

Protocol for AmbiSet 15


1. Sample Preparation

- a. Prepare the sample by thoroughly mixing the BioFix Buffer with your reagent of choice.

We recommend mixing your reagent of choice with the BioFix Buffer at a 1:1 ratio, with a maximum total volume per column type listed in the table below.

We do not recommend exceeding 1 μg of reagent per 1 μl of BioFix Buffer.

We also recommend adding a carrier protein, such as BSA, at 100 mg/mL to reduce protein loss to surfaces. This is especially important when working with protein concentrations of $< 100 \mu\text{g/mL}$. Make sure to mix gently until clear - do not use if the mixture is foamy or cloudy.

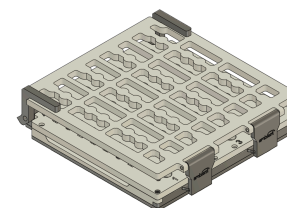
Column Type	Maximum Total Loading Volume (μl) (reagent + BioFix Buffer)	Maximum mass of reagent per column (μg)	Cycle Name	Cycle Duration
AmbiSet 15 	15	7.5	AS15	60 minutes

2. Stabilizer Initialization

- a. Press the white button to turn on the Stabilizer.
- b. Insert the SD card and press the green button to confirm. Use the red button to visualize different cycle options and use the green button to select a cycle. Once the cycle is confirmed, the Stabilizer will start to initialize. A series of beeps will indicate the chamber is ready for samples.

3. AmbiSet Column Preparation

- a. Open the white AmbiSet tray (included in the equipment box and pictured on the right) by unlatching the 2 locks at the front.
- b. Place the AmbiSet columns into the tray. Open each column.



- c. Pipette the BioFix Buffer/reagent mixture onto the scaffold at the bottom of each column. Press closed the column lids.
- d. Close the lid on the tray and secure it by locking the 2 latches on the front.

4. Run the Stabilization Cycle

- a. Confirm the chamber is ready by checking the screen says 'Start Cycle?'
- b. Place the tray into the chamber and close the chamber door, tightening the knobs clockwise.
- c. Press the green button to begin the cycle.

5. Sample Storage

- If you are using the samples immediately after stabilization, go to step 6.
 - a. Place the AmbiSet columns into the supplied 2 mL tubes.
 - b. Place the assembled column/tubes into the supplied mylar bag (with desiccant).
 - c. Zip seal the mylar bag shut. Clip seal using the provided closures.
 - d. The sample can now be stored at ambient temperature.

6. Sample Reconstitution

- a. The stabilized sample can be eluted from the column with 100-600 μ l using your desired eluent.
- b. Incubate at room temperature for ≥ 30 seconds.
- c. Centrifuge the spin column-tube assembly at $\geq 300 \times g$ for 2 minutes.
- d. Discard the column.
 - a. The flow-through now contains your reagent and is ready for use.

