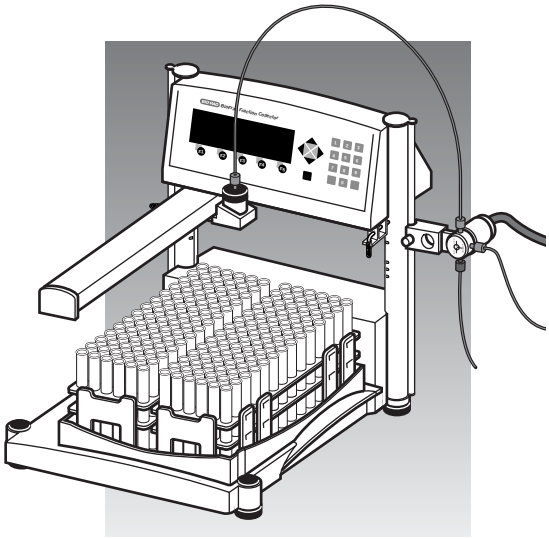

# BioFrac™ Fraction Collector

## Instruction Manual

For technical service
call your local Bio-Rad office or
in the U.S. call 1-800-4BIORAD
(1-800-424-6723)
On the Web at discover.bio-rad.com

BIO-RAD

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# Safety

**Caution/Warning**

Disconnect power to the fraction collector before servicing. No user-serviceable parts are inside. Refer servicing to Bio-Rad service personnel.

This Bio-Rad BioFrac fraction collector is designed and certified to meet EN 61010* safety standards, and meets the compliance requirements of EN 61326(1998)** for conducted and radiated electromagnetic emission. Certified products are safe to use when operated in accordance with the instruction manual. This safety certification does not extend to other chromatography equipment or accessories not EN 61010 certified, even when connected to this fraction collector.

This instrument should not be modified or altered in any way. Alteration of this instrument will void the manufacturer's warranty, void the EN 61010 certification, and create a potential safety hazard for the user.

Bio-Rad is not responsible for any injury or damage caused by the use of this instrument for purposes other than for which it is intended or by modifications of the instrument not performed by Bio-Rad or an authorized agent.

Inaccurate dispenser arm positioning or interruption of the programmed collection method may result if contact with the dispenser arm or diverter valve occurs while unit is in operation. The diverter valve should be handled only during installation setup or plumbing of the valve according to procedures outlined in the instruction manual.

* EN 61010 is an internationally accepted electrical safety standard for laboratory instruments.
**EN 61326 is an internationally accepted standard for conducted and radiated electromagnetic emission.

# Section 1
# Introduction

## 1.1 Overview

The BioFrac fraction collector provides automated collection options for analytical and preparative chromatography applications. It can be used as a stand-alone collector or as a companion to any chromatography system. The BioFrac fraction collector is capable of doing basic to complex fraction collection schemes and can be used at flow rates up to 100 ml/min. The fraction collector accommodates numerous rack options from microplates to bottles and carboys.

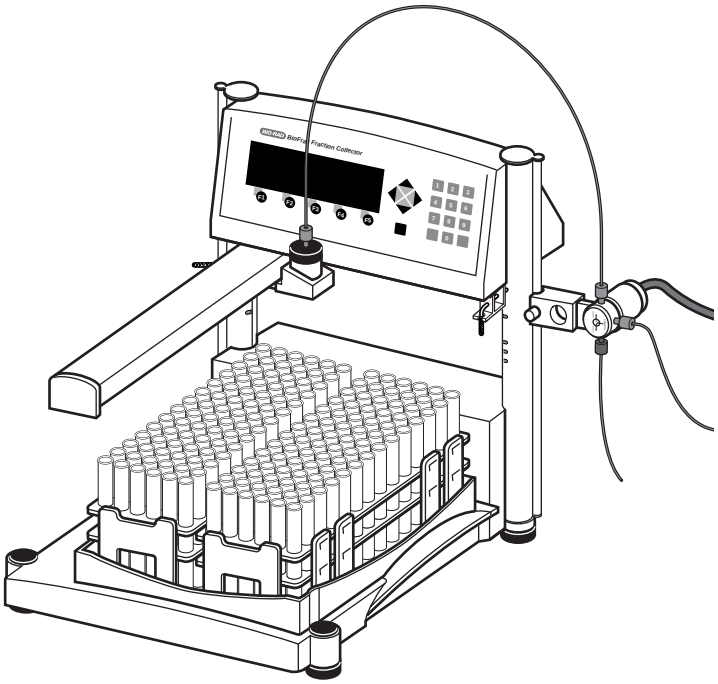


**Fig. 1. BioFrac fraction collector, shown with two F1 racks (12–13 mm Tubes).**

## 1.2 Features

Key features of the BioFrac fraction collector include:

- Microprocessor control, with easy-to-use front panel controls and a menu-driven software interface for method setup.
- Method library for saving up to 20 user-defined fraction collection methods
- Local and remote starting of the fraction collector.
- Collection by time, volume, or drops.

- Advanced fraction collection functions. Peak detection, Time, or Volume Windows (up to 20), or a combination of Peak Detection with Time or Volume Windows.
- Manually adjustable control module that accommodates tube heights up to 150 mm
- Compatibility with Bio-Rad and non-Bio-Rad chromatography systems.
- Modular. Can be stacked directly on top of the BioLogic DuoFlow and BioLogic LP chromatography systems.
- Accommodates several inexpensive, off-the-shelf racks for tubes (12–20 mm, and 30 mm diameter), Eppendorf/microtubes (0.5 ml, 1.5 ml, and 2 ml) and scintillation vials. Racks are autoclavable.
- Collection of up to 180 fractions in tubes or 384 fractions in 96-well microplates.
- Optional Prep-20 adaptor for preparative collection in up to 20 collection vessels of any size, from milliliter to liter collection volumes.
- Optional ice bath (with tube grips) that doubles as a holder for microplates. Tube grips hold tubes firmly in any position while decanting. Rack allows cooling of 13 mm tubes on ice. As a microplate holder it accommodates 12-, 24-, 48-, and 96-well microplates and Titertube™ tubes that adhere to SBS standards for microplates.
- Diverter valve, to divert flow and minimize spillage during tube changes. Diverts unwanted eluent to waste, and eliminates spills during fraction advances.
- Multirun capability for overlaying fractions or collection of experiments sequentially.
- Serpentine arm movement. Can be changed to a column or row pattern for microplates and Titertube tubes.
- Optional Drop Former that is optimized for small volume drop dispensing (recommended when collecting into microplates).
- Ability to start/stop an external pump and chart recorder.
- Screen sleep mode for longer display life.

## 1.3 Unpacking

When unpacking the fraction collector, carefully inspect the containers for any damage that may have occurred in shipping. Severe damage to a container may indicate damage to its contents. If you suspect damage to the contents may have occurred, immediately file a claim with the carrier in accordance with their instructions before contacting Bio-Rad Laboratories.

**Caution**
Do not lift the fraction collector by its dispenser arm!

Grip the base of the fraction collector and lift it slowly out of its packing. Do not lift the unit by its dispenser arm. Remove the remaining contents from each of the boxes and check all of the parts against the supplied packing list. The BioFrac fraction collector is shipped with the following:

- Fraction collector unit
- AC power cord

- Fittings kit (shown in Figure 2 below)
- Instruction manual
- Tube rack # F1 (2) for 12–13 mm tubes
- Diverter valve (including an Allen wrench)
- Combicon connector (included with instruction manual)

If any part is missing or damaged, contact Bio-Rad Laboratories immediately.

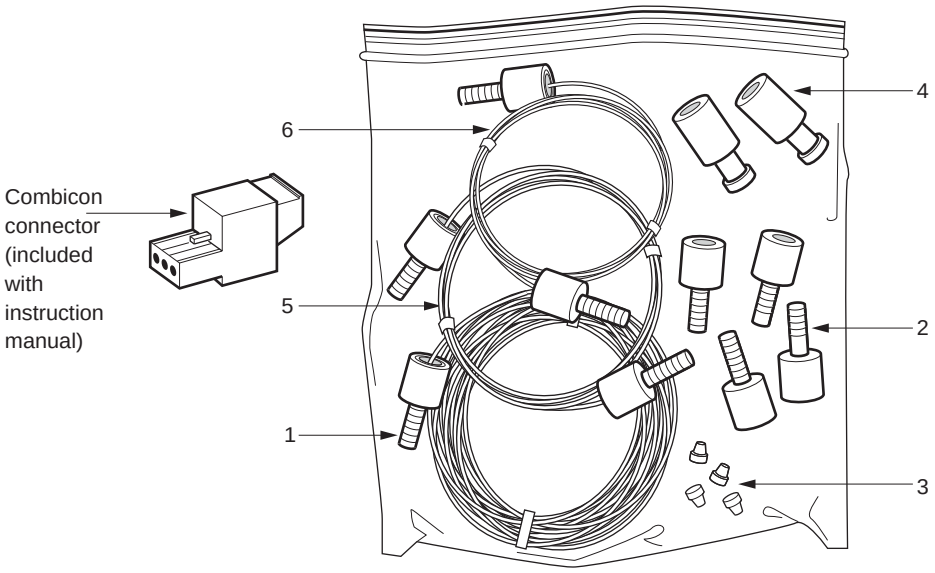



**Fig. 2. Fittings kit. (See Table 1 for key to numbered components.)**

## Table 1. Fittings Kit Contents

| Item Number | Quantity | Contents of Fittings Kit |
|---|---|---|
| 1 | 1 | 1.0 meters Tefzel Tubing, 1/16" OD, .030" ID with one 1/4-28 fitting |
| 2 | 4 | Fittings, 1/4-28, 1/16" OD |
| 3 | 4 | Ferrules, 1/16" OD |
| 4 | 2 | Union, luer to 1/4-28 |
| 5 | 1 | 26 inch PEEK ID 0.030" 1/16" OD, with two 1/4-28 fittings labeled collect HF |
| 6 | 1 | 26 inch PEEK ID 0.020" 1/16" OD, with two 1/4-28 fittings labeled collect |

## 1.4 Physical Description

Refer to Figure 3 for the following discussion.

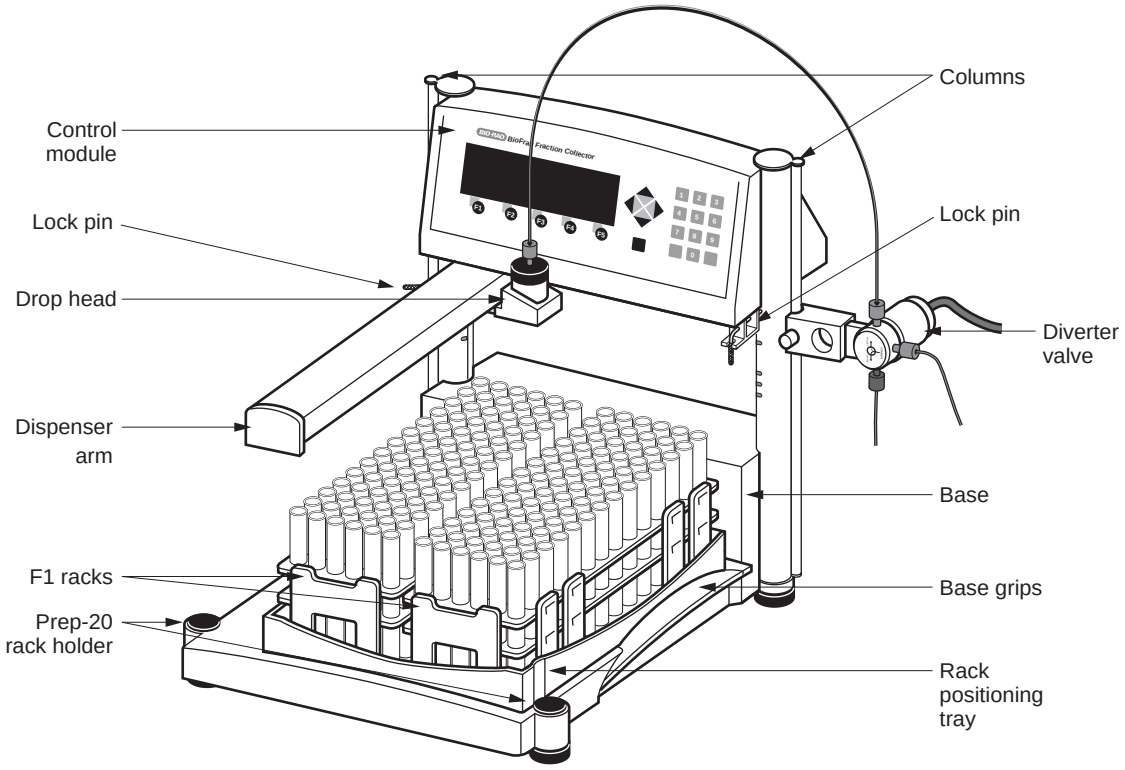



**Fig. 3. Physical features of the BioFrac fraction collector.**

**Control Module** — The fraction collector head contains the display, function keys, and alphanumeric keypad (see Section 2). Its height may be adjusted to accommodate tube heights up to 150 mm.

**Base** — The fraction collector base holds the rack positioning tray, power switch, and I/O connectors. It is designed so that it can be stacked on top of the BioLogic DuoFlow and LP workstations if benchspace is limited.

**Columns** — The columns hold the fraction collector head and serve as a rack for mounting the diverter valve or other devices such as pH probe and UV monitor using Bio-Rad bar clamps (Cat. #750-0265).

**Rack Positioning Tray** Accurate dispensing requires that racks be positioned using the rack-positioning tray. Molded indents on the tray allow the rack feet to be placed securely on the rack-positioning tray. The tray holds two full racks (F1, F2, or F3) on one face and can be inverted for positioning of four half racks (H1, H2, H3, or H4). See Section 3.3 for available rack options.

**Dispenser Arm** Moves the drop head in a serpentine X-Y motion over each of the collection tubes. Can be changed to a column or row pattern for microplates and Titertube tubes.

**Drop Head** Consists of the drop former, a photodiode cell for drop counting, and a clear glass tube that protects the photodiode cell from splashes. The inlet tubing is connected to the drop former, which provides uniform drop size. The drop former accepts 1/4-28 fittings. An optional drop former is available that has been optimized for small volume drop dispensing (25 µl drops).

**Diverter Valve** The diverter valve minimizes spillage during drop head movement. The BioFrac diverter valve is designed to have a minimal internal volume and is 100% flushed in order to minimize loss of chromatographic resolution.

**Lock Pin** The height adjustment lock pins, located on the left and right front of the control module, are used to secure the drop head height.

# Section 2
# Front and Rear Panel Controls and Connectors

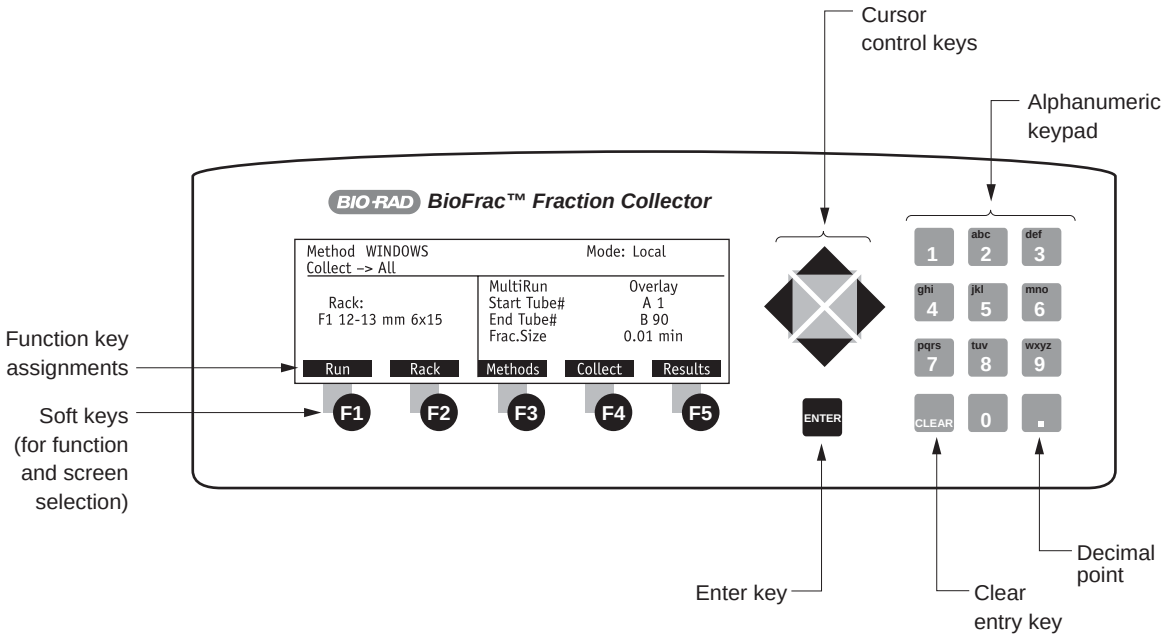



**Fig. 4. Front Panel Controls.**

## Table 2. Front Panel Controls

| Key | Description |
|---|---|
| Cursor keys | Used to move the cursor on the LCD display up, down, left, or right. |
| Function keys | These five keys are located directly below the LCD display. The function each key executes is displayed above it. |
| Clear Entry key | Clears a cursor field or closes a menu option list. |
| Decimal point key | Used to enter a decimal point. |
| Enter key | Accepts a numeric value that has been entered. |
| Alphanumeric keypad | Used to enter a decimal value in numeric fields and alphanumeric characters in text fields. In a text field the character displayed is incremented with each press of the keypad (i.e., 2→A→B→C→2, etc.). |

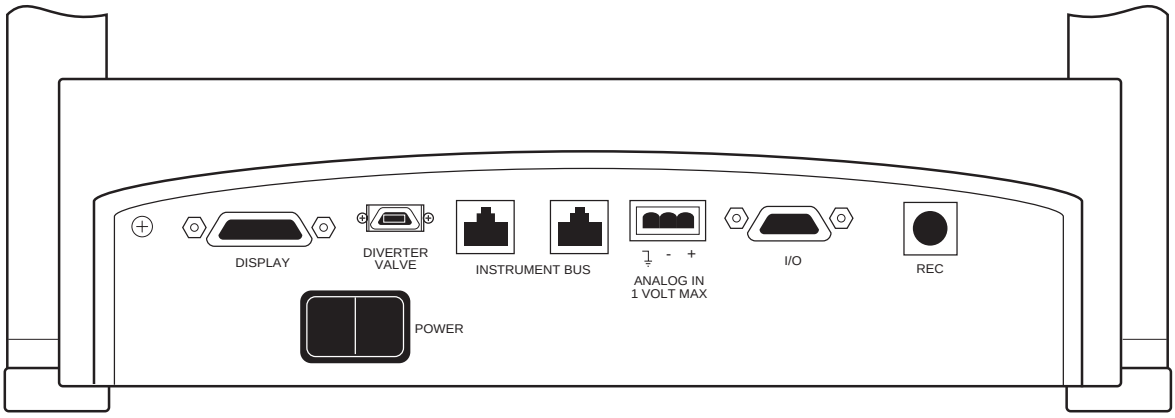
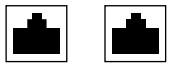



**Fig. 5. Rear Panel Connectors.**

## Table 3. Rear Panel Connectors

| Connector | Description |
|---|---|
| DISPLAY | **Display cable:** Connects the fraction collector control module to the power supply and other ports in the base of the fraction collector. |
| DIVERTER VALVE | **Diverter Valve Connector:** Used to connect the diverter valve to the fraction collector. |
| I/O | **I/O connector:** A 15-pin D connector used for connecting the following instruments to the BioFrac fraction collector:<br>• BioLogic LP system, Model EP-1 Econo pump, and BioLogic HR system. These systems control fraction advances.<br>• Econo gradient pump. The fraction collector controls all fraction collection parameters.<br>• External pump. With a BioFrac accessory cable (15-pin to bare wires) connected to the I/O port, the fraction collector can receive start, stop, and fraction advance signals from an external pump. It can also send a start or stop signal to a pump. (The pump must have compatible control circuitry logic. Refer to Appendix B.) |

| | |
|---|---|
|  | **Instrument Bus:** For connection of the BioFrac to a DuoFlow system using system cables 17, 18, 19, or 21. |
|  | **REC connector:** This 8-pin mini-DIN connector is for controlling the diverter valve or a chart recorder:<br>• When connected to a BioLogic LP system or Model EP-1 Econo pump, the connector receives diverter valve control signals.<br>• When connected to a chart recorder, the fraction collector controls paper feed, pen up/down and event marks. (The chart recorder must have compatible control circuitry logic. Refer to Appendix B.) |
|  | **Combicon connector:** Used to connect a UV monitor or other detector for Peak Detection by Threshold. (Refer to Section 3 for cable information.) |

# Section 3
# System Configuration and Plumbing

## 3.1 Fraction Collector Setup

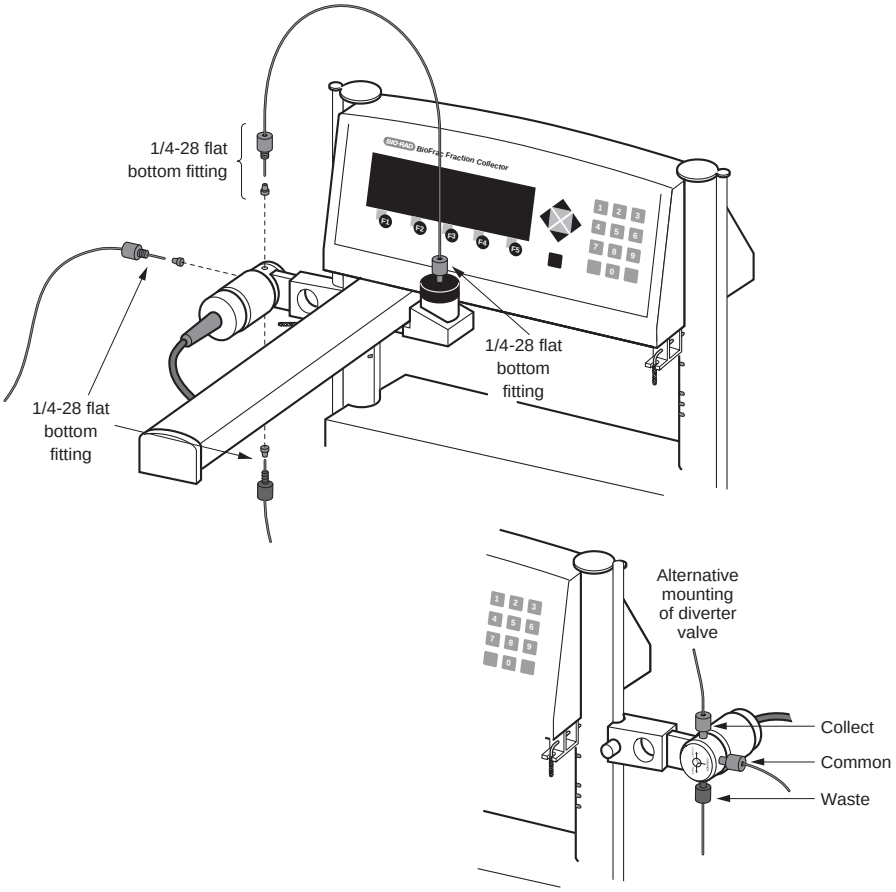



**Fig. 6. Plumbing the BioFrac fraction collector.**

The BioFrac fraction collector is shipped assembled and requires minimal plumbing and cabling to prepare it for use. To set up the BioFrac fraction collector:

1. Place it on a level surface on a laboratory or coldroom bench or in a cold cabinet. Alternatively, if benchspace is limited, the modular fraction collector can be stacked on top of the BioLogic DuoFlow or LP system workstations.

2. Connect the fraction collector display cable to the port labeled Display on the back of the fraction collector.

3. Connect the diverter valve cable to the Diverter Valve port on the back of the fraction collector.

4. Attach the diverter valve to either the right or left column. The diverter valve should be attached so that the Collect port is pointed up and Waste port is pointing down (see Figure 6). Use the included Allen wrench to secure the diverter valve to the column.

5. Connect all power cords to available grounded, surge protected outlets.

6. Connect the preassembled 26 inch PEEK 1/16" OD tubing, supplied in the BioFrac Fittings kit, to the Collect port of the diverter valve and to the drop head. Use the 0.02" ID (orange) tubing for flow rates less than 20 ml/min and the 0.03" ID (Green) tubing for flow rates greater than 20 ml/min.

   Note: the diverter valve is rated to a maximum pressure of 30 psi.

   If tubing other than that found in the fittings kit is used to plumb the fraction collector, make sure all tubing lengths are kept to a minimum. This reduces the delay volume (See Section 5.5) and backpressure. Tubing choice is dependent on the flow rate and pressure characteristics of the pumping system. For flow rates above 20 ml/min, use 0.03" ID or 1/8" OD, 0.062" ID tubing (available from most tubing/fitting suppliers).

7. Attach the 0.03" ID Tefzel tubing, supplied in the fraction collector fittings kit to the Waste port of the divert valve and place the other end into a waste collection vessel. Note, if using flow rates greater than 20 ml/min, be sure to use tubing that has a 0.03" ID or larger.

8. Attach the tubing from your chromatography system to the Common port on the diverter valve using either a 1/4-28 fitting or luer to 1/4-28 Union. The BioFrac fittings kit includes 1/4-28 fittings ( for 1/16" OD tubing) and luer to 1/4-28 Unions.

9. Connect the fraction collector to your chromatography system as described in Section 3.2.

## 3.2 Connecting Other Instruments and Devices

The BioFrac fraction collector can be operated in any of the following configurations:

- BioLogic DuoFlow system control. The DuoFlow system controls the operation of the fraction collector through the system bus. Refer to Section 3.2.1 for details.
- LP/Econo mode collection with Bio-Rad components such as the BioLogic LP system, Model EP-1 Econo Pump, and BioLogic HR system. The BioLogic LP system, Econo pump, and BioLogic HR system, control all aspects of fraction collection. Refer to Sections 3.2.2 and 3.2.3 for details.
- Stand-alone fraction collection (Local mode) with Bio-Rad components, such as the Econo gradient pump, a BioLogic QuadTec detector, a Model EM-1 Econo UV monitor, and/or a Model 1327 chart recorder. Note: the fraction collector provides complete control of fraction collection. Refer to Sections 3.2.4, 3.2.5, and 3.2.6 for details.
- Stand-alone fraction collection with non-Bio-Rad components, such as a pump, UV monitor and/or chart recorder. In Local mode, the fraction collector provides complete control of fraction collection and can start/stop an external device such as a pump or chart recorder. In LP/Econo mode, the non-Bio-Rad controller provides complete control of fraction
  collection. Non-Bio-Rad devices connected to the BioFrac must be capable of Transistor to Transistor Logic (TTL) control. Refer to Section 3.2.8 for details.

**Note**: Before proceeding, make sure power is turned off to each component to be connected. Be sure to use a grounded, surge protected outlet when plugging in the power cables.

### 3.2.1 Connection to Bio-Rad's BioLogic DuoFlow system

Connect the BioFrac fraction collector to the DuoFlow system using system cables 17, 18, 19, or 21.

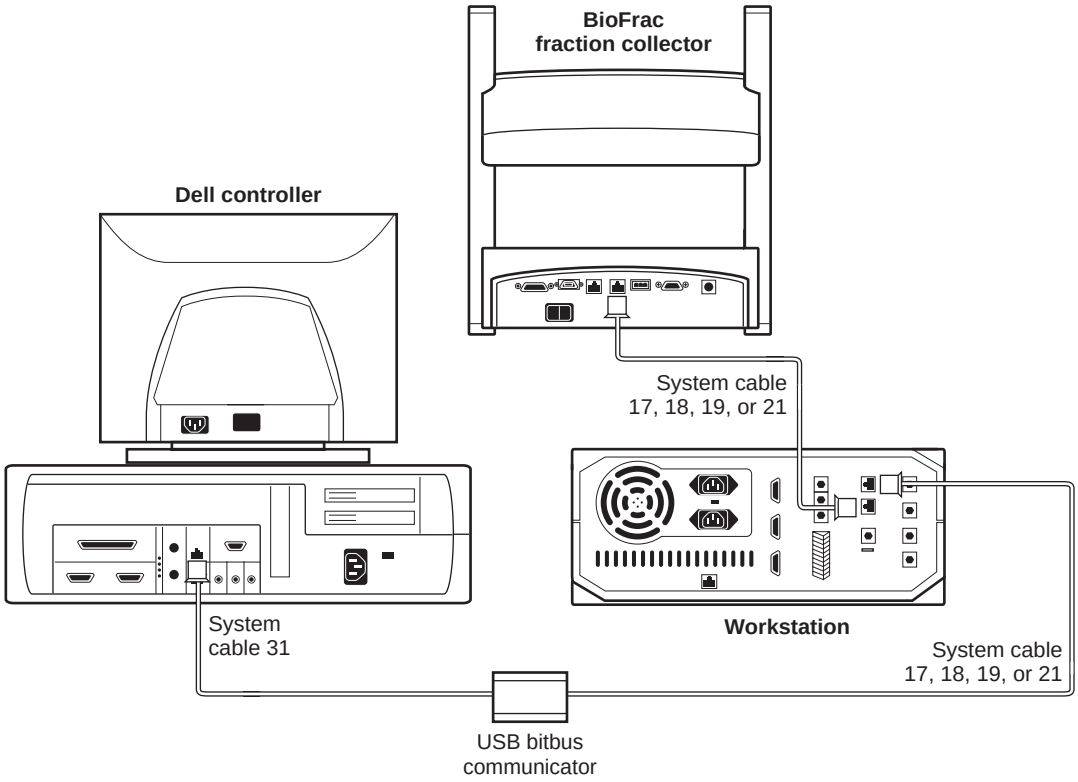



**Fig. 7. Connecting the fraction collector to a BioLogic DuoFlow System**

1. Connect the bus cable to the connector marked INSTRUMENT BUS on the base of the fraction collector.

2. Connect the other end of the bus cable to either of the INSTRUMENT BUS connector on the back of the DuoFlow workstation or to other Bio-Rad components that are daisy chained to the DuoFlow workstation by the instrument bus.

3. Place the BioFrac fraction collector in Local mode with the Main screen displayed (See Section 4.1). Start the DuoFlow software and press System in the manual screen fraction collector dialog box. Note, the BioFrac must be in Local mode and on the Main screen before the BioLogic DuoFlow system can control it.

### 3.2.2 Connection to Bio-Rad's BioLogic LP System or a Model EP-1 Econo Pump

To connect the fraction collector to the BioLogic LP system or Model EP-1 Econo pump, you will need system cables 3 and 15. System cable 15 relays the fraction advance signal to the BioFrac, and system cable 3 allows the BioLogic LP system or Model EP-1 Econo pump to control the BioFrac diverter valve.

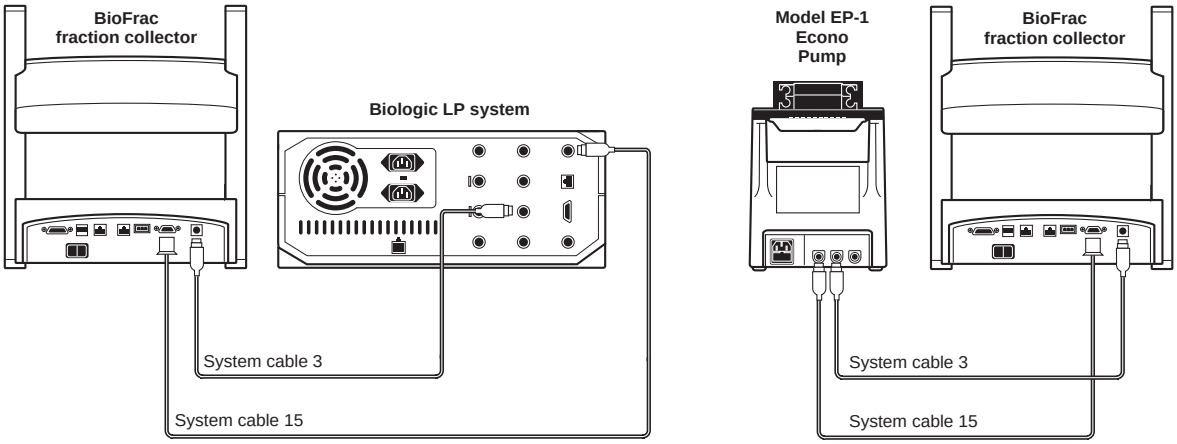



**Fig. 8. Connecting the fraction collector to a BioLogic LP system and Model EP-1 Econo pump.**

1. Connect the fraction collector to the BioLogic LP system or Model EP-1 Econo pump using system cable 15.
   a. Connect the cable's 15-pin D connector to the port labeled I/O on the fraction collector.

   b. Connect the mini-DIN connector to the port labeled Fraction Collector on the BioLogic LP system or the port labeled on the Model EP-1 Econo pump.

2. Connect the fraction collector to the BioLogic LP system or Model EP-1 Econo pump using system cable 3.
   a. Connect one of the cable's mini-DIN connector to the REC port on the fraction collector.

   b. Connect the other mini-DIN connector to the port labeled Diverter Valve on the BioLogic LP system or the port labeled on the Model EP-1 Econo pump.

3. If you are connected to a BioLogic LP press the Collector button on faceplate and then press the MODEL softkey followed by the BIOFRAC (or 2128) softkey. When asked if an external valve cable is connected answer "YES".

4. Follow the procedure in Section 6, LP/Econo Mode Operation.

### 3.2.3 Connection to Bio-Rad's BioLogic HR System

To connect the fraction collector to the BioLogic HR system, you will need a BioFrac accessory cable, system cable 7, and an SV3-2 diverter valve. In this configuration, the SV3-2 valve is used for control of windows and threshold collection by the BioLogic HR system.

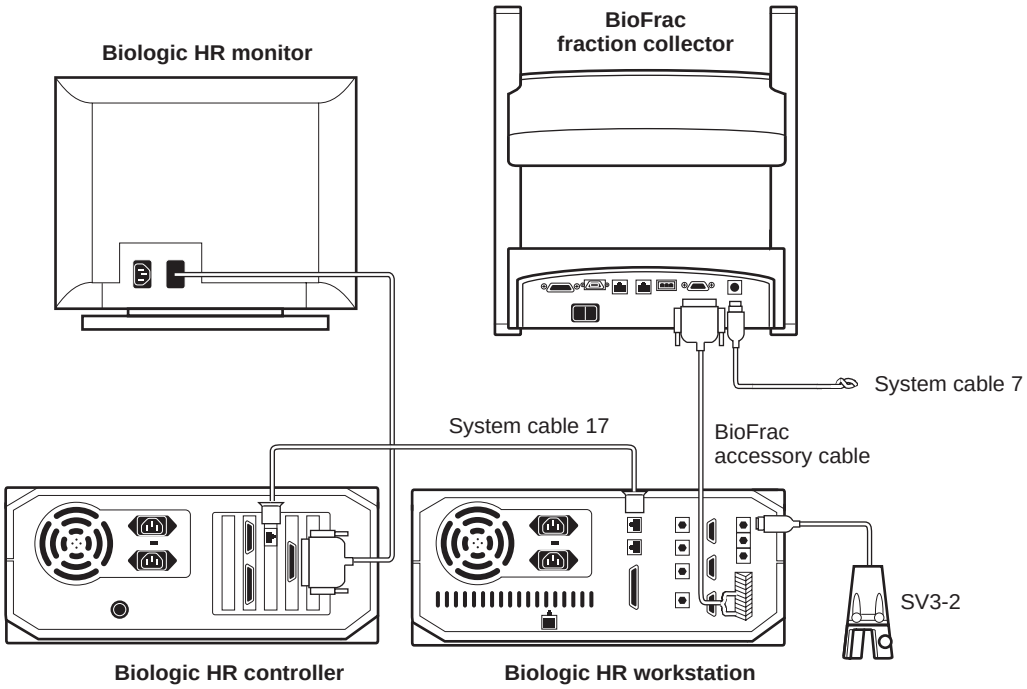



**Fig. 9. Connecting the fraction collector to a BioLogic HR system.**

1. Connect the fraction collector to the HR system using the BioFrac accessory cable.
   a. Connect the cable's 15-pin D connector to the I/O port on the fraction collector.
   b. Connect the black wire to the fraction advance pin on the BioLogic HR system Aux port (pin #5). Connect the blue/white wire to Aux port ground (pin #9) on the BioLogic HR system.

2. Connect the system cable 7 mini-DIN connector to the BioFrac REC Port. Connect the system cable 7 orange wire and green wire together. This allows the BioFrac diverter valve to open during fraction collection.

3. Connect an SV3-2 valve (catalog #750-0410) to the BioLogic HR system and mount it on the BioFrac fraction collector column next to the BioFrac diverter valve. Plumb the SV3-2 valve so that its Collect port is connected to the BioFrac diverter valve common port. Plumb the waste port on each valve to go to waste.

4. Program the fraction collector as described in Section 6, LP/Econo Mode Operation.

5. In the BioLogic software, define the BioFrac as a "Generic" fraction collector.

### 3.2.4 Connection to the Bio-Rad Model EM-1 Econo UV monitor and Model 1327 Chart Recorder

To connect the fraction collector to a Model EM-1 Econo UV monitor and a Model 1327 chart recorder you will need system cable 4, system cable 16, and a 2-wire shielded cable (26 gauge or larger, available from most electronics or hardware stores). This simple isocratic system allows full use of the fraction collector's advanced programming features. To remotely start the Model EP-1 Econo pump from the BioFrac, see Section 3.2.7.

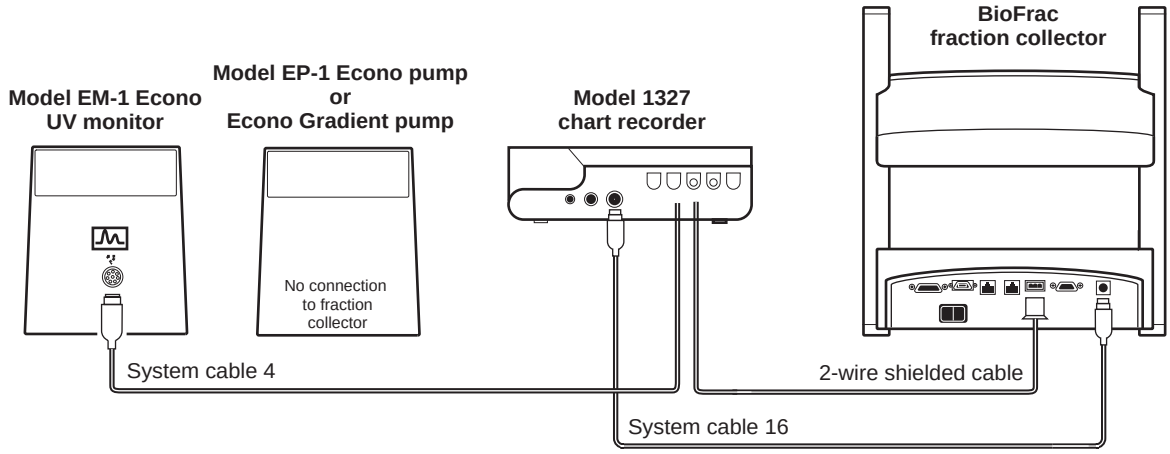



**Fig. 10. Connecting the fraction collector to Econo components.**

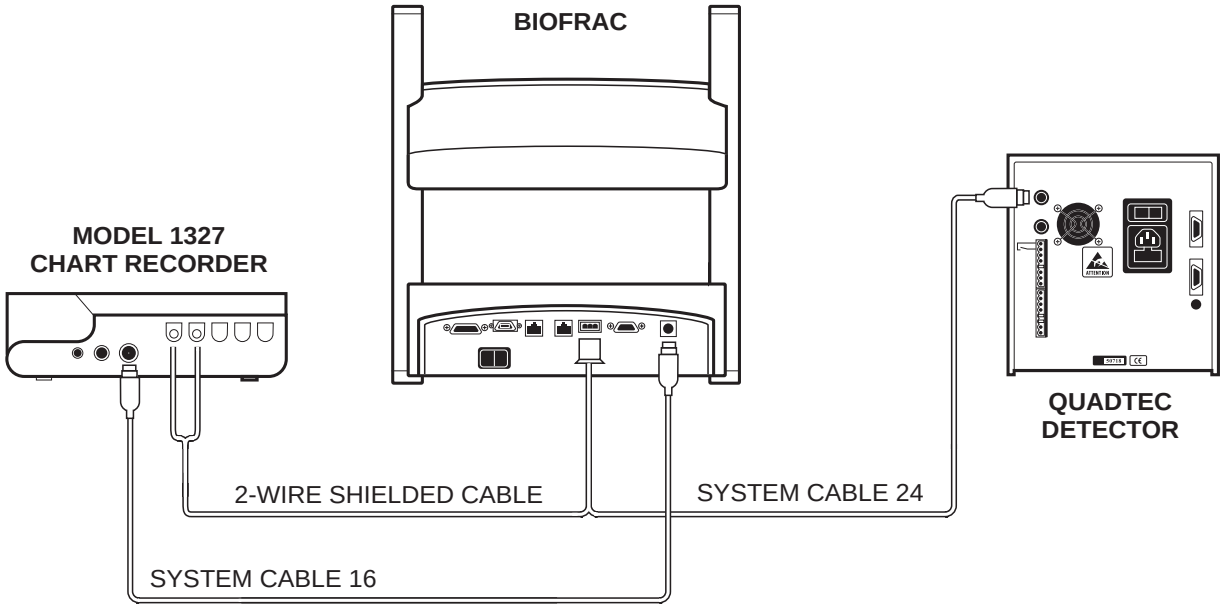
1. Connect the fraction collector to the Model 1327 chart recorder using system cable 16 (mini-DIN-to-DIN cable).
   a. Connect the 8-pin mini-DIN connector to the port labeled REC on the fraction collector.
   b. Connect the DIN connector to the DIN port on the chart recorder.
2. Connect the Model EM-1 Econo UV monitor to channel 1 of the Model 1327 chart recorder using system cable 4 (mini-DIN to banana plug cable).
   a. Connect the 8-pin mini-DIN connector to the Econo UV monitor's recorder port labeled ⎍.
   b. Connect the red (+) wire to the chart recorder's channel 1 (V) banana plug socket. Connect the black (–) wire to the channel 1 (⊥) banana plug socket. This provides 0 V to 1 V full scale. (Refer to the documentation for the Model 1327 chart recorder and the Model EM-1 Econo UV monitor for instructions on making connections to these units.)
3. Connect the fraction collector to the Model EM-1 Econo UV monitor via the Model 1327 chart recorder.
   a. Connect the fraction collector's UV input to the chart recorders signal input. To do this, connect one wire from the 2-wire shielded cable between the chart recorder's channel 1 (V) banana plug socket and the fraction collector's green plastic Combicon analog IN(+) connector. A Combicon plug is supplied with the BioFrac for this purpose (see Section 1.3).
   b. Connect the second wire from the 2-wire shielded cable between the chart recorder's channel 1 (⊥) banana plug socket and the analog IN(-) terminal on the green plastic Combicon plug.
   c. Connect the shield of the 2-wire cable to the analog ground on the Combicon plug.

d. Turn threshold collection on and set the full scale voltage to 1 V (see Section 4.6 of this manual).

4. Follow the procedures in Section 5, Stand-Alone Operation.

### 3.2.5 Connection to the Bio-Rad BioLogic QuadTec Detector and Model 1327 Chart Recorder

To connect the fraction collector to a BioLogic QuadTec detector and a Model 1327 chart recorder, you will need system cable 24 (You will need two system cable 24's if both QuadTec channels will be connected to the chart recorder), system cable 16 and 2-wire shielded cable (26 gauge or larger, available from most electronics or hardware stores). This system allows full use of the fraction collector's advanced programming features. To remotely start a Model EP-1 Econo pump or Econo Gradient pump from the BioFrac see section 3.2.7.




System Cable 24 and a 2-wire shielded cable are shown with their bare wires mated using a Combicon connector.

**Fig. 11. Connecting the fraction collector to a BioLogic QuadTec detector and a Model 1327 chart recorder.**

1. Connect the fraction collector to the Model 1327 chart recorder using system cable 16 (mini-DIN-to-DIN cable).
   a. Connect the 8-pin mini-DIN connector to the port labeled REC on the fraction collector.
   b. Connect the DIN connector to the DIN port on the chart recorder.
2. Connect the QuadTec UV/Vis detector to the Model 1327 by way of the BioFrac fraction collector.
   a. Connect the system cable 24 banana plug into one of the two integrator sockets on the rear of the QuadTec Detector.
   b. Connect the wires from system cable 24 to the screw terminals on the fraction collectors Combicon connector. The red wire should be attached to analog IN(+) and the black wire to analog IN(-) on the Combicon connector. A green Combicon plug is supplied with the

BioFrac for this purpose (see Section 1.3)

c. Connect one wire from the 2-wire shielded cable between the chart recorder's channel 1 (V) banana plug socket and the fraction collector's green plastic Combicon analog IN(+) connector.
d. Connect the second wire from the 2-wire shielded cable between the chart recorder's channel 1 (⊥) banana plug socket and the analog IN(-) terminal on the green plastic Combicon plug.
e. Connect the shield of the 2-wire cable to the Combicon connector analog ground.
f. Turn threshold collection on and set the full scale voltage to 1 V (see Section 4.6 of this manual)

3. Follow the procedures in Section 5, Stand-Alone Operation.
4. Optional, connect a second system cable 24 directly between the QuadTec and chart recorder.

### 3.2.6 Connection to the Bio-Rad Econo Gradient Pump and a Model 1327 Chart Recorder

The following configuration shows the fraction collector set up with an Econo gradient pump and a Model 1327 chart recorder. This system allows full use of the fraction collector's advanced programming features.

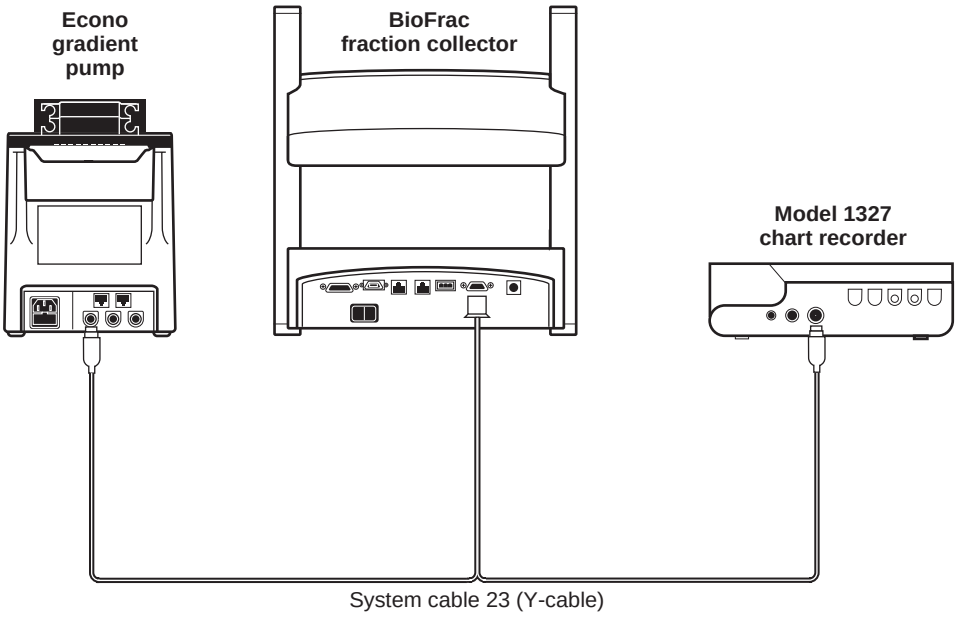



**Fig. 12. Connecting the fraction collector, Econo gradient pump and Model 1327 chart recorder.**

1. Connect the fraction collector to the Econo gradient pump (EGP) and Model 1327 chart recorder using system cable 23 (BioFrac Y-Cable).
   a. Connect the mini-DIN connector to the I/O connector on the Econo gradient pump.
   b. Connect the 15-pin D-connector to the I/O connector on the fraction collector.
   c. If a Model 1327 chart recorder is present, connect the cable's remaining end to the chart recorder DIN connector. If the chart recorder is not present, then place this portion of the cable aside.

2. If collecting fractions by threshold, connect a detector to the fraction collector and chart recorder as described in Sections 3.2.4 and 3.2.5.

3. Follow the procedures in Section 5, Stand-Alone Operation.

### 3.2.7 Connection and Remote Start of the Bio-Rad Econo Gradient Pump and Model EP-1 Econo Pump

The BioFrac fraction collector can start an Econo gradient pump and Model EP-1 Econo pump remotely. To make the necessary connections you will need a BioFrac accessory cable (catalog # 731-8290) and system cable 7 (catalog # 731-8267).

1. Connect the BioFrac accessory cable green/black and blue/white wires to the system cable 7 blue wire.
2. Connect the BioFrac accessory cables orange/black wire to the system cable 7 yellow wire if you are connecting to an Econo gradient pump or red wire if you are connecting to a Model EP-1 Econo pump.
3. Trim off all unused wires.
4. Connect the end of the BioFrac accessory cable with the 15-pin "D" connector to the I/O port on the BioFrac.
5. Connect the system cable 7 mini-DIN connector to the I/O port on the Econo gradient pump or Model EP-1 Econo pump. With the fraction collector in Local mode, press Run to start the fraction collector and pump. If you are collecting by volume, be sure to enter the pump flow rate into the BioFrac flow rate entry field.

Note: pressing stop on the BioFrac will not stop the Econo gradient pump run. The EGP program must be stopped from the Econo gradient pump faceplate.

### 3.2.8 Connection to Non-Bio-Rad Components

The BioFrac fraction collector is designed to work with a variety of non-Bio-Rad equipment. The BioFrac can control remote devices, such as pumps, chart recorders, and UV detectors, or can accept start/stop signals from other equipment such as pumps and UV detectors. Below is a description of the remote fraction collection control options. To connect the fraction collector to a non-Bio-Rad pump, UV monitor, and/or chart recorder, you will need the BioFrac accessory cable (15-pin to bare wires), system cable 7 (mini-DIN to bare wires), and 2-wire shielded cable (26 gauge or larger, available from most electronics or hardware stores). In addition, the pump and chart recorder circuit logic must be Start/closed circuit = ON; Stop/open circuit = OFF. The UV monitor's analog output must be 100 mV or 1 V. (Refer to Appendices B and C for details.)

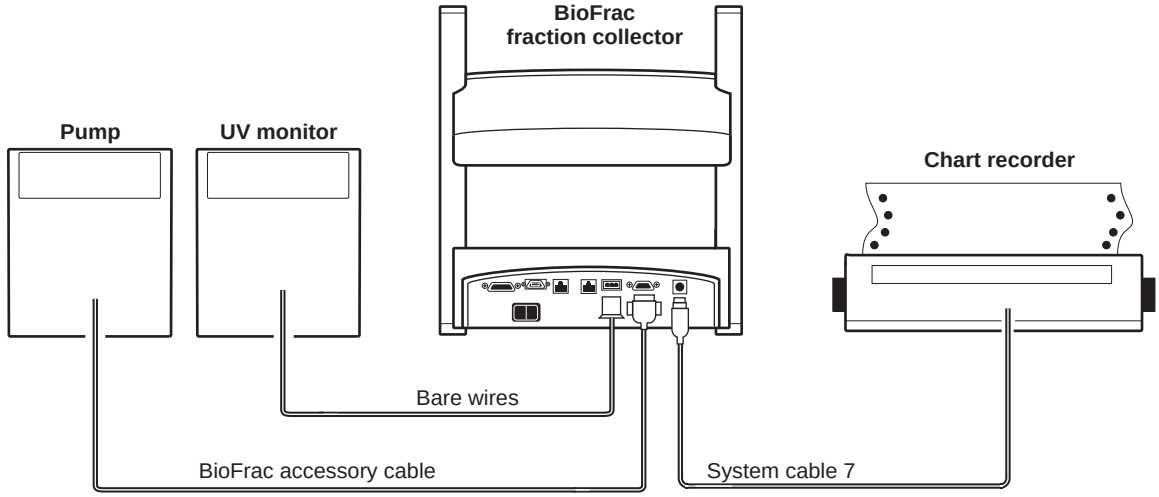



**Fig. 13. Connecting the fraction collector to components other than Bio-Rad's.**

Remote Start/Stop of an External Pump from the BioFrac.

1. As described in Section 5, set the fraction collector to Local mode. In Local mode, all fraction collection parameters are programmed on the BioFrac front panel.

2. Connect the BioFrac Accessory cable (15-pin to bare wires; See appendix B) to the I/O port on the back of the fraction collector.

3. Connect the BioFrac accessory cable green/black wire (pin #9) to the start pin on the remote instrument. Refer to your particular pump's documentation for further information.

4. Connect the orange/black wire (pin #10) to the remote instruments signal ground. When Run is pressed on the BioFrac, the remote start relay pins (pins #9 and #10) are connected and the start signal is relayed to the remote device.

**Remote Start/Stop of the Fraction Collector**

1. Set up the fraction collection parameters as described in Section 5 with the fraction collector in Local mode. In Local mode, the fraction collector controls all aspects of fraction collection.

2. Connect a BioFrac accessory cable (15-pin to bare wires, see Appendix B) to the I/O port on the back of the fraction collector.

3. Connect pin #5 (orange wire) to the signal ground, pin #15 (blue/white wire). Making this connection allows the BioFrac to listen for a start/stop signal.

4. Connect pin #6 (blue wire) to one terminal of the start/stop relay on the controlling device and connect pin #15 (blue/white wire) to the other terminal of the start/stop relay. Refer to the documentation for your particular controller before completing the setup. Pressing start on the controlling device starts the BioFrac by connecting pin #6 to ground. Fraction collection is stopped when the last tube is reached, Stop is pressed on the BioFrac or when the connection between pin #6 and ground is broken by pressing stop on the controlling device. When doing a remote start, we recommend that you test the collection parameters by pressing Run and then Stop on the BioFrac before running an experiment. The BioFrac does a series of error checks at the start of a run and will not start a run if it finds an error in the setup.

**Remote Control of a Chart Recorder.**

1. Set up the fraction collection parameters as described in Section 5 with the fraction collector in Local mode. In Local mode the fraction collector controls all aspects of fraction collection.

2. Connect system cable 7 (mini-DIN to bare wires; see Appendix B) to the REC connector on the back of the fraction collector.

3. Connect pins #3 through #8 to their corresponding counterparts on the chart recorder. See Appendix B for pin assignments and wire color code assignments. Refer to the documentation for your particular chart recorder before completing the setup.

4. If Collecting by threshold, connect the fraction collector to the UV monitor using the green plastic Combicon connector provided with the fraction collector (see Section 1.3) and the 2-wire cable. The UV monitor's (+) output connects to the Combicon's analog IN (+), and the UV monitor's (–) connects to the Combicon's analog IN (-) (See Appendix B). Connect the shield of the 2-wire cable to the analog ground. Ensure that the wires are securely held in the Combicon connector by tightening the screws with a small screwdriver.

5. Refer to the documentation for your particular chart recorder, pump, and UV monitor before completing the setup.

6. If your UV monitor has a selectable range of output settings, use either 100 mV or 1 V setting. From the fraction collector display (see Section 4.6), turn threshold collection on and set the full scale input voltage to either 100 mV or 1 V.

**Remote Control of the Drop Head and Diverter Valve**

1. Set up the fraction collection parameters as described in Section 6 with the fraction collector in LP/Econo mode. In LP/Econo mode, the remote instrument controls all aspects of fraction collection.

2. Connect a BioFrac accessory cable (15-pin to bare wires; see Appendix B) to the I/O port on the back of the fraction collector.

3. Connect the I/O port pin #1 (black wire) to the pin controlling the fraction advances on the remote instrument.

4. Connect the I/O port pin #15 (blue/white wire) to the signal ground of the remote instrument.

5. Connect system cable 7 (mini-DIN to bare wires; see Appendix B), to the REC connector on the back of the fraction collector.

    a. Connect REC pin #2 (orange wire) to the diverter valve control pin on the remote controller. Note, if you cannot control the BioFrac diverter valve remotely, connect REC pin #2 (orange wire) to REC pin #8 (green wire). This causes the BioFrac diverter valve to open when Engage is pressed on the fraction collector (see Section 4.1 LP/Econo mode). If you cannot control the BioFrac diverter valve, you will need to supply a diverter valve with your system. This second diverter valves collect port should be plumbed to the common port of the BioFrac diverter valve.

    b. Connect REC pin #8 (green wire) to the signal ground on the remote controller.

6. On the BioFrac LP/Econo Main screen press Engage (F1). The BioFrac will now wait for the fraction advance and diverter valve signals.

## 3.3 Setting Up the Fraction Collector Racks

The BioFrac fraction collector is designed to accommodate a variety of rack options, including both custom molded racks and off-the-shelf racks. Custom racks include the ice bath/microplate and Prep-20 racks. The top of the ice bath rack holds microplate (12-, 24-, 48-, and 96-well) and Titertube tube racks that adhere to Society of Biomolecular Screening (SBS) standards for microplates. Inexpensive off-the-shelf racks, which accommodate tubes, scintillation vials, and Eppendorf tubes can be purchased from Bio-Rad or most scientific product vendors. The rack options are shown in Figure 14 and listed in Table 4.

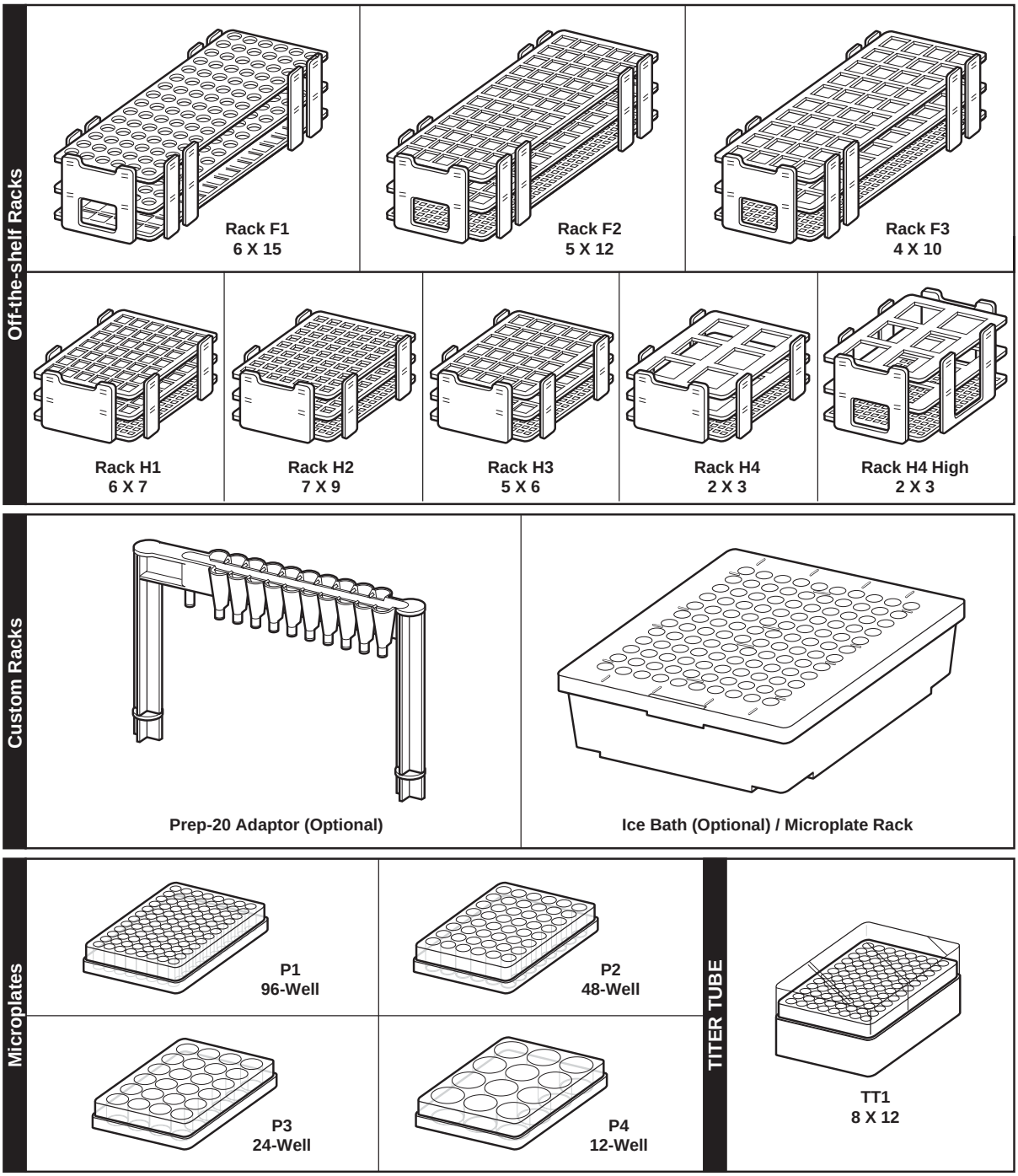



**Fig. 14. BioFrac fraction collector racks.**

## Table 4. BioFrac Fraction Collector Racks

| Rack ID | Bio-Rad Catalog # | Rack Description | Rack Capacity | Format Columns x Rows | Tubes Per Rack (Total) |
|---|---|---|---|---|---|
| F1 | 741-0010 | Holds 12–13 mm diameter tubes up to 150 mm in height | 2 racks | 6 x 15 | 90 (180) |
| F2 | 741-0011 | Grip rack, holds 15–16 mm diameter tubes up to 150 mm in height | 2 racks | 5 x 12 | 60 (120) |
| F3 | 741-0012 | Grip rack, holds 18–20 mm diameter tubes up to 150 mm in height | 2 racks | 4 x 10 | 40 (80) |
| H1 | 741-0013 | For 1.5–2.0 ml capless microtubes | 4 racks | 6 x 7 | 42 (168) |
| H2 | 741-0014 | For 0.5 ml capless microtubes | 4 racks | 7 x 9 | 63 (252) |
| H3 | 741-0015 | For 16 mm scintillation vials | 4 racks | 5 x 6 | 30 (120) |
| H4 | 741-0016 | For 30 mm scintillation vials | 4 racks | 2 x 3 | 6 (24) |
| H4-High | 741-0020 | For 50 mm tubes | 4 racks | 2 x 3 | 6 (24) |
| Ice bath/ microplate rack | 741-0017 | Ice bath for 13 mm diameter tubes. This rack also serves as a holder for 12-, 24-, 48-, and 96-well microplates and microtiter tubes that adhere to the SBS standard format. | 1 rack | 10 x 12 | 120 |
| P1 | 224-0096 | | 4 plates | 96-well | 96 (384) |
| P2 | | | 4 plates | 48-well | 48 (192) |
| P3 | | | 4 plates | 24-well | 24 (96) |
| P4 | | | 4 plates | 12-well | 12 (48) |
| TT1 | | | 4 racks | 96-well | 96 (384) |
| Prep-20 | 741-0018 | Preparative rack | 1 | 2 x 10 | 20 funnels |
| Bottle | | For 250 ml bottles | 1 | | 4 bottles |
| Additional racks, which are compatible with the BioFrac fraction collector, are available from Scienceware (www.belart.com). Compatible racks include the no-wire and no-wire grip full racks (248 x 105 x 64 mm and 246 x 104 x 64 mm) and half racks (128 x 105 x 43 mm and 128 x 105 x 43 mm) that have the format listed for racks F1-F3 and H1-H4. | | | | | |

To prepare the BioFrac fraction collector for fraction collection, set up the unit as follows:

- **Racks F1, F2 and F3**: Place the rack positioning tray into the base of the fraction collector, deep side facing up. Position each rack so that its legs are in the rack guides. Adjust the fraction collector's control module appropriately for the height of the tubes being used. The height adjustment lock pins (see Figure 15) can be used to lock the drop head at the desired height. Tube number 1 is located in the front-left corner of the each rack.
- **Racks H1, H2, H3, H4, and H4-HIgh**: Place the rack positioning tray into the base of the fraction collector, deep side facing down. Position each rack so that its legs are in the rack guides. Adjust the fraction collector control module head appropriately for the height of the tubes being used. The height adjustment lock pins (see Figure 15) can be used to lock the drop head at the desired height. Tube number 1 is located in the front-left corner of the each rack. For racks H1 and H2, use only capless microtubes.
- **Ice Bath**: Remove the lid of the ice bath/microplate rack and fill the tub approximately ½ full with crushed ice, replace the lid and insert 13 x 100 mm culture tubes. Remove the rack positioning tray from the fraction collector and replace it with the ice bath rack. Adjust the fraction collector control module appropriately for the height of the tubes being used. The height adjustment lock pin (see Figure 15) can be used to lock the drop head at the desired height. Tube number 1 is located in the front-left corner of the rack.
- **Microplates and Titertube™ Tubes (Racks P1, P2, P3, P4 and TT1):** Remove the rack positioning tray from the fraction collector and replace it with the ice bath/microplate rack. Position the ice bath/microplate rack such that rack tube number 1 is in the front left corner. Mount the plates on the rack using the plate positioning tabs located on the top of the rack. Adjust the fraction collector control module appropriately for the height of the plates being used. The height adjustment lock pins (see Figure 15) can be used to lock the drop head at the desired height. Position the plates such that tube A1 is in the left-front corner. In order to obtain a uniform fraction size when collecting fractions smaller that 0.5 ml, we strongly suggest that fraction size be specified in drops rather than time or volume. Alternatively, the microplate Drop Head Kit (catalog # 741-0088) can be used to allow finer resolution of fraction sizes since it delivers an approximately 25 µl drop.
- **Preparative (Prep-20) Adaptor**: Attach Tygon tubing to each funnel that is long enough to reach the collection vessels (20 feet of 3/8" OD, 1/4" ID Tygon tubing is provided for this purpose.) Ensure that no kinks constrict flow in the tubing. Gravity flow from the prep adaptor requires that the container used for collection be mounted below the Prep-20 adaptor funnels. A drain trough is provided as added security in the event the tubing becomes plugged or kinked, preventing sample loss. The drain trough tubing should be inserted into a clean empty collection vessel.

  To install the Prep-20 adaptor, remove the two green rubber plugs on the left front and right front of the fraction collector. After attaching the Tygon tubing insert the Prep-20 adaptor into the preparative rack holder slots. Notice that the drain trough slopes slightly towards the drain funnel. The collection port tubing extends down the front of the fraction collector.
  **Note**: The Prep-20 adaptor is rated for use with flow rates up to 100 ml/min. (For discussion of the plumbing for high flow rates, refer to Section 3.1).
- **Bottles**: Place the rack positioning tray, deep side face up, and place the bottles in the circles labeled A, B, C, and D. Adjust the fraction collector head appropriately for the height of the bottles being used.

**Adjusting the Drop Head Height**. The BioFrac accomodates collection vessel heights up to 150 mm. Detents, arranged at predefined heights, are positioned along both columns for added security. The detent positions accomodate all standard tube and microplate heights. Lock pins fit snugly within the detents to secure the control module height and prevent the control module from slipping. To adjust the drop head height:

a. Disengage the lock by pulling both Lock Pins outward from the fraction collector. If desired, the lock pins can be disabled by turning the handles sideways (Figure 15A).

b. Move the control module to the desired height (Figure 15B).

c. Move the control module up or down until the lock pins click into position.

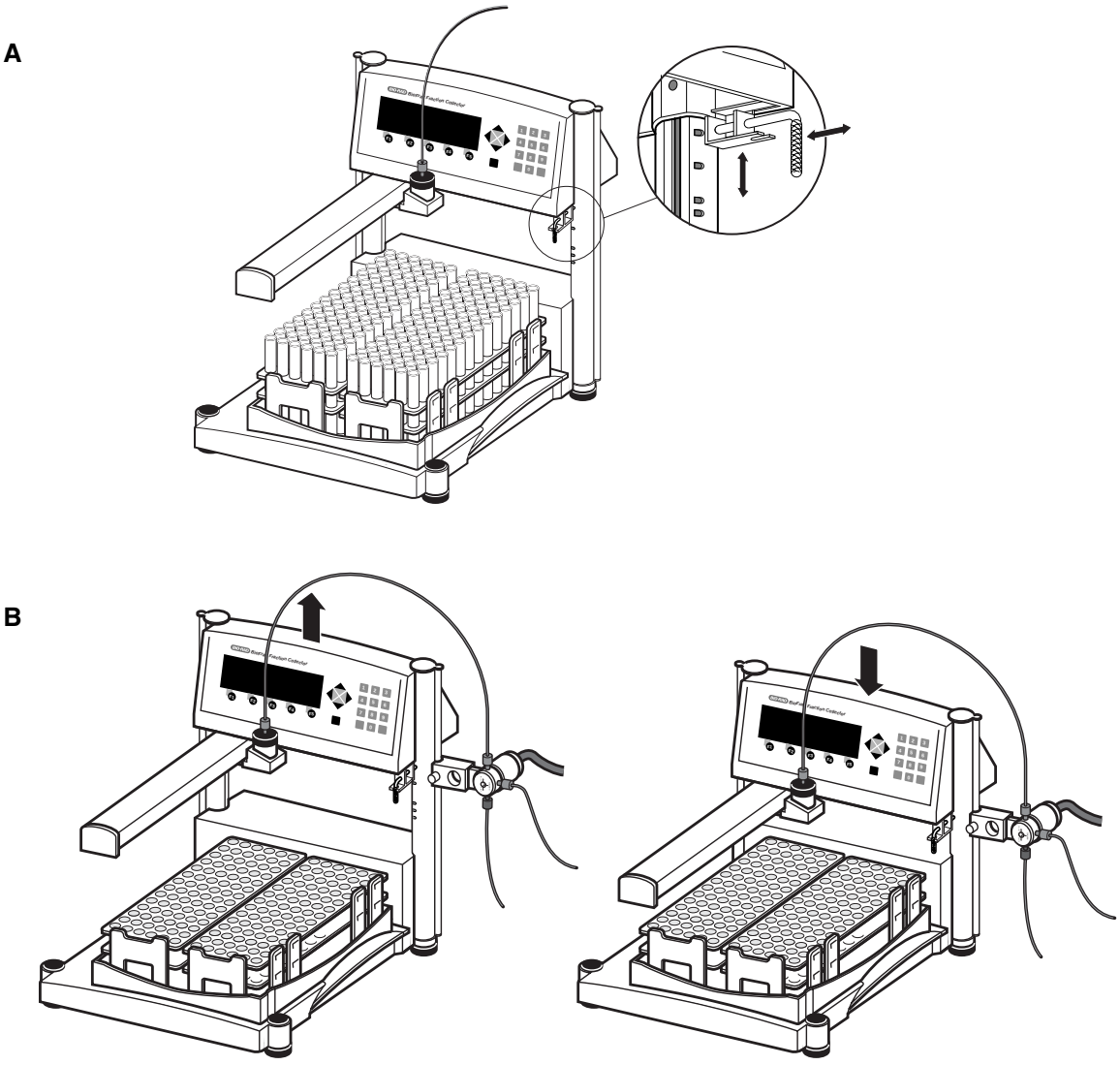



**Fig. 15. BioFrac dispenser arm tube height adjustment. A, lock pin adjustment. B, height adjustment.**

# Section 4
# Introduction to the User Interface

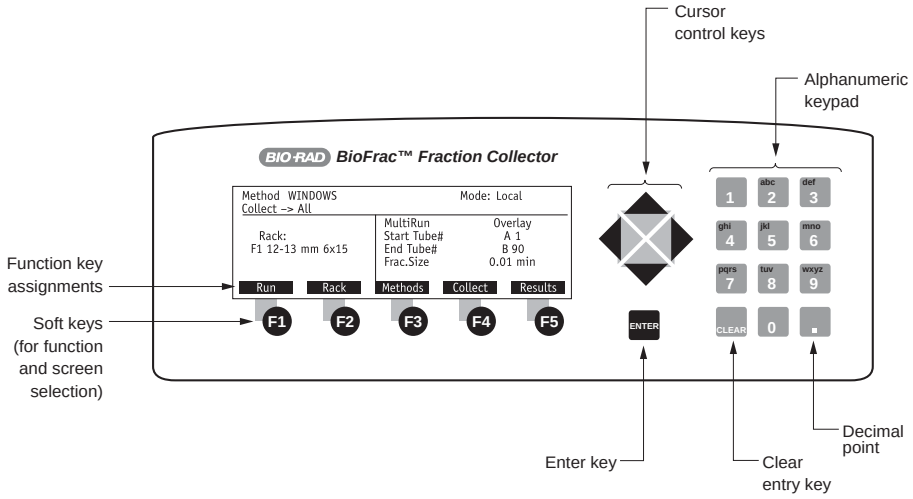



**Fig. 16. BioFrac User Interface.**

Control of the fraction collector interface is through the arrow, ENTER, CLEAR, and alphanumeric keys that are located to the right of the display or through the function keys located below the display. Navigation between the fields is accomplished using the arrow keys. Numbers and/or letters are entered into the alphanumeric fields using the alphanumeric keypad. ENTER is used to accept the changed parameter and CLEAR deletes the entered data. The left arrow key can also be used to backspace and delete one character at a time. Each button on the keypad shows the characters assigned to it. In numeric fields only numbers are accessible. In text fields sequential pressing of a key toggles through the available characters (i.e., 2→ A→ B→ C→ 2, etc.). The five function keys are used to move between different screens or perform predefined functions. When the cursor is in a menu selection field, pressing ENTER causes the menu to be displayed. Once a menu is displayed, the Up/Down arrow keys are used to navigate through the menu items. Pressing ENTER selects the menu item shown at the cursor position.

## 4.1 Main Screen

The Main screen provides information about the operation mode (Local or LP/Econo), method name, rack, and fraction collection parameters. The parameter fields displayed depend on whether the fraction collector is in Local or LP/Econo mode (see Figures 17 and 18 and Tables 5 and 6).

In Local mode, the Main screen is used to set the multirun mode, start and end tube numbers, fraction size, fraction size units and flow rate (see Figure 17 and Table 5). Function keys are used to start a run, (see Section 4.2, Local Mode), or switch to the Rack (see Section 4.3), Method Library (see Section 4.4), Advanced Collection (see Section 4.6), and Results (see Section 4.8) screens.

In LP/Econo mode, the Main screen is used to set the multirun mode and start and end tube numbers (see Figure 18 and Table 6). All other fraction collection parameters are controlled remotely. Function keys are used to engage the fraction collector so that it will accept remote divert and fraction advance signals (see Section 4.2, LP/Econo Mode), or to select a rack from the rack screen (see Section 4.3).

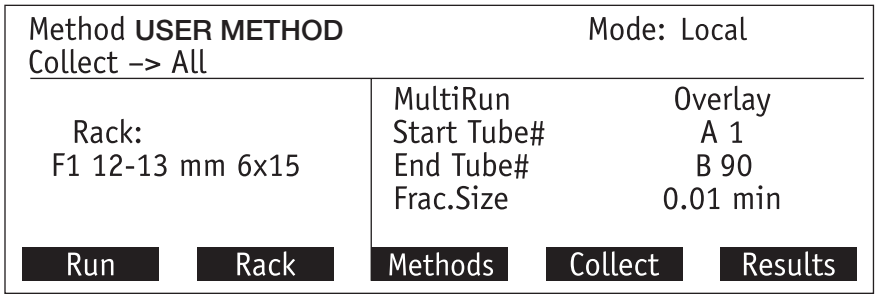



**Fig. 17. Main screen (Local mode).**

## Table 5. Main Screen Parameters and Function Keys (Local Mode)

| Parameter | Function |
|---|---|
| Method | Displays the current method name (text only). |
| Collect | Displays the current collection mode: All, Threshold, Windows, Windows/Threshold (text only). |
| Rack | Displays the currently selected rack (text only). |
| MultiRun | Menu for choosing the a multiple run function:<br>**Overlay***:* Causes collection to occur in the same tubes for each subsequent experiment. In this mode the initial end tube should be equal to or greater than the number of tubes required for each experiment.<br>**Seq. Tube +1***:* Increments the start and end tube #'s at the end of each run so that one tube is skipped between runs. In this mode the initial end tube should be set to reflect the number of tubes required for each experiment rather than the number of tubes in the rack. If the incremented start or end tube # exceeds the rack's tube capacity, a message appears stating that there are not enough tubes for the next run.<br>**Seq. Rack**: Increments the start and end tube #'s at the end of each run so that each run starts at a different rack. In this mode, the initial end tube should be set to reflect the number of tubes required for each experiment rather than the total number of tubes available. If the incremented start or end tube # exceeds the rack's tube capacity, a message appears stating that there are not enough tubes for the next run. |
| Start Tube #<br>End Tube # | Used to set the start tube and end tube. A, B, C, and D correspond to the specific rack position and the associated number corresponds to the tube position within each rack. Start tube and end tube are automatically updated at the end of an experiment if MultiRun is set to "Seq. Tube+1" or "Seq. Rack". |
| Frac. Size | Defines the current fraction size in Time (min), Volume (ml), or Drops. |
| min/ml/drop | Menu for choosing the fraction size units, Time (min), Volume (ml), Drops.<br>Note: if collecting by volume, the flow rate must be entered in the flow rate field. |
| Flow Rate | Used to define the flow rate. Must be set if collecting by Volume. This field is not displayed if collecting in Time or Drop mode. |

| Function Keys | Function |
|---|---|
| Run | Starts an experiment and displays the Run screen (See Section 4.2). |
| Rack | Displays the Rack screen (see Section 4.3). |
| Method | Displays the Method Library screen (see Section 4.4). |
| Collect | Displays the Advanced Collection parameter screen (see Section 4.6). |
| Results | Displays the Results screen. This screen displays the run results of the last completed experiment (see Section 4.8).<br><br>Note: The contents of this screen are lost when Run is pressed or the fraction collector is turned off. |

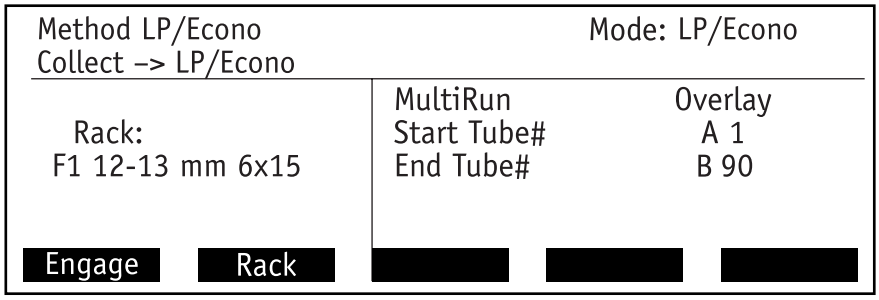



**Fig. 18. Main screen (LP/Econo mode).**

## Table 6. Main Screen Parameters and Function Keys (LP/Econo Mode)

| Parameter | Function |
|---|---|
| Method | LP/Econo (text only). |
| Collect | LP/Econo (text only). |
| Rack | Displays the currently selected rack (text only). |

| | |
|---|---|
| MultiRun | Menu for choosing the multiple run function:<br><br>**Overlay***:* Causes collection to occur in the same tubes for each subsequent experiment. In this mode, the fraction collector moves back to the start tube when the end tube is filled. Stop is used to disengage the experiment.<br><br>**Seq. Tube +1***:* Increments the start and end tube #'s at the end of each run so that one tube is skipped between runs. In this mode, the initial end tube should be set to reflect the number of tubes required for each experiment rather than the number of tubes in the rack. If the incremented start or end tube # exceeds the rack's tube capacity a message appears stating that there are not enough tubes for the next run. The start tube and end tube are incremented each time the fraction collector is disengaged by pressing stop or by filling the end tube.<br><br>**Seq. Rack**: Increments the start and end tube #'s at the end of each run so that each run starts on a different rack. In this mode, the initial end tube should be set to reflect the number of tubes required for each experiment rather than the total number of tubes available. If the incremented start or end tube # exceeds the rack's tube capacity a message appears stating that there are not enough tubes for the next run. The start tube and end tube are incremented each time the fraction collector is disengaged by pressing stop or by filling the end tube. |
| Start Tube #<br>End Tube # | Used to set the start and end tube. A, B, C, and D correspond to the rack position. The associated number corresponds to the tube position within each rack. Start tube and end tube are automatically updated at the end of an experiment if MultiRun is "Seq. Tube+1" or "Seq. Rack". |
| **Function Keys** | **Function** |
| Engage | Moves the drop head to the start tube and causes the fraction collector to listen for fraction advance and diverter valve signals. |
| Rack | Displays the Rack Selection screen (see Section 4.3). |

## 4.2 Run Screen

The Run screen provides information about the progress of the current run. The information displayed on the screen depends on the type of method being used and whether the fraction collector is in Local or LP/Econo mode.

In Local mode, the information displayed includes, run status, run time, fraction filled, fraction size and drop head position. In addition, UV (%AUFS) and threshold values are displayed during threshold collection (see Figure 19 and Table 7). Run volume is displayed when collecting by volume. Four function keys are available during a run to stop or pause an experiment, do tube advances and control the divert valve.

In LP/Econo mode the Run screen shows the current divert valve status, and drop head position (see Figure 20 and Table 8). This screen also contains a function key that is used to stop fraction collection.

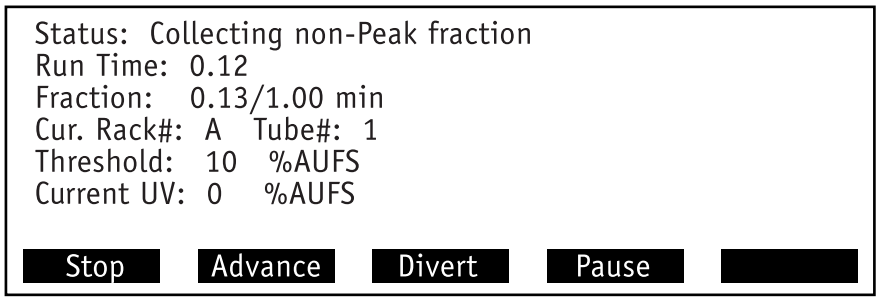



**Fig. 19. Run screen (Local mode).**

## Table 7. Run Screen Parameters and Function Keys (Local Mode)

| Parameter | Function |
|---|---|
| **Status** | Displays the current fraction collection status:<br><br>Collecting: The diverter valve is in the collect position and fractions of a user-specified size are being collected.<br><br>Diverting: The diverter valve is in the Waste position.<br><br>Run Paused!: The run is currently paused and the diverter valve is at Waste. A warning is displayed that the diverter valve is Diverting to Waste.<br><br>Collecting Non-Peak: The divert valve is in the collect position but is collecting fractions of non-peak size.<br><br>Will Collect at End of Delay: The fraction collector is waiting for the delay time before it switches the diverter valve to Collect.<br><br>Will Divert at End of Delay: The fraction collector is waiting for the delay time before it switches the diverter valve to Waste. |
| **Run Time** | Displays the current run time (in hours and minutes). Stops incrementing during a pause. If collecting by volume, the run volume is also displayed. |
| **Cur. Rack # Tube #** | Displays the current rack position and tube number. |
| **Threshold** | Displays the current threshold setting as a percentage of full scale (%AUFS). (Only displayed if collecting by threshold.) |
| **Current UV** | Displays the current UV signal value as a percentage of full scale (%AUFS). (Only displayed if collecting by threshold.) |

| Function Key | Function |
|---|---|
| Stop | Stops the current experiment and displays the Results screen. |
| Advance | Advances the drop head by one tube. |
| Divert/Collect | Toggles the diverter valve between Collect and Waste. |
| Pause | Pauses fraction collection and switches the diverter valve to Waste during the pause. |
| Resume | Resumes fraction collection after a pause. |

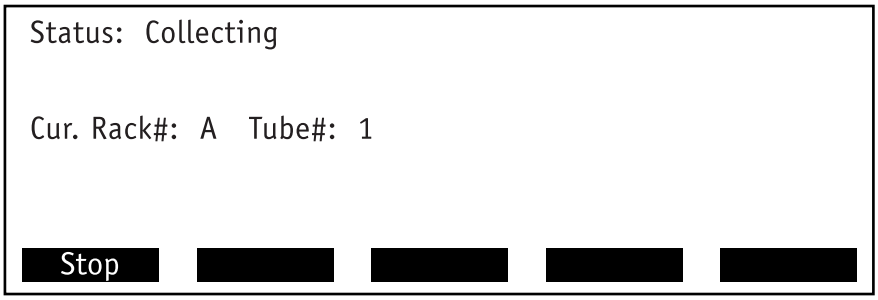



**Fig. 20. Run screen (LP/Econo mode).**

## Table 8. Run Screen Parameters and Function Keys (LP/Econo Mode)

| Parameter | Function |
|---|---|
| Status | Displays the current fraction collection status:<br><br>Collecting: The diverter valve is in the Collect position.<br><br>Diverting : The diverter valve is in the Divert to Waste position. |
| Cur. Rack # Tube # | Displays the current rack position and tube number. |
| Overlay Cycle | The number of times that the fraction collector has overlaid fractions in the current set of runs. (Only displayed in Overlay mode.) |
| **Function Key** | **Function** |
| Stop | Causes the fraction collector to stop listening for fraction advance and diverter valve signals and returns the drop head to the home position. |

## 4.3 Rack Screen

The Rack screen is used to select the rack type and collection pattern used in a method (see Figure 21 and Table 9). The default collection pattern is serpentine, however, it may be changed to collection by row or column when collecting in microplates or Titertube tubes. The Divert Between Tubes feature can be used to reduce the amount of liquid spilled during tube advances. When this option is turned on (default), the diverter valve switches to waste during tube advances. However, if this option is turned off (recommended when collecting in drop mode at low flow rates), the diverter valve switches to Waste only when the drop head is moving between non-adjacent tubes.

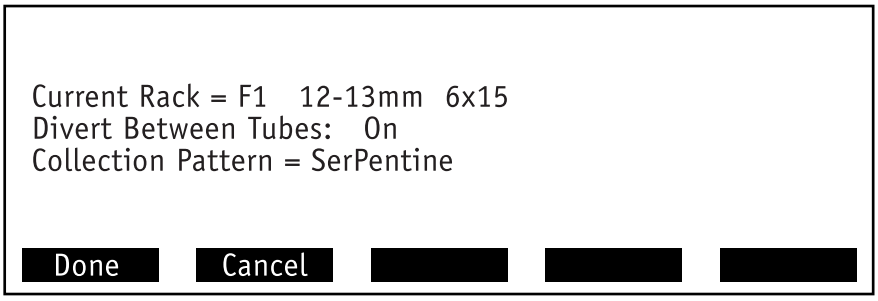



**Fig. 21. Rack screen.**

### Table 9. Rack Screen Parameters and Function Keys

| **Parameter** | **Function** |
|---|---|
| **Current Rack** | Menu used for rack selection. |
| **Divert Between Tubes** | Allows the user to turn off the Divert Between Tubes function. The fraction collector still diverts when the drop head is moving between non-adjacent tubes. |
| **Collection Pattern** | Displays the current fraction collection pattern. This pattern can be changed to row or column for microplates and Titertube tubes. |
| **Function Key** | **Function** |
| **Done** | Accepts all changes and returns to the Main screen. |
| **Cancel** | Aborts all changes and returns to the Main screen. |

## 4.4 Method Library Screen

The Method Library screen is used to retrieve, save, or delete user defined collection methods (see Figure 22 and Table 10). Up to 20 methods may be stored. Function keys are used to load, save, and delete methods or to enter the Calibration screen (see Section 4.5).

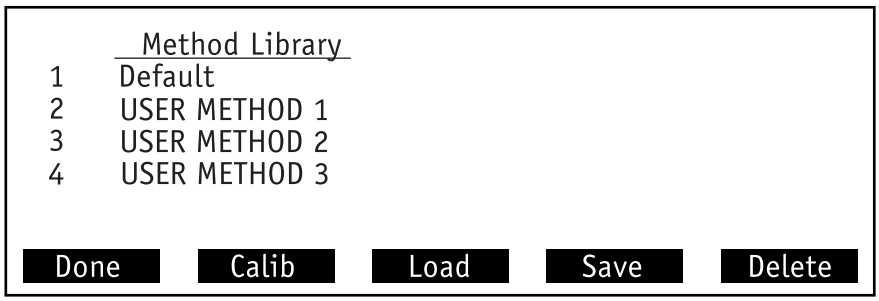



**Fig. 22. Method Library Screen.**

### Table 10. Method Library Screen Parameters and Function Keys

| **Parameter** | **Function** |
|---|---|
| **Method Library** | Displays the Default method and user-defined methods. |
| **Function Key** | **Function** |
| **Done** | Returns the display to the Main screen. |
| **Calib** | Displays the Calibration screen (see Section 4.5). |
| **Load** | Loads the method pointed to by the cursor. |
| **Save** | Allows the current fraction collection parameters to be saved as a method. A method name can be up to 16 characters long. |
| **Delete** | Deletes the method pointed to by the cursor. |

## 4.5 Calibration Screen

The Calibration screen is used to adjust the drop head calibration, screen contrast, sleep mode, and to zero the analog-to-digital converter (AtoD). Refer to Table 11 for more information about the use of each function key.

## Table 11. Calibration Screen Function Keys

| Function Keys | Function |
|---|---|
| **Done** | Returns the display to the Method Library screen. |
| **Contrst** | Allows the user to set the display contrast. The up and down arrow keys are used to increase and decrease the screen brightness, respectively. |
| **X-Y axis** | Places the fraction collector in rack calibration mode. Allows the user to calibrate the fraction collector X-Y arm. Calibration should rarely be required. Calibration is done on an F1 rack that has 13 x 100 mm glass tubes in positions A1, A15, and B90 (plastic tubes should not be used). To do the calibration:<br>a. Remove the drop former top by twisting it counter counterclockwise and gently lifting it out.<br>b. Press X-Y Axis and then Next.<br>c. While looking through the drop head, center it over tube A15 using the arrow keys, press Next.<br>d. Center the drop head over tube A1 using the arrow keys, press Next.<br>e. Center the drop head over tube B90 using the arrow keys, press Save.<br>f. Replace the top of the drop former by inserting it into the drop head and twisting it clockwise<br><br>Caution: Using this function overwrites the previous calibration. |
| **Zero AD** | Allows the user to zero any voltage offset in the analog-to-digital (AtoD) converter. The AtoD is used to convert the analog UV signal used for threshold collection to a digital signal. In the event of an offset in the AtoD voltage, the Current UV signal (see Figure 19) displayed on the run screen may be in error. This calibration should rarely be required. To zero the AtoD - press Zero AD, connect a shorting plug (Combicon connector with the analog IN(+) and IN(-) pins connected to the analog IN (ground) and then press Next. |
| **Sleep** | Allows the user to select how many minutes the display should wait before going into sleep mode. Pressing any button will wake up the display. |

## 4.6 Advanced Collection Screen

The Advanced Collection screen is used to turn threshold and windows collection on or off and to set threshold and delay parameters (see Figure 23 and Table 12). Threshold parameters are displayed only when threshold is turned on. From this screen, the threshold level and UV detector input voltage (100 mV or 1 V) can be entered and the user can specify whether non-peak fractions are to be collected or not. A bubble filter time constant can also be entered. During threshold collection the bubble filter function suppresses unwanted fraction advances due to air bubbles passing through the UV detector. See Appendix A for more information about the bubble filter function.

When windows collection is turned on, the Table function key, F3, is used to display the Collection Windows Table screen (see Section 4.7). The Collection Windows Table screen is used to enter all collection windows parameters. If you are collecting by threshold and windows, you should turn on threshold prior to entering the Collection Windows Table screen.

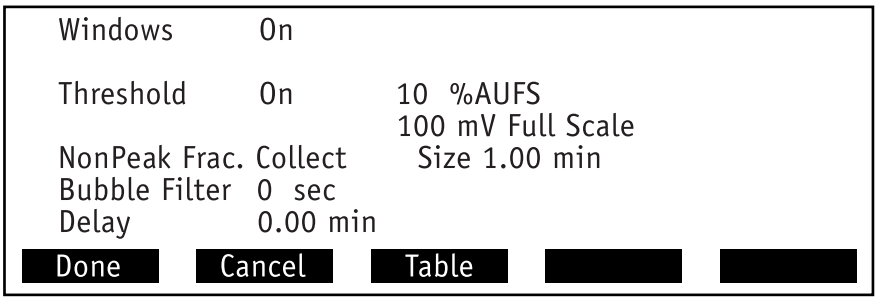



**Fig. 23. Advanced collection screen.**

## Table 12. Advanced Collection Screen Parameters and Function Keys

| Parameter | Function |
|---|---|
| **Windows** | Menu for turning Collection by Windows on or off. |
| **Threshold** | Menu for turning Collection by Threshold on or off. |
| **%AUFS** | Current threshold setting. (Only displayed if Threshold is on.) |
| **Full Scale** | Menu for defining the UV detectors output voltage. (Only displayed if Threshold is on.) The detector input voltage can be set to either 100 mV or 1 V. |
| **Non-Peak Frac.** | Turns collection of non-peak fractions on or off. (Only displayed if Threshold is on.) |
| **Size** | The non-peak fraction size. The non-peak fraction size has the same units as the fraction size and delay parameters. (Only displayed if Threshold is on.) |
| **Bubble Filter** | Time constant entered in seconds, used to filter false peaks above a threshold due to electrical noise or air bubbles. Typically a bubble filter time of 0 or 1 second will suffice. For a discussion of bubble filter time, refer to Appendix A. (Only displayed if Threshold is on.) |
| **Delay** | A delay time used to precisely synchronize the UV signal with event marks on the chart recorder and tube advances. Delay must be less than or equal to the fraction size. Note: using delay will result in a timing offset between tube advances and the displayed fraction fill volume shown on the run screen. Delay has the same units as the fraction size parameter. For a discussion of the Delay function, refer to Section 5.5. |
| **Function Keys** | **Function** |
| **Done** | Accepts all changes and returns to the Main screen. |
| **Cancel** | Aborts all changes, including changes made in the Collection Windows Table screen (See the Table function key below). |
| **Table** | Changes the screen to the Collection Windows Table screen (see Section 4.7). (Only visible if Windows is on). |

## 4.7 Collection Windows Table Screen

The Collection Windows Table screen is used to enter the collection windows parameters (see Figure 24 and Table 13). Up to 20 different collection windows can be defined, each with a different fraction size and threshold if desired. The threshold parameter column (Thold) shown in Figure 24, is displayed only if threshold collection has been turned on in the Advanced Collection screen (see Section 4.6). Windows start and end parameters are entered in minutes (when collecting by time or drops) or in milliliters (when collecting by volume). The menu at the top of the fraction size column can be used to change the fraction size units. The global fraction size and threshold entered on the Main screen (see Section 4.1) and Advanced Collection screen (see Section 4.6), respectively, are the default fraction size and threshold for the first window.

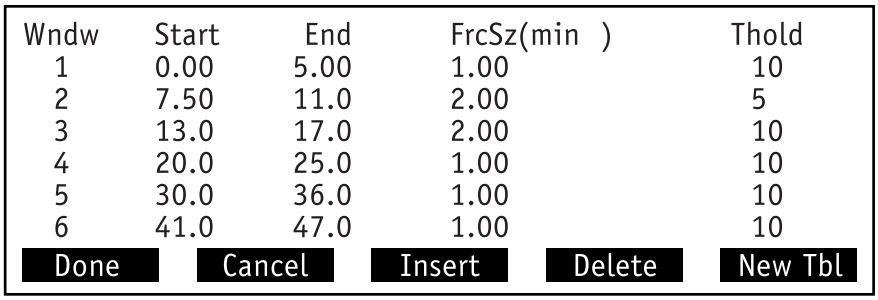



**Fig. 24. Collection Windows Table screen.**

## Table 13. Collection Windows Table Screen Parameters and Function Keys

| Parameter | Function |
|---|---|
| Start<br><br>End | The Collection Window start and end times are in units of time (min) or volume (ml).<br>The Collection Windows start and end times are in units of time if collecting fractions by drop. |
| FrcSz | The fraction size for the current Window (min, ml, drops). The header for this column is also a menu that can be used to change the fraction size units to min, ml, or drops. |
| Thold | The threshold value for each Collection Window. |
| **Function Key** | **Function** |
| Done | Saves any changes, contingent upon the pressing Done on the Advanced Collection screen and exits the Collection Windows Table screen. |
| Cancel | Aborts all changes and returns to the Advanced Collection screen. |
| Insert | Inserts a new window at the position pointed to by the cursor.<br>Caution: if 20 windows are defined, pressing insert causes window #20 to be deleted. |
| Delete | Deletes a window at the position pointed to by the cursor. |
| New | Deletes all windows. |

## 4.8 Results Screen

The Results screen provides a list of the tubes associated with each peak or window. When collecting by threshold, a peak is defined as the tubes collected while the UV signal was above threshold. The Results screen is displayed at the end of each run, if the run was not started remotely, or can be viewed by pressing F5 on the Main screen (see Section 4.1). The information on this screen is lost when a new run is started or the fraction collector is turned off.

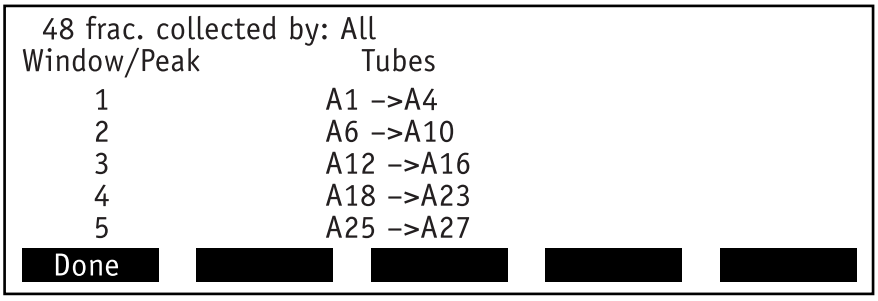



**Fig. 25. Results screen.**

### Table 14. Results Screen Parameters and Function Keys

| **Parameter** | **Function** |
|---|---|
| **Window/Peak** | If collecting by windows, the window number is displayed. If collecting by Threshold or by Windows and Threshold the peak number is displayed. |
| **Tubes** | List of the tubes associated with each window or peak. |
| **Function Key** | **Function** |
| **Done** | Returns to the Main screen. |

# Section 5
# Stand-Alone Operation

Normal operation of the BioFrac fraction collector is in Local mode (Stand-alone mode). As a stand-alone fraction collector, the BioFrac may be used to collect fractions based on time (minutes), volume (milliliters), or drops (up to a flow rate of 5.0 ml/min). In Local mode, the fraction collector controls all aspects of fraction collection and is not in communication with Bio-Rad's BioLogic DuoFlow system, BioLogic LP system, or Model EP-1 Econo pump. However, it may be connected to separate components such as a UV monitor, a chart recorder, and if desired, a pump as described in Section 3.2.

There are four modes of collection, each of which can include a Delay function:

- Collect All: Allows you to collect an entire run without diverting any fluid to waste. This is discussed in Section 5.1.
- Peak Detection by Threshold: Allows you to collect peaks by defining a threshold value (percent of full scale) above which fractions will be collected. This method can be used only when the fraction collector is connected to a UV monitor. When the UV signal is less than the threshold, fluid is diverted to waste. Alternatively, it may be collected in tubes with a non-peak fraction size. A slope function is built into the Threshold function so that double peaks above the set threshold level are detected and collected separately. This collection method is discussed in Section 5.2.

  Note: False peaks above a threshold (such as electrical noise or air bubbles) may be filtered using the Bubble Filter Time function. See Appendix A for further discussion.
- Windows Collection: Allows you to specify periods of time or volume (windows) during which fractions are to be collected. For example, a window can be defined to start after an initial void volume. The BioFrac fraction collector lets you define up to 20 different time or volume windows. The liquid delivered during a collection window is collected into tubes, whereas the liquid delivered outside of a collection window is diverted to waste. This collection method is discussed in Section 5.3.

- Peak Detection by Threshold with Time or Volume Windows: Allows you to combine the Peak Detection by Threshold and the Time or Volume Windows methods discussed above. This method can be used only when the fraction collector is connected to a UV monitor. You can program up to 20 different time or volume windows, each with its own threshold level. This is a useful feature to compensate for baseline drift. This collection method is discussed in Section 5.4.

  Note: False peaks above a threshold (such as electrical noise or air bubbles) may be filtered using the Bubble Filter Time function. See Appendix A for further discussion.

- Delay Function: The purpose of the Delay function is to synchronize the fraction collection event marks and the signal from the UV monitor (output on a chart recorder) with the actual delivery of liquid into collection tubes. This feature allows easy post-run analysis of a chromatogram. The Delay function is slightly different for each of the collection modes: All, Threshold, Windows, and Threshold with Windows. As a consequence, the event marks on the chart recorder will vary depending on the individual application. This collection method is discussed in Section 5.5.

Regardless of the type of collection method programmed, the method will end once the end tube number is reached. Always ensure that the start and end tube numbers are set to allow completion of your collection method.

## 5.1 Collect All

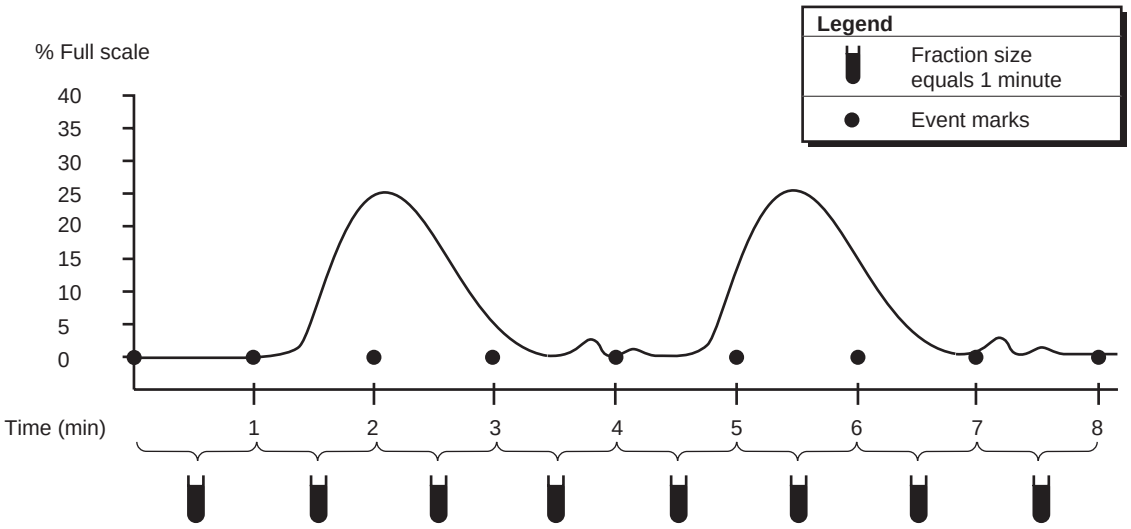



**Fig. 26. Running a Collect All method.**

1. In the Main screen, ensure that Collect is set to Collect->All. If the screen is not set to Collect->All press the Collect button (F4) and turn Windows and Threshold off. Alternatively, press Method (F3) and load the method Default.
2. To select a rack, press the Rack function key (F2) and choose a rack from the rack menu. Choose whether or not you want the diverter valve to divert to waste during fraction advances. If you are collecting in microplates or Titertube tubes, choose the desired collection pattern: Serpentine, Row, or Column. Note, that whenever a new rack is selected the start tube and end tube values are updated to reflect the maximum number of tubes available for the selected rack.

3. Choose the appropriate MultiRun mode for your experiment (see Table 5 for a description of the MultiRun function).

4. Set the start tube and end tube rack position and tube number. The fraction collector will stop when the last tube is reached unless a stop command is received prior to reaching the last tube. At the end of a run, the start tube # and end tube # will be automatically updated according to the MultiRun mode selected.

5. Set the fraction size units to the desired units (min, ml, or drops) and enter the fraction size. If collecting by volume, you must enter a flow rate.

6. (Optional) Press the Collect function key (F4) and set the delay time (see Section 5.5).

7. (Optional) Press the Methods function key (F3) and save your method.

8. Press the Run function key (F1) to start the experiment.

9. At any point during the run you may use the Run screen function keys (see Table 7).

## 5.2 Peak Detection by Threshold

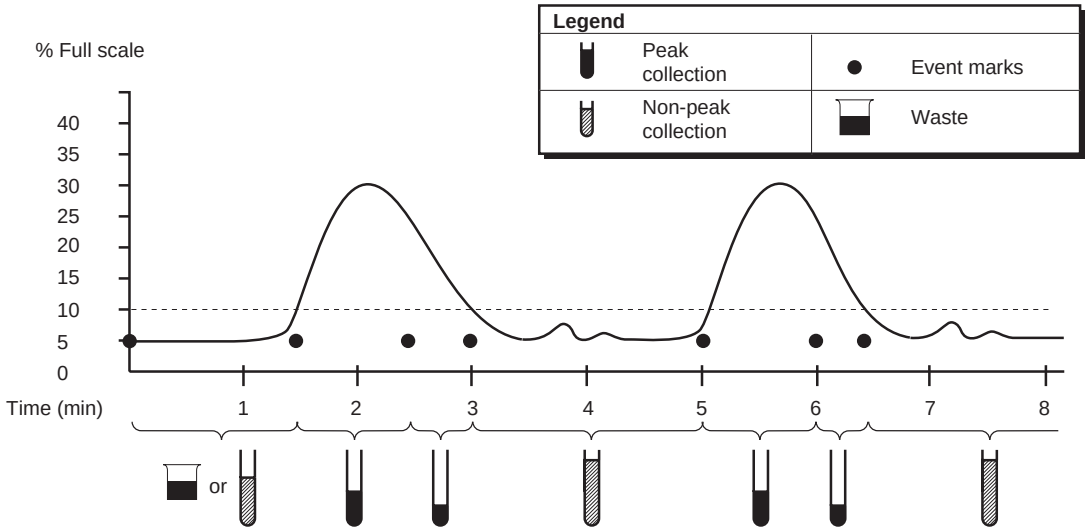



**Fig. 27. Running a Collect by Threshold method.**

1. To select a rack, press the Rack function key (F2) and then choose a rack from the Rack menu. Choose whether or not you want the diverter valve to divert to waste during fraction advances. If you are collecting in microplates or Titertube tubes, choose the desired collection pattern: Serpentine, Row, or Column. Note, that whenever a new rack is selected the start tube and end tube values are updated to reflect the maximum number of tubes available for the selected rack.

2. Choose the appropriate MultiRun mode for your experiment (see Table 5 for a description of the MultiRun function).

3. Set the start tube and end tube rack position and tube number. The fraction collector will stop when the last tube is reached unless a Stop command is received prior to reaching the last tube. At the end of a run, the start tube # and end tube # will be automatically updated according to the MultiRun mode selected.

4. Set the fraction size units to the desired units (min, ml, drops) and set the fraction size. If collecting by volume, you must enter a flow rate.

5. Press the Collect function key (F4) and turn Threshold on. Set the global %AUFS (absorbance units full scale) as desired and set the detector input voltage to either 100 mV or 1 V full scale depending on your detector's output voltage. Press Done. On the Main screen, Collect should now read Collect->Threshold.

6. If Collection of non-peak fractions is desired, change NonPeak Frac. from Divert to Collect and enter a size for the non-peak fractions.

7. (Optional) set the bubble filter time. This function detects and filters false peaks (such as electrical noise or air bubbles). Typically, a bubble filter Time of 0 or 1 second will suffice. (For discussion of the Bubble Filter Time Function, refer to Appendix A.)

8. (Optional) press the Collect function key (F4) and set the Delay time (see Section 5.5).

9. (Optional) press the Method function key (F3) and save your method.

10. Press the Run function key (F1) to start the experiment.

11. At any point during the run you may use the Run screen function keys (see Table 7).

## 5.3 Collect by Windows

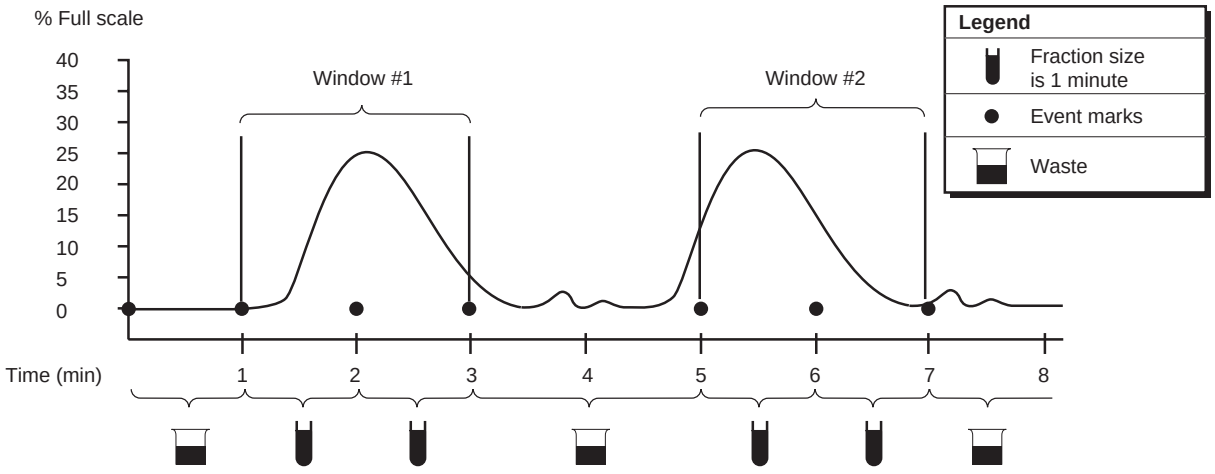



**Fig. 28. Running a Collect by Windows method.**

1. To select a rack, press the Rack function key (F2) and then choose a rack from the rack menu. Choose whether or not you want the diverter valve to divert to waste during fraction advances. If you are collecting in microplates or Titertube tubes, choose the desired collection pattern: Serpentine, Row, or Column. Note, that whenever a new rack is selected, the start tube and end tube values are updated to reflect the maximum number of tubes available for the selected rack.
2. Choose the appropriate MultiRun mode for your experiment (see Table 5 for a description of the MultiRun function).
3. Set the start tube and end tube rack position and tube number. The fraction collector will stop when the last tube is reached unless a Stop command is received prior to reaching the last tube. At the end of a run, the start tube # and end tube # will be automatically updated according to the MultiRun mode selected.
4. Set the fraction size units to the desired units (min, ml, or drops) and set the fraction size. If collecting by volume, you must enter a flow rate.
5. Press the Collect function key (F4) and turn Windows on.
6. Press the Table function key (F3) and enter the desired windows and fraction size in the Collection Windows Table screen. Note that the fraction size entered in the Main screen is the default size. The fraction size can be changed for each window, if desired. When finished, press Done. On the Main screen, Collect should now read Collect->Windows.
7. (Optional) press the Collect function key (F4) and set the delay time (see Section 5.5).
8. (Optional) press the Method function key (F3) and save your method.
9. Press the Run function key (F1) to start the experiment.
10. At any point during the run you may use the Run screen function keys (see Table 7).

## 5.4 Collect by Windows and Threshold

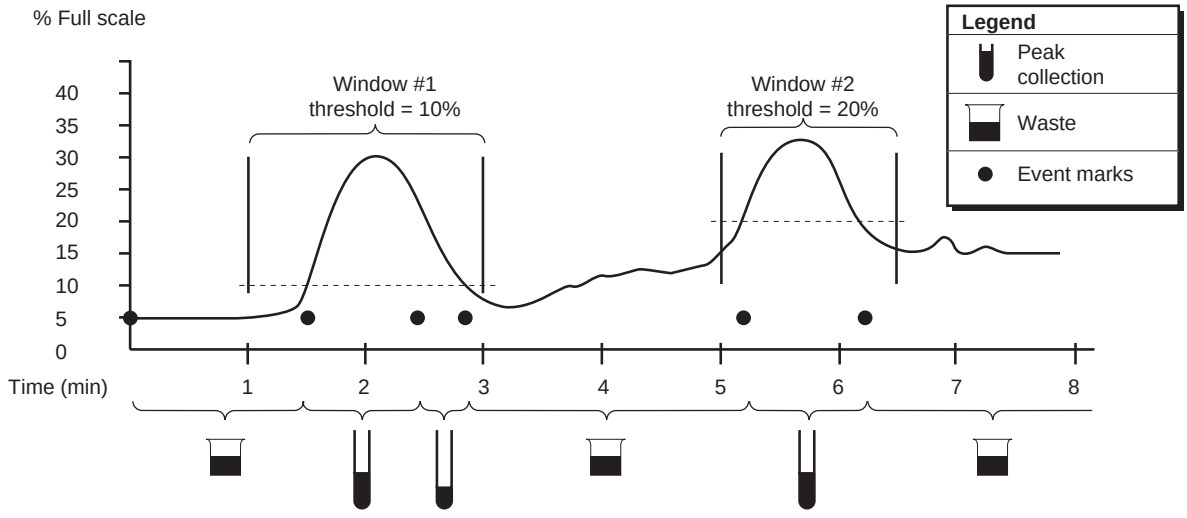



**Fig. 29. Running a Collect by Windows and Threshold method.**

1. To select a rack, press the Rack function key (F2) and then choose a rack from the rack menu. Choose whether or not you want the diverter valve to divert to waste during fraction advances. If you are collecting in microplates or Titertube tubes choose the desired collection pattern: Serpentine, Row, or Column. Note that whenever a new rack is selected, the start tube and end tube values are updated to reflect the maximum number of tubes available for the selected rack.

2. Choose the appropriate MultiRun mode for your experiment (see Table 5 for a description of the MultiRun function).

3. Set the start tube and end tube rack position and tube number. The fraction collector will stop when the last tube is reached unless a Stop command is received prior to reaching the last tube. At the end of a run, the start tube # and end tube # will be automatically updated according to the MultiRun mode selected.

4. Set the fraction size units to the desired units (min, ml, or drops) and set the fraction size to the desired size. If collecting by volume you must enter a flow rate.

5. Press the Collect function key (F4) and turn Windows and Threshold on.

6. Set the detector input voltage to either 100 mV or 1 V full scale depending on your detector's output voltage.

7. Press the Table function key (F3) and enter the desired windows, fraction size, and threshold in the Collection Windows Table screen. Note that the global fraction size entered in the Main screen and the global threshold entered on the Advanced Collection screen are automatically entered into the first window. The fraction size and threshold can be set for each window, if desired. When finished, press Done.

8. If Collection of non-peak fractions is desired, change Non-Peak Frac. from Divert to Collect and enter a size for the non-peak fractions.

9. (Optional) set the delay time (see Section 5.5).

10. (Optional) set the bubble filter time. This function detects and filters false peaks (such as electrical noise or air bubbles). Typically, a bubble filter time of 0 or 1 second will suffice. (For discussion of the bubble filter time, refer to Appendix A.)

13. Press Done (F1). On the Main screen, Collect should now read Collect->Windows/Threshold.

14. (Optional) press the Method function key (F3) and save your method.

15. Press the Run function key (F1) to start the experiment.

16. At any point during the run you may use the Run screen function keys (see Table 7).

## 5.5 Collection Using a Delay Function

The delay volume is the volume of fluid contained in the path between the significant detector, typically the UV monitor, and the fraction collector drop head. When plumbing the system there is typically a length of tubing between the detector(s) and the fraction collector. The length of tubing defines the delay volume. Thus the fluid in the path of the detector will not actually arrive at the fraction collector until it passes through this tubing. Although the volume in the tubing is generally small, it may be significant when collecting a small fraction size. To synchronize the detector signal with the fraction collector, the fraction collector advance can be delayed while the fluid passes through the tubing. The fraction collector advance can occur when the fluid reaches the drop head of the fraction collector. Although this is most important when using Threshold fraction collection, it is advantageous to have this synchronization at all times. Delay can be entered in units of time, volume, or drops and has the same units as the fraction size. The delay volume can be determined by either of the following methods:

a. Using a syringe, fill the tubing (from the UV monitor outlet to the fraction collector drop head) with water. Then expel the water into a separate container and weigh it.

b. Using the known volume of any inline devices and measuring the length of the tubing between the UV detector and the drop head. Table 15 lists the volume of commonly used tubing and devices.

### Table 15. Volume of Commonly Used Tubing and Inline Devices

| Tubing Dimensions | PEEK™ Tubing Color | Volume |
|---|---|---|
| 0.005 inch (0.127mm) ID | Red | 0.322 µl/inch |
| 0.010 inch (0.254mm) ID | Blue | 1.288 µl/inch |
| 0.020 inch (0.508 mm) ID | Orange | 5.145 µl/inch |
| 0.030 inch (0.762 mm) ID | Green | 11.577 µl/inch |
| 0.040 inch (1.016 mm) ID | - | 20.581 µl/inch |
| 0.050 inch (1.270 mm) ID | - | 32.160 µl/inch |
| 0.062 inch (1.575 mm) ID | - | 49.474 µl/inch |
| **Devices** | | |
| Backpressure regulator (40 psi) | | 80 µl |
| Bio-Rad pH probe/flow cell | | 80 µl |
| Diverter Valve | | 12 µl |
| Conductivity cell | | 6 µl |

On the Main screen, set the fraction size units as desired (min, mls, or drops). From the Advanced Collection screen, press the Collect function key (F4). If collecting by volume the delay volume can be entered directly. If collecting by time the delay time is calculated from the delay volume divided by the flow rate. To calculate the delay in drops, divide the delay volume by the drop volume (1 drop ≈ 50 µl for the standard drop head or ≈ 25 µl for the microplate drop head).

Below are some examples that demonstrate the use of the Delay function:

### 5.5.1 Collect All with Delay

Figure 30 shows an example in which the delay is 0.2 minutes and the fraction size is 1 minute. The diverter valve remains at divert during the delay time. The chart recorder makes the first tick mark (following the start/run mark) at 1.0 minute, and each subsequent tick mark at 1-minute intervals. The drop head advances at 1.2 minutes and continues advancing at 1-minute intervals. Each fraction size is still 1 minute.

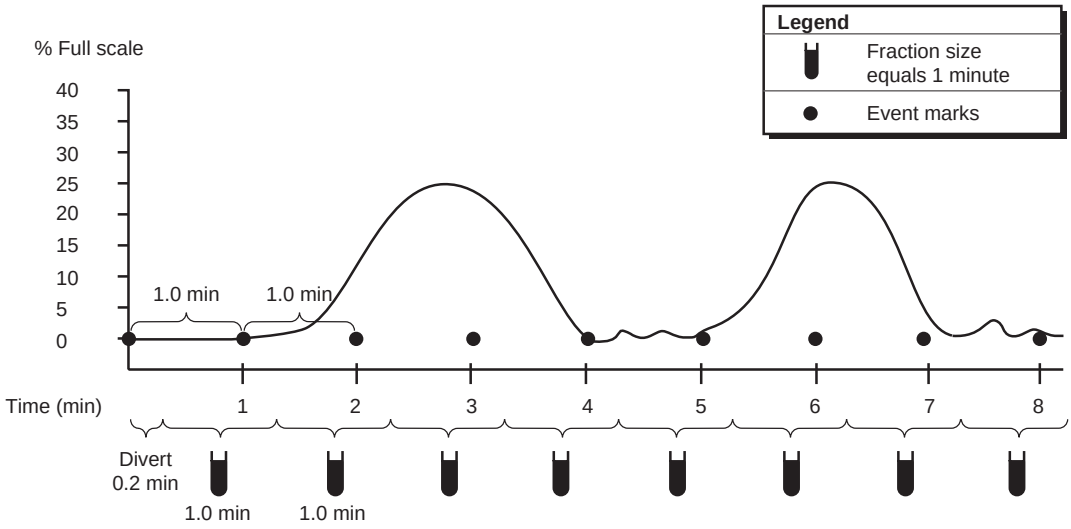



**Fig. 30. Example showing Collect All with a delay of 0.2 minutes.**

Figure 31 shows an example of a Collect All experiment in Drop mode with a 2 drop delay and a 10 drop fraction size. In this case the chart recorder makes its first tick mark (following the start/run mark) at 8 drops (fraction size – delay = 10 drops – 2 drops = 8 drops). Each subsequent tick mark occurs at 10 drop intervals. The drop head advances at 10 drop intervals following the start of the run. Note that if the delay size equals the fraction size, the start/run mark and the first event mark are superimposed. This means that the first observed tick mark (following the start/run mark) corresponds to the second fraction advance.

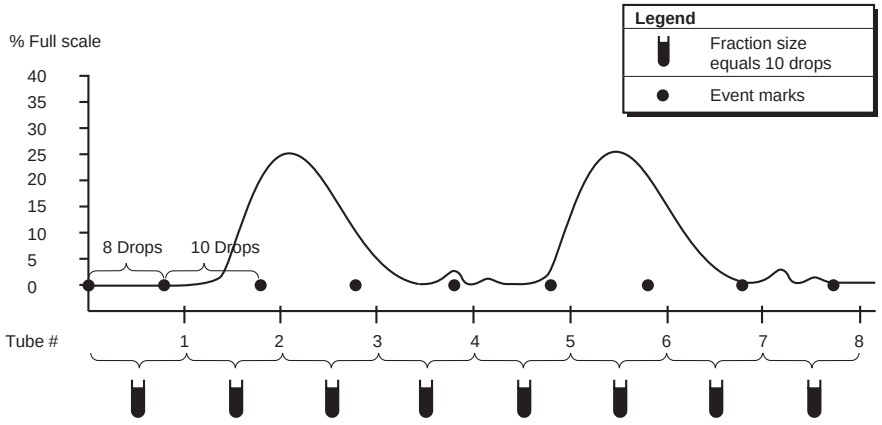



**Fig. 31. Example showing Collect All with a delay of 2 drops.**

### 5.5.2 Peak Collection by Threshold and Delay

Figure 32 shows an example in which Delay is used during the collection of peaks by threshold. Assume the following for the run:

| | |
|---|---|
| Flow rate | 1 ml/minute |
| Peak fraction size | 1 minute |
| Delay time | 0.25 minutes |
| Threshold value | 10% AUFS |
| Non-peak destination | Waste |

At the start of the run an event mark is recorded. Initially the liquid is diverted to waste because the monitor signal is below threshold. When the UV signal rises above the specified threshold of 10%, an event mark is recorded and the volume corresponding to the delay time (or the delay volume) is collected in tube 1. At the end of this time, there is a tube advance and the peak is collected in 1-minute fractions. As the falling edge of the peak passes through the Threshold, an event mark is recorded, but there is no tube advance until the volume corresponding to the delay time is collected. All subsequent peaks follow this same pattern, with a tube containing the Delay Volume separating each peak.
Note: If the delay time is set to zero, an empty tube will separate the peaks.

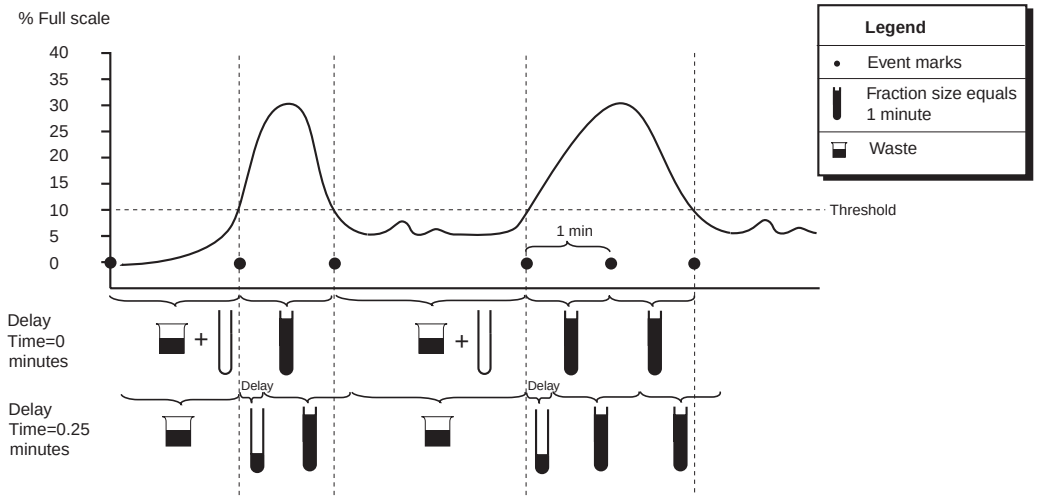



**Fig. 32. Example showing Peak Collection by Threshold using a delay of 0.25 minutes.**

When the non-peak (below Threshold) liquid is diverted to tubes, the delay volume (at the rising edge) of the peak is collected as part of the non-peak fraction. To clearly distinguish between the two types of fractions in this situation, we suggest programming a much larger non-peak fraction size.

### 5.5.3 Collection Using Time Windows and Delay

Case 1, where Start Time = 5 and Delay Time = 0
Case 2, where Start Time = 5 and Delay Time = 0.25

Assume the following:

| | |
|---|---|
| Flow rate | 1 ml/minute |
| Peak fraction size | 1 minute |
| Time window #1 | Start 5, End 8 |
| Time window #2 | Start 10, End 12 |

Note that each time or volume window is separated by a tube containing the delay volume. If delay time is zero, then tube #1 will be empty and subsequent time or volume windows collected are separated by empty tubes.

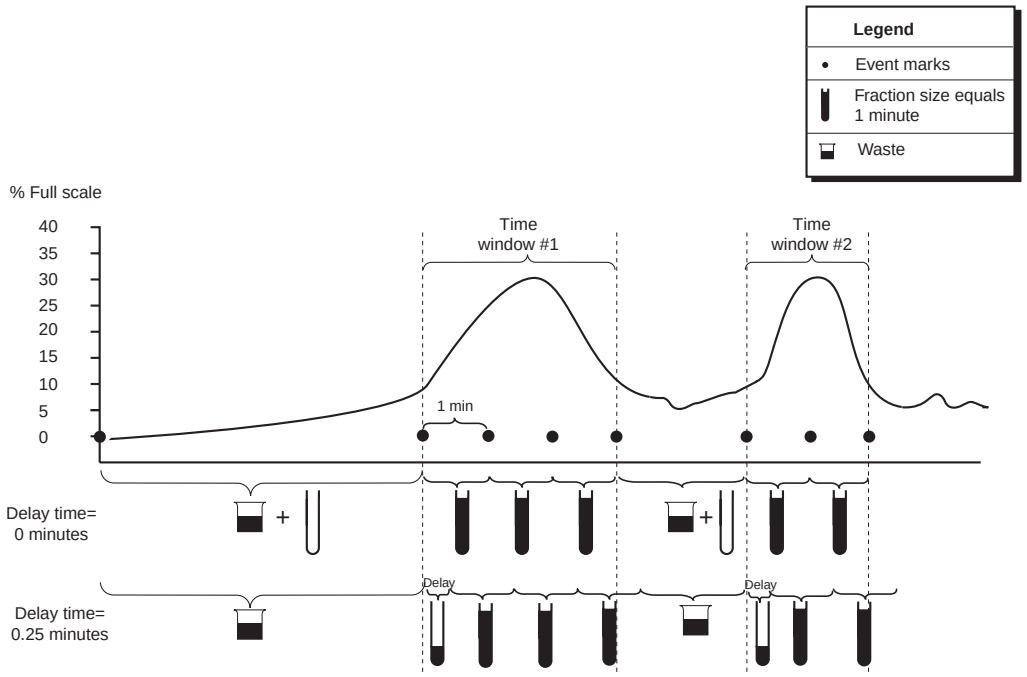



**Fig. 33. Example showing Collection Windows and a delay.**

# Section 6
# LP/Econo Mode Operation

In LP/Econo mode all aspects of fraction collection are controlled by an external controller. In this mode, only Rack, MultiRun, Start Tube, and End Tube parameters may be set from the fraction collector Main screen. Time and Volume Windows, Threshold, and Delay functions are not programmable from the fraction collector in this mode. However, if the fraction collector is connected to a BioLogic LP system (or a BioLogic HR with an SV3-2 diverter valve), Collect All, Threshold, Collection Windows, and Threshold+Collection Windows (all with delay volume, if desired) can be controlled through the BioLogic LP system (or BioLogic HR). When the BioFrac is connected to a Model EP-1 Econo pump, only Collect All (with delay volume using void (Vo)) is available. Any controller capable of sending TTL fraction advance and diverter valve signals can be used in this mode, if it has compatible control circuitry logic (see Appendix B).

To run in LP/Econo mode, set up the fraction collector as follows:

1. Change mode to LP/Econo and ensure the appropriate cable is connected to the fraction collector. (See sections 3.2.2, 3.2.3 and 3.2.7 for cabling information.) Connect system cable 15 to the I/O port on the fraction collector and to the BioLogic LP system or Model EP-1 Econo pump.

2. Connect system cable 3 to the Rec port on the BioFrac and to the BioLogic LP System or Model EP-1 Econo pump (see Section 3.2.2, 3.2.3 and 3.2.7 for cabling information).

3. To select a rack, press the Rack function key (F2) and then choose a rack from the Rack menu. Choose whether or not you want the diverter valve to divert to waste during fraction advances. If you are collecting in microplates or Titertube tubes choose the desired collection pattern: Serpentine, Row, or Column. Note that whenever a new rack is selected, the start tube and end tube values are updated to reflect the maximum number of tubes available for the selected rack.

4. Choose the appropriate MultiRun mode for your experiment (see Table 6 for a description of the MultiRun function).

5. Set the start tube and end tube rack position and tube number as follows:

   If you are overlaying fractions. Set the start tube and end tube numbers on the fraction collector to correspond to the required number of fractions derived from programming the fraction size on the BioLogic LP system, or Model EP-1 Econo pump or other controller. For example, assuming the number of fractions calculated is 30:

   i) If start tube # is A1, then end tube # is A30
   ii) If start tube # is B16, then end tube # is B45

If you are doing sequential fraction collection. Set Multi Run = Seq. Tube +1 or Seq. Rack and set the start tube and end tube numbers on the fraction collector to be greater than or equal to the number of fractions required for the run. When the run is ended by pressing Stop or by reaching the last tube, the BioFrac will automatically increment start tube and end tube according to the MulitRun function selected (see Table 6).

Alternatively, sequential tube collection can be controlled by the BioLogic LP system. In this case, however, no tubes are skipped between runs. To run in this mode, set MultiRun to Seq. Tube+1 or Seq. Rack and set end tube to be greater than or equal to the total number of tubes required for all runs. For example, if you will be doing 3 runs requiring 25 tubes each, then 75 tubes are required. Set the MultiRun parameter on the BioLogic LP as described in the BioLogic LP system manual.

6. Press the Engage function key (F1) to cause the fraction collector to move to the start tube and listen for fraction advance and diverter valve signals. The BioFrac will now wait for fraction advance commands from the external controller.

7. At any point during the run, you may use the Run screen's Stop function key (see Table 8) to stop the fraction collector.

# Section 7
# Maintenance and Troubleshooting

## 7.1 Maintenance

The BioFrac fraction collector requires little maintenance to ensure reliable operation. To clean the case, first unplug the fraction collector. Use a damp cloth to wipe down the outer case. Avoid getting the power switch and rear panel connectors wet.

Over time the clear glass ring inside the drop head may require cleaning. This ring protects the drop detector from splashes or dust buildup. To clean the clear glass ring, simply unscrew the inlet fitting and lift out the clear inside ring that protects the detector. Wipe the ring clean using a damp cloth.

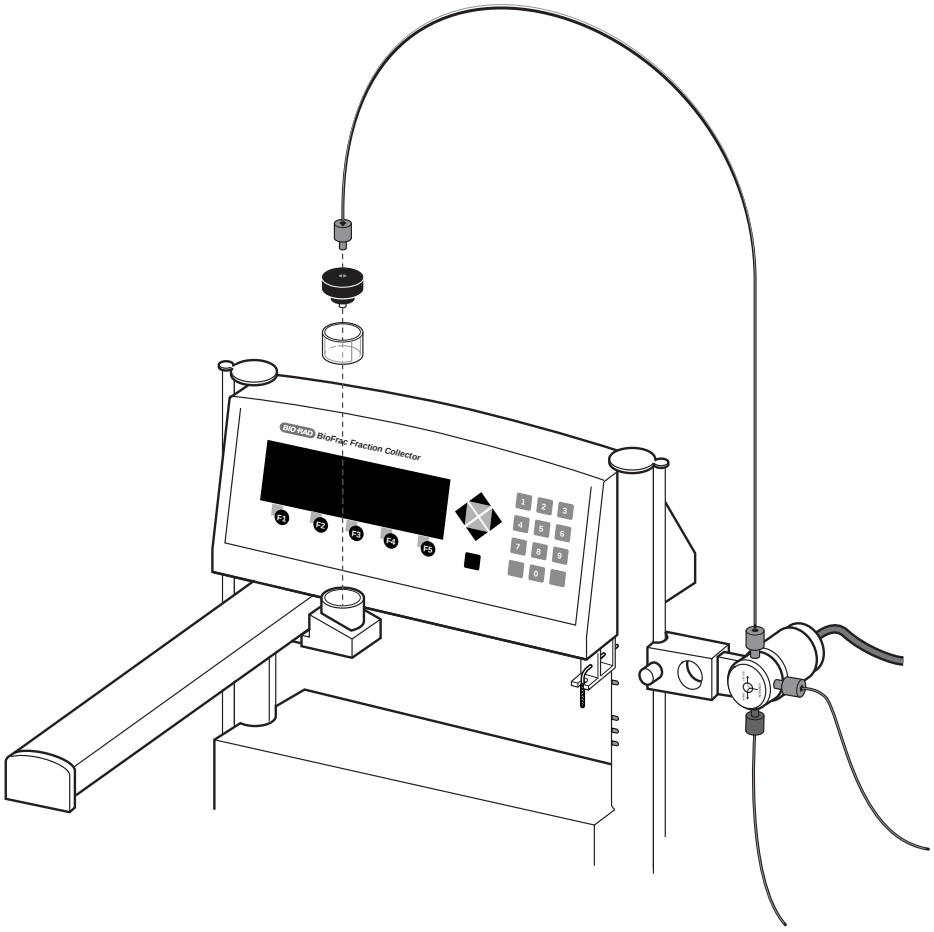



**Fig. 34. Cleaning the drop head window.**

When finished using the BioFrac fraction collector, be sure to rinse all salts from the diverter valve with water. Leaving salt solution in the valve may cause salt crystals to form, which could plug or damage the valve. Rinsing salt from the valve will increase the valve's lifetime.

## 7.2 Troubleshooting

| Problem | Possible Cause | Solution |
|---|---|---|
| No LCD display | No power to unit. | Check the power switch to be sure it is on.<br>Check the power cord connections.<br>Check the power at the outlet.<br>Make sure the fraction collector display cable is plugged into the base unit.<br>If problem persists, contact Bio-Rad.<br>**Note**: the unit contains no fuses to replace or circuit breakers to reset. |
| LCD display is difficult to read | LCD setting needs adjustment. | Increase or decrease the brightness as described in Section 4.5. |
| Drops miss tube | Rack type is incorrect.<br>Tube rack is misaligned.<br>Drop former missing.<br>Unit not level.<br>Drop head misaligned. | Select the correct rack type.<br>Reposition tube rack.<br>Insert drop former.<br>Place the unit on level surface.<br>Turn the unit off and on to reposition the arm.<br>Recalibrate the drop head as described in Section 4.5. |
| Drops not counted | Flow rate is too high.<br>Drop head is dirty. | When collecting by drops, do not exceed 5 ml/min (3 ml/min for the microplate drop head).<br>Clean the drop head as described in Section 7.1. |
| Flow continues after pump stops | Excessive backpressure. | Change to tubing with larger inner diameter (ID). |
| During peak detection bubbles are not being filtered | Bubble Filter setting is not appropriate.<br><br>Bubbles are too slow to trigger the Bubble Filter. | Check the Bubble Filter setting.<br><br>Reset the bubble filter time according to the guidelines in Appendix A.<br><br>No corrective action. |
| Fraction collector causes a high back pressure | Tubing is kinked.<br>Wrong tubing is being used.<br><br>Divert valve or tubing is plugged. | Replace tubing.<br>Connect the appropriate sized tubing for the flow rate being used (see Section 3.1).<br>Rinse the diverter valve with water to remove salt crystals or other particulate matter. |
| Fraction collector will not go to system mode when connected to a BioLogic DuoFlow system | BioFrac is not in Main screen.<br>BioFrac is not in Local mode.<br>Bus cable is not connected | Place the fraction collector in Local mode and make sure the Main screen (start-up screen) is displayed. |

# Appendix A
# Bubble Filter Time

The Bubble Filter Time function is used in conjunction with the Peak Collection by Threshold. The purpose of the Bubble Filter Time function is to distinguish true chromatographic peaks from unwanted signals such as electrical spikes or the passage of an air bubble. Such false signals are characterized by extremely fast rise and fall times.

The Bubble Filter Time function is entered at the end of the threshold programming sequence. The default value is off (bubble filter = 0 seconds).

A signal that exceeds the set threshold value with a rise time of > 6.0% full scale (FS) in 0.1 seconds is perceived by the fraction collector as possibly signifying a false peak. In such cases, the fraction collector looks for a bubble filter time set by the user. If the signal falls to 2% of the pre-rise FS level within the set bubble filter time, the signal is rejected and the false peak is not collected. Signals that exceed the threshold value, and are of a longer duration than the bubble filter time, are collected as true peaks.

Very sharp, true chromatographic peaks (such as those from an HPLC application) may rise at a sufficient rate to trigger this function. As a consequence, if an unsuitable bubble tilter time has been set, then the early part of such peaks may not be collected.

The bubble filter time may be set from 0 to 10 seconds. For most chromatographic applications, a filter time of 0 or 1 second will suffice. The actual setting depends upon both the flow rate and peak duration and should be optimized for each type of separation. Generally speaking, a very short bubble filter time should be used with sharp peaks of short duration.

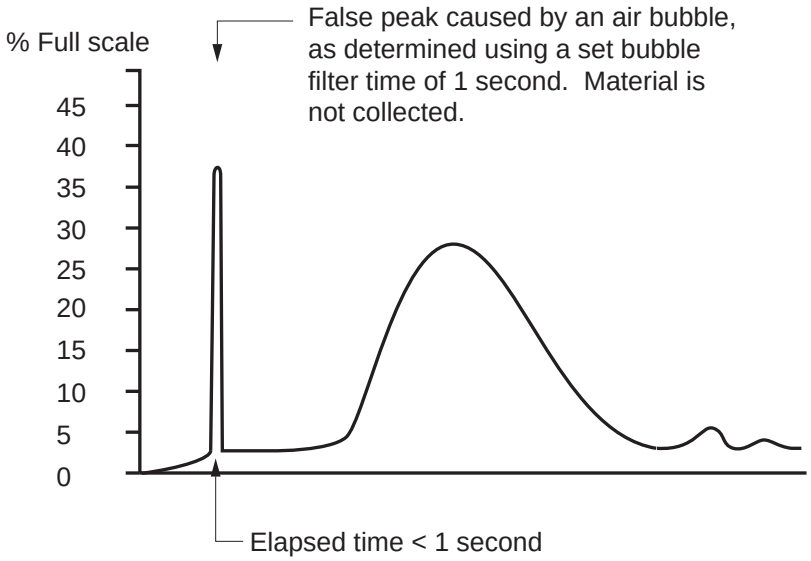
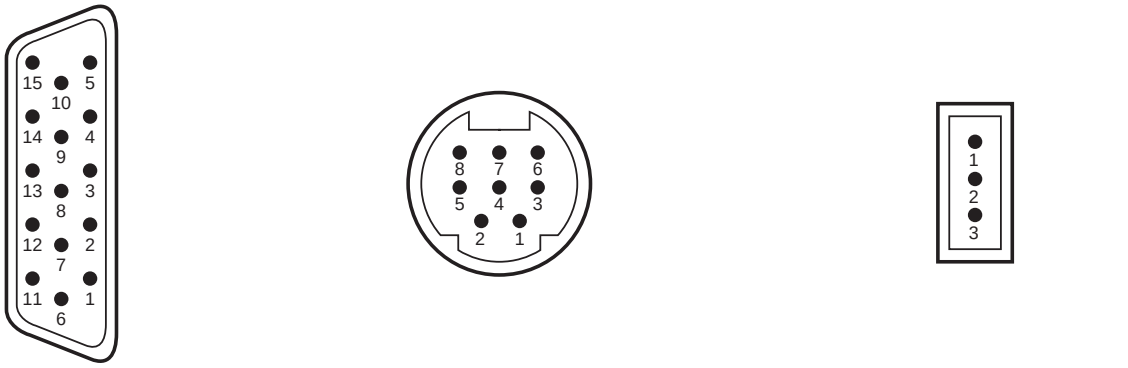



**Fig. 35. Bubble filter time.**

# Appendix B
# Rear Panel Connector Information




**15 - pin**
**D connector**

**8 - pin**
**mini - DIN connector**

**3 - pin**
**Combicon connector**

**Fig. 36. 15-pin D, 8-pin mini-DIN, and 3-pin Combicon connectors.**

## 15-Pin D Connector

| Pin # | Input Signals | Type | Active | BioFrac Accessory Cable Wire Color | Description |
|---|---|---|---|---|---|
| 1 | ADVANCE | Pulse | Low | Black | Receives remote fraction advance signals (100 ms pulse). Active in LP/Econo mode. |
| 5 | LISTEN FOR REMOTE START | Level | Low | Orange | Connection of pin #5 and #15 (signal Ground) tells the fraction collector to wait for a remote start signal on pin #6. Active in Local mode. |
| 6 | RUN/STOP | Level | Low | Blue | Grounding of pin #6 by the start/stop relay starts the currently programmed method. Pins #6 and #15 should be connected to the Run/Stop relay on the controlling device. Active in Local mode. |
| **Pin #** | **Output Signals** | **Type** | **Active** | **Wire Color** | **Description** |
| 4 | READY | Level | Low | Green | Indicates that the fraction collector diverter valve is connected and functioning properly. |
| 9<br>10 | START REMOTE<br>DEVICE | Relay | Closed | Green/black<br>Orange/black | Relay used for starting a remote device such as a pump. Relay is closed when Run is pressed and opened when Stop is pressed. Active in Local mode. |
| 11 | EVENT MARK | Pulse | Low | Blue/Black | Sends a negative pulse (200 ms) for each fraction advance. If the delay function is being used, the drop head advance will be separated from the event mark by the delay time, volume, or drops. |
| 15 | SIGNAL GROUND | | | Blue/White | Signal ground |

All input and output pins, except the relay terminals, are TTL compatible. All input pins are with 10 kΩ pull-up. The relay pins are used to control low-voltage DC contact-closure type devices, such as 12 V, 10 mA. **Caution**: Do not apply 110 V AC or 220 V AC directly to the relay terminal pins.

## 8-Pin Mini-DIN Connector

| Pin # | Input Signals | Type | Active | System Cable 7 Wire Color | Description |
|---|---|---|---|---|---|
| 2 | WASTE/COLLECT | Level | Low | Orange | Controls diverter valve. Connection of pin #2 to pin #8 (signal ground) causes the diverter valve to change to collect. Controlled by a relay on a remote device. Active in LP/Econo mode. |
| **Pin #** | **Output Signals** | **Type** | **Active** | **Wire Color** | **Description** |
| 3 | EVENT MARK | Pulse | Low | Yellow | Sends a negative pulse (200 ms.) for each fraction advance. If the delay function is being used, the drop head advance will be separated from the event mark by the delay time, volume, or drops. Active in Local mode. |
| 4 | PAPER FEED/STOP | Level | High | Red | Chart recorder control, paper feed/stop. Active in Local mode. |
| 5 | PEN UP/DOWN | Level | Low | Brown | Chart recorder control, pen up/down. Active in Local mode. |
| 6<br>7 | ADS TERMINAL | Level | Low | Purple<br>Blue | Event marks for Method Start and Fraction Advance by way of a relay. Pins #6 and #7 are closed for 200 ms with each fraction advance. Active in Local mode. |
| 8 | SIGNAL GROUND | | | Green | Signal ground |

All input and output pins, except the relay terminals, are TTL compatible. All input pins are with 10 kΩ pull-up. The relay pins are used to control low-voltage DC contact-closure type devices, such as 12 V, 10 mA. Caution: Do not apply 110 V AC or 220 V AC directly to the relay terminal pins.

## 3-Pin Combicon Connector

| Pin # | Input Signals | Type | Active | Description |
|---|---|---|---|---|
| 1 | Analog ground | Common | Low | UV monitor input: analog ground |
| 2 | Analog IN(-) | Input | N/A | UV monitor input 100 mV and 1 V (–) |
| 3 | Analog IN(+) | Input | N/A | UV monitor input 100 mV and 1 V (+) |

# Appendix C
# Specifications

| | |
|---|---|
| Fractionation | |
| Time | 0.02 to 9999.9 minutes |
| Drop | 1 to 99999 drops; flow rate 5.0 ml/min (max.) |
| Volume | 0.02 to 99999 mls. |
| Collection methods | Time, Drop, Time Windows and Volume Windows (up to 20 windows), Peak Detection, Time or Volume Windows plus Peak Detection. |
| Peak detection | Threshold, including slope detection algorithm for collection of double peaks above a set threshold |
| Drop counting capability | Up to 5 ml/min flow rates |
| Maximum collection volume | Virtually unlimited when used with the Prep-20 Adaptor |
| Tube change time | <350 ms, with rack F1<br><350 ms, with rack F2 |
| Detector input | ±0 to 100 mV; and ±0 to 1 V |
| Event marker | TTL diverter/peak mark; contact closure fraction mark |
| Input power | 100-240 VAC, ±10%, 50/60 Hz, single phase |
| Input current | 1.2 A (max.); fuse rating inside the power supply is 2 A |
| Power consumption | 45 W |
| Fusing | No external fusing |
| 3-way diverter valve | 3-way diverter valve mounts on either the left or right column. Minimizes spillage during fraction advances |
| External operation | Can be controlled by the BioLogic DuoFlow system, BioLogic LP system, Model EP-1 Econo pump or other systems via external commands |
| Wetted parts | All nonmetal, including Tefzel, PEEK |
| Safety certification | EN 61010-1 |
| EMI certification | EN 61326 |
| Environmental | |
| Operating temp. | 4 to 40°C |
| Storage temp. | 4 to 40°C |
| Humidity | 5 - 95%, noncondensing |
| Weight | 6.9 kg; 15.3 lb |
| Dimensions | 44.5 x 35.6 x 38.7 cm<br>17.5 x 14.0 x 15.2 in |

# Appendix D
# Warranty and Ordering Information

The BioFrac fraction collector is warranted for 1 year against defects in materials and workmanship. If any defects should occur during this warranty period, Bio-Rad Laboratories will replace the defective parts without charge. However, the following defects are specifically excluded:

1. Defects caused by improper operation.
2. Repair or modification done by anyone other than Bio-Rad Laboratories or their authorized agent.
3. Use with fittings or other spare parts not specified by Bio-Rad Laboratories.
4. Damage caused by deliberate or accidental misuse.
5. Damage caused by disaster.
6. Damage due to use of improper solvent or sample.
7. Tubing and fittings.

## Warranty Information

Model: _____

Serial Number: _____

Date of Delivery: _____

Warranty Period: _____

## Ordering Information

| Catalog Number | Product Description |
|---|---|
| 741-0002 | BioFrac Fraction Collector, includes a 110 V power cord, rack F1(2), Econo system cable 15, and fittings kit |
| 741-0010 | Rack Set F1, accepts 12 to 13 mm diameter tubes (90 tubes max) |
| 741-0011 | Rack Set F2, accepts 15 to 16 mm diameter tubes (60 tubes max) |
| 741-0012 | Rack Set F3, accepts 18 to 20 mm diameter tubes (40 tubes max) |
| 741-0013 | Rack Set H1, accepts 42 capless 1.5 ml Eppendorf/microtubes (168 tubes max) |
| 741-0014 | Rack Set H2, accepts 63 capless 0.5 ml Eppendorf/microtubes (252 tubes max) |
| 741-0015 | Rack Set H3, accepts 30 reduced volume scintillation vials, 16 mm (120 vials max) |
| 741-0016 | Rack Set H4, accepts 6 scintillation vials, 30 mm (24 vials max) |
| 741-0020 | Rack Set H4-High, accepts 6 tubes, 30 mm (24 tubes max.) |
| 741-0017 | BioFrac Ice Bath/Microplate Rack, holds 120 tubes on ice, 12–13 mm or up to 4 microplates/Titertube racks (SBS standard format) |
| 741-0018 | BioFrac Prep-20 rack, for preparative collection in up to 20 collection vessels, includes drain tubing and 20 feet of Tygon tubing |
| 741-0008 | BioFrac Diverter Valve |

| Catalog Number | Product Description |
|---|---|
| 741-0088 | BioFrac Microplate Drop Head Kit, includes drop head which delivers 25 µl drops compared to standard drop head delivery of 50 µl |
| 731-8286 | System cable 15, 15-pin D to mini-DIN, for connecting the fraction collector to the BioLogic LP system controller or the Model EP-1 Econo pump |
| 731-8263 | System cable 3, mini-DIN to mini-DIN cable, for connecting the fraction collector to a BioLogic LP system controller or a Model EP-1 Econo pump (for control of the diverter valve) |
| 731-8290 | BioFrac Accessory Cable, 15-pin D to bare wires, for connecting the fraction collector to non-Bio-Rad equipment |
| 731-8287 | System cable 16, mini-DIN to DIN cable, for connecting the fraction collector to a Model 1327 chart recorder |
| 731-8264 | System cable 4, 8-pin mini-DIN to banana plug cable, for connecting the Model EM-1 Econo UV monitor to the Model 1327 chart recorder |
| 731-8267 | System cable 7, 8-pin mini-DIN to bare wires cable, for connecting the BioFrac fraction collector to non-Bio-Rad equipment |
| 741-0007 | BioFrac Fittings Kit |

**Collection Tubes***

| | |
|---|---|
| 223-9500 | 1.5 ml Capless Micro Test Tubes, polypropylene, natural, 500/box |
| 223-9750 | 13 x 100 mm Clear Polystyrene Test Tubes, 1000/box |
| 223-9750 | 13 x 100 mm Natural Polypropylene Test Tubes, 1000/box |
| 224-0096 | Costar 96-Well Flat-bottom EIA Plate, polystyrene, 5 per package, box of 100 |

*Additional tubes sizes are available from Bio-Rad. Contact your local Bio-Rad representative for a liquid handling catalog.

**Bio-Rad Laboratories, Inc.**

*Life Science Group*

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***Brazil*** *Ph. 55 21 507 6191* ***Canada*** *Ph. (905) 712-2771, Fx. (905) 712-2990* ***China*** *Ph. (86-21) 63052255, Fx. (86-21) 53964775*
***Denmark*** *Ph. 45 44 52-1000, Fx. 45 4452 1001* ***Finland*** *Ph. 358 (0)9 804 2200, Fx. 358 (0)9 804 1100* ***France*** *Ph. 01 47 95 69 65, Fx. 01 47 41 9133*
***Germany*** *Ph. 089 318 84-177, Fx. 089 318 84-123* ***Hong Kong*** *Ph. 852-2789-3300, Fx. 852-2789-1257*
***India*** *Ph. (91-124)-6398112/113/114, 6450092/93, Fx. (91-124)-6398115/6450095* ***Israel*** *Ph. 03 951 4127, Fx. 03 951 4129*
***Italy*** *Ph. 39 02 216091, Fx. 39 02 21609399* ***Japan*** *Ph. 03-5811-6270, Fx. 03-5811-6272* ***Korea*** *Ph. 82-2-3473-4460, Fx. 82-2-3472-7003*
***Latin America*** *Ph. 305-894-5950, Fx. 305-894-5960* ***Mexico*** *Ph. 52 5 534 2552 to 54, Fx. 52 5 524 5971*
***The Netherlands*** *Ph. 0318-540666, Fx. 0318-542216* ***New Zealand*** *Ph. 64-9-4152280, Fx. 64-9-443 3097* ***Norway*** *Ph. 47-23-38-41-30, Fx. 47-23-38-41-39*
***Portugal*** *Ph. 351-21-472-7700, Fx. 351-21-472-7777* ***Russia*** *Ph. 7 095 721 1404, Fx. 7 095 721 1412* ***Singapore*** *Ph. 65-2729877, Fx. 65-2734835*
***South Africa*** *00 27 11 4428508, Fx. 00 27 11 4428525* ***Spain*** *Ph. 34 91 590 5200, Fx. 34 91 590 5211* ***Sweden*** *Ph. 46 (0)8-55 51 27 00, Fx. 46 (0)8-55 51 27 80*
***Switzerland*** *Ph. 061 717-9555, Fx. 061 717-9550* ***United Kingdom*** *Ph. 0800-181134, Fx. 01442-259118*

*Sig 1001*

4006211 Rev B