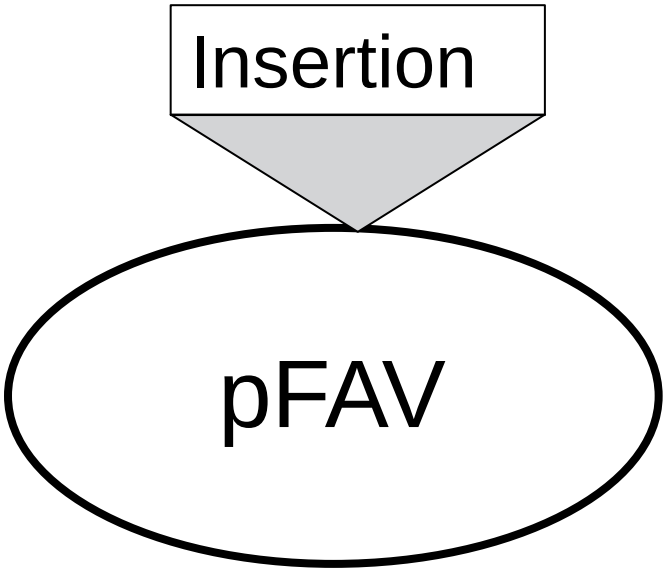
that puts you in control of your samples, in your own time, in your own way.

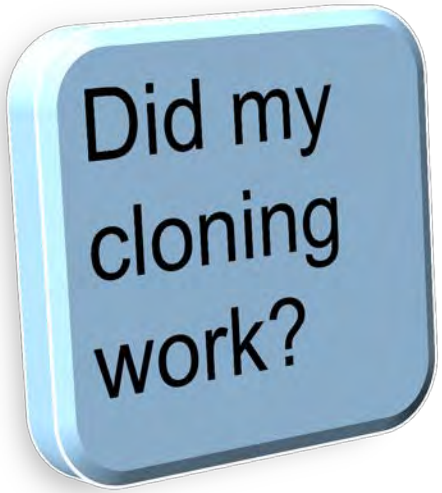
Set up the cartridge, click and run, check your data remotely

- **Easy set up from wherever you are**
- **Monitor, share your protocols or data using any device**
- **From anywhere, with Thermo Fisher Cloud**

Sequencing Plasmid DNA: Clone Checking

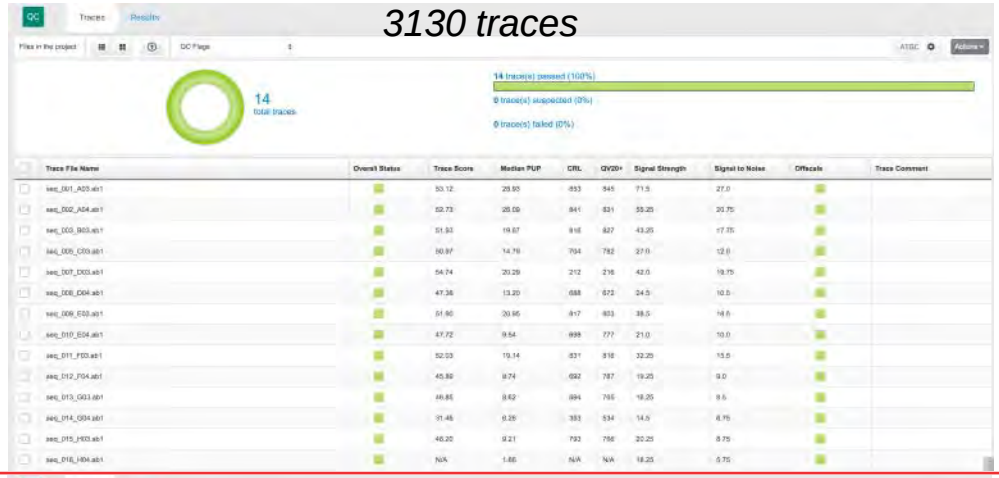
Application	Sample Type	Chemistry	Module	Analysis Software
Clone Confirmation	Plasmid DNA	BDTv3.1	Long Seq	VA Cloud Module

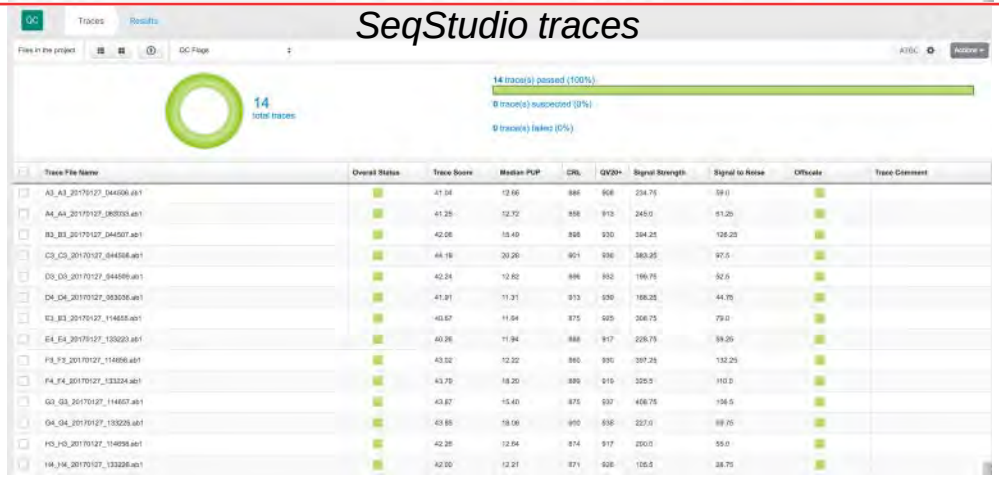




Simple and direct cloud workflow for plasmid verification

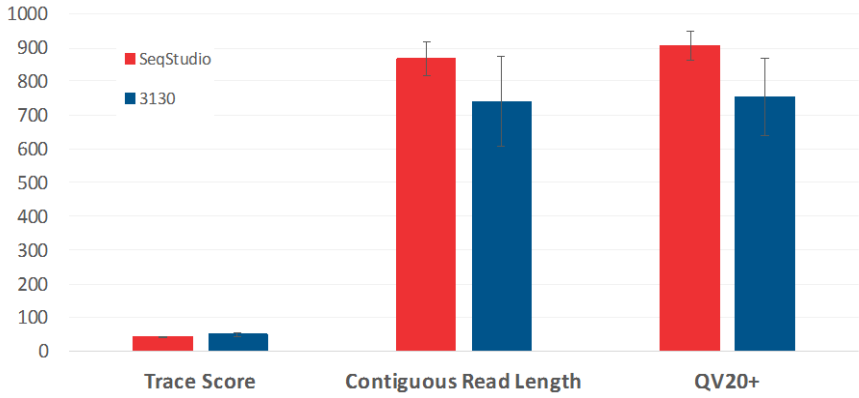
Sequencing Plasmid DNA: Clone Checking





Experiment:

- pGEM7zf(+) plasmid was sequenced using M13 primers and BDTv3.1
- Data analyzed on Sanger QC Cloud app and SeqScanner



Sanger sequencing quality metrics were very similar

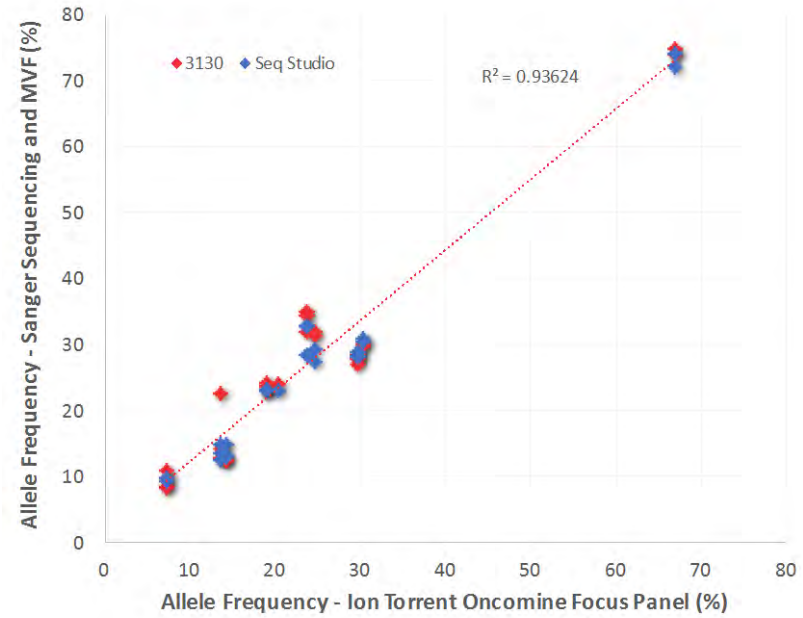
Oncology Research: Sequencing FFPE DNA

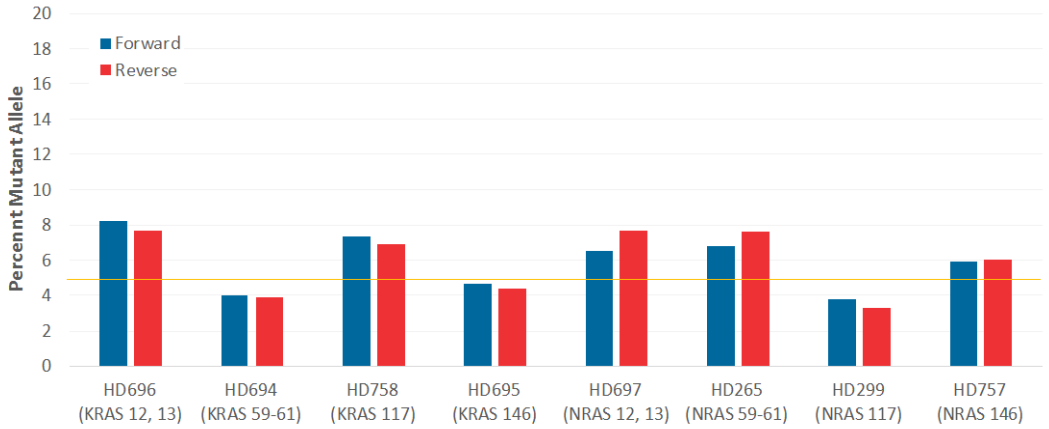
Application	Sample Type	Chemisty	Module	Analysis Software
Oncology Research	FFPE Tissue	BDD/BDX	Short Seq.	Minor Variant Finder (MVF)

Questions:

- What mutations are present?
- Interesting or relevant alleles?
- What are the allele frequencies?

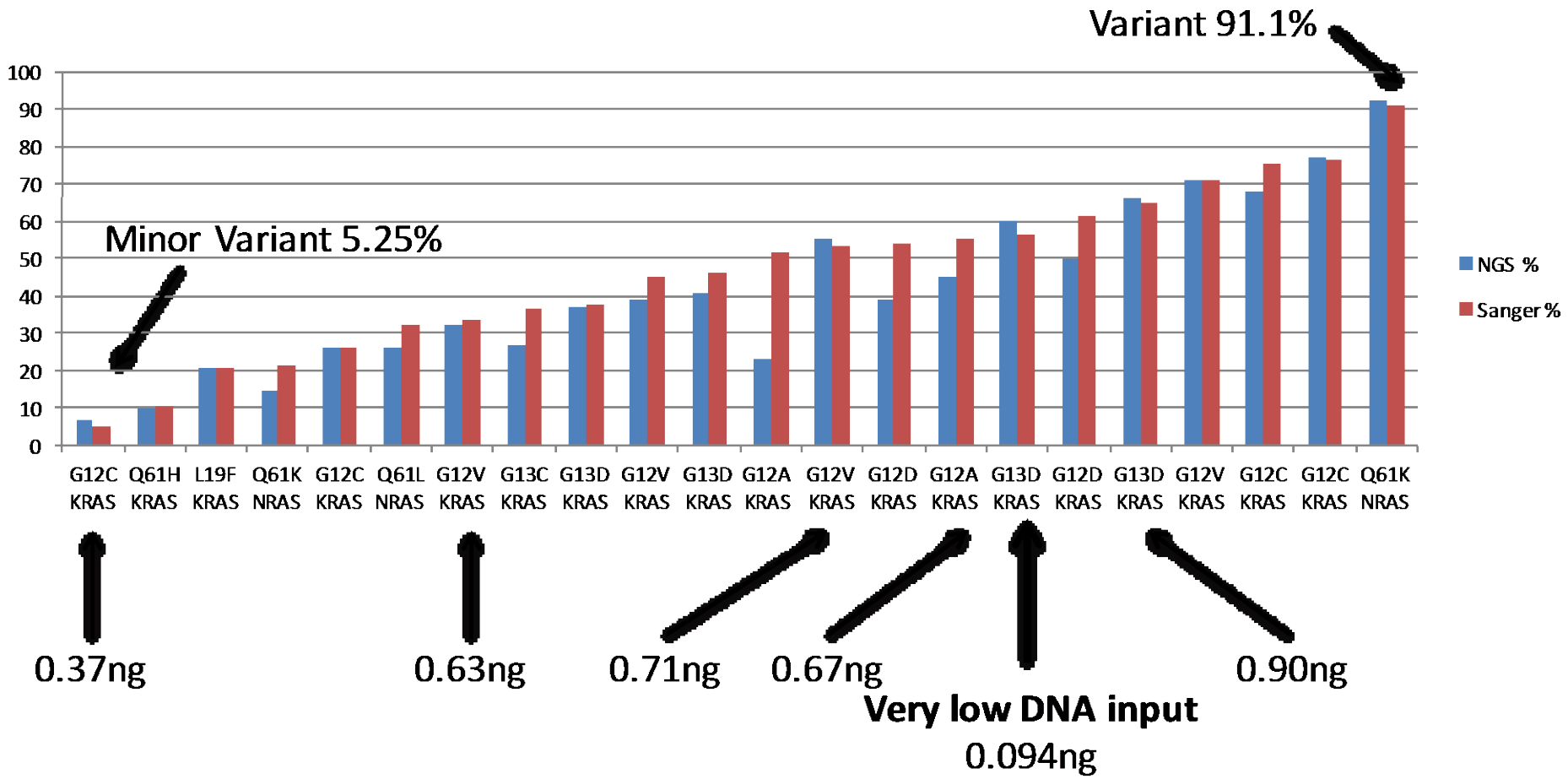
Minor Variant Finder: Next Generation Confirmation





- *Correlation of 10 different tissue samples with seven different hotspot loci at different frequencies was excellent*
- *SeqStudio and MVF can measure allele frequencies as low as 5%*

Early Access Customer Data: Next Gen Confirmation



6 samples below 1 ng / reaction input amount

Data courtesy Dr Luca Quagliata, University of Basel

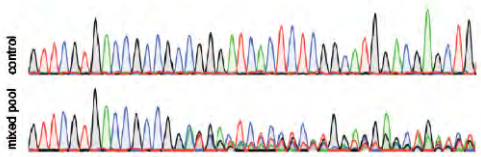
Sanger Sequencing in Genome Editing Workflow

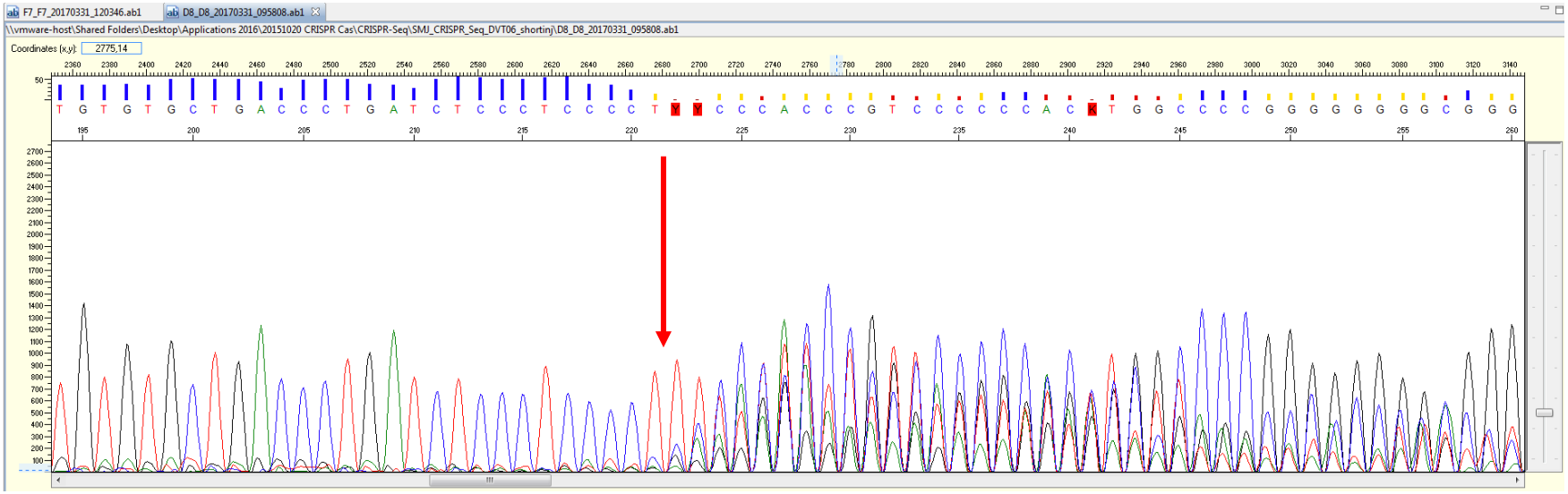
Experiment:

- HEK293 cells treated for random deletions at RELA and HPRT loci
- Primary culture with heterogeneous population of mutations was sequenced
- Mutation profile was determined using TIDE software

TIDE: Tracking of Indels by DEcomposition

NETHERLANDS CANCER INSTITUTE
ANTONI VAN LEEUWENHOEK





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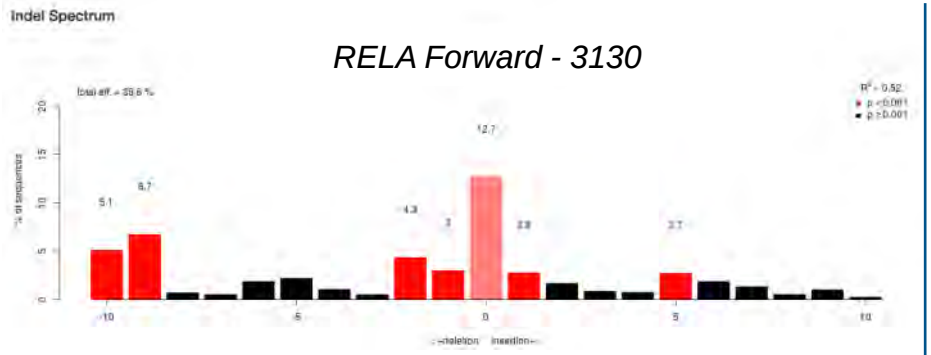
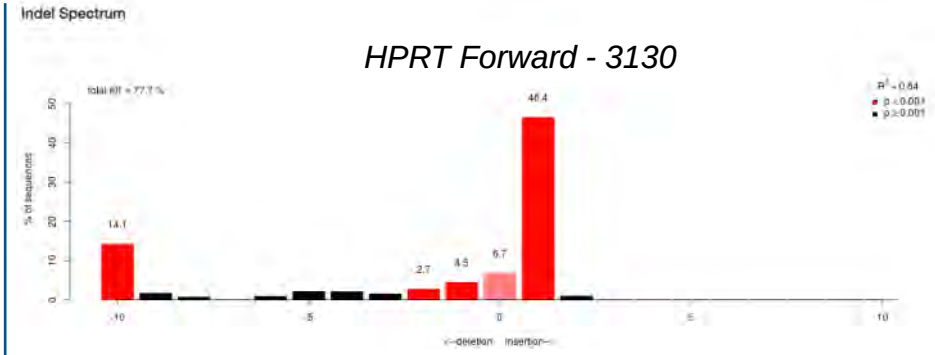
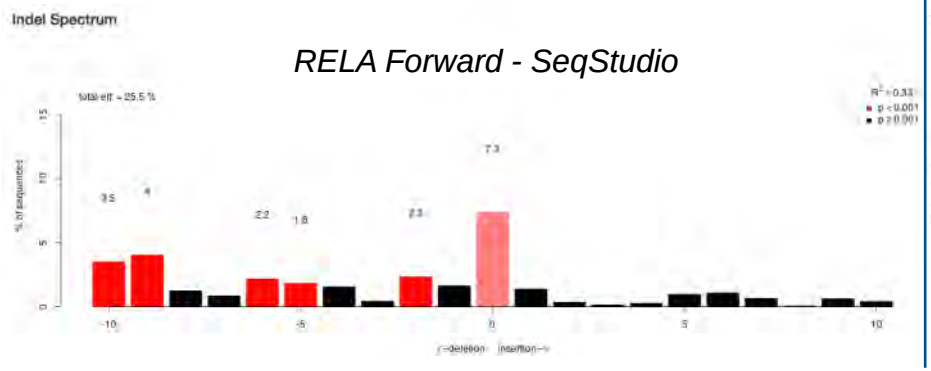
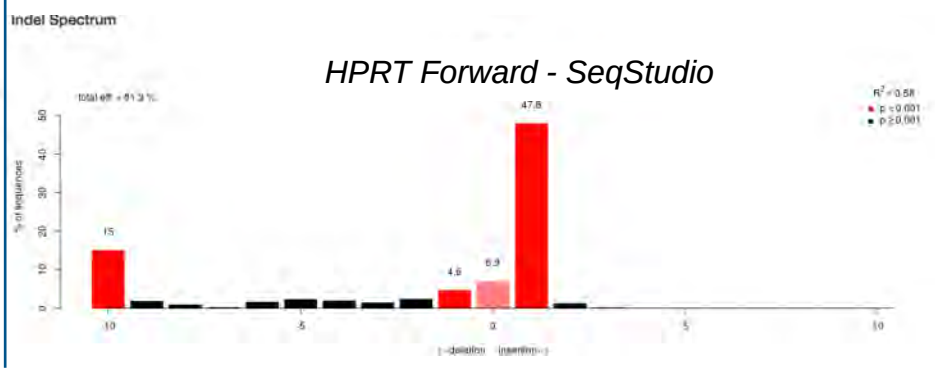
Sanger Sequencing in Genome Editing Workflow









- *Spectrum and frequency of mutations detected were similar among the two platforms*
- *Total efficiencies correlated well with other methods used to determine editing efficiency (genomic cleavage assay, TOPO subcloning and sequencing)*

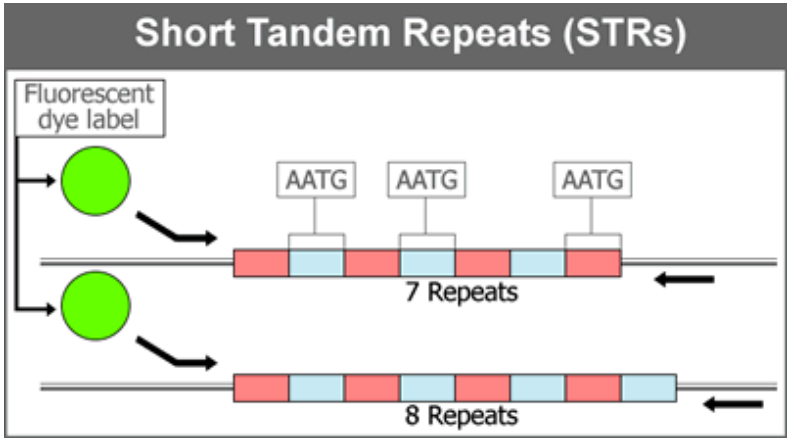
Cell Line Authentication on SeqStudio Genetic Analyzer

Problem

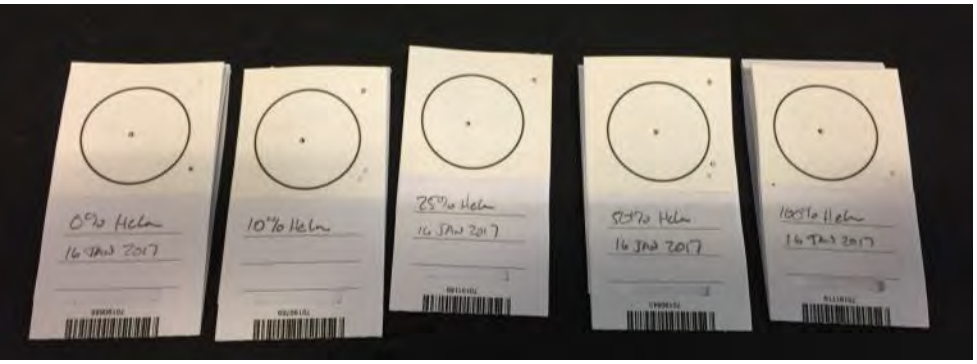
- Model cell lines are not always what investigators think they are
- Cell cultures might be mixed populations of different cell types
- iPS samples need to be matched after induction
- Human samples (Biobanks, milk repositories) need to be tagged and authenticated

Solution

- Non-HID analysis of STRs (short tandem repeats) by fragment analysis



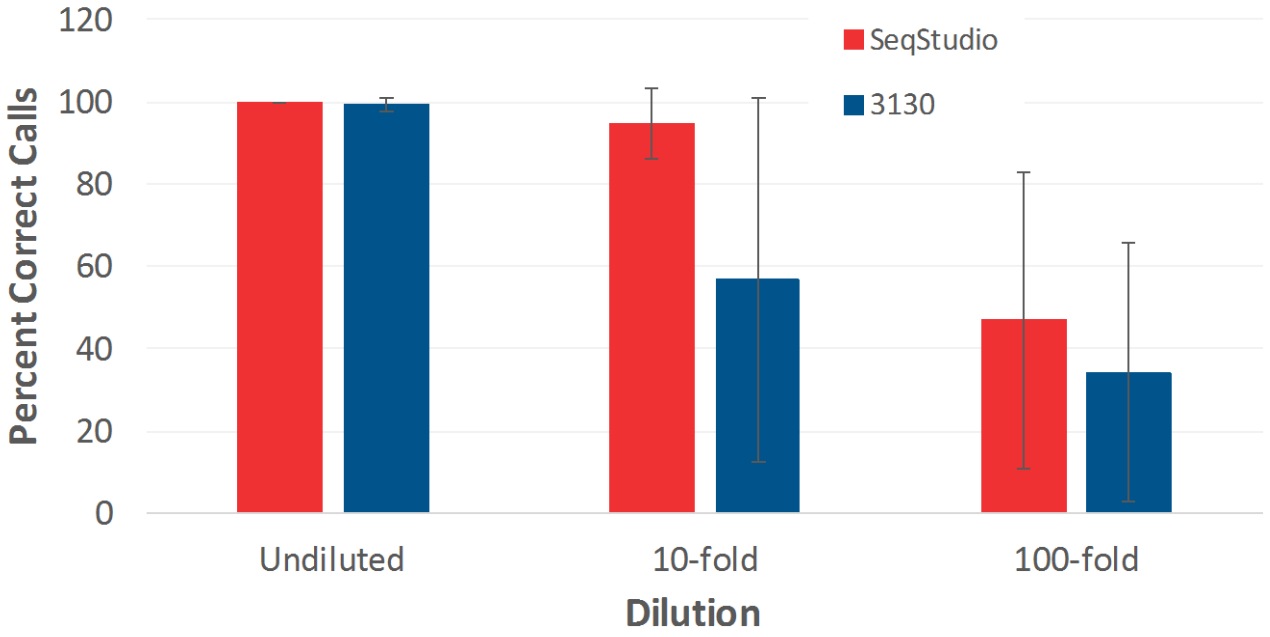
Cell Line Authentication with Identifiler Direct, NUCLEIC-CARDS



Experiment:

- Suspensions of cultured cells (starting concentration average 4x10^5 cells/ml) were spotted onto *NUCLEIC-CARDS*
- 1.2mm punches were collected and analyzed using Identifiler Direct kit
- Results analyzed with GeneMapper and compared with predicted allelic profile

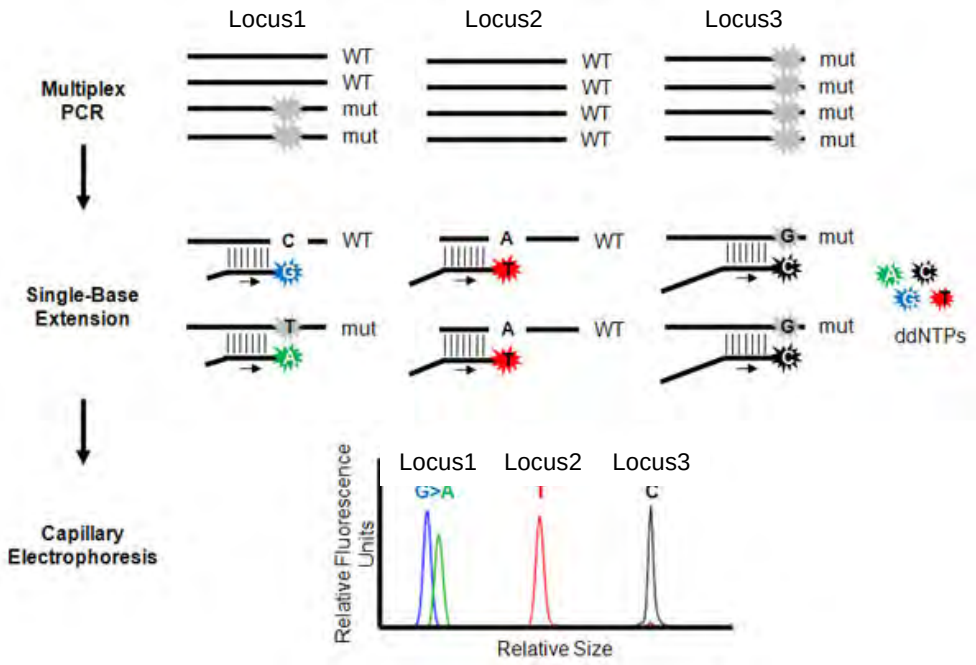
- *Correct profiles can be generated from around 5x10^5 cells/ml spotted onto NUCLEIC-CARDS*



Allelic Analysis using SNaPshot Multiplex Kit

Application	Sample Type	Seq. Chem.	Frag. Chem.	Module	Analysis Software
SNaP Shot	FFPE Tissue		SNaP Shot	SNaPshot	Gene Mapper

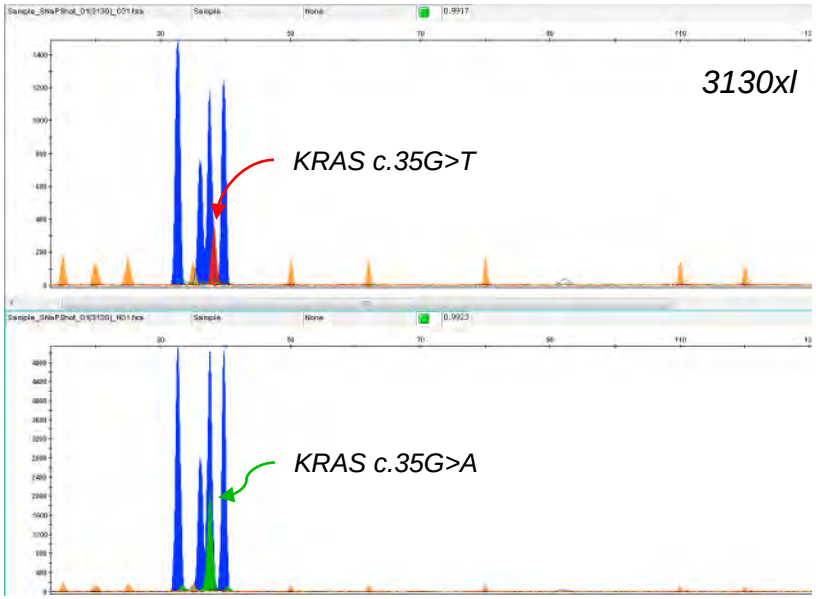
Used to screen and confirm SNPs, detect minor sequence variations, assess DNA methylation

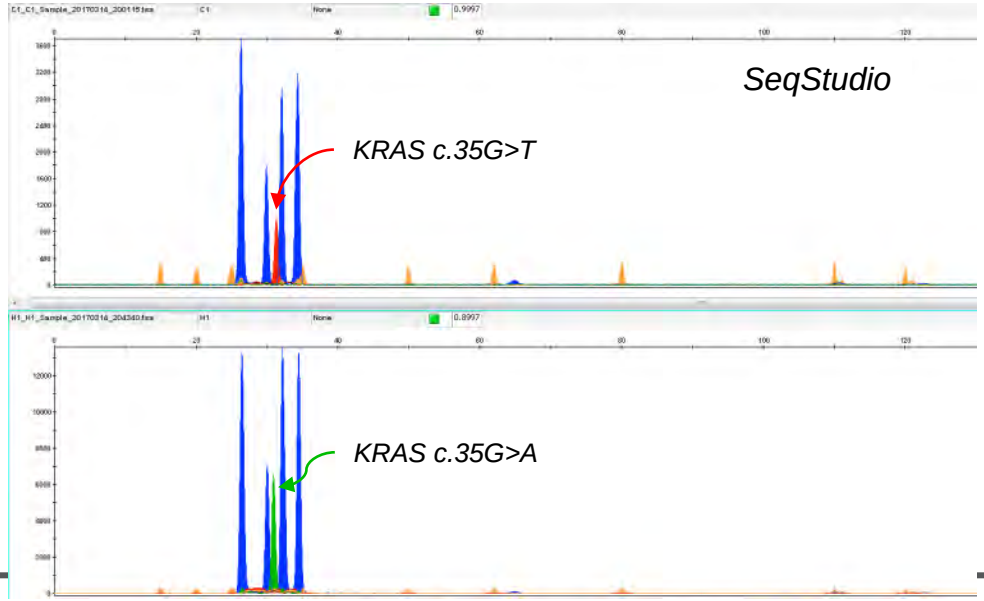


Experiment:

- SNaPshot primers ordered that query KRAS c.34, c.35, c.37 and c.38 (G12X or G13X)
- FFPE samples identified that contain mutations at indicated positions (verified by MVF and NGC)
- Extracted DNA was analyzed with SNaPshot Multiplex kit and Gene Mapper

Allelic Analysis using SNaPshot Multiplex Kit

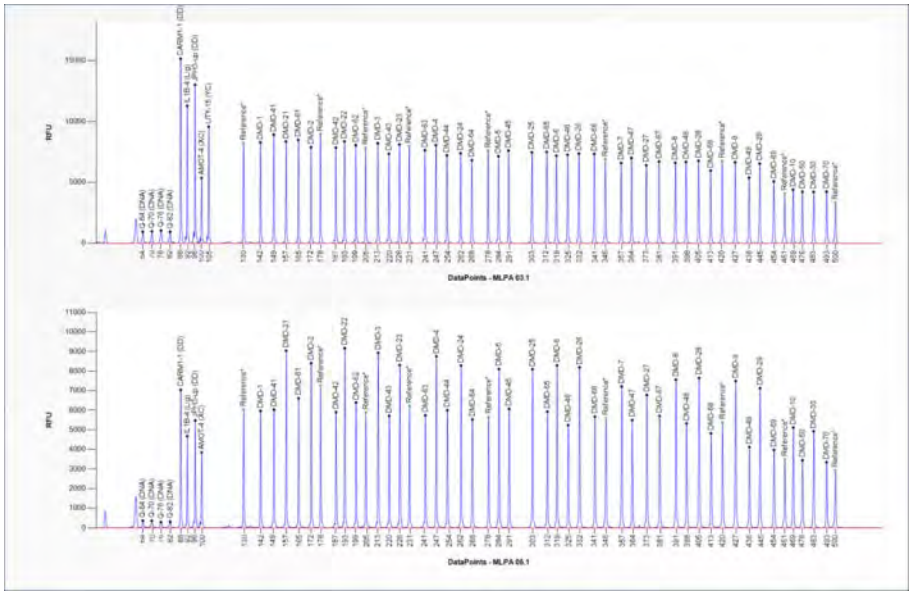




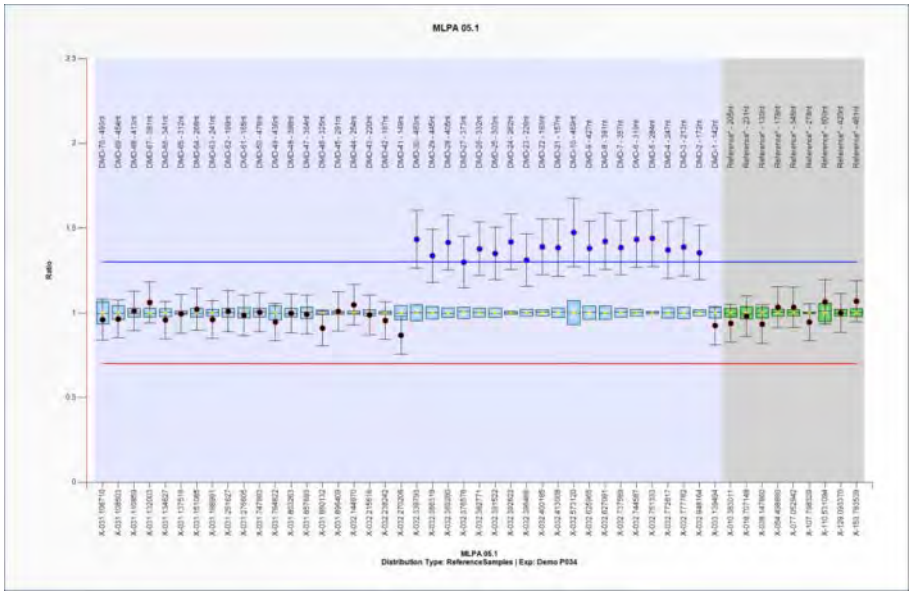
- blue peaks: wild-type alleles at KRAS c.34, c.35, c.37 and c.38.
- red peak (top graph): a KRAS c.35G>T mutation is also present
- green peak (bottom graph): a different allele is present at the same position in a different sample.
- Similar results seen with other samples.

- *Peak detection was equivalent on the two platforms*
- *Absolute migration position will be different, since polymers are different on the two platforms since polymers are different*

Inherited Disease Research by MLPA



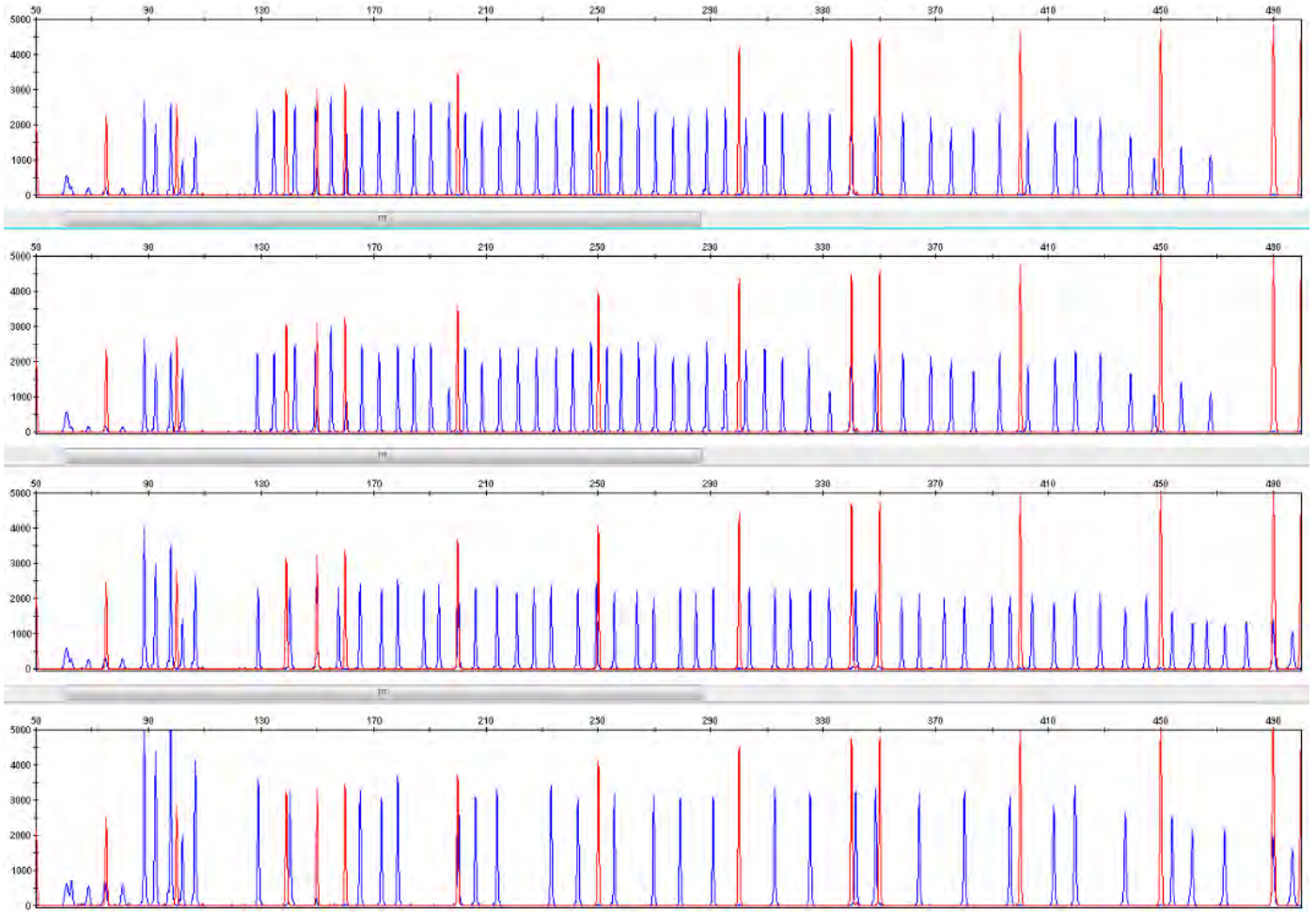
MLPA data analyzed by the Coffalyser.net software.
Upper panel: the annotated peak profile of a normal reference samples
Lower panel: proband test sample where a duplication is suspected.



Ratio chart showing analyzed peak heights of reference and proband sample.

Loci above the blue line are duplications of exon; loci below the red line are deletions.

Inherited Disease Research by MLPA



Courtesy of Jan Schouten, MRC Holland, Amsterdam (Booth 660)

Introducing the New SeqStudio Genetic Analyzer

- The new SeqStudio Genetic Analyzer provides you an **integrated experience** to put you back in control of your lab life
  - ***all-in-one cartridge*** for easy set up and reduced hands-on time
  - with the ***flexibility*** for both sequencing and fragment analysis in a single run.
  - ***Thermo Fisher Cloud-based connectivity options*** for remote monitoring, data transfer and analysis.
  - ***run time of as little as 30 minutes*** for fast turnaround time

- Get an all-new, **state of the art experience** in an incredibly affordable package.

Experience the SeqStudio at the Thermo Fisher Scientific booth!

Acknowledgements

Arpad Gerstner
Edgar Schreiber
Scott Higdon
Kamini Varma

Ion Torrent Applications team (Oncomine Focus results)

Luca Quagliata, University of Basel
Jan Schouten, MRC Holland

Exceptional Convenience: SeqStudio Innovative Consumables

Key Feature	Benefit
Integrated consumables cartridge containing capillaries, buffer, polymer, and array pump.	Easy to use, helps reduce instrument hands-on time & user error, sequencing and fragment analysis applications in the same run
4 Months on instrument cartridge stability	Efficient use of polymer and buffers for low throughput needs
Universal polymer for fragment analysis and sequencing	No need to change polymers for different applications
Remote consumables usage tracking	Monitor usage on consumable during, before and after runs from anywhere

SeqStudio Features Put You In Control of Your Work Time

Key Feature	Benefit
Instrument auto calibration	Helps reduce hands-on time; running time for spectral and spatial calibration reduced from hours to minutes
Integrated/ web based user interface	Removes the need for separate computer.
Interactive touch screen with graphical user interface	Intuitive and easy to use; no computer required
Connected ecosystem: remote monitoring and remote plate setup with Thermo Fisher Cloud	Set up and keep track of the run when outside of the lab
Small Footprint (49.5 x 64.8 x 44.2 cm W x D x H)	Takes up less space in the lab than older genetic analyzers
Accepts 96-well standard plate and 8-well strip tubes	Accommodates multiple sample formats for your CE needs