Hydrodynamic addressing in Biacore 4000 when using pre-immobilized sensor chips

This protocol describes how the hydrodynamic addressing (HA) procedure in Biacore 4000 should be run when using pre-immobilized sensor chips like Sensor Chip Protein A. For other sensor chips, the normal HA protocol should be run.

Preparation of solutions

1. **Prepare running buffer HBS-EP+:**

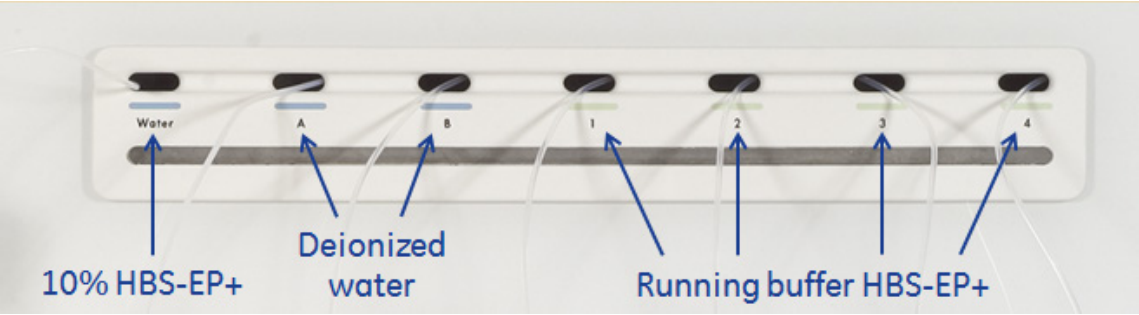
   Mix 100 ml 10x HBS-EP+ (BR-1006-69) with 900 ml deionized water.

2. **Prepare 10% HBS-EP+:**

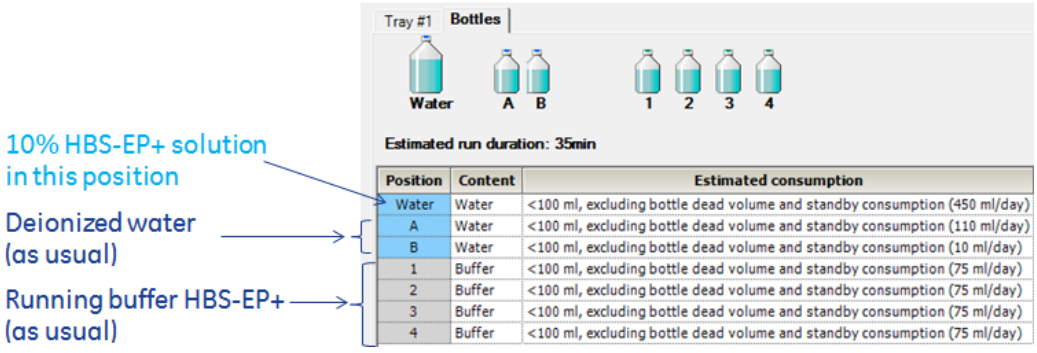
   Mix 100 ml running buffer HBS-EP+ with 900 ml deionized water.

Hydrodynamic addressing

1. Dock a chip and select **Hydrodynamic Addressing with normalize**.
2. Place bottles with 10% HBS-EP+, deionized water and running buffer in the instrument and insert the tubing according to the picture below.



The only difference compared to the instructions in the software is that the tubing marked "Water" should be placed in the bottle with 10% HBS-EP+ solution:



Position	Content	Estimated consumption
Water	Water	<100 ml, excluding bottle dead volume and standby consumption (450 ml/day)
A	Water	<100 ml, excluding bottle dead volume and standby consumption (110 ml/day)
B	Water	<100 ml, excluding bottle dead volume and standby consumption (10 ml/day)
1	Buffer	<100 ml, excluding bottle dead volume and standby consumption (75 ml/day)
2	Buffer	<100 ml, excluding bottle dead volume and standby consumption (75 ml/day)
3	Buffer	<100 ml, excluding bottle dead volume and standby consumption (75 ml/day)
4	Buffer	<100 ml, excluding bottle dead volume and standby consumption (75 ml/day)

3. Start the Hydrodynamic Addressing run.
4. **Important!** To avoid salt precipitation in the instrument, the 10% HBS-EP+ solution must be replaced with deionized water after the hydrodynamic addressing run and the tool **Change Buffers 1-4** should be run.

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