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BGISEQ-500 Sequencing

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Protocol status: Working
We use this protocol and it's working

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Abstract

BGISEQ-500 is a new desktop sequencer developed by BGI. Using DNA nanoball and combinational probe anchor synthesis developed from Complete Genomics™ sequencing technologies, it generates short reads at a large scale.

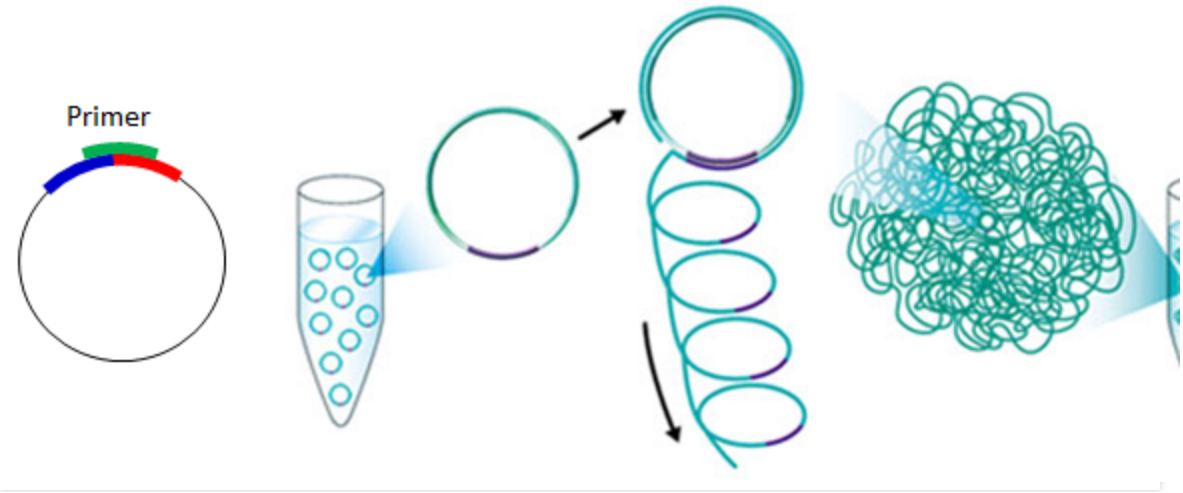
1 Test the quality of the sequencing library (see protocol for library preparation) by the Qubit® ssDNA Assay Kit and homogenized at 6ng total amounts.

2 Carry out Rolling circle amplification（RCA）for 10 minutes in an 80 ul reaction volume with pure water, buffer and DNB polymerase

00:10:00

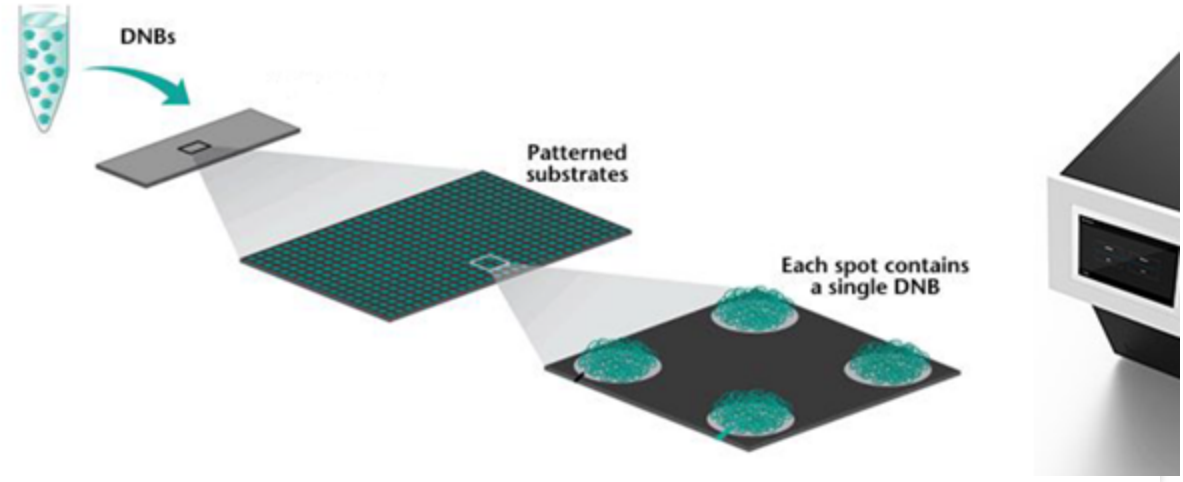
3 Add 20 ul DNBs stopping buffer to stop the RCA reaction

4 Check the quality of the DNBs using the Qubit® ssDNA Assay Kit , concentration should be above 10 ng/μL



5 Add 33ul DNBs loading buffer to the DNBs product from the last step, and place it in the BGIDL-50 (the DNBs loading machine)

6 install the sequencing chip and selected the DNBs loading process (Version: sample load 2.0) to load the DNBs.



7 after loading, take the sequencing chip out and install in the BGISEQ-500 sequencing machine

8 load the reagent sequencing kit and open the sequence control software (Version 1.1.0.10003), and select the sequence process Version 1.0.06 and Zebracall process Version 0.5.0.13875 for sequencing.

9 Sequencing is initiated after the sequencing reagents preloaded and sequencing chip was installed, and this process is finished in about 72 hours. When the whole sequencing was finished, the binary file with bases and quality score were converted into FASTQ format with Phred+33 quality score.