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**Proyecto “Implementación de tecnologías de última generación para detección simultánea de múltiples patógenos a gran escala, aplicada a la vigilancia fitosanitaria y diagnóstico temprano de enfermedades en Arándano y Yuca”**



**Oxford Nanopore Lima-Perú Octubre-2022**



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## Grupo de Virología y Protección de cultivos

# Introducción: Secuenciación Nanopore



**Ana Maria Leiva Sandoval**  
**Dr. Wilmer Cuellar**

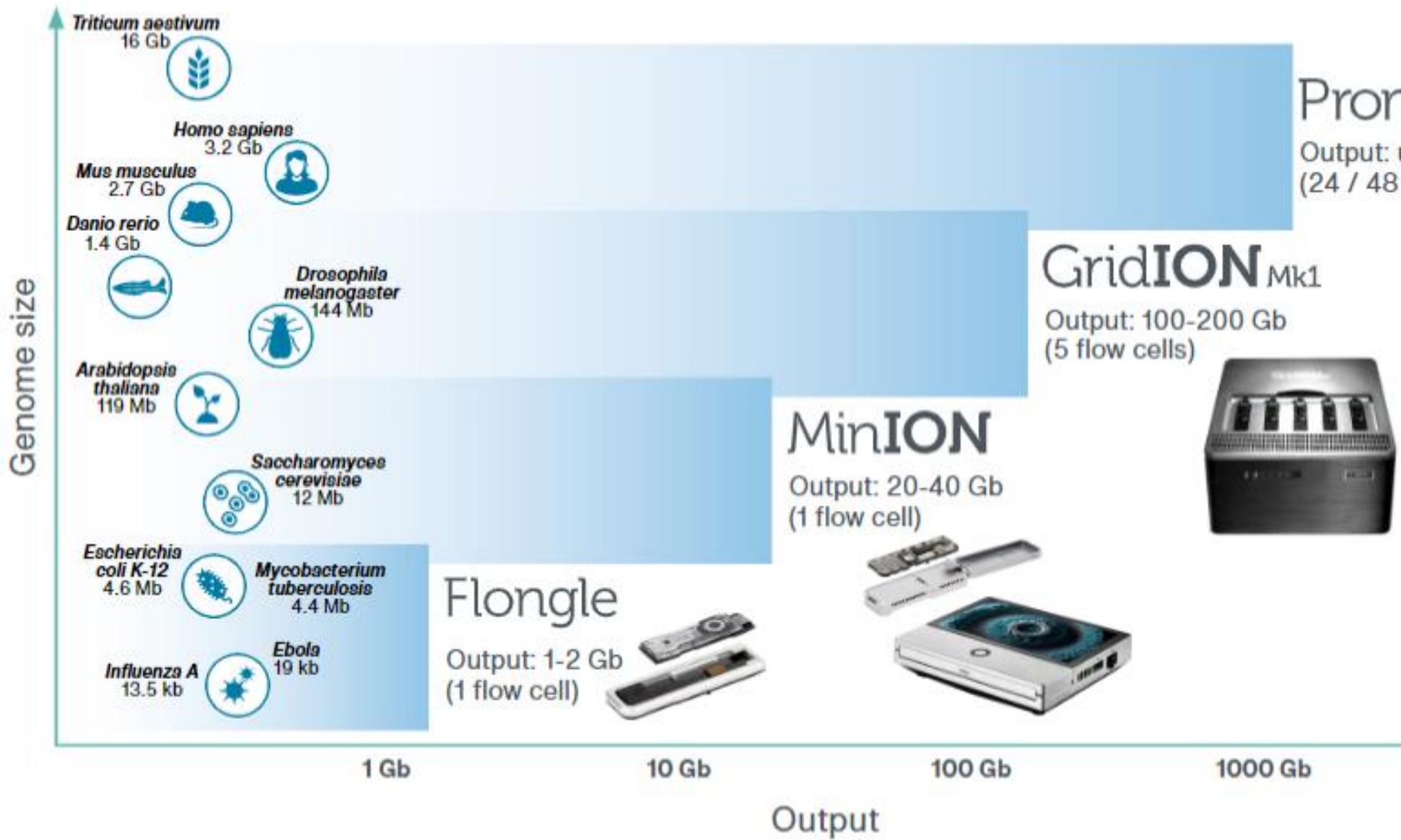
Lima-Perú Octubre-2022

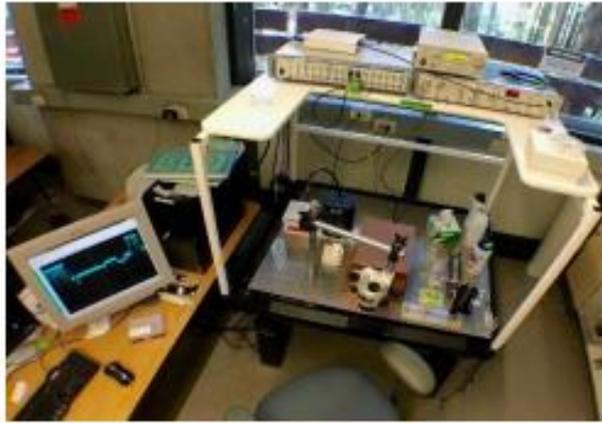
La secuenciación de próxima generación (NGS, por sus siglas en inglés) es el término general utilizado para describir una serie de tecnologías modernas de secuenciación masiva de alto rendimiento.

Las tecnologías permiten la secuenciación de ADN y ARN mucho más rápidamente que la secuenciación de Sanger por tanto revolucionaron el estudio de la genómica y la biología molecular.

# Tecnologías de secuenciación

	Tecnologia	Enfoque	Read	objetivos	Ideal para:
1 <sup>st</sup> Generación (Sanger)	Applied Biosystems 	Sequencing by synthesis (SBS) -baja eficiencia	1 kb	Secuenciación PCR. Usada para primer genoma humano	PCR
2 <sup>nd</sup> Generación	Illumina (IonTorrent) 	SBS - masivo -alta eficiencia -alto costo	100-150 bp	Secuenciación económica y rápida de millones de genomas	WGS Metagenomas
3 <sup>rd</sup> Generación	Oxford Nanopore (PacBio) 	-Secuenciación de una sola molécula (SMS) -Alta eficiencia	100 bp – 500 Mbp	Secuenciación portatil/ reads Largos. Bajo costo	PCR-WGS Metagenomes





MinION  
Flow Cell



MinION  
device



PromethION  
Flow Cell



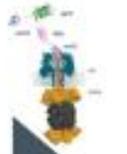
P48 device  
Runs up to 48  
Flow Cells



Pipeline of lower  
power and higher  
density  
flow cells



Protein and other  
molecule sensing  
on same  
hardware



channel

**Axopatch**  
1 Nanopore channel  
~1 experiment per person per week



up to

channels

**MinION**  
512 nanopore sequencing channels  
Running in parallel



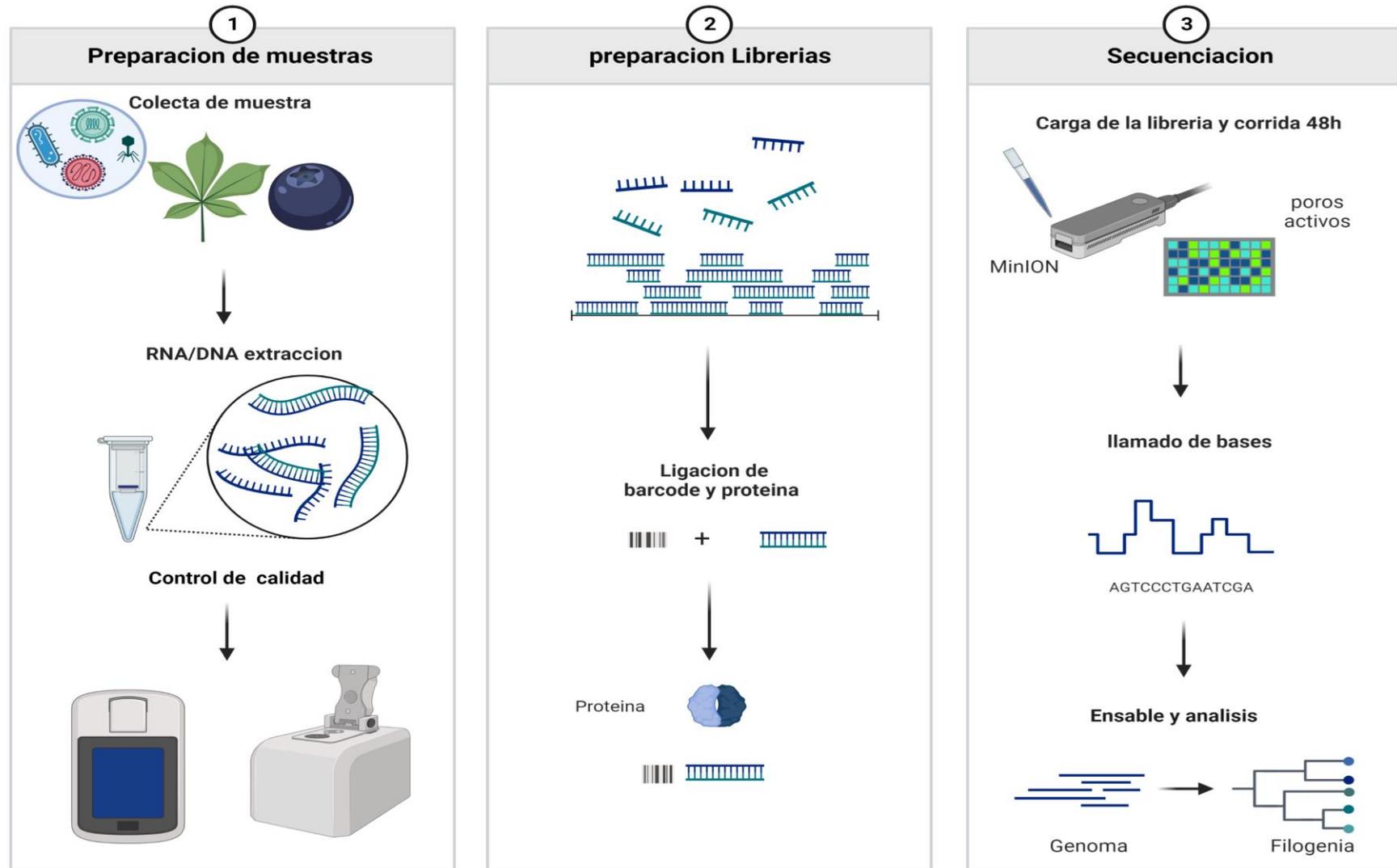
up to

channels

**Range of devices**  
from 126 to  
128,400 channels

# Flujo de Trabajo

## Secuenciación usando tecnología Oxford Nanopore



# Partes de la Flowcell





# Secuenciador Portatil



**Todo ser vivo**



**Cualquier Persona**

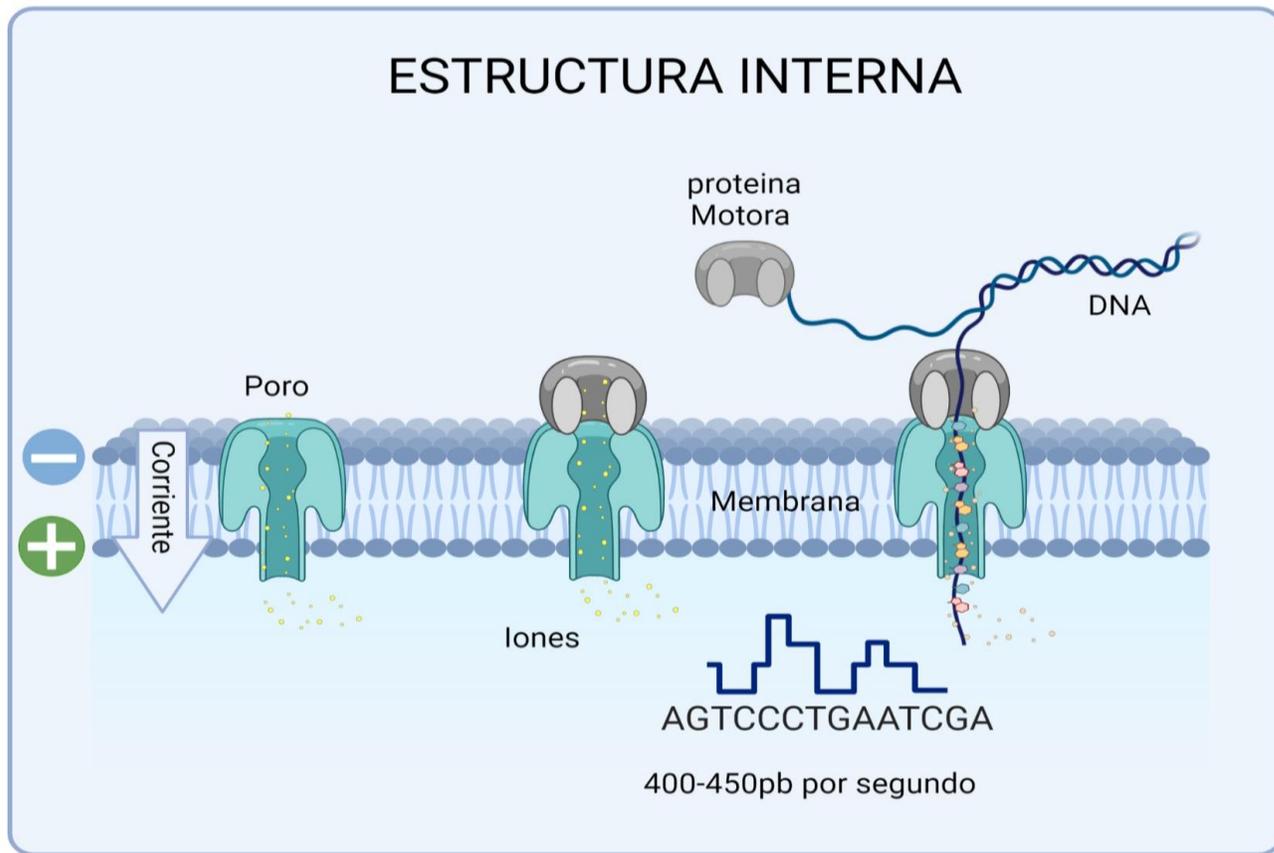
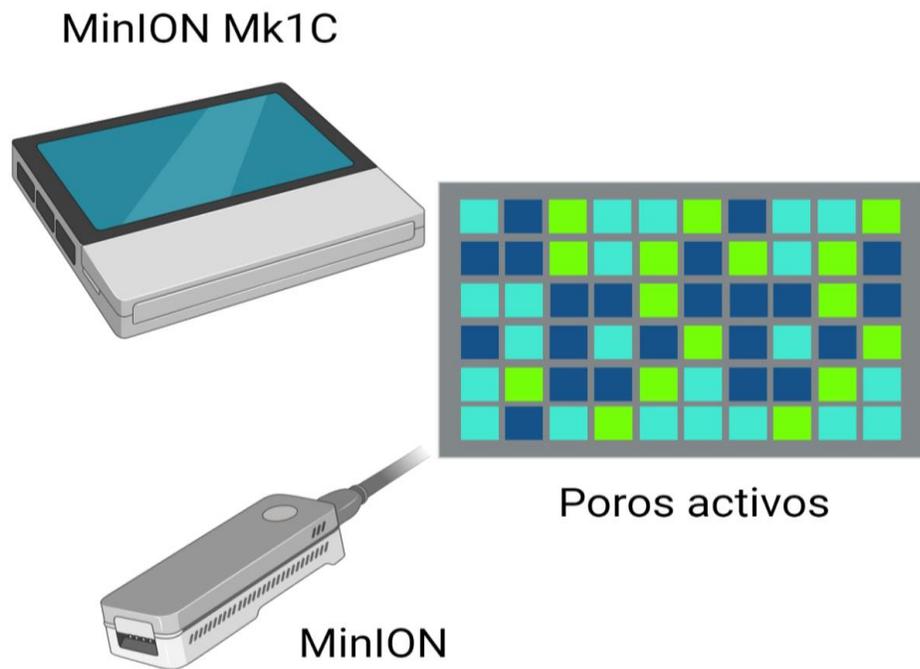


**Cualquier Lugar**





# Principio de funcionamiento de un secuenciador de ADN de Nanopore



# MinION

- Herramienta portátil de análisis biológico alimentada por USB
- Análisis de 12 a 96 muestras a la vez usando códigos de barras
- Hasta 512 canales disponibles en cualquier momento Condiciones mínimas:

I7

**16Gb RAM**

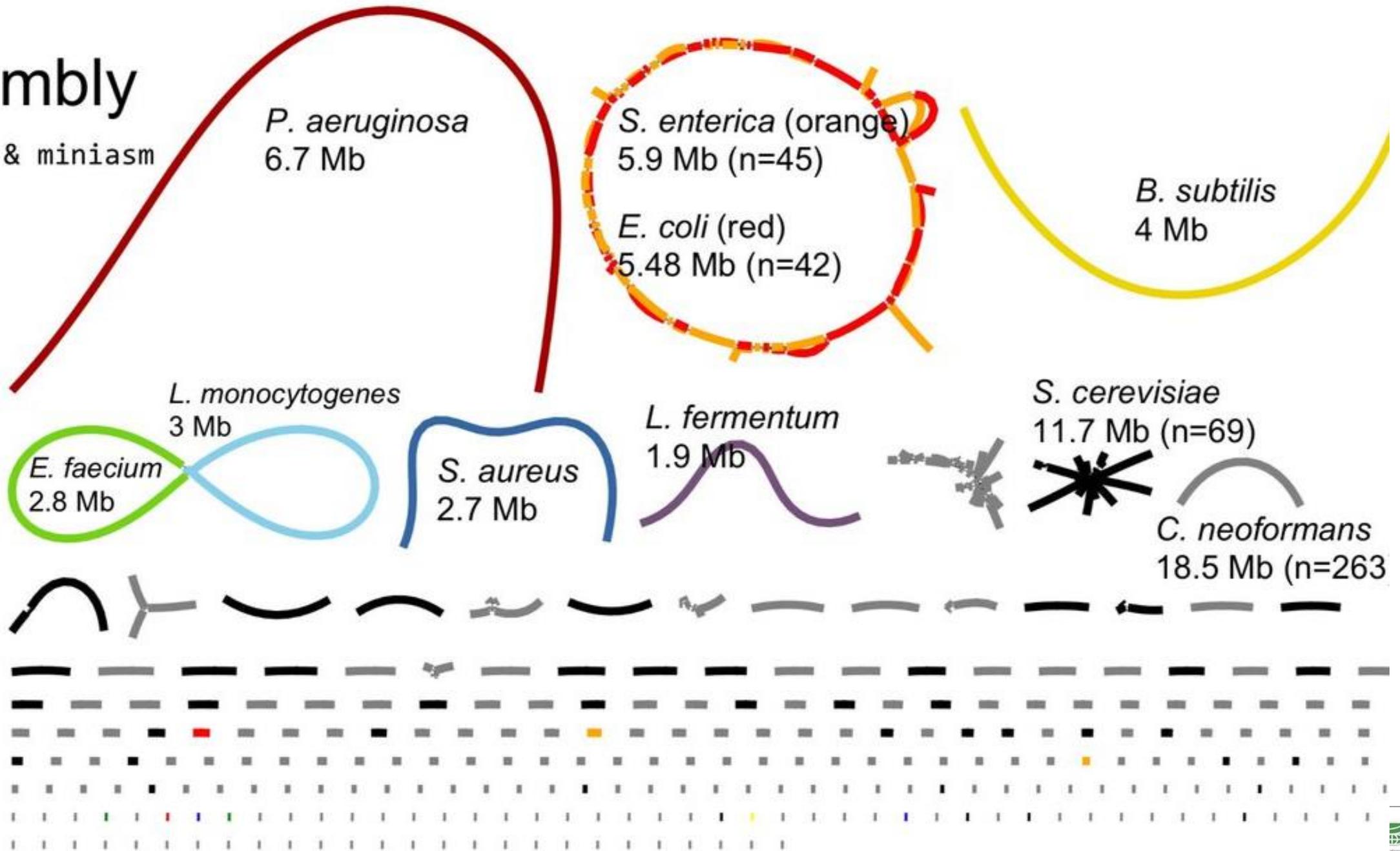
**1TB SSD**



System capacity	Theoretical maximum	Internal best	Current customer best
Bases per 72 hour run per flow cell (@ 450 bps)	60 Gb	>50 Gb	>40 Gb

# Assembly

minimap2 & miniasm



DOCS HELP LOGIN

Dataset

community

pestdisplace

CMDASIA1

Date Range

1927-04-22 2020-06-19

PLAY RESET

Color By

Country

Filter Data

Type filter query here...

Tree Options

Layout

RECTANGULAR

RADIAL

UNROOTED

CLOCK

SCATTER

Branch Length

TIME DIVERGENCE

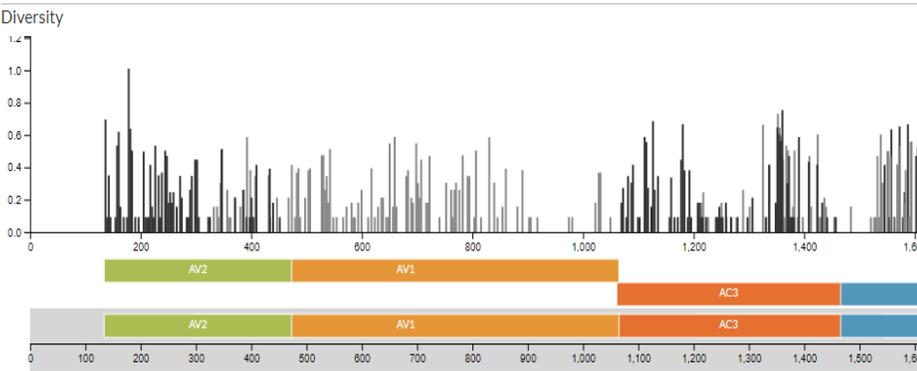
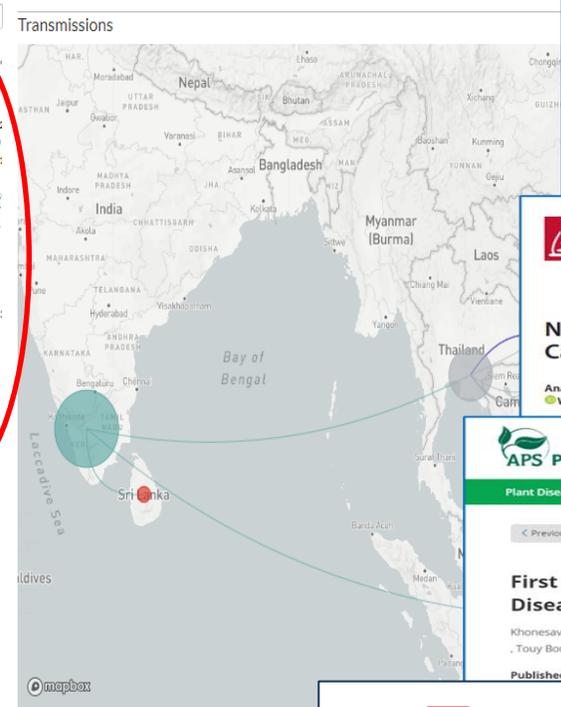
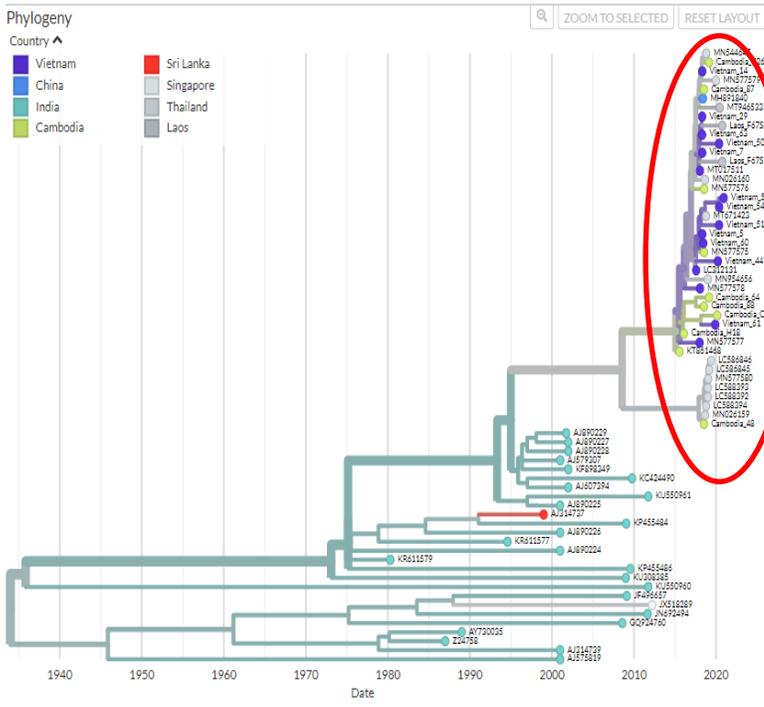
Show confidence intervals

Branch Labels

none

Tip Labels

Sample Name



ELSEVIER

Virus Research

Volume 285, August 2020, 197959

Short communication

Surveillance and diagnostics of the emergent Sri Lankan cassava mosaic virus (Fam. Geminiviridae) in Southeast Asia

AMERICAN SOCIETY FOR MICROBIOLOGY

Microbiology Resource Announcements

GENOME SEQUENCES

Nanopore-Based Complete Genome Sequence of a Sri Lankan Cassava Mosaic Virus (*Geminivirus*) Strain from Thailand

Ana M. Leiva,<sup>a</sup> Wanwisa Siriwan,<sup>b</sup> Diana Lopez-Alvarez,<sup>a</sup> Israel Barrantes,<sup>a</sup> Nuannapa Hemniam,<sup>b</sup> Kingkan Saokham,<sup>d</sup> Wilmer J. Cuellar<sup>a</sup>

APS Publications

Plant Disease Home About Submit Journals Books Publisher's Home

First report of Sri Lankan cassava mosaic virus and Cassava Mosaic Disease in Laos

Khonesavane Chittarath, Jenyfer Jimenez, Pinkham Vongphachanh, Ana Maria Leiva, Somkhit Sengsay, Diana Lopez-Alvarez, Touy Boumvilayvong, Derlyn Lourido, Viengvilay Vorlachith, and Wilmer Jose Cuellar

Published Online: 12 Jan 2021 | <https://doi.org/10.1094/PDIS-09-20-1868-PDN>

AMERICAN SOCIETY FOR MICROBIOLOGY

Microbiology Resource Announcements

GENOME SEQUENCES

Complete Genome Sequence of the Plant Pathogen *Ralstonia solanacearum* Strain CIAT-078, Isolated in Colombia, Obtained Using Oxford Nanopore Technology

Diana López-Alvarez<sup>a,b</sup>, Ana M. Leiva<sup>a</sup>, Israel Barrantes<sup>c</sup>, Juan M. Parde<sup>a</sup>, Viviana Domínguez<sup>a</sup>, and

AMERICAN SOCIETY FOR MICROBIOLOGY

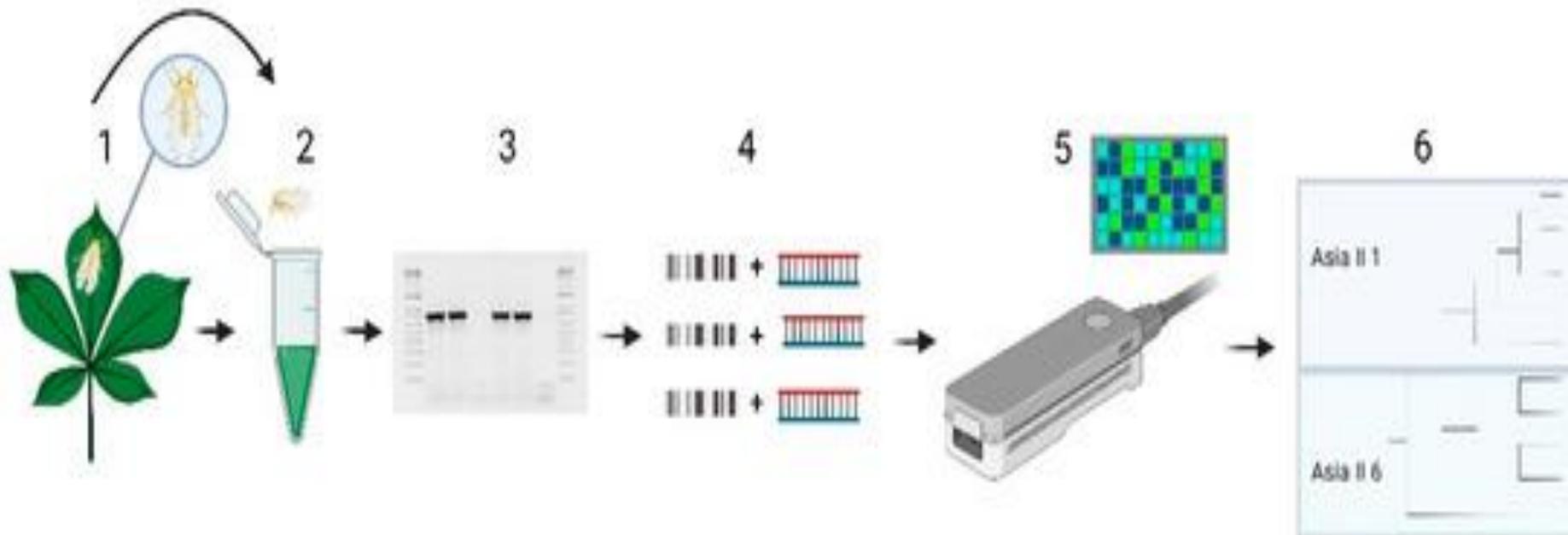
Microbiology Resource Announcements

GENOME SEQUENCES

Draft Genome Sequence of *Fusarium oxysporum* f. sp. *cubeense* Tropical Race 4 from Peru, Obtained by Nanopore and Illumina Hybrid Assembly

Ana M. Leiva<sup>a</sup>, Mathieu Rouard<sup>b</sup>, Diana Lopez-Alvarez<sup>a,c</sup>, Alberto Cenci<sup>b</sup>, Catherine Breton<sup>b</sup>, Rosalyn Acuña<sup>d</sup>, Juan Carlos Rojas<sup>e</sup>, Miguel Dita<sup>f</sup>, Wilmer J. Cuellar<sup>a</sup>

<https://nextstrain.org/community/pestdisplace/CMDASIA1>



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## Mitochondrial Genetic Diversity of *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae) Associated with Cassava in Lao PDR

by Ana M. Leiva<sup>1</sup>, Khonesavanh Chittarath<sup>2</sup>, Diana Lopez-Alvarez<sup>3</sup>, Pinkham Vongphachanh<sup>2</sup>, Maria Isabel Gomez<sup>1</sup>, Somkhit Sengsay<sup>2</sup>, Xiao-Wei Wang<sup>4</sup>, Rafael Rodriguez<sup>1</sup>, Jonathan Newby<sup>5</sup> and Wilmer J. Cuellar<sup>1,\*</sup>

<sup>1</sup> Cassava Program, Crops for Nutrition and Health, International Center for Tropical Agriculture (CIAT), The Americas Hub, Km 17 Recta Cali-Palmira, Cali 763537, Colombia

<sup>2</sup> Plant Protection Center (PPC), Department of Agriculture, Ministry of Agriculture and Forestry, Vientiane P.O. Box 811, Laos

<sup>3</sup> Department of Biological Sciences, Universidad Nacional de Colombia UNAL-Palmira, Palmira 763533, Colombia

<sup>4</sup> Institute of Insect Sciences, Zhejiang University, Hangzhou 310058, China

<sup>5</sup> Cassava Program Asia Office, Crops for Nutrition and Health, International Center for Tropical Agriculture (CIAT), Laos Country Office, Vientiane P.O. Box 783, Laos

\* Author to whom correspondence should be addressed.

Academic Editors: Vincenzo Cavalieri and Sabrina Bertin

*Insects* **2022**, *13*(10), 861; <https://doi.org/10.3390/insects13100861>

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# Nuevos avances

## MinION Mk1D

An accessory keyboard with integrated sequencer for tablet devices



## Ubik™

Rapid and portable, single-tube sample preparation



## Plongle™

High-throughput analysis of smaller, frequently performed tests and assays in a 96-well plate format

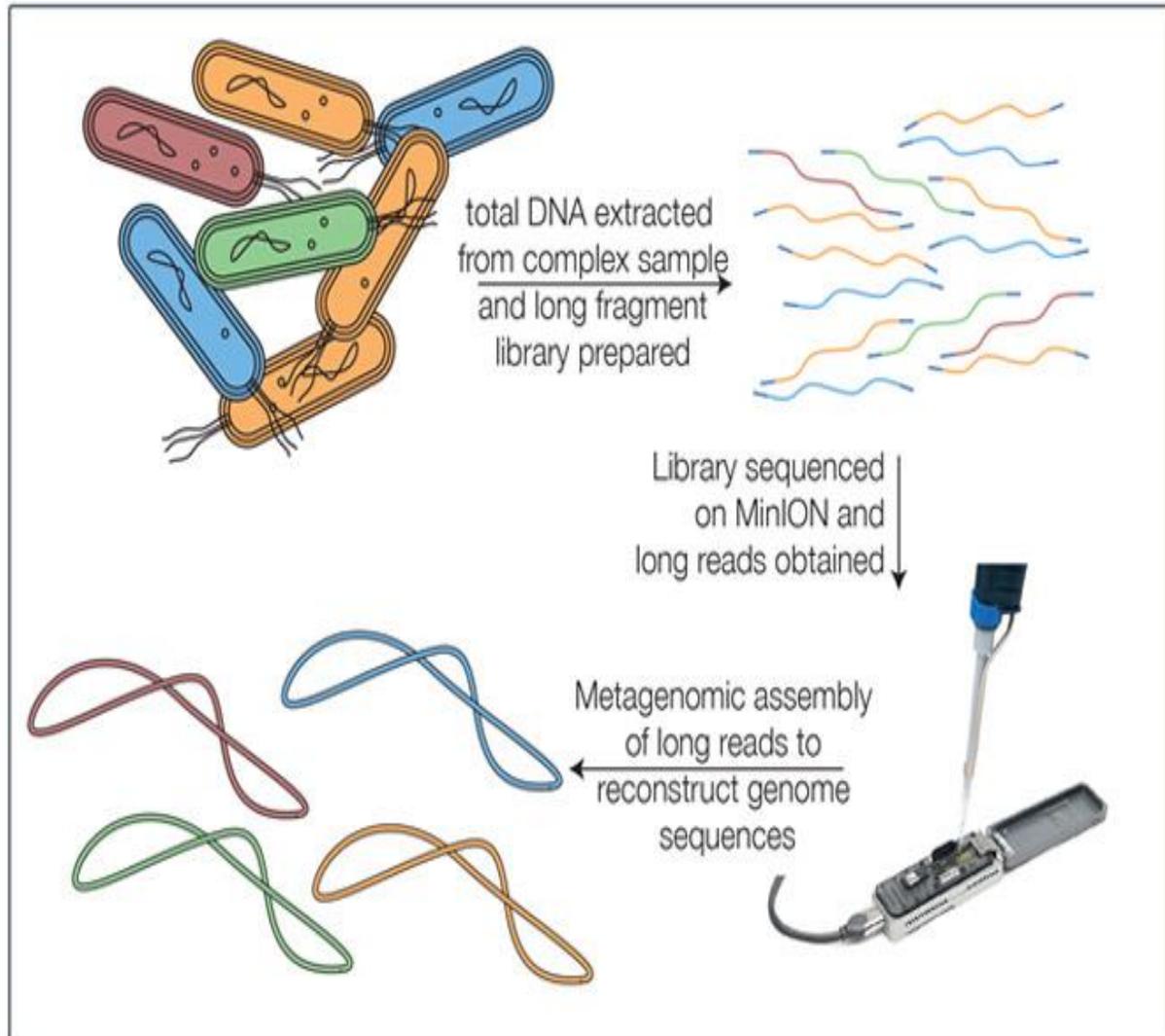


## SmidgION™

Real-time nanopore sequencing and analysis on a smartphone



# Metagenómica



**16S Barcoding Kit**  
SQK-RAB204

**16S Barcoding Kit 1-24**  
SQK-16S024

# Nanopore Learning & Support

## Nanopore Learning

Explore our online courses and video library to assist with your Nanopore experience.

### Train online

Online courses to support your future experiments from sample to result

[View courses](#)

### Lesson library

Learn about nanopore sequencing by browsing our collection of short videos

[Browse all lessons](#)

### On-site training

We offer training on all of our sequencing devices including workshops and bespoke training packages either at your institution or an Oxford Nanopore lab



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SUPPORT

## Nanopore Community Support

ALL

Search support, or ask a question



Getting Started



Library Preparation



Sequencing



Data Analysis



Devices



Software



Troubleshooting



Customer Services

#### Top Customer Services Items >

- Can I upgrade my MinIT to a MinION Mk1C?
- Flongle production and shipment update
- Changing the delivery schedule of your order
- What is the warranty period on my flow cells?
- Where do I submit my flow cells

#### Top Technical Support Items >

- What is the warranty period on my flow cells?
- How do I basecall on the MinIT?
- Which protocols are currently compatible with Flongle?
- How do I refuel my flow cell?

#### Top Tags

MINION

GRIDION

PROMETHION

ORDERS

FLOW CELL



**Colab** son archivos que permiten combinar código ejecutable y texto enriquecido en un solo documento, junto con imágenes, HTML, LaTeX y más. Cuando creas tu propio Colab. Colab, o "Colaboratorio", le permite escribir y ejecutar Python en su navegador.

- Se requiere configuración cero
- Acceso a las GPU sin cargo
- Fácil de compartir
- Tanto si es estudiante, científico de datos o investigador de inteligencia artificial, Colab puede facilitarle el trabajo.



[https://colab.research.google.com/?utm\\_source=scs-index#scrollTo=-Rh3-Vt9Nev9](https://colab.research.google.com/?utm_source=scs-index#scrollTo=-Rh3-Vt9Nev9)

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Ministerio de Agricultura y Riego



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de Internet**

