Identification of China's first case of mixed infection of *Chikungunya* virus and *Zika* virus

MGI Microbiological Detection Total Package

Highlights

Up-to-date Microbial Database

- A comprehensive database of 27, 830 microbial genomics-enabling massive screening at once

Streamlined Workflow

- Complete the whole process from sample to report with one instrument in 24 hours

Proprietary Platforms

- Fully-automated sample preparation system, high-throughput sequencing platform and various supported reagent kits

Flexible Package

- Users can flexibly choose hardware and matching reagent consumables according to their needs

Introduction

Arboviruses are one of the most common infectious pathogens in the world, among which Dengue virus, *Chikungunya* virus and *Zika* virus can be transmitted through the common host, Aedes. From the analysis of transmission routes, humans are very likely to be infected with multiple arboviruses simultaneously through mosquito bites, which increases the difficulty of clinical diagnosis and treatment.

A passenger departed from Manila, Philippines, to Guangzhou on January 5, 2019, had been having a fever and coughing for 2 days when entering at Guangzhou Baiyun Airport. Epidemiological investigation and blood samples were taken immediately.

The laboratory staff first conducted fluorescence RT-PCR detection of various arboviruses, and the results showed that the nucleic acid amplification of virus was positive (Ct value was about 20), while *Chikungunya* virus was positive (Ct value was about 26). It was preliminarily identified that this passenger had mixed infection of *Zika* virus and *Chikungunya* virus. To further confirm the mixed infection results, MGI Microbiological Detection Total Package was used to sequence the virus metagenomic sequence (FIG. 1).

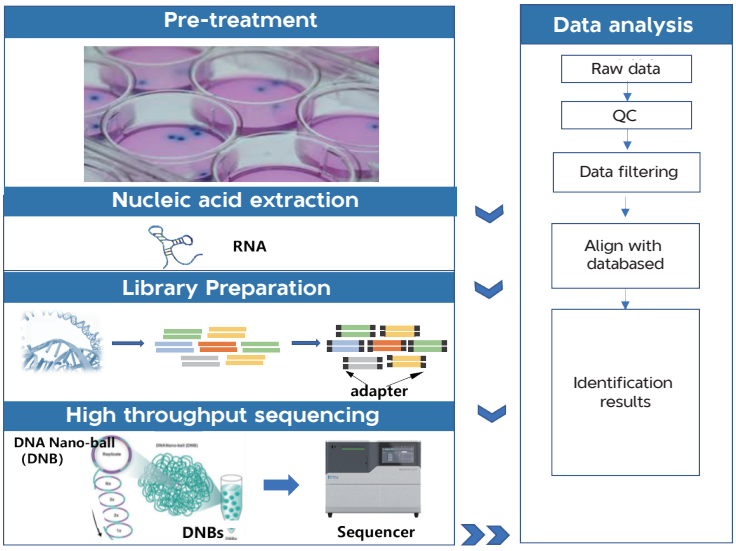

Figure 1. Virus metagenomic sequencing and analysis

Experimental Method

Library preparation

The samples were inoculated with Vero cells for virus isolation and culture, and the cells showed obvious lesions after 72 hours. After the nucleic acid was extracted from cell supernatant, the library was prepared using MGIEasy RNA library prep set (16 RXN, Cat. No. 1000006383) and the host rRNA was removed using MGIEasy rRNA depletion kit (32 RXN, Cat. No. 1000005953). The automated sample preparation system MGISP-100 (MGI, Cat. No. 900-000051-00) can also be used to prepare library automatically.

Sequencing

After each sample library was built, multiple sample libraries with different label sequences were mixed together and paired-end sequencing of 2×50 bp was performed on the DNBSEQ-G50RS* (Cat. No. 900-000354-00) sequencer. In addition, MGI also provides higher throughput sequencing platforms, DNBSEQ-G400* (Cat. No. 900-000170-00) and DNBSEQ-T7* (Cat. No. 900-000128-00), as other options for fast and flexible sequencing packages for pathogen detection.

Data analysis

MGI has developed a database containing the genetic information of 27, 830 microbes, including bacteria, fungi, viruses, and parasites, and the software named as PFI (Pathogen Fast Identification, Cat. No. 510-000164-00). The integrated system can quickly generate reliable analyses of microbial genome information and the identification report automatically.

Results

Verification of mixed infection of *Chikungunya virus* and *Zika virus*

The results of viral metagenomic sequencing showed that partial reads were related to *Chikungunya virus*, the number of reads was 114,226,369. A small number of reads were related to *Zika virus*; and the number of reads was 3,320. The results confirmed that the passenger was infected with both *Chikungunya virus* and *Zika virus* (Table 1).

Table 1. PFI software reports the results of virus metagenomic sequencing

NO.	Species	Reads Number	Relative Abundance(%)
1	*Chikungunya virus*	114,226,369	99.657
2	*Barmah Forest virus*	282,111	0.246
3	*Ross River virus*	32,645	0.028
4	*Zika virus*	3,320	0.003
5	*Western quine encephalitis virus*	2,052	0.002

Summary

In this application guide, we introduce the application cases of MGI Microbiological Detection Total Package in the identification of mixed infections. This method combines the library preparation by MGIEasy RNA library prep and the DNBSEQ-G50RS* sequencer. It also uses the Pathogen Fast Identification software (PFI) for rapid identification of multiple unknown viruses. The whole process (from samples to reports) can be completed in less than 24 hours.

Based on the proprietary high-throughput sequencing platform and rapid identification system of pathogen infection, MGI has developed a microbiological detection total package, which can achieve fast, accurate and comprehensive pathogen screening for clinical diagnoses (FIG. 2). Moreover, MGI provides a variety of hardware and compatible reagent kits for the system to support an extensive range of pathogen testing.

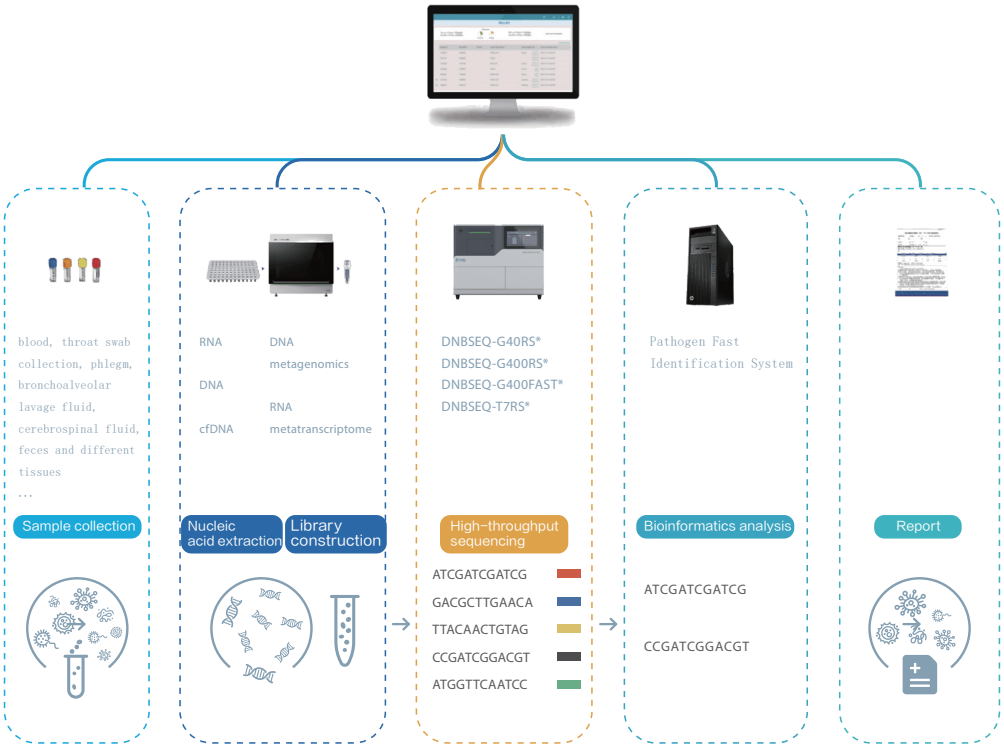

Figure 2. MGI Microbiological Detection Total Package

MGI

Microbiological Detection Total Package

Ordering information

Equipment

Product Name	Catalog No.	Configuration
DNA Sequencing Library Preparation System MGISP-100	900-000051-00	RUO
Genetic Sequencer DNBSEQ-G50RS*	900-000354-00	RUO
Genetic Sequencer DNBSEQ-G400RS*	900-000170-00	RUO
Genetic Sequencer DNBSEQ-T7RS*	900-000128-00	RUO

Reagents

Product Name	Catalog No.	Configuration
MGIEasy RNA Library Prep Set	1000006383	16 RXN, RUO
MGIEasy RNA Library Prep Set	1000006384	96 RXN, RUO
MGIEasy rRNA Depletion Kit	1000005253	32 RXN, RUO

Analysis

Product Name	Catalog No.	Configuration
Fast identification of pathogenic infections (PFI, with server)	510-000164-00	Server + software, RUO
Fast identification of pathogenic infections (PFI)	057-000060-00	software, RUO

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*Unless otherwise informed, StandardMPS and CoolMPS sequencing reagents, and sequencers for use with such reagents are not available in Germany, USA, Spain, UK, Hong Kong, Sweden, Belgium, Italy, Finland, Czech Republic, Switzerland and Portugal.

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