NOTE #8

ZIKA in Brazil Real Time Analysis (ZiBRA-2): an RRI experience

By Marta Giovanetti, Fernanda Khouri, Luiz Alcantara

RRI IMPLEMENTATION IN BIOSCIENCE ORGANISATIONS

GUIDELINES FROM THE STARBIOS2 PROJECT

Andrea Declich with the STARBIOS2 partners

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Advances in DNA sequencing technology have ushered in a new era of pan-genomics and genomic surveillance, in which traditional molecular diagnostics and genotyping methods are being enhanced and even replaced by genomics-based methods to aid epidemiologic investigations of communicable diseases (Gardy et al., 2018). The ability to compare and analyse entire pathogen's genomes has allowed unprecedented resolution into how and why infectious diseases spread. The rapid development of these technologies has made sequencing of viral genomes possible and even routine (Shendure et al., 2008).

There are currently two major ways in which high-throughput sequencing technologies are used in public health and diagnostic applications, (i) to track outbreaks and epidemics in order to call for public health responses and (ii) to characterize individual infections to tailor treatment decisions (Theze et al., 2018; Faria et al., 2017). Focusing on these aims, genome sequencing has been successfully used to describe unique and detailed insights into the transmission, biology, and epidemiology of many health care-associated viral pathogens.

Considering the improvements on portability and quality of sequencing, and the acceleration and standardization of analytical pipelines, the applicable routine of genome sequencing may soon become the common *de facto* method for infectious disease control. In the context of virus investigations, pan-genomics and bioinformatics in general face great challenges. Rapid extraction of genomic features with an evolutionary signal facilitates

evolutionary analyses ranging from the reconstruction of species phylogenies to tracing epidemic outbreaks.

In February 2016, the World Health Organisation declared a Public Health Emergency of International Concern in response to the transmission of ZIKV in the Americas. In that context, the ZiBRA-2 project was launched as a multicentre collaboration between the University of Oxford, University of Birmingham, Evandro Chagas Institute, University of São Paulo and Oswaldo Cruz Foundation employing a promising approach to generating a substantial number of complete genome sequences for Zika virus (ZIKV) through MinION in a mobile laboratory trip.

The ZiBRA-2 project is based on principles of ethics, social engagement and open access to the information obtained. We consider that it is necessary to present the ZIKV results to other scientific communities and try to increase the participation of the public and civil society in bioscience research. Thus, during the project all the sequences and information generated are published in real-time on the ZiBRA-2 websites (http://www.zibraproject.org; https://www.zibra2project.org), and the final results are made available to society through scientific publications in open access journals.

Based on a previous genomic surveillance trip during the Ebola outbreak in Guinea in 2014-2015, the ZiBRA-2 project aimed to generate a large number of ZIKV complete genome sequences from the Northeast of Brazil covering a broad geographical region including historical samples, and from patients with a range of clinical presentations. The method consisted of genome-tiling PCR to enrich ZIKV material in clinical samples followed by library preparation prior to MinION loading (Faria et al., 2016; Quick et al., 2017).

The ZiBRA-2 team working together with the Central Laboratory of Public Health (LACEN) personnel, tested 1349 clinical samples for ZIKV RNA across Rio Grande do Norte, Paraíba, Recife, Maceió, and Bahia states and captured 850 mosquitoes from urban

and peri-urban fields in each place along the trip. The project also involved capacity building as each local team was trained to perform the whole protocol on subsequent trips. It is important to note that the team is composed of men and women who participate from the design of the study until the final publication and are trained at all stages, to reduce the gender discrimination. (Faria et al., 2016).

After the original trip that took place in June 2016, the ZiBRA-2 project has been extended and up to now trained teams to track not only ZIKV, but also other arboviruses circulating in Brazil including emerging and re-emerging strains. The team was also employed to investigate the dispersion of the CHIKV - East-Central South African genotype spreading in North Brazil (Naveca et al., 2019) as well as to characterize the largest Yellow Fever outbreak registered in Southeast Brazil in December 2016. By analysing 64 new yellow fever virus genomes the virus transmission pattern was revealed to originate in non-human primates, rejecting the hypothesis of urban transmissions.

As the mobile trips occur, more people are being trained to continue performing genomic surveillance throughout the country and also in some places in Africa like Angola and Cabo Verde (Hill et al., 2019). Also, the productivity of these trips is increasing each time, with generation of around 60 complete genome sequences in five days. Besides that, the development of faster protocols and more than 12 barcodes per run suggests this number will increase soon. A single flow cell used in MinION can run up to 96 genomes and produces reads up to 200 Kb in length, with a throughput of 1.5 Gb, and more than 100,000 reads at a single run. Ongoing improvements to the launched barcoding kits in the nanopore sequencing technology had the potential to increase the number of generated genomes per sequencing run from 12 to 96, which could also increase the number of genome sequences derived from affected regions and allow more detailed investigations of the

association between pathogen mutations and environmental context with less costs.

The participation of ZiBRA-2 in STARBIOS2 provides an ideal environment to showcase these research projects, and highlights the practice of Responsible Research and Innovation (RRI) in the context of this unique bioscience endeavour.

ABOUT THE STARBIOS2 GUIDELINES

This guideline aims to help readers formalize and trigger structural change aimed at introducing appropriate RRI-related practices to their own organisations. This is not a series of prescriptions, but an itinerary of reflection and self-interpretation addressed to different actors within the biosciences. To support this itinerary of reflection and self-interpretation, the document provides...

- a description of a general RRI Model for research organisations within the biosciences, that is a set of ideas, premises and "principles of action" that define the practice of RRI in bioscience research organisations,
- some practical guidance for designing interventions to promote RRI in research organisations in the Biosciences, putting into practice the RRI Model,
- a set of useful practices in implementing the structural change process,
- and information on particular STARBIOS2 cases and experiences, as well as materials, tools and sources, are also provided in the Appendix and in the Annex.