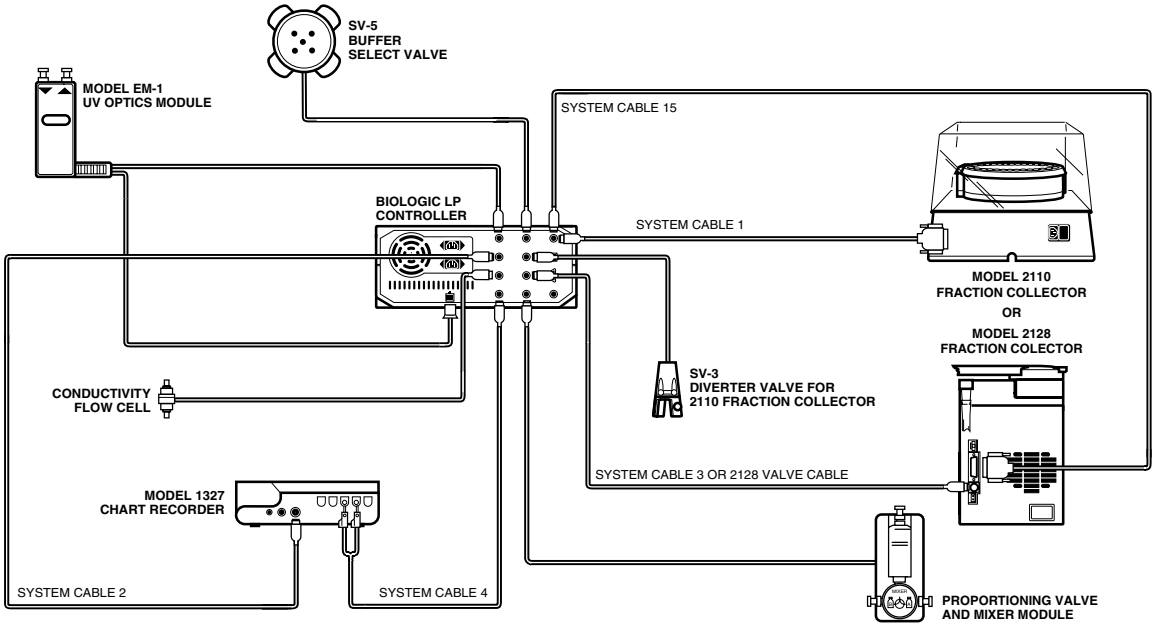
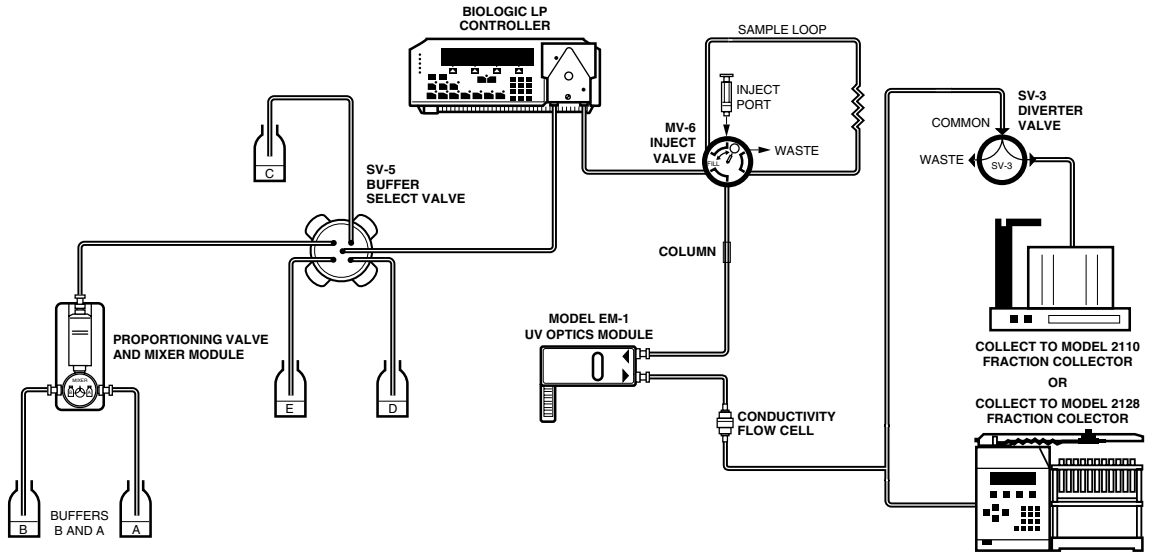
BioLogic LP Quick Start Tutorial

The BioLogic LP Starter Kit (Catalog # 731-8350) allows separation of a standard anion exchange mixture using the Econo Pac High Q cartridge. Cabling and plumbing, platen adjustment, UV setup, and conductivity min and max settings are to be completed prior to programming; instructions can be found in the BioLogic LP Starter Kit Instruction Manual, page 3–9. The illustrations below show the suggested cabling and tubing connections for the BioLogic LP system.

SYSTEM CABLING



SYSTEM TUBING



1. Buffer and Sample Preparation.

a. Buffer "A": Empty the contents of the bottle labeled Buffer "A" into a 500 ml graduated cylinder. Add degassed, deionized water to 500 ml volume. Stir briefly. Label container as Buffer "A".
b. Buffer "B": Prepare Buffer "B" and label container as described above.
c. Sample: Dissolve the anion exchange standard in Buffer "A" using the syringe to measure 6.5 ml of buffer into the bottle of protein standards. Replace the stopper and shake gently to dissolve.

2. Press the Program mode key; select New Method.

3. Select Time programming mode.

4. Program the Pump.

a. Press ADD.
b. Select Buffer "A" with the Previous/Next keys; press OK.
c. Enter the step length of 1 minutes; press OK.
d. Enter flow rate of 3.0 ml/min; press OK.
e. You have now entered the first step of the method (Buffer "A", step length of 1 minute, flow rate of 3.0 ml/min.) Enter the remaining steps.
Step 2. Gradient 0% to 50% "B", step length 5 minutes, flow rate 3.0 ml/minute.
Step 3. Buffer "B", step length 3 minutes, flow rate 3.0 ml/minute.
Step 4. Buffer "A", step length 3 minutes, flow rate 3.0 ml/minute.
f. After entering Step 4; press OK.

5. Set the Alarm.

a. Press the Alarm soft key.
b. Press ADD.
c. Enter a time of 1 minute for Alarm 1 and check that Hold Methods is set to NO.
d. Press OK twice.
The alarm sounds after 1 minute to remind you to turn the valve back to the load position. If left in the Inject position, the gradient will flow through the loop first instead of going directly to the column.

6. Enter the fraction collector program.

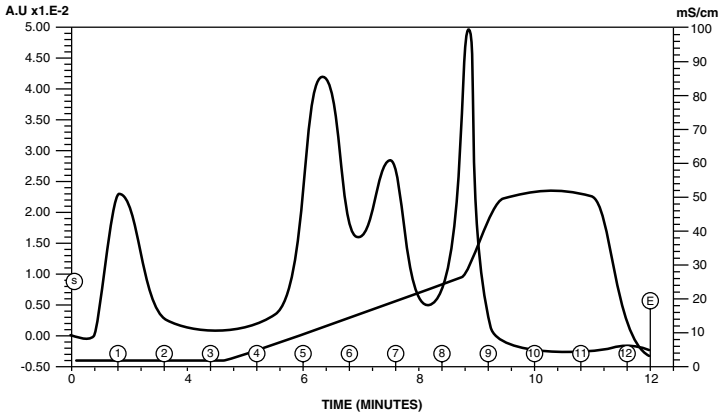
a. Press the Frac Coll soft key.
b. Select ALL.
c. Enter a fraction size of 0.5 minute.
d. Press OK twice, then select DONE.
e. Press SAVE, name the method "DEMO 1", and press DONE.

7. Load Sample loop with protein standard.

a. Turn the MV-6 Injector valve knob counter-clockwise as far as it will go.
b. Draw 2 ml of the protein standard mixture into the syringe.
c. Insert the syringe in the top port of the MV-6 injector valve, and fill the sample loop. Leave the syringe in the port when you have injected the sample — this prevents the sample from siphoning out of the loop.

8. Start Method.

a. Press the Run mode key. The system will count down 10 seconds, then start the method.
b. When the method starts, turn the injector valve knob to the right as far as it will go. When the alarm sounds, turn the valve knob to the left.
c. If you are using a Bio-Rad 2128 fraction collector, press "Park", then "Yes" when method is complete.



RUN CONDITIONS

Column: Econo Pac High Q Cartridge (Catalog #732-0028)

Buffer A: 25 mM Tris HCl, pH 8.1

Buffer B: 25 mM Tris HCl, pH 8.1 + 0.5 M NaCl

Standard: Anion Exchange Std. (Catalog #125-0561)

Equine myoglobin is not retained on the column — it elutes in the void volume (first peak). The remaining three proteins bind to the column and elute separately as the salt concentration increases.

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