

Supplementary Laboratory User guide for sequencing on Element AVITI™ and AVITI24™ platforms

For use with biomodal duet multiomics solution duet evoC & duet +modC

This addendum supplements the duet evoC and duet +modC User guides (compatible with whole-genome sequencing workflow). It provides specific instructions for sequencing on Element AVITI™ and AVITI24™.



This user guide is specifically compatible with **Product Numbers: 6205/5205 (8 reactions)** and **6206/5206 (24 reactions)**. If you have a different product number, please refer to the appropriate guide or contact biomodal support for assistance.

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I. Pre-requisites

- Sequencing duet evoC and duet +modC libraries on Element AVITI™ and AVITI24™ requires a custom sequencing recipe. Contact your Element Field Application Scientist for access to the EnhancedShortInsert.rec custom recipe.
- This protocol uses reagents already provided in the Cloudbreak™ or Cloudbreak Freestyle™ sequencing kits. For frequent users of the Individually Addressable Lanes add-on, confirm you have at least 1.3mL of Library Loading Buffer available (in addition to the amount needed for library dilution) before proceeding.

 **Note:** this protocol is for use with the whole-genome sequencing workflow. If you're planning on running targeted, some optimization might be required.

II. How to use this addendum

- Follow the standard duet evoC or duet +modC User Guide until the Sequencing section. At the point of sequencing, replace the standard sequencing instructions with the steps below.
- This addendum provides all additional instructions required for sequencing on Element Cloudbreak™ or Cloudbreak Freestyle™ systems.

III. Sequencing

1. Libraries set-up

- 1.1 Libraries must be sequenced on an Element AVITI™ or AVITI24™ sequencing platforms as paired-end sequencing runs. Perform a 2 x 151 bp paired-end sequencing analysis. Include 2 x 8 bp UDI index reads for sample pooling and deconvolution.

-  **IMPORTANT:** Spike-in of a base balanced library, such as PhiX, is required due to the C to T conversion during the deamination step. **2% PhiX** is recommended for best results. If running 1X coverage, no phiX is required.
- For best results, we recommend quantifying the PhiX before calculating the amount needed to spike-in.

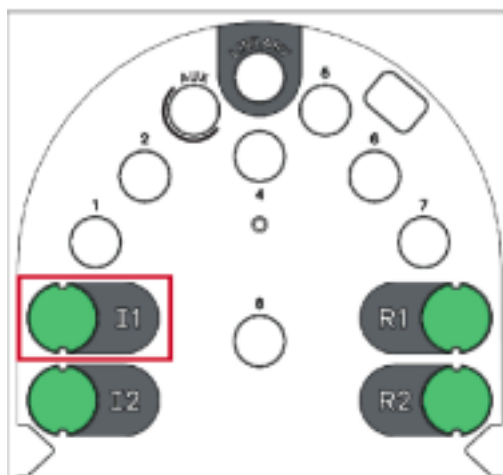
- 1.2 Optimise loading concentration for the sequencer used. 14.5 pM should be used as a starting point.
- 1.3 Index sequences: they are provided separately and can be found in the sequences for UDIs document, on the customer documentation portal.

2. Custom recipe set-up

- 2.1 Obtain the EnhanceShortInsert.rec custom recipe file.
- 2.2 Load the custom recipe file during instrument set-up:
 - For planned runs, the custom recipe can be uploaded and specified in the Advanced Run Settings section of the planned run form in ElemBio™ Cloud.
 - For manual runs, the recipe can be loaded onto the instrument with a USB, then selected from the Custom Recipe dropdown in the Advanced Settings menu.

3. Sequencing run set-up

- 3.1 Prepare the sequencing run with your sequencing kit according to the Cloudbreak Sequencing User Guide. Prior to loading the sequencing cartridge into the instrument reagent bay, pierce the Index 1 primer tube (highlighted in the red box in the figure below). Add 1.3 mL of Library Loading Buffer from your sequencing kit to the Index 1 tube. Pipette 10 times with a 1 mL pipette to mix.



- 3.2 Confirm that the EnhancedShortInsert custom recipe has been selected (in ElemBio Cloud for planned runs or during run setup for manual runs). Sequencing run setup can proceed according to the Cloudbreak Sequencing User Guide with no additional changes.

IV. Software guide for duet sequencing on Element AVITI™ sequencing platforms

- To analyse duet evoC or duet +modC sequencing data generated on Element AVITI™, an additional run parameter must be included when launching the pipeline.

```
--additional-params prelude.phred_binning_strategy=element_aviti_4_bins
```

- This is described in the “inferred instrument type” section of the duet software installation and running guide (CLI).
- This functionality is supported from CLI 2.0.0 and pipeline 1.5.0.
- If you are planning on merging data from multiple runs, instructions on sample ID naming conventions is provided in the section “merging data from multiple runs” in the duet software installation and running guide.

V. Revision history

Version	Date	Description
1	February 2026	Protocol release