

Wellcome Genome Campus | OC4_2-10_Illumina_seq

The Illumina Sequencing System enables a broad array of applications in genomics, transcriptomics, and epigenomics. In this short video, we will go through all the steps of a next-generation sequencing (NGS) workflow.

Libraries need to undergo a denaturation step to allow the binding of single-strand DNA to the flow cell.

To allow denaturation, we mix libraries with a solution of 0.2 N NaOH, or sodium hydroxide.

Complete denaturation of double-stranded DNA to single-stranded DNA occurs in five minutes.

After five minutes, the denatured libraries are diluted in the sequencing buffer (HG 1) to an intermediate concentration of 20 picomolar.

The sample is vortexed and centrifuged.

The denatured library is then diluted to the final concentration of eight picomolar.

The second step in an NGS workflow is the sequencing. During this step, the libraries are loaded onto a flow cell and placed on the sequencer.

The flow cell is rinsed in distilled water first.

The flow cell is then rinsed one more time using ethanol, then it is dried thoroughly.

The old flow cell is replaced with the new one that has just been rinsed.

The sequencing buffer is loaded.

The library is loaded into the new cartridge.

The old cartridge is removed, and the new cartridge containing the library is loaded.

The run settings are reviewed, and the sequencer is started.