



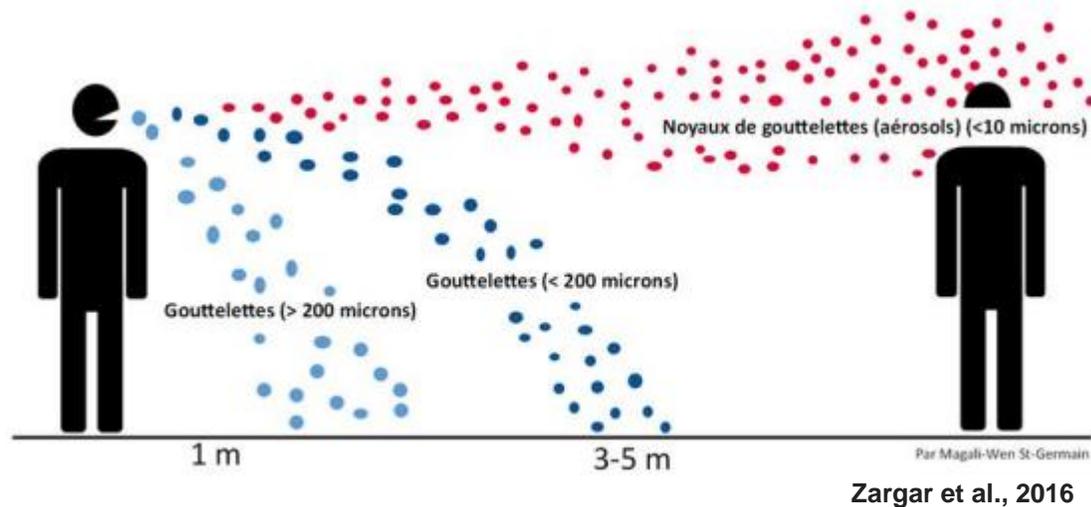
## « Applications du NGS, du microbiome au metabarcoding »

*Le pouvoir de notre solution AmpliSeq au service  
d'Ion Torrent*

Bouchera DOUAH & Emeric ROUX  
Product Specialist NGS France

 The world leader in serving science

# Precept of the composition of the air



Covid crisis has further highlighted the presence of **airborne microorganisms**

Highlighting also **the importance of daily mask use** during this outbreak

## Le niveau de filtration des différents masques en France

Le port de certains masques en tissu est déconseillé et la distance de sécurité passe de un mètre à **deux mètres** entre chaque personne\*\*



# Bioaerosol and Surface Analysis in Subway Station



Article

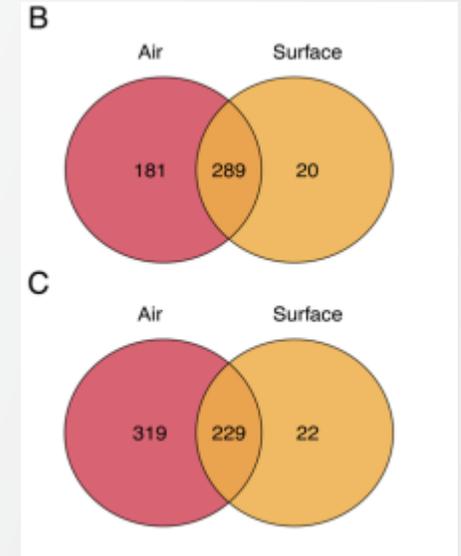
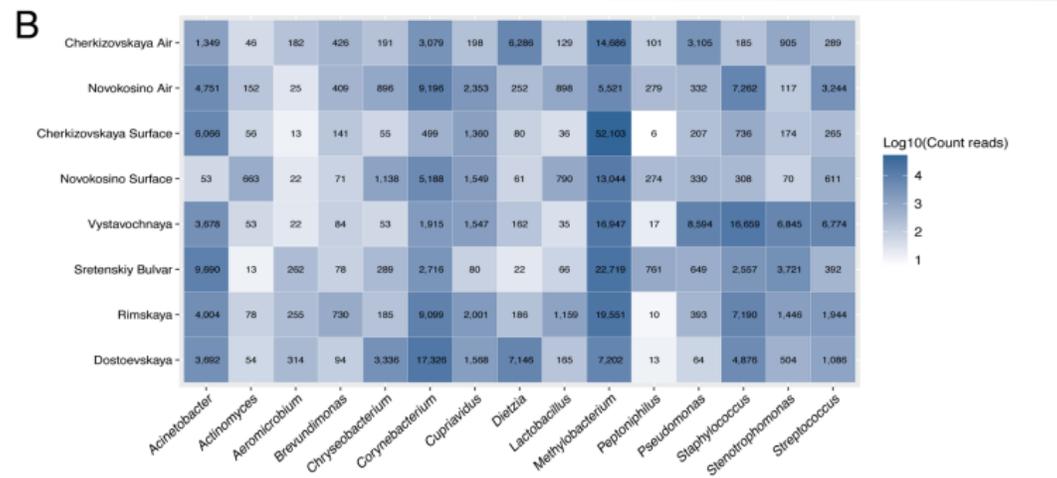
## Microbiological Characteristics of Some Stations of Moscow Subway

Andrei A. Pochtovyi <sup>1,2,\*</sup>, Daria V. Vasina <sup>1</sup>, Bakhtiyar I. Verdiev <sup>1</sup>, Alexey M. Shchetinin <sup>1</sup>, Anton G. Yuzhakov <sup>1,3</sup>, Roman S. Ovchinnikov <sup>1,4</sup>, Artem P. Tkachuk <sup>1</sup>, Vladimir A. Gushchin <sup>1,2,\*</sup> and Alexander L. Gintsburg <sup>1,5</sup>

<sup>1</sup> Federal State Budget Institution "National Research Centre for Epidemiology and Microbiology Named after Honorary Academician N.F. Gamaleya" of the Ministry of Health of the Russian Federation, 123098 Moscow, Russia; d.v.vasina@gmail.com (D.V.V.); yuryevpolskei@yandex.ru (B.I.V.); shchetinin.am@gmail.com (A.M.S.); anton\_oskol@mail.ru (A.G.Y.); rsovchinnikov@mail.ru (R.S.O.); artem.p.tkachuk@gmail.com (A.P.T.); gintsburg@gamaleya.org (A.L.G.)

In this study, they compared the composition of the **microbiome in the air compared to the microbiome on the surfaces of the metro.**

performed using the  
**Ion 16S™**  
**Metagenomics Kit**



They focused on **two different stations** of the Moscow. The obtained data indicated the **formation of a core microbiome at each station**, depending on the characteristics of the station and the level of passenger traffic. It also identified the distribution of microorganisms associated with human microflora, **which are not susceptible to seasonal fluctuations.**



AerosolSense by ThermoFisher Scientific

# The Foundation of Ion Torrent NGS Solutions



Gene panels or  
exomes

As low as **1 ng**  
DNA and RNA

**Transcriptome**  
and gene  
**expression**

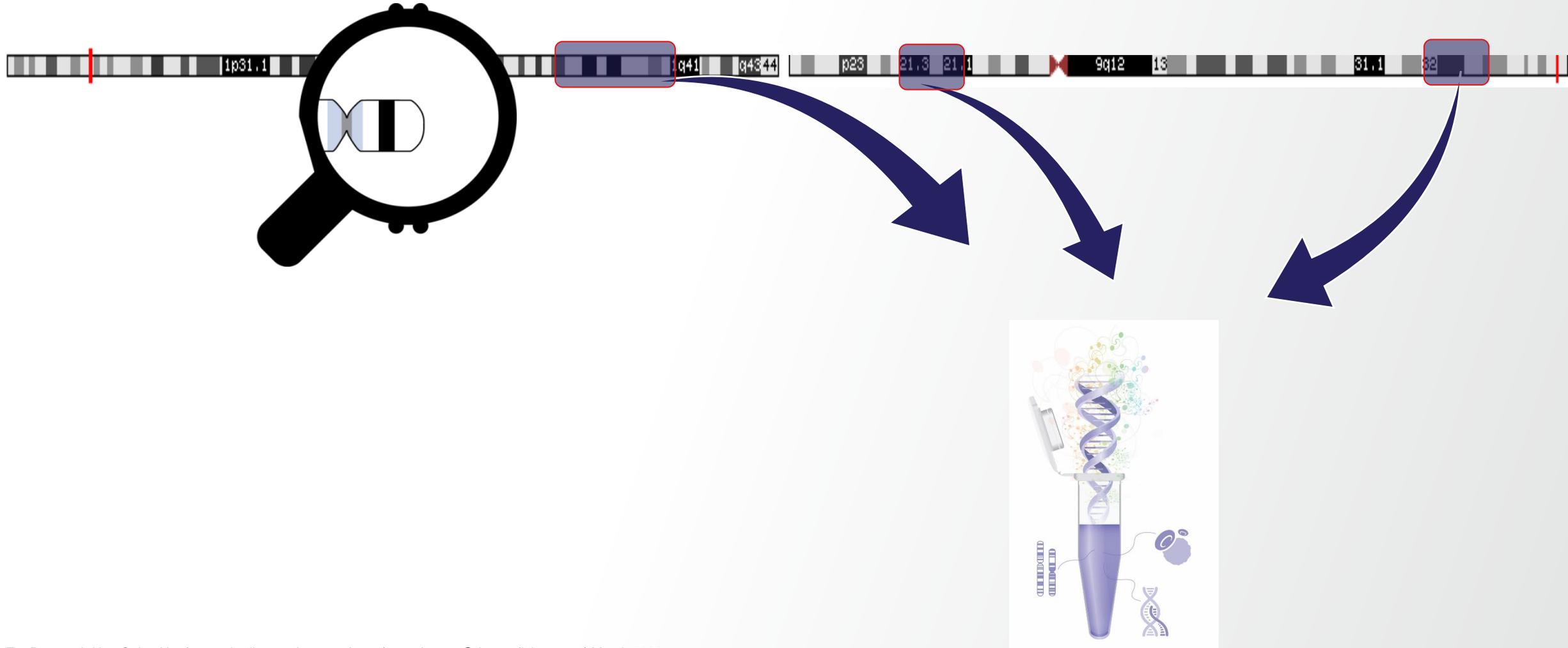
**Multiple**  
applications

SNPs,  
indels,  
CNVs, fusions

**Single-tube**  
workflow

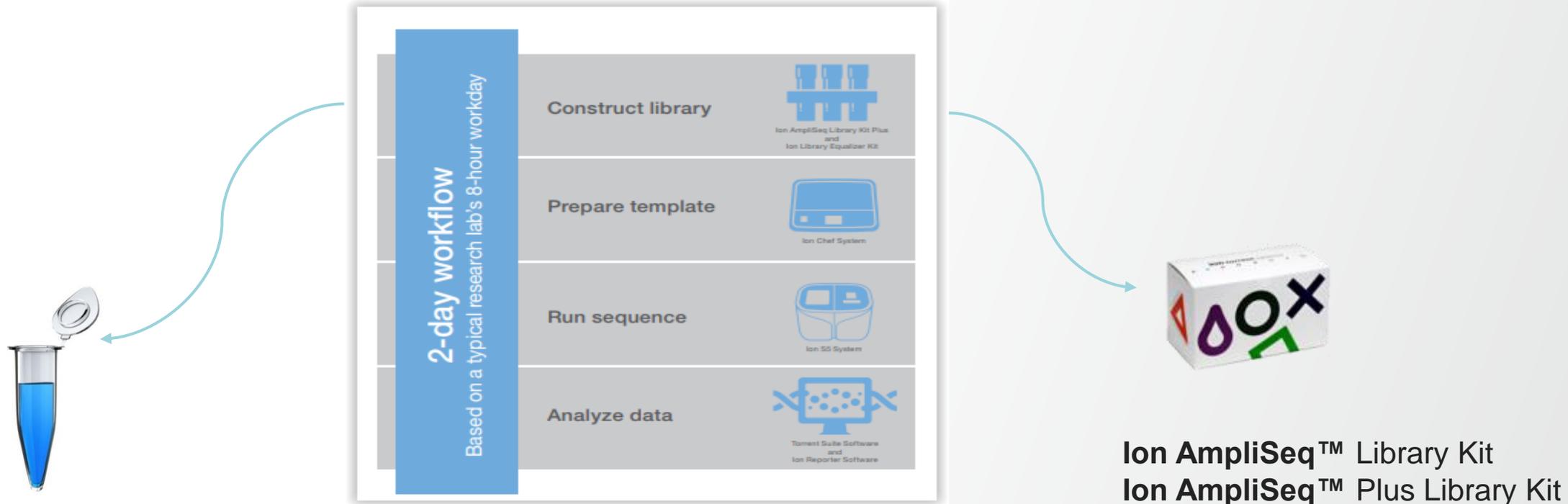
# What is Targeted Sequencing?

**Targeted Sequencing** isolates and **focuses** your sequencing on **specific genes or genomic regions** of interest rather than surveying the whole genome.



# Ion AmpliSeq™ Technology: As Simple As PCR

Single-tube, ultra-high multiplex for targeted resequencing



**12-24 576 primers** pairs per tube

*Ion AmpliSeq™ Custom Panels*  
OR  
*Ion AmpliSeq™ Ready-to-Use Panels*

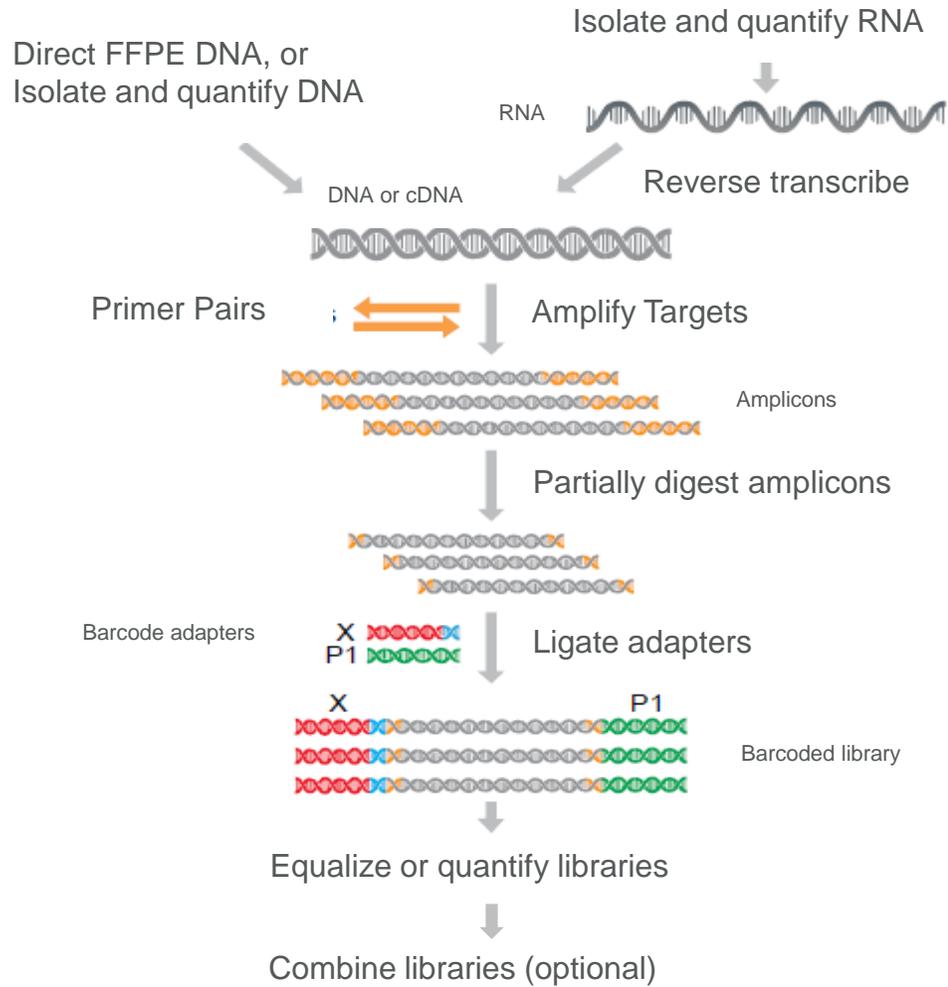
+



+

**1-100 ng DNA** per tube

# Targeted Sequencing Ciblé- Ion AmpliSeq Technology



- Custom Designs with **any Genome**
- As little as **1-100 ng** of input DNA, enabling FFPE analysis
- Rapid results with 3-5-hour manual library construction
- Library construction **automation** on the Ion Chef instrument



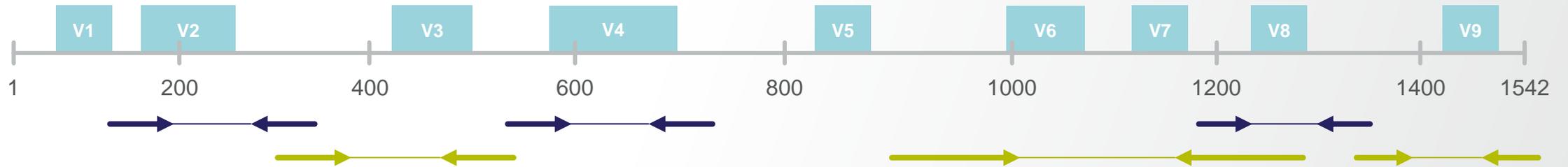
# Ion 16S Metagenomics Kit

## Primer sets

- Set V2-4-8
- Set V3-6,7-9



## 16S gene



- **Stringent Bioinformatic Assay Design:** Analyses against the entire GreenGenes database as well as the 26K species of primary metagenomic interest.
- **Broad Coverage Primer Sets:** Addresses multiple groups of organisms using multi-amplicon approach (amplicon sizes 200–300bp range)
- **Maximum Read Length:** Uses 400bp sequencing chemistry

# Study of SarsCov2 mutation in WasteWater



OMICS DATA SETS



## Genome Sequencing of Wastewater Confirms the Arrival of the SARS-CoV-2 Omicron Variant at Frankfurt Airport but Limited Spread in the City of Frankfurt, Germany, in November 2021

Shelesh Agrawal,<sup>a</sup> Laura Orschler,<sup>a</sup> Simona Tavazzi,<sup>b</sup> Robert Greither,<sup>c</sup> Bernd Manfred Gawlik,<sup>b</sup> Susanne Lackner<sup>a</sup>

<sup>a</sup>Technical University of Darmstadt, Department of Civil and Environmental Engineering Sciences, Institute IWAR, Chair of Water and Environmental Biotechnology, Darmstadt, Germany

<sup>b</sup>European Commission, Joint Research Centre, Ispra, Varese, Italy

<sup>c</sup>Life Technologies, Darmstadt, Germany

In November 2021, they found all characteristic mutations of Omicron in wastewater originating from Frankfurt Airport before the first confirmed clinical report from an arriving passenger on 26 November 2021.

the library was prepared using the **Ion AmpliSeq SARS-CoV-2 research panel** on Ion S5 Sequencer

They show that **the surveillance of wastewater** from transportation hubs, such as airports, using sequencing can support **tracking of the import of SARS-CoV-2 variants.**

TABLE 1 Summary of the sequencing data for the samples

Sample location and collection date (day-mo-yr)	Total no. of reads	No. of mapped reads	Avg target base coverage depth (X)	Avg read identity vs target (%) <sup>a</sup>	GC content (%)	BioSample accession no.	SRA accession no.
<b>Airport</b>							
4-11-2021	1,169,001	345,149	319.5	95.61	62.9	SAMN24156791	SRR17258655
23-11-2021	8,299,055	3,245,669	9,693	98.36	55.8	SAMN24156792	SRR17258654
<b>City</b>							
4-11-2021	1,972,935	620,663	753.4	98.72	62.2	SAMN24156793	SRR17258653
23-11-2021	9,908,030	3,825,450	11,834	98.02	56.8	SAMN24156794	SRR17258652

<sup>a</sup> The target sequence was the SARS-CoV-2 reference genome (Wuhan-Hu-1 [GenBank accession numbers NC\_045512 and MN908947.3]).

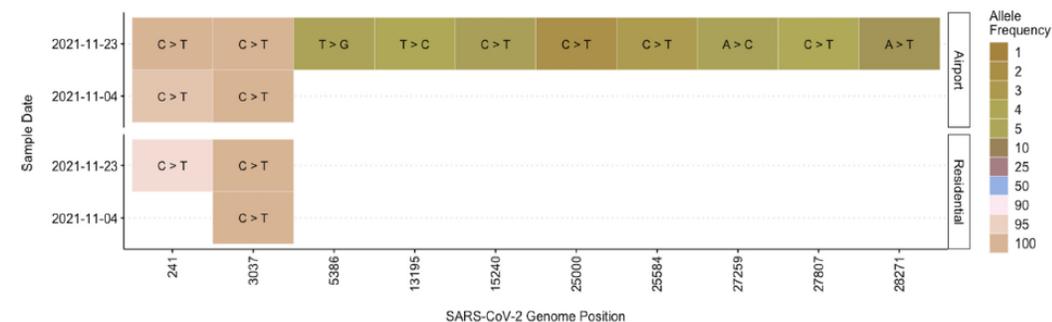


FIG 1 (Top) Occurrence (i.e., the numbers of reads corresponding to each mutation) of characteristic S, E, M, and N protein mutations and ORF1ab mutations of the Omicron variant. (Bottom) Heatmap showing the allele frequency and the alternate nucleotide of the characteristic synonymous mutations of the Omicron variant. Airport, wastewater sample with a significant fraction from Frankfurt Airport; Residential, influent wastewater sample collected from the WWTP of Frankfurt City.

# Ion AmpliSeq SARS-CoV-2 Insight Research Panel

**Higher sensitivity** for earlier identification of variants in **low titer samples (Ct>28)** to inform public health guidance and intervention strategies

- **237 amplicons specific** to SARS-CoV-2 with **addition of variant-tolerant primers for emerging variants**
- **5 newly-designed human expression controls** for sample **confirmation** and **quality** assessment
- Amplicon length range: **125-275 bp**
- **>99% coverage** of SARS-CoV-2 genome (~30kb)
- **All potential serotypes covered**



- **Increase productivity** by running **more samples** per chip with the insight panel

Chip type	Samples*/chip
510	2-3
520	4-6
530	16-32
540	80-160
550	130-200

\* We recommend that you allow 500,000–1,000,000 reads per sample for the Ion AmpliSeq SARS-CoV-2 Insight Research Panel dependent on sample quality

# Ampliseq Designer tool: custom panel design

**i** Community Ion AmpliSeq SARS-CoV-2 Insight Research Assay now available to support COVID-19 research

[Getting Started with AmpliSeq](#)

## In stock

Ships in 4 – 10 working days; fewer reactions **i**

[Check gene availability](#)

## Made-to-order

Ships in 2 – 3 weeks; more reactions **i**

Product and description	Oncomine™ tumor specific panels <b>New!</b>	Ion AmpliSeq™ On-Demand panels	Ion AmpliSeq™ Custom panels	Ion AmpliSeq™ HD Custom panels
	Select from catalog of solid tumor panels from > 301 pretested genes and fusion drivers	Select from catalog of > 5,000 pre-tested genes	Customizable for any genome	Customizable with ultrahigh allelic sensitivity for applications including liquid biopsy
	<a href="#">Browse tumor types</a>	<a href="#">Create On-Demand panel</a>	<a href="#">Create Custom panel</a>	<a href="#">Create Custom HD panel</a>
	<a href="#">Or Add gene list</a>	<a href="#">Or browse disease research areas</a>		
Genome	Human	Human	Any	Human
Nucleic acid	DNA / RNA	DNA only	DNA / RNA	DNA / RNA
Sample type	FFPE	Genomic DNA	Any	cfDNA / FFPE
Target type	Genes	Genes	Genes, regions, hotspots, amplicons, expression, fusions	Genes, regions, hotspots, amplicons, fusions
Limit of detection <b>i</b>	5%	5 – 20%	5%	0.1%
Pre-curated <b>i</b>	●	●		
Pre-tested <b>i</b>	●	●		

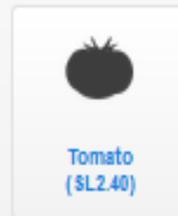
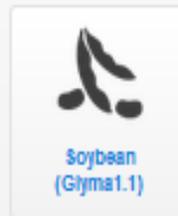
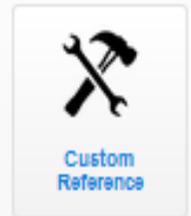
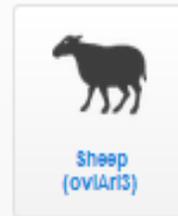
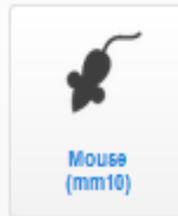
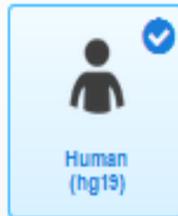
# AmpliSeq™ technology: Chose target on any Genome

## 2 Application type \*

DNA designs may include a mix of gene and region targets, including small hotspot regions.



## 3 Select genome to use \*



# Metabarcoding AmpliSeq Custom

SHORT REPORT

Open Access



## Metabarcoding reveals low prevalence of microsporidian infections in castor bean tick (*Ixodes ricinus*)

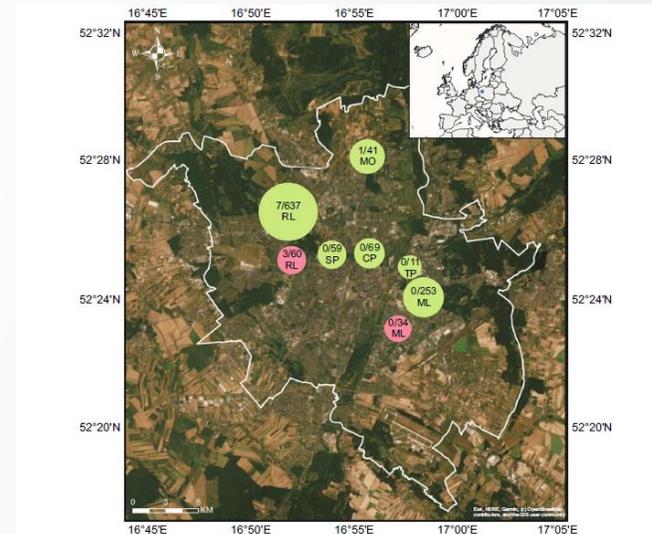
Artur Trzebny<sup>1\*</sup>, Justyna Liberska<sup>1</sup>, Anna Slodkiewicz-Kowalska<sup>2</sup> and Mirosława Dabert<sup>1</sup>

in this study, they applied a recently developed DNA metabarcoding method for efficient microsporidian DNA identification to assess the role of ticks as potential vectors of microsporidian species causing diseases in humans.

They did a **DNA barcode** covering 322 bp of the COI Gene for sequencing **using the Ion Torrent system**. We totally can imagine design this panel with the help of our **AmpliSeq Designer** and our Bio – informatic Team.

**Table 2** DNA sequences used in phylogenetic analysis

Microsporidium species	Host species	Host class	GenBank no	References
<i>Endoreticulatus bombycis</i>	<i>Bombyx mori</i>	Insecta	AY009115	[41]
<i>Endoreticulatus itili</i>	<i>Listronotus bonariensis</i>	Insecta	KJ755828	[42]
<i>Endoreticulatus poecilimonae</i>	<i>Poecilimon thoracicus</i>	Insecta	KJ755827	[43]
<i>Endoreticulatus schubergi</i>	<i>Lymantria dispar</i> , <i>Hyphantria cunea</i> , <i>Choristoneura fumiferana</i>	Insecta	L391109	[44, 45]
<i>Endoreticulatus</i> sp. CHW-2004 Bulgaria	<i>Lymantria dispar</i>	Insecta	AY502945	[46]
<i>Endoreticulatus</i> sp. CHW-2004 Taiwan	<i>Ocinara lida</i>	Insecta	AY502944	[46]
<i>Endoreticulatus</i> sp. CHW-2008 Austria	<i>Thaumetopoea processionea</i>	Insecta	EU260046	[47]
<i>Endoreticulatus</i> sp. isolate JMM 2007	<i>Loxostege sticticalis</i>	Insecta	M8929470	[48]
<i>Endoreticulatus</i> sp. Melnik	<i>Euproctis chrysothoea</i>	Insecta	KU900486	[43]
<i>Endoreticulatus</i> sp. PL01	<i>Ixodes ricinus</i>	Arachnida	MT1911425	This study
<i>Endoreticulatus</i> sp. Shengzhou	<i>Bombyx mori</i>	Insecta	JN792450	[49]
<i>Endoreticulatus</i> sp. Sofia	<i>Euproctis chrysothoea</i>	Insecta	KU900485	[43]
<i>Endoreticulatus</i> sp. Zhenjiang	<i>Bombyx mori</i>	Insecta	FJ772431	[50]
<i>Endoreticulatus</i> sp. isolate EC	<i>Eilema complana</i>	Insecta	KY615713	[51]
Microsporidium sp. clone Chula Myositis 1	<i>Homo sapiens</i>	Mammalia	JN619406	[52]
<i>Cystosporogenes legeri</i>	<i>Lobesia botrana</i>	Insecta	AY233131	[53]
<i>Cystosporogenes operophterae</i>	<i>Operophtera brumata</i>	Insecta	AJ302320	[54]
<i>Cystosporogenes</i> sp. GKK-2009	<i>Agrilus anxius</i>	Insecta	GQ379703	[55]
<i>Cystosporogenes</i> sp. KCI-2	<i>Operophtera brumata</i>	Insecta	GU299511	[56]
<i>Chytridiopsis typographi</i>	<i>Ips typographus</i>	Branchiopoda	MH728789	[57]



**Fig. 1** Sampling places of unfed host-seeking ticks (red circles) and feeding (green circles) ticks in the city of Paznan (blue dot on the Europe map). The size of circle corresponds to the number of analyzed ticks. Numeric value inside the circles: number of microsporidian-positive ticks/number of analyzed ticks. For sampling place characteristics, see Additional file 1: Table S1 and Additional file 2: Table S2

Our data suggest that *I. ricinus* is **not involved** in transmission of zoonotic microsporidian species. Moreover, we noticed a **very low prevalence** of these parasites in the tested ticks

# Equipments



# Combined with Novel Innovation in Library Preparation Ion Torrent Platform



## On Demand: Flexibility



### Ion AmpliSeq™ technology

Foundational technology  
for targeted sequencing

## Made to order: Sensitivity



### Ion AmpliSeq™ HD technology\*

Novel core technology for next-  
generation targeted sequencing

The content provided herein may relate to products that have not been officially released and are subject to change without notice.

# Flexible Portfolio Configurable to Your Needs

Ion GeneStudio™ S5



**Fast.**

Ion GeneStudio™ S5 Plus



**Flexible.**

Ion GeneStudio™ S5 Prime



**Powerful.**



**Ion 510™ Chip**  
2–3 M reads  
Up to **400 bp**



**Ion 520™ Chip**  
3–6 M reads  
Up to **600 bp**



**Ion 530™ Chip**  
15–20 M reads  
Up to **600 bp**

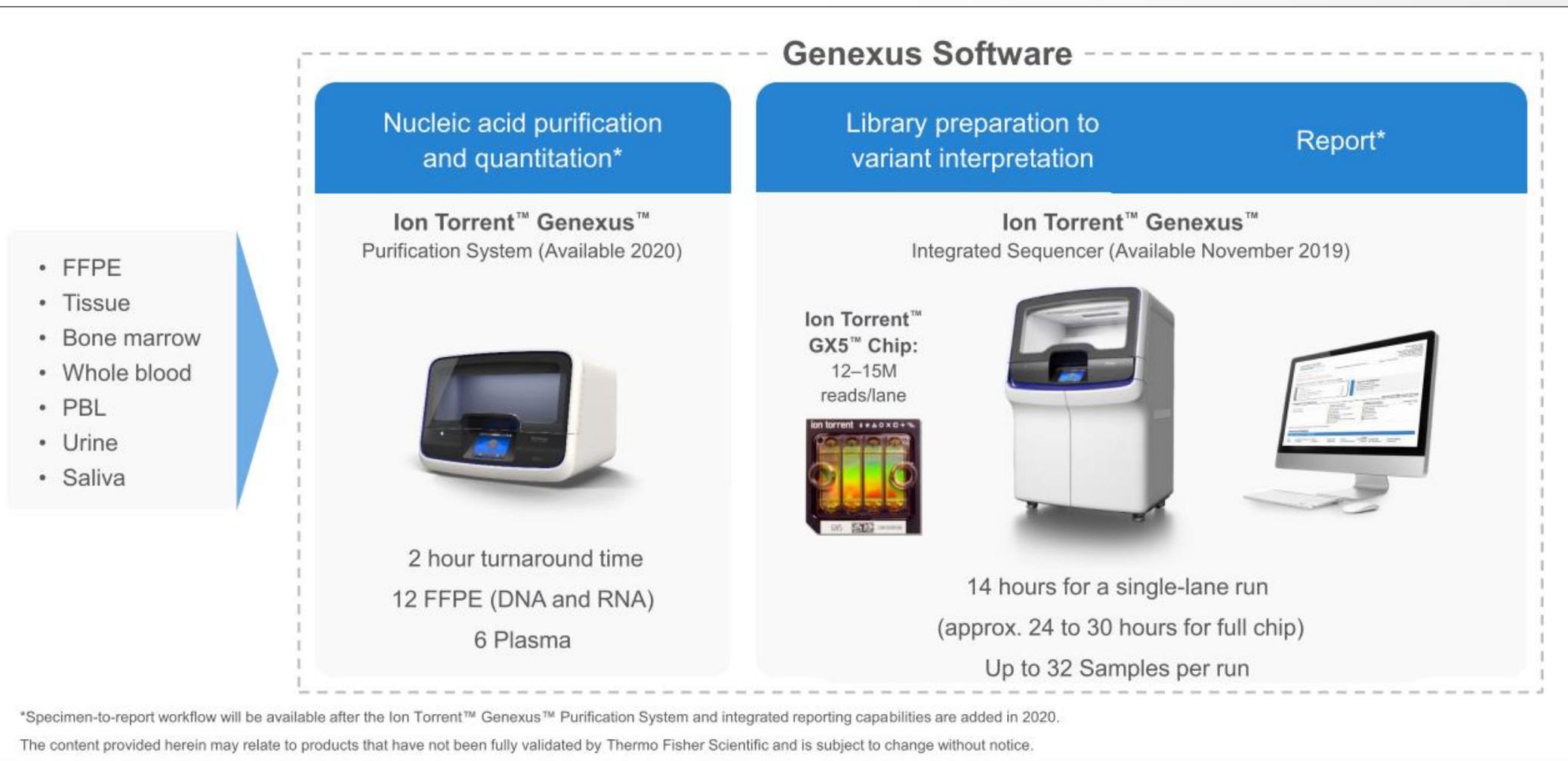


**Ion 540™ Chip**  
60–80 M reads  
Up to **200 bp**



**Ion 550™ Chip**  
100–130 M reads  
Up to **200 bp**

# Genexus System End to End Workflow



# We are here for you !!!



- Bouchera DOUAH  
[Bouchera.douah@thermofisher.com](mailto:Bouchera.douah@thermofisher.com)  
Tel :+33 (0) 6 16 22 04 04  
[Bouchera DOUAH | LinkedIn](#)



- Emeric ROUX  
[Emeric.roux@thermofisher.com](mailto:Emeric.roux@thermofisher.com)  
+33 (0) 7 87 20 84 38  
[Emeric ROUX | LinkedIn](#)





Thank you

**ThermoFisher**  
S C I E N T I F I C

For Research Use only. Not for use in diagnostic procedures.

© 2020 Thermo Fisher Scientific Inc. All rights reserved. All trademarks are the property of Thermo Fisher Scientific and its subsidiaries unless otherwise specified.

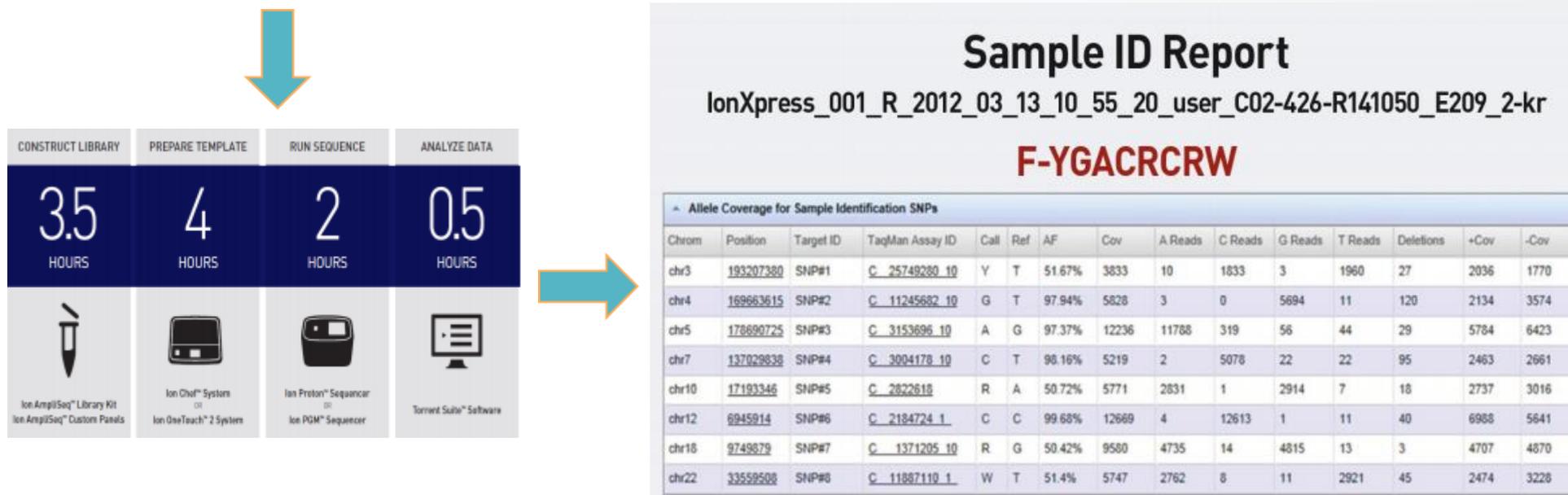
# Strain-Typing and Antibiotic Resistance Profiling

Strain-Typing and Antibiotic Resistance Profiling From Research Samples Using Highly Multiplexed Targeted Library Construction with High Throughput Semi-Conductor Based Sequencing.

John Bishop, Loni Pickle, Miro Dudas, Guoying Liu, Melanie Baumann, and Astrid Ferlinz

Thermo Fisher Scientific, Carlsbad, CA, USA

- They want to identify **Mycobacterium** from culture and sputum samples to **genotype specific loci** and antibiotic Resistance in **M. Tuberculosis**



“We demonstrated this process with a panel specific [Ion AmpliSeq™ TB Research Panel](#) to full gene coverage of eight known antibiotic resistance loci in M. tuberculosis, however this process is scalable and extensible to simultaneously assess thousands of loci across multiple organisms per sample”

