

SEQAFRICA Virtual Training Course

Course information

Title: SARS-CoV-2 whole genome sequencing

Language of instruction: English.

Offered as: Webinar with lectures and exercises.

Duration of course: 4 x ½ days. Originally held 17 – 28 May 2021.

Responsible: Rene S. Hendriksen (DTU), rshe@food.dtu.dk

Course co-responsible: Pernille Nilsson (DTU), Anthony Smith (NICD, South Africa), Jinal Bhiman (NICD, South Africa), Marco van Zwetselaar (KCRI, Tanzania), Beverly Eygir (NMIMR, Ghana), Iruka N. Okeke (UI, Nigeria).

General course objectives:

The course introduces and cover all aspects of the entire WGS workflow for SARS-CoV-2 starting with a respiratory sample and finishing with completely analysed DNA sequence/genome.

The participant will upon completion of the course know all the steps included in preparing and conducting WGS using Illumina short read technology, which will include the wet laboratory procedures, computational genome assembly and the use of online tools to assess phylogenies and mutations in the global context.

Learning objectives:

A participant who has met the objectives of the course will be able to:

- Describe wet laboratory and computational workflow to generate and assemble SARS-CoV-2 genomes
- Perform wet laboratory protocols
- Perform drag-and-drop bioinformatics using online analysis tools and interpret the results

Content:

The course covers a detailed run through the sequencing workflow, starting with a respiratory sample to completed analysis of raw reads or assembled genome and will give the participants a thorough understanding of the process for short read technologies.

The course comprise of lectures and hands-on exercises that the participants can chose to submit.

Course literature:

No literature required.

Audience:

Users with some experience/knowledge of WGS and WGS data (e.g. attended SEQAFRICAs Introduction to WGS in AMR surveillance).

Day 1: Sequencing workflow overview and wet lab methods
 Links to [Presentation](#) and [Recording](#)

Speaker/Presenter	Content/Activity	Slides	Video
Pernille Nilsson (DTU, Denmark)	Welcome and Introduction		
Jinal Bhiman (NICD, South Africa)	[1] SARS-CoV-2 genomic surveillance: From respiratory sample to SARS-CoV-2 genome. (30 min)	PDF	Video
Noxolo Ntuli (NICD, South Africa)	[2] RNA extraction: From respiratory sample to RNA. (20 min)	n/a	Video
Boitshoko Mahlangu (NICD, South Africa)	[3] cDNA synthesis and tiling PCR: Method and background for cDNA synthesis using random hexamers followed by tiling PCR using primer pools with >170 individual primers (25 min)	n/a	Video
Frank Oteng/Noguchi team (NMIMR, Ghana)	[4] Amplicon Generation and Quantification (15 min)	n/a	Video Video
Frank Oteng/Noguchi team (NMIMR, Ghana)	[5a] Library preparation: Going from high quality DNA to sequencing libraries. “Hands-on” and theory. Description of library preparation using the Nextera Flex kit (20 min)	n/a	Video Video
Thabo Mohale, Zama Khumalo (NICD, South Africa)	[5b] Library preparation: Going from high quality DNA to sequencing libraries. “Hands-on” and theory. Description of library preparation using the COVIDSeq (30 min)	PDF	Video
	Q&A and Wrap-up		

Day 2: Illumina sequencing and accessing data
 Links to [Presentation](#) and [Recording](#)

	Welcome and Introduction		
Zama Khumalo (NICD, South Africa)	[6a] Illumina sequencing: Hands-on how to load the NextSeq machine with your prepared libraries (20 min).	PDF	Video
Frank Oteng/Noguchi team (NMIMR, Ghana)	[6b] Illumina sequencing: Hands-on how to load the MiSeq machine with your prepared libraries (15 min).	n/a	Video

Stanford Kwenda (NICD, South Africa)	[7] Downloading data: Once the sequencing run is finished, how do you get your data? (20 min)	PDF	Video
Frank Oteng/Noguchi team (NMIMR, Ghana)	[7] Exporting data from MiSeq. (2 min)		Video
Jinal Bhiman (NICD, South Africa)	[7Q] Quiz: Introduction to quiz covering the wet lab steps. QC of samples through in-house and COVIDSeq methods following and/or prior to library prep. (5-minute introduction to the quiz)		Quiz
	Q&A and Wrap-up		
Day 3: Friday SARS-CoV-2 WGS bioinformatics workflows and online tools Links to Presentation and Recording			
	Welcome and Introduction		
Jinal Bhiman (NICD, South Africa)	[8] Recap on the wet lab workflow. (30 min)	n/a	Video
Cathrine Scheepers, (NICD, South Africa)	[9] Galaxy pipeline: Bioinformatics pipeline used for SARS-CoV-2 genome assemble using a reference. QC of sequence outputs from Galaxy (50 min)	PDF	Video
Simon Travers (Hyrax Biosciences)	[10] Exatype pipeline: Bioinformatic pipelines used for SARS-CoV-2 genome assemble using a reference. QC of sequence outputs from Exatype (25 min)	n/a	Video
Bright Adu (NMIMR, Ghana)	[11a] Introduction to Nextstrain online tool: Demonstration of data analysis at Nextstrain. (35 min)	n/a	Video
Daniel Amoako (NICD, South Africa)	[11b] Introduction to PANGOLin and COVDB online tools: Demonstration of data analysis at PANGOLin and Stanford University – implication of mutations reported in literature (vaccine escape etc.) (40 min)	PDF	Video

Cathrine Scheepers, (NICD, South Africa)	[9_10E] Introduction to Exercise: Going from raw reads to analyzed genome assemblies Assemble WGS, generate FASTA, look at QC, assign lineage and clade, compare Galaxy and Exatype outputs. Low, medium and high-quality test set for generating the QC report will be provided.	PDF Example report	Video
	Q&A and Wrap-up		
Day 4: Mutant impact assessment, data sharing and review of exercises/quizzes Links to Presentation and Recording			
	Welcome and Introduction		
Constantinos Kurt Wibmer, (NICD, South Africa)	[12] Introduction to PyMoL*: Predictive assessment of impact of mutations based on spike structure (For novel mutations) (40 min)	PDF	Video
Constantinos Kurt Wibmer, (NICD, South Africa)	[12E] Exercise: Plot given set of mutations on structure and suggest functional role for these.	PDF Pse-file	Video
Anne von Gottberg (NICD, South Africa)	[13] Data sharing and uploading to GISAID: Overview of the GISAID database, polices and uploading (including compulsory metadata) (30 min).	n/a	Video
	Going through results from Quiz 7Q (20 min)	Video	
	Going through results from 11E (35 min)	Video	
	Q&A and Close		

PyMoL is **free for educational use and you do not need to purchase a license. Download PyMoL here: <https://pymol.org/2/>*