

**TITLE OF INVENTION: [MULTIPLE MIXED SAMPLE DIRECT RNA NANOPORE
SEQUENCING METHOD AND SYSTEM]**

ABSTRACT

The present invention relates to the field of sequencing technologies, and specifically to a multiplexed mixed-sample direct RNA nanopore sequencing method and system. The method is used for sequencing n test-sample RNA polynucleotides, wherein $2 \leq n \leq 24$ and n is an integer. The method comprises performing nanopore sequencing on sequencing constructs corresponding to the n test-sample RNA polynucleotides and extracting corresponding electrical-current signal data; segmenting electrical-current signal data of a barcode sequence therein; and analyzing the segmented electrical-current signal data using a classification model to obtain sequence information of the barcode sequence, thereby determining classification information of the n test-sample RNA polynucleotides. The barcode sequence is specific to each test-sample RNA polynucleotide and is selected from any one of SEQ ID NOs: 1–24. The method and system provided in the present invention enable simultaneous sequencing of multiple biological samples on a single flow cell, reduce the initial input amount of biological samples, and significantly lower the cost of multi-sample direct RNA sequencing.

[Fig. 1]