iontorrent

Ion OneTouch[™] 2 System USER GUIDE

For installation, setup, and maintenance of the Ion OneTouch $^{^{\text{TM}}}$ 2 Instrument and Ion OneTouch $^{^{\text{TM}}}$ ES Instrument

Catalog Number 4474779

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Revision A.0



The information in this guide is subject to change without notice.

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About this guide



CAUTION! ABBREVIATED SAFETY ALERTS. Hazard symbols and hazard types specified in procedures may be abbreviated in this document. For the complete safety information, see the "Safety" appendix in this document.

IMPORTANT! Before using this product, read and understand the information in the "Safety" appendix in this document.

Revision history

Revision	Date	Description
A.0		Instrument user guide that includes instructions for installation, setup, and maintenance.

Purpose of the guide

The *Ion OneTouch*^{$^{\text{IM}}$} 2 *System User Guide* (Pub. no. MAN0014388) provides reference information for the installation, setup, and maintenance of the Ion OneTouch $^{^{\text{IM}}}$ 2 System (Cat. no. 4474779), which includes the Ion OneTouch $^{^{\text{IM}}}$ 2 Instrument and the Ion OneTouch $^{^{\text{IM}}}$ ES Instrument. For information about how to run these instruments, see the template kit-specific user guides.



Product information

Instrument system and components

The Ion OneTouch[™] 2 System is an automated system used for the generation of template-positive Ion Sphere [™] Particles (ISPs) for semiconductor sequencing.

The Ion OneTouch[™] 2 System (Cat. no. 4474779) contains the following boxes and components:

Description	Catalog No.
Ion OneTouch [™] 2 Instrument ^[1]	4474778, 4474779
Ion OneTouch [™] ES Instrument	4474779
AC Power Supply and Cords	

^[1] Sold separately or as part of the system.

Note: See the sequencer-specific site preparation guide for information on site preparation for the sequencer and the Ion OneTouch $^{\text{\tiny TM}}$ 2 System.

Note: We recommend using an uninterruptable power supply (UPS) for laboratories that experience frequent power outages or line voltage fluctuations. The UPS must be rated for 1500 W output or higher. The 1500 VA unit from APC provides several minutes of backup power for the Ion OneTouch $^{\text{TM}}$ 2 Instrument and Ion OneTouch $^{\text{TM}}$ ES Instrument. Use a surge protector or line conditioner as needed.

Required materials and equipment (not provided)

Installation, setup, and maintenance of the Ion OneTouch[™] 2 System uses common molecular biology equipment, supplies, and reagents. Unless otherwise indicated, all materials are available through **thermofisher.com**. MLS: Fisher Scientific (**www.fisherscientific.com**) or other major laboratory supplier.

Item ^[1]	Source
 Ion OneTouch[™] template preparation solutions (provided in your Ion OneTouch[™] Template Kit): Ion OneTouch[™] Oil Ion OneTouch[™] Recovery Solution 	See product web pages
Ion OneTouch [™] template preparation supplies (provided in your Ion OneTouch [™] Template Kit): • Ion OneTouch [™] Reagent Tubes • Ion OneTouch [™] Recovery Routers • Ion OneTouch [™] Recovery Tubes • Ion OneTouch [™] Sippers • Ion OneTouch [™] 2 Amplification Plates	See product web pages
One of the following control library kits:	
For PGM [™] Sequencers: E. coli DH10B Control Library from the Ion PGM [™] Controls Kit v2	4482010
For Ion Proton [™] Sequencers: Human CEPH Control 200 Library from the Ion PI [™] Controls 200 Kit	4488985
For Ion S5 [™] Sequencers: Human CEPH Control 200 Library from the Ion S5 [™] Controls Kit	A27760
Dynabeads [™] MyOne [™] Streptavidin C1 Magnetic Beads	65001
DynaMag [™] -2 magnet	12321D
GeneAmp [™] PCR System 9700 thermal cycler or equivalent	N8050200 (Base) 4314443 (Block)
Category 6 Ethernet Cable	MLS
Surge protector or line conditioner	MLS
1.5-mL Eppendorf LoBind [™] tubes	MLS
0.2-mL PCR tubes	Axygen PCR-02-L-C
	BioExpress T-3035-1
Microcentrifuge ^[2]	MLS
Pipettors (P2, P20, P200, P1000) and appropriate low- retention tips	MLS
Vortexer with a rubber platform	MLS
Tube rack to fit 15-mL conical tube	MLS
Tube rack for 50-mL conical tube	MLS

Item ^[1]	Source
1 M NaOH	MLS
Bleach	MLS
Xiameter [™] PMX-200 Silicone Fluid ^[3]	Neely Industries PMX200-12500PT
Benchtop absorbent paper or mat	MLS
1/8-inch L-wrench (hex wrench) or equivalent tool	MLS

^[1] We have verified this protocol using these specific materials. Substitution may adversely affect system performance.

Must fit standard 1.5- and 0.2-mL microcentrifuge tubes; must generate 15,500 \times g.

^[3] Material required for periodic maintenance of the Ion OneTouch™ ES.



Ion OneTouch[™] 2 Instrument installation, setup, and maintenance

Instrument clearances

Position the instrument so that the front is a minimum of 12 in. (30.5 cm) from the front of the laboratory bench. Place the instrument at least 40 in. (1 meter) away from major sources of electronic noise such as refrigerators or microwaves. For more information, refer to the appropriate site preparation guide specific for your sequencing system.

Unpack and install the Ion OneTouch[™] 2 Instrument

For detailed instructions on site preparation and installation of the Ion OneTouch [™] 2 Instrument, refer to the site preparation and installation requirements in your sequencing system site preparation guide.

1. Unpack and install the Ion OneTouch[™] 2 Instrument in a location different from the location used to prepare the amplification solution. Remove the instrument by laying the shipping box sideways on a table, then sliding out the instrument.

IMPORTANT! Do not lift the instrument by the metal handle used to access the Ion OneTouch[™] 2 Amplification Plate.

- 2. Ensure that the power switch is turned **Off**, then plug the power cord into the instrument
- **3.** Plug the other end of the power cord into an electrical outlet of the appropriate voltage.

IMPORTANT! The Ion OneTouch^{TM} 2 Instrument draws 6 amps of current. Do not exceed the circuit breaker limit for current. If necessary, plug multiple instruments into different circuits.

4. Turn the power switch **On**. Initial start-up takes ~3 minutes. You may hear sounds from the instrument. This is normal.

IMPORTANT! Leave the Ion OneTouchTM 2 Instrument **On** indefinitely. If the instrument is turned off, critical log files are lost. If you need to turn off the instrument or if a run fails, download the log files (see "Download the log files from the Ion OneTouchTM 2 Instrument" on page 26).

Checking and updating the firmware on the Ion OneTouch[™] 2 Instrument

Firmware updates to the software controlling the Ion OneTouch™ 2 Instrument are periodically released. To update the firmware to the appropriate version, use either a USB flash drive or an ethernet connection, as described in the next sections.

IMPORTANT! Ensure that the latest firmware is installed on the Ion OneTouch[™] 2 Instrument. However, if you are using the Ion PGM[™] Template OT2 200 Kit together with the Ion PGM[™] Hi-Q[™] Sequencing Kit, do not upgrade to Torrent Suite[™] Software v5.0 or later until you no longer require the mixed workflow. The OT2 program script enabling the mixed workflow is discontinued in v5.0. We recommend using the Ion PGM[™] Hi-Q[™] OT2 Kit (Cat. no. A27739) together with the Ion PGM[™] Hi-Q[™] Sequencing Kit.

Check the firmware version

In the instrument touch screen, touch **Options**, then touch **Info**.

Install a firmware update with a USB flash drive

Firmware updates can only be installed by Administrator-level users on the Torrent Server connected to your Ion OneTouch™ 2 Instrument.

1. Ensure that the USB flash drive is FAT32-formatted. If your USB is not FAT32 formatted, follow the instructions below to reformat it.

Note: Reformatting will erase on the data on the USB.

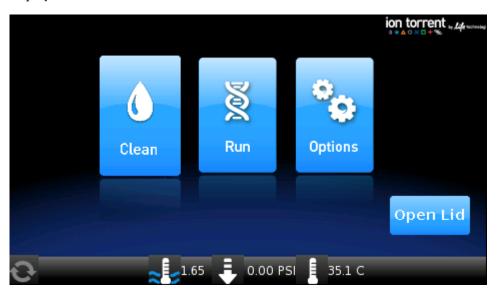
Operating system	Procedure	
Mac OS [™]	 Insert the USB flash drive into a USB port, then double-click the Macintosh HDD icon. 	
	 Navigate to the Applications folder, then click: Utilities ➤ Disk Utility. 	
	3. Double-click the Disk Utility application.	
	4. Select the drive (the <i>USB flash drive</i>) to format, then click Erase .	
	From the volume format, select MS-DOS (FAT), then enter a name for the USB flash drive.	
	6. Click Erase , then confirm the FAT32 formatting.	
Windows [™]	Insert the USB flash drive into a USB port, then double-click My Computer.	
	2. Right-click the USB flash drive icon, then select Format .	
	 Select FAT32 from the drop-down menu, click Start, then confirm the FAT32 formatting. 	

2. Log in to the Torrent Browser with an Administrator account user name and password.



Checking and updating the firmware on the Ion OneTouch™ 2 Instrument

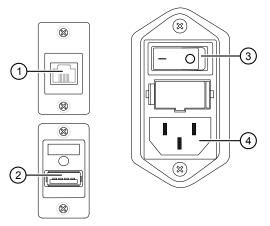
- **3.** Click the **Settings** icon at top-right of the screen, then select **Configure** from the drop-down menu.
- **4.** Click **Admin Interface Device**, then download the appropriate update file (.bz2) to your formatted USB flash drive.
- **5.** On the Ion OneTouch[™] 2 Instrument, turn the power switch **On**.
- **6.** Ensure that the instrument is not performing a run and that the home screen displays:



7. On the home screen, touch **Options** • **Upgrade**:



8. Insert the USB flash drive into the USB port, located on the back of the Ion OneTouch[™] 2 Instrument:



- 1 Ethernet connection
- ② USB port
- (3) Power switch (on/off)
- (4) Power socket
- **9.** On the Upgrade screen, touch **Yes**.
- **10.** Wait 10 seconds for the screen to display the software status update:
 - If the status update displays, proceed to step 11.
 - If the Options screen displays, then there are no firmware updates.
- **11.** Touch **Options** ▶ **Info** to ensure that the new version of the firmware update is installed.
- **12.** Turn the power switch **Off**, wait several seconds, then remove the USB flash drive.
- **13**. Turn the power switch **On**.

Alternative method: Install a firmware update using an ethernet connection **Note:** Use a shielded Category 6 ethernet cable to connect to the Ion OneTouch [™] 2 Instrument.

- 1. Turn the power switch Off.
- **2.** Connect the Ion OneTouch[™] 2 Instrument to the Torrent Server via a Category 6 ethernet cable.
- **3.** Turn the power switch **On**. Initial start-up takes ~3 minutes.

Note: You may hear sounds from the instrument. This is normal.

- **4.** Log in to the Torrent Browser with an Administrator account user name and password.
- **5.** Click the **Settings** icon at top-right of the screen, then select **Configure** from the drop-down menu.

6. Click **Admin Interface** ▶ **Update OneTouch Device** ▶ **Update** then wait for the update to complete and the instrument to reset.

Note: A progress bar appears on the instrument during the update.

7. Touch **Options** on the Ion OneTouch[™] 2 Instrument home page to ensure that the new version of the firmware update is installed.

Ion OneTouch[™] 2 Instrument touchscreen messages

If the on-screen message is	Then the instrument is
Priming and Filling Recovery Solution	Priming the filter and filling the collection tubes with Ion OneTouch™ Recovery Solution.
Sample Injection	Injecting sample and starting emulsification.
Amplification	Amplifying the sample by PCR.
Filling Recovery Solution	Completely filling the Ion OneTouch [™] Recovery Tubes with Ion OneTouch [™] Recovery Solution.
ISP Collection	Breaking the emulsion and collecting the Ion Sphere [™] Particles (ISPs).
Wash Cycles	Washing the instrument and draining the washes.
Final Spin	Performing final centrifugation to pellet the ISPs in the Ion OneTouch™ Recovery Tubes.

Set up the Ion OneTouch[™] **2 Instrument**

Before operating the Ion OneTouch $^{\scriptscriptstyle{\text{TM}}}$ 2 Instrument for the first time, you must perform the one-time initialization on the instrument. Initialization primes the pumps and tubing lines for reliable operation. Perform initialization at any time before the first run.



Ion OneTouch™ 2 Instrument layout

- (1) Ion OneTouch™ Reaction Filter
- (2) Clamp handle to access the Amplification Plate in the heat block
- (3) Waste Container
- (4) Ion OneTouch™ Oil
- (5) Ion OneTouch™ Recovery Solution 💍
- (6) Pinch valve to hold disposable tubing
- (7) Oil waste tray
- (8) Centrifuge to spin the Recovery Tubes and Recovery Router
- (9) Ion OneTouch™ DL Injector Hub
- (10) Instrument display

Materials required for this procedure

Provided in your template preparation supplies kit:

- 2 Ion OneTouch[™] Reagent Tubes
- Ion OneTouch $^{\text{\tiny TM}}$ Recovery Router
- 2 Ion OneTouch[™] Recovery Tubes
- Ion OneTouch[™] 2 Amplification Plate
- 2 Ion OneTouch[™] Sipper Tubes

Provided in your template preparation solutions kit:

- Ion OneTouch[™] Oil
- Ion OneTouch[™] Recovery Solution

Note: This protocol has been verified using only the materials specified. Substitution may adversely affect performance and safety.

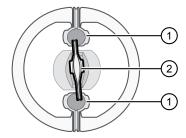
Install the Ion OneTouch[™] Recovery Tubes and Ion OneTouch[™] Recovery Router 1. On the instrument touchscreen, touch **Open Lid**, wait until the lid clicks open, then lift and hold the side of the centrifuge lid.

IMPORTANT! Do *not* lift the lid by the tubing attached to the Ion OneTouch^{TM} DL Injector Hub.

- Insert an Ion OneTouch™ Recovery
 Tube into each slot in the centrifuge,
 making the sure the tube arm is
 inserted into the slot next to each
 hole.
- Pinch the sides of the Ion OneTouch™ Recovery Router and push the router down into the center slot of the centrifuge until it is seated flat and secure in the center of the rotor.



Note: The router is not directly aligned with the recovery tubes, but is intentionally positioned at a angle.



- (1) Ion OneTouch™ Recovery Tube
- (2) Ion OneTouch™ Recovery Router
- **4.** Close the centrifuge lid.

Install the Ion OneTouch™ 2 Amplification Plate 1. If there is a used Ion OneTouch™ Cleaning Adapter on the instrument, remove and appropriately discard it.

Note: The Cleaning Adapter may be filled with Ion OneTouch[™] Oil.

2. Push the handle back to open the heat block.



CAUTION! Hot Surface. Use care when working around this area to avoid being burned by hot components.



WARNING! Safety Hazard. Do not use the instrument with flammable or explosive materials. Use only the materials specified for use with the instrument to ensure safety.

- **3.** Insert the Amplification Plate:
 - **a.** Inspect the Ion OneTouch^m 2 Amplification Plate to ensure the plate port is straight and perpendicular to the plate.

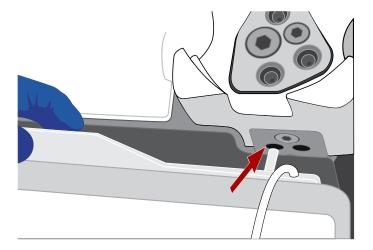
IMPORTANT! The disposable tubing and disposable injector are attached to the Amplification Plate. Do *not* disconnect tubing from the top plate port. If you have questions about the plate, contact Technical Support.

b. Hold the disposable injector, connected to the disposable tubing, in one hand and the Amplification Plate in the other hand.



CAUTION! PHYSICAL INJURY HAZARD. The pointed end of the disposable injector can puncture your skin. Keep your hand away from the point of the disposable injector.

c. Insert the Amplification Plate into the open instrument heat block, being careful to align the plate port with the *left* outlet hole on the Ion OneTouch™ 2 Instrument:



4. Pull the handle forward to secure the amplification plate. The disposable tubing should be under the handle.



 Thread the disposable tubing through the Ion OneTouch™ DL Tubing Catch, as shown below. Make sure that the tubing is not twisted.



6. Align the disposable tubing with the slot that runs along the bottom of the pinch valve. Gently pull the tubing up into the slot until it is secured in the notch, as shown below.



7. Adjust the disposable line so that it is straight, but not too taut. Allow enough line to install the disposable injector.

Install the disposable injector

Note: The long metal shaft of the disposable injector may be slightly bent, which is normal. If you have questions about the disposable injector, contact Technical Support.

1. Ensure that the needle of the injector is screwed tightly onto the rubber tubing. Hold the centrifuge lid down with one hand, and with other hand install the disposable injector by inserting it straight down into the injector hub. Push down until it just touches the router.



CAUTION! PHYSICAL INJURY HAZARD. The pointed end of the injector can puncture your skin. Keep your hand away from the point of the injector.





Note: The color of the injector may vary.

2. The spring-loaded top of the injector hub will click upon release, automatically adjusting the tip to the correct distance from the router surface. You can test this by gently pushing the injector down again and releasing. You should hear a click from the hub.







IMPORTANT! If the Injector Hub remains in the down position, see Appendix A, "Troubleshooting".

IMPORTANT! If you raise the centrifuge lid, do not hit the injector against the instrument. If you damage the disposable injector, appropriately dispose of the injector, amplification plate, and tubing. Use a new injector and Ion OneTouch™ 2 Amplification Plate.

Install the Ion OneTouch™ Oil

- 1. Fill a new Ion OneTouch™ Reagent Tube with Ion OneTouch™ Oil on the *left* front port 🙏:
 - a. Use fresh gloves to attach the Luer-Lok[™] end of a new Ion OneTouch[™] Sipper Tube to the *left* front port. Do *not* let the Sipper Tube touch any surface.
 - b. Invert the on Ion OneTouch™ Oil bottle (450-mL size) 3 times to mix, then fill a new Reagent Tube half-full with oil. Minimize bubbles.
- 2. Insert the filled Reagent Tube into the left front port, and screw the tube firmly into place, one-quarter turn on the instrument.

Install the Ion OneTouch™ **Recovery Solution**

IMPORTANT! Use only the Ion OneTouch[™] Recovery Solution in the Ion OneTouch[™] kit that you are currently using. Do not use a different recovery solution from another kit.

- 1. Inspect the Recovery Solution. If the solution is not clear, heat the bottle in a 30°C bath until the solution is clear.
- 2. Fill a new Ion OneTouch™ Reagent Tube with Ion OneTouch™ Recovery Solution and install it on the right front port δ :
 - a. Use fresh gloves to attach the Luer-Lok[™] end of a new Ion OneTouch[™] Sipper Tube to the right front port. Do *not* let the Sipper Tube touch any surfaces. Minimize bubbles.

2

- **b.** Invert the Recovery Solution bottle 3 times, then fill the Reagent Tube a quarter-full with solution.
- **3.** Insert the filled Reagent Tube into the right front port, and screw the tube firmly into place, one-quarter turn on the instrument.

Empty the Waste Container

- 1. Pull the external tubing from the port of the Waste Container.
- **2.** Empty the Waste Container into the appropriate receptacle.
- 3. Reinstall the empty Waste Container.

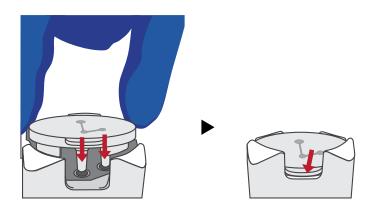
Inspect the oil waste tray

- 1. Slowly pull out the oil waste tray but do not remove it completely from the slot underneath the center of the instrument.
- **2.** Check for oil in the oil waste tray:
 - If there is little or no oil, push the tray back fully into the instrument.
 - If there is excessive oil, remove the tray, then appropriately dispose of the oil. Reinsert the oil waste tray into the slot, then push the tray back fully into the instrument. Contact Technical Support.

Install the Ion OneTouch[™] 2 Cleaning Adapter

Note the orientation of the ports and tabs on the Ion OneTouch[™] 2 Cleaning Adapter:

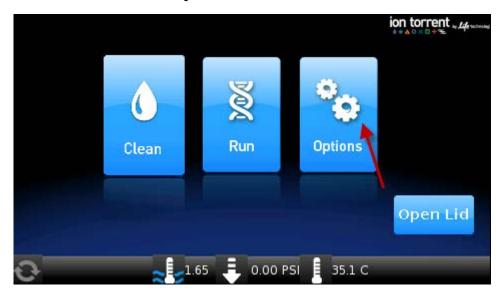
Firmly insert the 3 ports of the Cleaning Adapter into the three holes on the top stage of the Ion OneTouch $^{\text{\tiny TM}}$ 2 Instrument. One of the two tabs protruding from the outer edge of the Cleaning Adapter fits into the front notch of the stage:



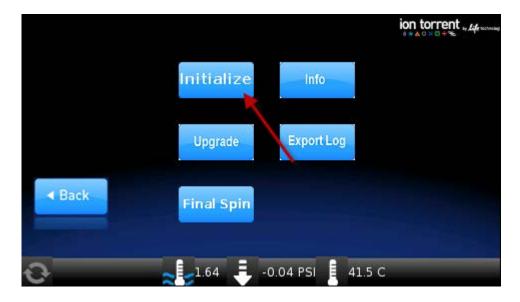
Initialize the instrument

Initialize the instrument

1. On the home screen, touch **Options**:



2. Touch Initialize:



Initialization takes 10 minutes.

Complete the set up

- Remove and retain the Ion OneTouch[™] Cleaning Adapter. You can reuse the Cleaning Adapter one time for the instrument maintenance protocol after the first run, but you must appropriately dispose of the used Cleaning Adapter after maintenance (see "Ion OneTouch™ 2 Instrument maintenance" on page 23).
- 2. Keep all of the disposable components that were used for initialization in place, including the Ion OneTouch™ 2 Amplification Plate, disposable injector and tubing, Ion OneTouch™ Router, and Ion OneTouch™ Recovery Tubes.

IMPORTANT! After the first run on the Ion OneTouchTM 2 Instrument, appropriately dispose of all components as directed.

3. Proceed to "Perform a verification run".

Perform a verification run

To ensure optimal use of the Ion OneTouch^{TM} 2 System, we recommend first preparing and enriching template-positive ISPs on the system with a control library.

- 1. Select the control library from the appropriate control kit for your sequencing system (see "Required materials and equipment (not provided)" on page 8).
- 2. In an Eppendorf LoBindTM tube, dilute 1 μ L of control library into 259 μ L of Nuclease-free Water. Use 100 μ L of the dilution in the amplification solution.
- 3. Prepare and enrich the template-positive ISPs on the Ion OneTouch™ 2 System as described in your template preparation kit user guide.
- **4.** If you have a Qubit[™] 2.0 or 3.0 Fluorometer, determine the percent template-positive ISPs. If not, proceed to step 5.
- 5. Sequence the enriched ISPs as described in your sequencing guide.
- 6. After sequencing, review the run report in the Torrent Browser and confirm successful results with the control library.
 If you have questions about the results, contact technical support.

The instrument is now ready for use.

Ion OneTouch[™] 2 Instrument maintenance

Follow the cleaning procedure in this section to clean the Ion OneTouch^{$^{\text{TM}}$} 2 Instrument with the Ion OneTouch^{$^{\text{TM}}$} Cleaning Adapter. The cleaning procedure is performed according to the steps displayed on the instrument after removing the Recovery Tubes.

IMPORTANT! Perform the cleaning procedure after every run. Do not skip this procedure.

Note: To set up the Ion OneTouch[™] 2 Instrument when switching between sequencing platforms and/or template preparation kits, see Chapter 5, "Demonstrated protocol: Switching between older and newer template preparation kits".

Clean the Ion OneTouch[™] 2 Instrument

Determine the appropriate reagents to use for maintaining the Ion OneTouch[™] 2
 Instrument:

If you are	Then
Switching to a new template preparation kit?	See Chapter 5, "Demonstrated protocol: Switching between older and newer template preparation kits".
Using the same template preparation kit?	Proceed to step 2.

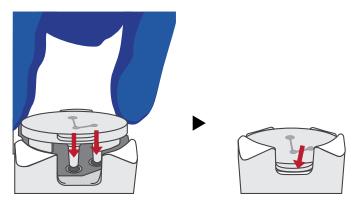
- 2. Check the level of Ion OneTouch™ Oil in the Reagent Tube:
 - **a.** Ensure that the *left* Reagent Tube has ≥20 mL of oil:
 - **b.** If the Reagent Tube has <20 mL of oil, pour oil into the Reagent Tube until it is half-full.
- 3. Remove and appropriately discard the used Ion OneTouch™ Reaction Filter. Remove the assembly from the instrument by grasping the *filter*.

Note: The Reaction Tube is filled with Ion OneTouch[™] Oil.

4. Keep the Ion OneTouch[™] 2 Amplification Plate in the heat block.

Ion OneTouch™ 2 Instrument maintenance

5. Firmly insert the 3 ports of a new single-use Cleaning Adapter into the three holes on the top stage of the Ion OneTouch™ 2 Instrument (see the following illustration). One of the tabs protruding from the outer edge of the Cleaning Adapter fits into the front notch of the stage:

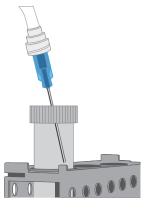


- **6. Note:** Steps 6–9 are only necessary if you have not already removed the disposable injector before removing the Recovery Tubes from the instrument. Place a 50-mL conical tube in a tube rack, then place the tube rack with the tube adjacent to the instrument.
- 7. Gently pull the disposable tubing downwards on the both sides of the pinch valve until the disposable tubing is out of the valve.
- **8.** Remove the disposable injector from the Ion OneTouch[™] DL Injector Hub:
 - a. Place one hand on the centrifuge lid.
 - **b.** With the other hand, firmly grip the rigid plastic connector at the top of the disposable injector.
 - **c.** Slowly and steadily withdraw the disposable injector straight from the port of the Injector Hub.

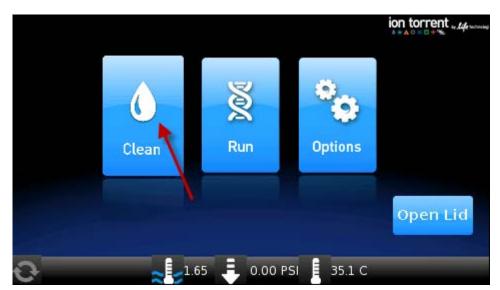


CAUTION! PHYSICAL INJURY HAZARD. The pointed end of the disposable injector can puncture your skin. Keep your hand away from the point of the disposable injector.

9. Place the used, disposable injector into the empty 50-mL conical tube in the tube rack. The conical tube will be used to collect waste:



10. On the home screen of the instrument, touch **Clean**:



- 11. Complete each task displayed on the screen, then touch **Next**. After you touch **Next** on the last task, the cleaning begins.
- 12. At the end of the cleaning run, the screen displays "Time Remaining 00:00:00, Cleaning Run Complete". Press Next, then ensure that the task in bold displays: "Remove plate, injector, conical tube, and waste".

Note: Keep the used Cleaning Adapter on the instrument between runs.

- **13.** Appropriately dispose of the waste in the 50-mL conical tube.
- **14.** Remove and appropriately dispose of the used Amplification Plate, disposable injector, and tubing from the instrument:
 - a. Push the handle to open the heat block.
 - **b.** Remove the disposable tubing from the Ion OneTouch[™] DL Catch.
 - **c.** Gently pull back the Amplification Plate from the inlet and outlet holes of the instrument.
 - **d**. Remove the Amplification Plate from the heat block, and appropriately dispose of the used Amplification Plate, injector, and tubing.
 - e. Leave the heat block open.



CAUTION! Hot Surface. Use care when working around this area to avoid being burned by hot components.

- 15. On the instrument touchscreen, touch **Open Lid**, wait until the lid clicks open, then open the centrifuge lid. Wipe the residue from the centrifuge lid with dry Kimwipes[™] disposable wipers and close the centrifuge lid.
- **16.** Touch **Next** to return to the home screen on the instrument.

Ion OneTouch[™] 2 Instrument decontamination

Before returning the instrument for service, decontaminate it according to the procedure below.

Materials and equipment required

- Disposable rubber gloves
- Safety glasses
- Lab coat
- Bleach
- Water
- Paper towels

IMPORTANT! This procedure does not guarantee total decontamination of the Ion OneTouch[™] 2 Instrument.

Decontaminate the instrument

- 1. Wear disposable rubber gloves, safety glasses, and a lab coat.
- 2. Use a cleaning pad wetted with a solution of 1 part chlorine bleach in 9 parts water (10% bleach solution) to clean all outside surfaces of the Ion OneTouch™ 2 Instrument. Use care to avoid getting bleach solution inside the chassis.
- **3.** Dry the surfaces of the Ion OneTouch[™] 2 Instrument with paper towels or other disposable wipes.
- 4. Use cotton swabs to clean and dry areas that are difficult to reach.
- **5.** Properly dispose of used cleaning materials to ensure that no one becomes exposed to contaminants.

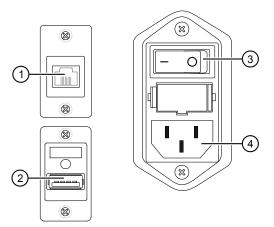
Download the log files from the Ion OneTouch[™] 2 Instrument

Log files capture important information regarding instrument operation and may be used to troubleshoot the instrument.

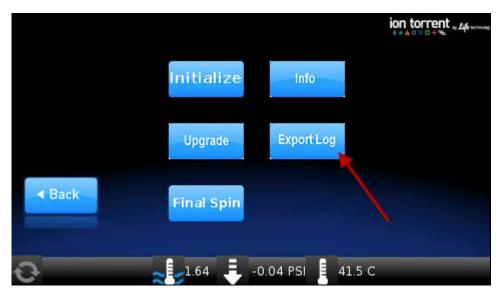
IMPORTANT! Log files are automatically deleted from the instrument when the instrument is turned off. To preserve the log files, leave the instrument on until you can download them.

- 1. Ensure that the Ion OneTouch[™] 2 Instrument is not performing a run.
- **2.** Format a USB flash drive with a FAT32 file system, if necessary (see "Install a firmware update with a USB flash drive" on page 11). Confirm that the formatted USB flash drive has no data files on it.

3. Insert the USB flash drive into the USB port, located on the back of the Ion OneTouch[™] 2 Instrument:



- 1) Ethernet connection
- ② USB port
- (3) Power switch (on/off)
- (4) Power socket
- 4. In the home screen, touch **Options**, then touch **Export Log**:



- **5.** In the Export log screen, touch **Yes**. The log files export to the USB flash drive and the status update displays. After export, the Options screen displays.
- **6.** Remove the USB flash drive from the instrument, then verify on a computer that the instrument downloaded the log files to the flash drive. If the files are:
 - Downloaded: Proceed to the next step.
 - Not downloaded: Repeat steps 3–6 until the log files are downloaded.
- **7.** Save the log files to a computer.



Ion OneTouch[™] ES Instrument installation, setup, and maintenance

IMPORTANT! Ensure that the fuse module for the Ion OneTouchTM ES Instrument is properly installed, according to 110–120 V or 240 V line voltages. See "Unpack and install the Ion OneTouchTM ES Instrument".

Instrument clearances

Position the instrument so that the front is a minimum of 12 in. (30.5 cm) from the front of the laboratory bench. Place the instrument at least 40 in. (1 meter) away from major sources of electronic noise such as refrigerators or microwaves. For more information, refer to the appropriate site preparation guide specific for your sequencing system.

Unpack and install the Ion OneTouch™ ES Instrument

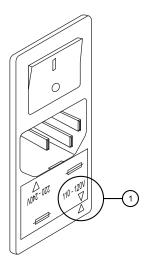
For detailed instructions on site preparation and installation of the Ion OneTouch $^{\text{\tiny M}}$ ES Instrument, refer to the site preparation and installation requirements in your sequencing system site preparation guide.

Note: Ensure that the Ion OneTouch^{$^{\text{TM}}$} ES is in a room at an operating temperature of 15°C to 25°C (60°F to 77°F).

- Unpack and install the Ion OneTouch[™] ES Instrument in a location different from the location used to prepare the amplification solution.
- **2.** Remove the accessories and AC line cords from the box.
- **3.** Place hands *under* the front and rear of the instrument and lift to remove instrument from the carton.

IMPORTANT! Do *not* lift the instrument by holding the syringe located at the back of the instrument.

- **4.** Install the AC line voltage fuse module, see page 30 for more information.
 - **a.** For 120-line voltage: Align the small arrow below the "110–120V" shown on the fuse module with the white arrow on the power entry module.



- (1) Arrows aligned
- **b.** For 220-line voltage: Align the small arrow below the "220–240V" shown on the fuse module with the white arrow on the power entry module.
- **5.** Ensure that the power switch is **Off**, then plug the power cord into the instrument.
- **6.** Plug the other end of the power cord into the surge protector or line conditioner.
- 7. Plug the surge protector or line conditioner into an electrical outlet.

IMPORTANT! The voltage of the electrical outlet must match the voltage of the surge protector and the selected voltage of the installed AC line voltage fuse module in the Ion OneTouch^{TM} ES.

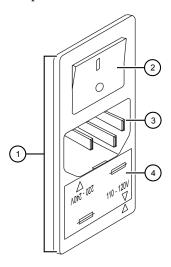


Install the AC line voltage fuse module

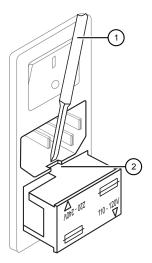
The fuse module must be installed into the power entry module so that the proper voltage is applied to the instrument.

Line voltage	Replacement fuse type
110/120 VAC	375 mA TT (Slow Blow) 1/4" × 1-1/4"
220/240 VAC	160 mA TT (Slow Blow) 5 × 20 mm

1. Turn the power switch to the **Off** position, then disconnect the power cord from the power cable socket.

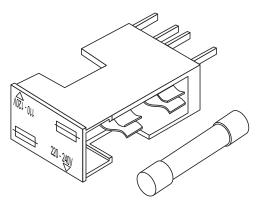


- 1 Power entry module
- 2 Power switch
- 3 Power cable socket
- (4) Fuse drawer
- **2.** Insert a small screw driver (or equivalent) into the slot at the top-middle of the fuse drawer, then pry open the fuse drawer.

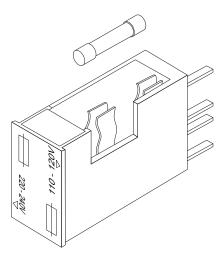


- 1 Small screw driver
- 2 Slot for opening fuse drawer

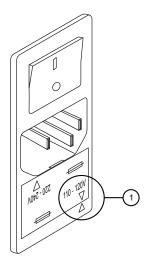
3. Orient the fuse drawer with 220–240V in the bottom-right corner, then install a 375 mA fuse into the clips on the right hand side.



4. Orient the fuse drawer with 110–120V in the top-right corner, then install a 160 mA fuse into the clips.



5. Re-install the fuse drawer oriented for the proper supplied operating voltage.



Fuse drawer aligned for 110–120 VAC service.

Set up the OneTouch[™] **ES Instrument**

Before operating the instrument for the first time, you must perform the residual volume test and calibrate the instrument in both the vertical and horizontal axes.

Materials required for this procedure

Provided in your template preparation solutions:

• Nuclease-free Water

Provided with your template preparation supplies:

- 8-well strip
- Eppendorf[™] LoRetention Dualfilter Tips (P300)

Provided with the Ion OneTouch[™] 2 System (Part no. 4474779):

• Corning Brand 96-Well Strip Ejector (Thermo Fisher Cat. no. 07-200-22), unless 8-well strips are supplied loose and out of the frame

Other Materials and Equipment:

- Elbow fitting for Ion OneTouch[™] ES
- 0.2-mL PCR tubes
- Pipettes
- Vortexer
- (Optional) Felt-tipped pen

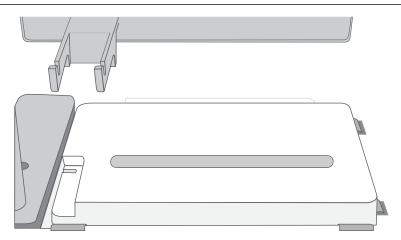
Set up the instrument

Ensure that the power switch is turned **Off** and the AC line voltage fuse module is installed in the proper orientation. See "Unpack and install the Ion OneTouch^{$^{\text{TM}}$} ES Instrument" on page 28.

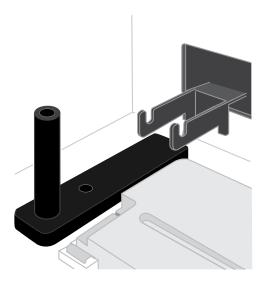
- 1. Locate the Tray, Tip Arm, and Tip Loader of the Ion OneTouch™ ES Instrument.
- **2.** Install the Tray:
 - **a.** Wipe the instrument and the bottom of the Tray with a damp lab wipe to remove any packaging debris.
 - **b.** Place the Tray on the Ion OneTouch[™] ES with the calibration shelf on the left as shown in the following illustration.

c. Push the Tray down firmly so that the Tray fits snugly in the clamps, then confirm that the Tray is level:

IMPORTANT! For proper operation of the Ion OneTouchTM ES, the tray must be firmly and uniformly seated in the cutout for the tray.

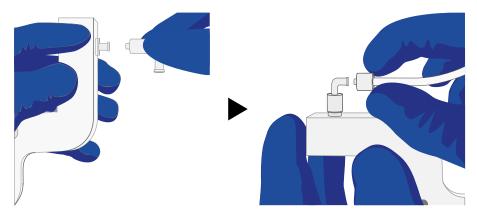


3. Place the Tip Loader in the slot on the left side of the instrument deck:



Set up the OneTouch™ ES Instrument

- **4.** Install the supplied elbow fitting and place the Tip Arm in the cradle.
 - **a.** Insert the male end of the elbow fitting into the female connector on the Tip Arm, then finger tighten the lock ring on the elbow fitting:



- **b.** Connect the tubing to the elbow fitting on the top of the Tip Arm:
- **c.** Finger-tighten the connectors *at both ends* of the elbow.

IMPORTANT! Do not twist or coil the tubing.

d. Tilt the Tip Arm back. Align the pins with the round notches in the cradle, then lower the Tip Arm into position. Move the Tip Arm forward into the working position (see the following illustration):



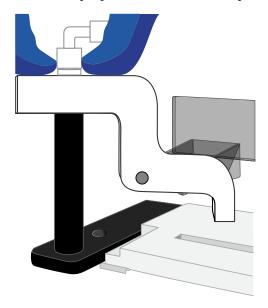
Note: Ensure that the back/bottom end of the Tip Arm is not resting on top of the thumb screw, causing the Tip Arm to tilt forward. Do *not* adjust the angle of the arm unless you need to calibrate the instrument.

5. Turn the power switch **On**. (*Optional*) Leave the instrument **On** continuously during daily and weekly use.

Install a new pipette tip on the tip arm

Before every calibration, residual volume test, or enrichment performed on the Ion OneTouch $^{\text{\tiny TM}}$ ES Instrument, install a new Eppendorf $^{\text{\tiny TM}}$ LoRetention Dualfilter P300 pipette tip.

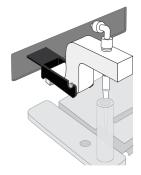
- 1. Place a new tip in the Tip Loader. Remove the Tip Arm from the cradle and align the metal fitting of the Tip Arm with the tip.
- 2. Keeping the fitting on the Tip Arm vertical, firmly press the Tip Arm down onto the new tip until the Tip Arm meets the Tip Loader. Hold the Tip Arm to the Tip Loader for ~1 second to ensure proper installation of the tip.



- 3. Lift the Tip Arm *straight* up to pull the installed tip from the Tip Loader tube.
- **4.** Return the Tip Arm to the cradle (see the following illustration).
 - **a.** Tilt the Tip Arm back (below left) and align the pins with the round notches in the cradle (below middle).
 - **b.** Lower the Tip Arm into position (below center).
 - **c.** Move the Tip Arm forward into the working position (below right).



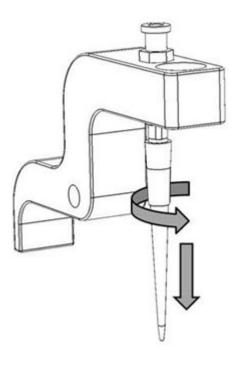




Set up the OneTouch™ ES Instrument

IMPORTANT! Ensure that the back/bottom end of the Tip Arm is not resting on top of the thumb screw, causing the Tip Arm to tilt forward.

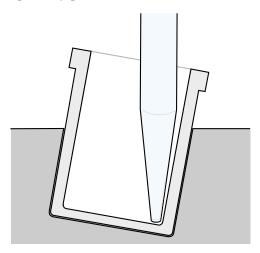
Note: To remove the used tip, keep the Tip Arm in its cradle and while standing above the Tip Arm, twist the tip *counterclockwise* and pull it downward to remove and discard the tip.



IMPORTANT! Improper removal of tips can loosen the metal tip adapter fitting on the Tip Arm and affect instrument operation.

Ion OneTouch[™] ES Instrument calibration

Perform horizontal and vertical calibrations so that during operation the tip is optimally positioned in the well (see the following illustration) of the 8-well strip:



Note that the 8-well strip is always tilted at a fixed 10-degree angle in the slot. The pipette tip is vertical. When the tip is aligned properly during calibration so that it is in line with the notch in the calibration shelf, the tip touches the bottom of the well during the run, close to the front bottom edge of the well.

IMPORTANT! If you use more than one Ion OneTouch[™] ES Instrument, do not switch Trays or Tip Arms between instruments. Each Tray and Tip Arm is calibrated with a particular instrument. To track the Tray and Tip Arm, each component has a printed label with the matching serial number of the instrument.

Calibrate the vertical axis

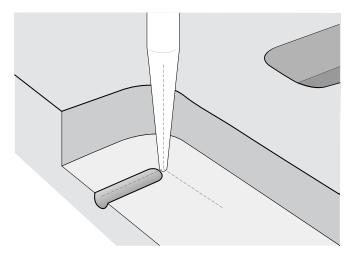
- 1. Install a new tip on the tip arm. See page 35.
- 2. Power Off the instrument, then wait 3 seconds.
- **3.** While holding down **Vert. Adjust**, power **On** the instrument to restore the factory default settings.
- **4.** Power **Off** the instrument.
- **5.** While holding down **Select/Calibrate**, power the instrument **On**. Keep holding down **Select/Calibrate** until "P1" is displayed.
- **6.** Press **Select/Calibrate** for ~3 seconds until the instrument beeps 2 times and "CAL" is displayed to put the instrument into calibration mode.

Note: The instrument will cycle through several values before "CAL" is displayed.

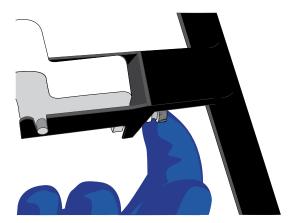
7. Press **Vert. Adjust**. The instrument displays "ASP" (Aspirate or z-bottom position).

Ion OneTouch™ ES Instrument calibration

- **8.** Press **Start/Stop**. The Tip Arm lowers to bring the tip near the notch in the calibration shelf on the left side of the Tray.
- **9.** Adjust if needed the position of the bottom of the pipette tip:



a. To adjust the alignment of the tip with the slot, turn the thumbscrew at the back of the Tip Arm.



b. To adjust the height of the tip, press the (minus) button repeatedly until the tip touches the shelf. Press the (minus) button eight more times to lower the tip further. This will account for variations in tip lengths and installation.

Note: It is better to have the ASP (aspiration) height be too low than too high.

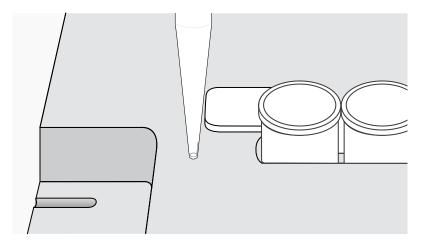
10. Press **Start/Stop**, then wait for the Tip Arm to stop moving and for "P1" to display.

Calibrate the horizontal axis

1. Press **Select/Calibrate** for ~3 seconds until the instrument beeps 2 times and "CAL" is displayed.

Note: The instrument will cycle through several values before "CAL" is displayed.

- 2. Press Horiz. Adjust. Instrument displays "FLA", then press Start/Stop.
- 3. Place an empty 8-well strip in the slot in the Tray, with the square tab on the left.
- **4.** Push the 8-well strip as far to the left in the slot as possible.
- 5. Observe the position of the 8-well strip relative to the position of the tip. When properly calibrated, the 8-well strip is ≤1 mm of touching but not pushing on the tip. To clearly see the relationship between the pipette tip, calibration shelf, and notch during calibration, mark each of them with a felt-tip pen:



- **6.** Adjust the horizontal position of the Tip Arm so that the tip just touches the left tab of the 8-well strip when the 8-well strip is pushed to the far left of the slot in the Tray:
 - a. Apply slight pressure to keep the 8-well strip to the far left.
 - b. Press the \triangle (plus) button repeatedly until the tip touches the 8-well strip. Each press of the \triangle (plus) key moves the Tip Arm to the *right* by ~0.002 inches (~50 μ m), which may be difficult to detect.
- 7. Press **Start/Stop** to save the setting and for "P1" to display.
- **8.** Power the instrument **Off**, wait >3 seconds, then power the instrument **On** to return to normal operating mode.
- **9.** Perform a residual volume test (see "Perform a residual volume test" on page 40).

Perform a residual volume test

If the condition is	Then
First use of the instrument and during monthly maintenance	Perform a residual volume test (see instructions below).
Routine use and residual volume in Well 1 and Well 8 is >5.0 µL	
Routine use and residual volume in Well 1 and Well 8 is ≤5.0 µL	Operate the instrument without performing the residual volume test.

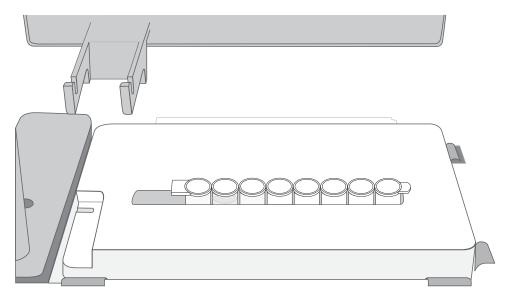
IMPORTANT! Before running the residual volume test, carefully read and familiarize yourself with this procedure. During the test, confirm that the pipette tip is centered between the sides of the wells when moving in or out of a well.

1. Pipet 80 µL water or Ion OneTouch™ Wash solution into the second well (Well 2) from the square-tabbed end of the 8-well strip.



- 1 Square-shaped tab
- (2) Well 2
- (3) Rounded tab

2. Load the 8-well strip into the slot of the Tray so that the square-tabbed end is to the *left* and the 8-well strip is pushed all the way to the right until it touches the end of the slot.



- **3.** Turn the instrument **On**, then wait for the instrument to initialize. The Tip Arm performs a series of movements and returns to the home position (~5 seconds), then "rdy" is displayed on the instrument screen.
- 4. Press Start/Stop.
- **5.** Wait for the instrument to aspirate the solution from Well 2 and completely remove the tip from Well 2, then *manually* push the 8-well strip to the left so that Well 4 is positioned directly under the Tip Arm.
- **6.** Wait for the instrument to dispense the tip contents into Well 4.
- **7.** Press **Start/Stop** to stop the test run, then press **Start/Stop** again to return the Tip Arm to the home position.
- **8.** Using a P10 pipette, aspirate the entire residual water or Ion OneTouch™ Wash Solution from well 2, then estimate the residual volume.
- 9. Remove and discard the used pipette tip and 8-well strip.



Perform a residual volume test

10. After performing the residual volume test, take one or more of the following actions:

Observation	Possible cause	Recommended actions
Residual volume in Well 2 is ≤5 µL	_	No action is necessary.
Residual volume in Well 2 is >5 µL	The tip height is too high during aspiration.	Restore defaults, then calibrate the instrument (see "Ion OneTouch™ ES Instrument calibration" on page 37).
Aspiration is irregular	This instrument out of calibration.	Restore defaults, then calibrate the instrument (see "Ion OneTouch™ ES Instrument calibration" on page 37).
The 8-well strip lifts as the tip rises to the top of the well	The tip is angled too far forward or the tip height is set too low.	Verify that the tip is vertical and positioned directly over the notch in the calibration shelf. If the tip is positioned correctly. Restore defaults, then calibrate the instrument (see "Ion OneTouch™ ES Instrument calibration" on page 37).

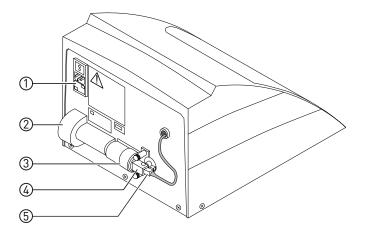


Ion OneTouch[™] ES maintenance

Materials and equipment required

Xiameter[™] PMX-200 Silicone Fluid

Ion OneTouch[™] ES back panel layout



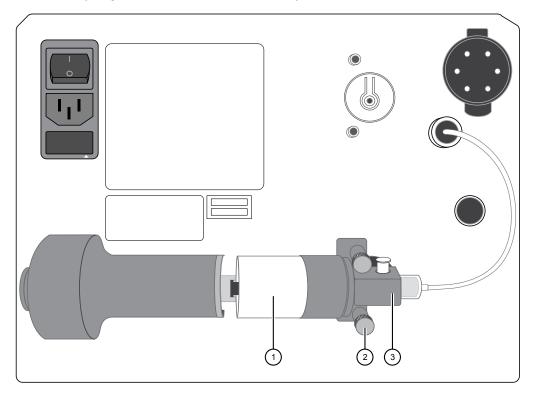
- 1 Fuse module
- ② Syringe pump
- 3 Syringe assembly

- 4 Syringe valve & valve cover
- **5** Tubing connection



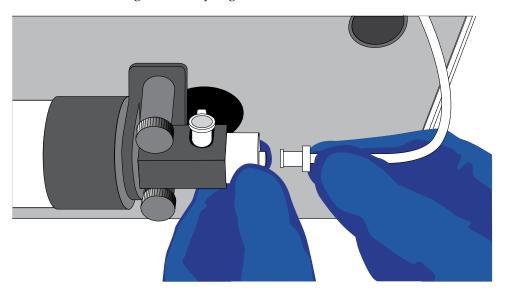
Disassemble and lubricate the syringe

Note: The syringe should be lubricated annually.

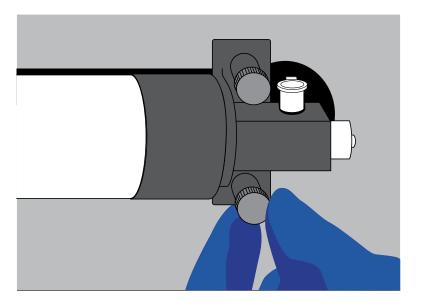


Ion OneTouch[™] ES Instrument rear view.

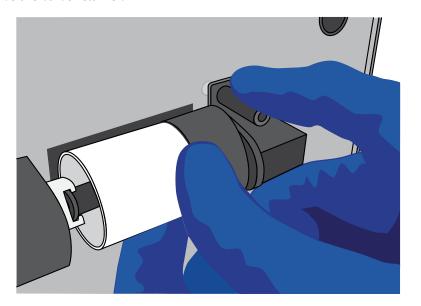
- ① Syringe
- 2 Retainer screw
- 3 Valve retainer
- 1. Disconnect the tubing from the syringe.



2. Remove the 2 retainer screws.

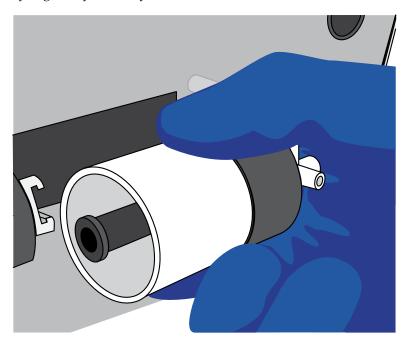


3. Remove the valve retainer.





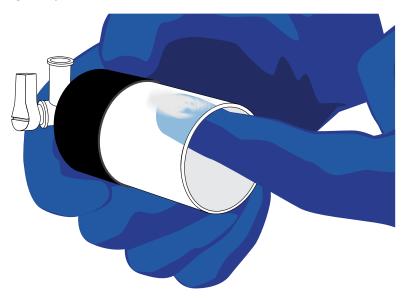
4. Pull the syringe body toward you to remove from the instrument.



5. Remove the plunger from the syringe body.

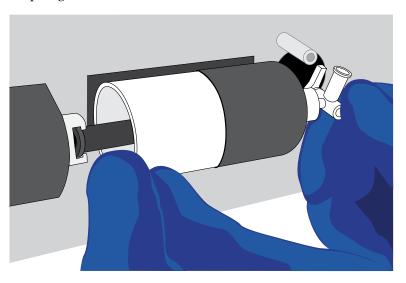


6. Apply a thin layer of Xiameter[™] PMX-200 Silicone Fluid evenly to the inside of the syringe body.

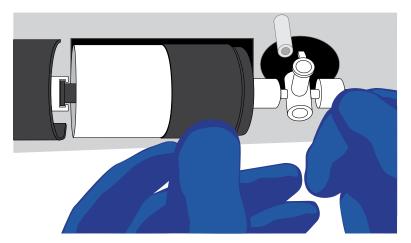


Reassemble the syringe

- 1. Push the plunger all the way into the syringe body, then pull back out approximately ¼ inch.
- **2.** Engage the plunger with its mate end.



3. Insert the valve into its docking position.



- **4.** Replace the retainer and the 2 retainer screws (finger-tighten).
- **5.** Reconnect the tubing.

Ion OneTouch™ ES decontamination

Before returning the instrument for service, decontaminate it according to the procedure below.

Materials and equipment required

- Disposable rubber gloves
- Safety glasses
- Lab coat
- Bleach
- Water
- Paper towels

IMPORTANT! This procedure does not guarantee total decontamination of the Ion OneTouch[™] ES.

Decontaminate the instrument

Wear disposable rubber gloves, safety glasses, and a lab coat.

- 1. Wipe all outside surfaces of the Ion OneTouch™ ES with 10% bleach solution. Avoid getting bleach solution inside the chassis.
- 2. Dry the surfaces of the Ion OneTouch™ ES with paper towels or other disposable wipes.
- **3.** Use cotton swabs to clean and dry areas that are difficult to reach.
- **4.** Properly dispose of used cleaning materials.



Set up and test the Ion Chip™ Minifuge

The Ion Chip[™] Minifuge (Cat. no. 4479672 or 4479673) is used to load sequencing chips for use on Ion PGM[™], Ion Proton[™], and Ion S5[™] sequencing platforms. To accommodate the larger chip size, Ion Proton[™], and Ion S5[™] sequencer users must:

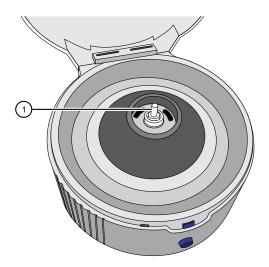
- install the Ion S5[™]/Ion Proton[™] Rotor and Buckets (Cat. no. 4482578).
- test the minifuge to confirm that no liquid is lost during centrifugation.

before using the minifuge to load chips for the first time.

Note: The following protocols may also be used to convert the Ion ChipTM Minifuge back for use with Ion PGMTM sequencing chips.

Install the Ion S5[™] /Ion Proton[™] Rotor and Buckets

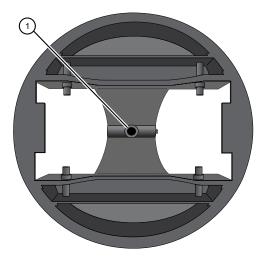
1. Grasp the existing rotor and pull straight up to remove the rotor from the motor shaft.



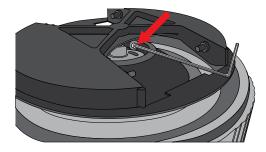
(1) Motor shaft



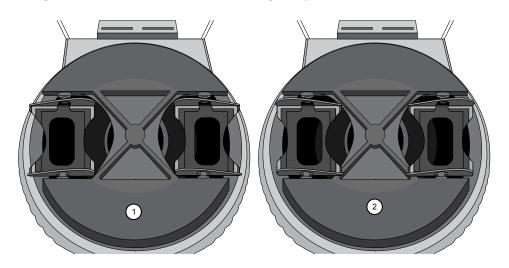
2. Press the Ion S5[™]/Ion Proton[™] Rotor down onto the motor shaft to install.



- (1) Insert motor shaft here
- **3.** Tighten the set screw (arrow) with a 1.5-mm hex wrench.



4. Install the two buckets. Position the buckets with the larger semi-circular cut-outs facing out, and ensure that the buckets hang freely.



- 1 Correct orientation
- 2 Incorrect orientation

Test the minifuge

- 1. Prepare two previously-used chips:
 - a. Inject $100~\mu L$ of isopropanol two times into the loading port of each chip. After each injection, remove the expelled liquid from the opposite port.

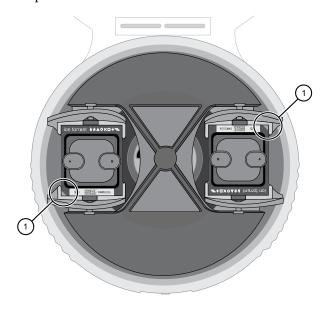
Note: Use 50 μ L volume of isopropanol if testing Ion PGMTM sequencing chips.

b. Aspirate the remaining isopropanol from the flow cells for 5–10 seconds. Confirm that the chips are dry.

Note: To aspirate the isopropanol, attach a P200 pipette tip to a vacuum line, then place the pipette tip in the chip loading port.

2. Place the two chips prepared in step 1 in the centrifuge buckets, with the chip notch pointing out. Add 55 μ L of nuclease-free water to each chip loading well (do not inject into the chip loading port).

Note: Use 35 μL volume of nuclease-free water if testing Ion PGMTM sequencing chips.



1 Chip notch

3. Centrifuge for 5–10 seconds, then examine each chip.

The flow cell in each chip should be completely filled with liquid with no air bubbles. A small volume of liquid will remain in the loading well; this is normal.

Result	Action
The chips are NOT completely filled	Contact Technical Support.
The chips ARE completely filled	Centrifuge the chips for an additional 10 minutes, then check the chips again for air bubbles, especially near the inlet and outlet ports.
The chips have air bubbles after the additional 10 minute centrifugation	Contact Technical Support.
The chips remain completely filled	The centrifuge is ready to use for chip loading.



Demonstrated protocol: Switching between older and newer template preparation kits

IMPORTANT! Thermo Fisher Scientific Demonstrated Protocols have been successfully demonstrated by Thermo Fisher Scientific research and development but not verified. There are no technical specifications for demonstrated protocols. Users assume all risk when using these protocols, and recognize that support for demonstrated protocols occurs through community discussion. All customers are encouraged to discuss and contribute via the Ion Community.

ioncommunity.thermofisher.com

When switching is required

Perform the following procedure when switching between older template preparation kits (Group 1) and newer kits (Group 2). If switching between kits listed within a group there is no requirement to perform this protocol.

Group 1 - Older kits	Group 2 - Newer kits
 Ion PGM[™] Template 0T2 400 Kit (Cat. no. 4479878) Ion PGM[™] Template 0T2 200 Kit (Cat. no. 4480974) 	 All Ion Proton[™] templating kits All Ion 520[™] & 530[™] and Ion 540[™] templating kits All Ion PGM[™] Hi-Q[™] templating kits

Required materials and equipment

- Ion OneTouch[™] 2 Instrument
- Ion OneTouch[™] 2 template preparation kit(s)
- Kimwipes disposable wipes
- 50-mL conical tube
- Tube rack for 50-mL conical tube

Before you begin

- 1. Complete the current run on the Ion OneTouch[™] 2 System.
- 2. Process the ISPs according to the template preparation user guide currently in use.

IMPORTANT! *Save* the following for re-use during the instrument initialization (see "Initialize the Ion OneTouch^{TM} 2 Instrument" on page 55):

- Both Ion OneTouch[™] Recovery Tubes
- Ion OneTouch[™] Router
- Ion OneTouch[™] 2 Amplification Plate
- 3. Obtain the appropriate kit for preparation of templated ISPs for the sequencing system you are switching to. The Ion OneTouch™ Oil and Ion OneTouch™ Recovery Solution in the kit are used to clean the instrument.

Clean the Ion OneTouch[™] 2 Instrument after the last run

IMPORTANT! Ensure that the latest firmware is installed on the Ion OneTouch^{$^{\text{TM}}$} 2 Instrument. See "Checking and updating the firmware on the Ion OneTouch^{$^{\text{TM}}$} 2 Instrument" on page 11. The latest firmware for the Ion OneTouch^{$^{\text{TM}}$} 2 Instrument is required to perform the cleaning procedure with the Ion OneTouch^{$^{\text{TM}}$} 2 Cleaning Adapter (refer to the Options screen on the instrument display).

Note: Visually inspect the rotor assembly and casing periodically to ensure there are no cracks or signs of other physical damage.

1. Discard or keep the following materials from the last run:

Discard	Keep
Reaction Filter Assembly	Both Ion OneTouch [™] Reagent Tubes (Store for re-use with the kit.)
Ion OneTouch™ Oil	Both Ion OneTouch [™] Sippers (Store for re-use with the kit.)
Ion OneTouch [™] Recovery Solution	Ion OneTouch [™] 2 Amplification Plate (Store clamped in the heat block.)

- 2. Install the materials and solutions from the appropriate template preparation kit you are switching to:
 - 2 Ion OneTouch[™] Sippers
 - Ion OneTouch[™] Reagent Tube filled half-full with Ion OneTouch[™] Oil
 - Ion OneTouch[™] Reagent Tube filled a quarter-full with Ion OneTouch[™] Recovery Solution
- **3.** Empty the waste container.

- **4.** Place a 50-mL conical tube in a tube rack, then place the tube rack adjacent to the instrument.
- 5. Firmly insert the 3 ports of a single-use Ion OneTouch™ Cleaning Adapter into the three holes (see page 20) on the top stage of the Ion OneTouch™ 2 Instrument. Ensure the tab protruding from the cleaning adapter fits into the front notch of the stage.
- **6.** Gently pull the disposable tubing downwards on both sides of the pinch valve until the disposable tubing is out of the valve.
- **7.** Remove the disposable injector from the Ion OneTouch[™] DL Injector Hub:
 - **a.** Place one hand on the centrifuge lid.
 - **b.** With the other hand, firmly grip the rigid plastic connector at the top of the disposable injector.
 - **c.** Slowly and steadily withdraw the disposable injector straight from the port of the Injector Hub.



CAUTION! PHYSICAL INJURY HAZARD. The pointed end of the disposable injector can puncture your skin. Keep your hand away from the point of the injector.

- **8.** Place the used, disposable injector into the empty 50-mL conical tube in the tube rack. The conical tube will be used for waste.
- **9.** In the Ion OneTouch[™] 2 Instrument home screen, touch **Clean**.
- **10.** Follow the on-screen prompts, touch **Next** after each. After you touch **Next** on the last task the cleaning begins.
- 11. Ensure that the task "Remove plate, injector, conical tube, and waste" displays at the end of the cleaning run.

IMPORTANT! Keep the used Ion OneTouch^{$^{\text{IM}}$} 2 Amplification Plate and Ion OneTouch^{$^{\text{IM}}$} 2 Cleaning Adapter on the instrument to use for initialization of the instrument. Keep the disposable injector in the 50-mL conical tube to collect waste.

Initialize the Ion OneTouch[™] 2 Instrument

- Ensure that the used Ion OneTouch[™] 2 Amplification Plate and Ion OneTouch[™] 2
 Cleaning Adapter are on the instrument. Keep the disposable injector in
 the 50-mL conical tube.
- 2. In the instrument display, touch **Open Lid**, then open the centrifuge lid.

IMPORTANT! Do *not* lift the lid by the tubing attached to the Ion OneTouch^{TM} DL Injector Hub. Do *not* force the lid open.



- **3.** Insert a *used* Ion OneTouch[™] Recovery Tube into each of the slots in the centrifuge.
- 4. Pinch the sides of the used Ion OneTouch™ Recovery Router then push the router down into the center slot of the centrifuge so that it is seated flat and secure in the center of the rotor.

Note: The Ion OneTouch[™] Recovery Router is intentionally offset from the Ion OneTouch[™] Recovery Tubes.

- **5.** In the home screen, touch **Options**.
- **6.** In the Options screen, touch **Initialize**, then touch **Next** until initialization of the instrument begins. Initialization takes ~10 minutes.

IMPORTANT! For this initialization after cleaning the instrument, do not install the disposable injector in the Ion OneTouch™ DL Injector Hub. Keep the disposable injector in the 50-mL conical tube.

Set up the Ion OneTouch[™] 2 Instrument with the appropriate kit

- After initialization, remove and appropriately discard the following from the instrument:
 - Ion OneTouch[™] 2 Cleaning Adapter
 - Ion OneTouch[™] 2 Amplification Plate, disposable tubing, and injector
 - Both Ion OneTouch™ Recovery Tubes
 - Ion OneTouch™ Router
- 2. In the instrument display, touch **Open Lid**, wait until the lid opens, then lift and hold the side of the centrifuge lid.

IMPORTANT! Do *not* lift the lid by the tubing attached to the Ion OneTouch^{TM} DL Injector Hub. Do *not* force the lid open.

- **3.** Insert two new Ion OneTouch™ Recovery Tubes from the appropriate template preparation kit into the slots of the centrifuge.
- **4.** Pinch the sides of the *new* Ion OneTouch[™] Recovery Router then push the router down into the center slot of the centrifuge so that it is seated flat and secure in the center of the rotor.
- **5.** Insert a *new* Ion OneTouch[™] Amplification Plate from the appropriate template preparation kit.
- **6.** Pull the handle to close the heat block.

Set up the Ion OneTouch $^{\mathtt{m}}$ 2 Instrument with the appropriate kit

- 7. Close the centrifuge lid, and thread the disposable tubing through the catch and pinch valve, then insert the disposable injector in the Ion OneTouch™ DL Injector Hub.
- **8.** Refer to the appropriate template preparation kit user guide to prepare and install the amplification solution. Complete the remaining tasks to run the sample on the Ion OneTouch™ 2 Instrument to prepare template-positive ISPs for sequencing.



Troubleshooting

Ion OneTouch[™] 2 Instrument

Observation	Possible cause	Recommended action
Firmware does not update or the status update screen does not display ≤10 seconds	Firmware not updating on the instrument	 Power the instrument OFF then ON. Ensure that the USB flash drive is FAT32-formatted and that the file is in the root directory. Remove then reinsert the USB flash drive immediately after the main menu displays. Repeat steps 1–3 as needed.
Disposable injector remains in "down" position in the Ion OneTouch™ DL Injector Hub	 Reagent build-up New part 	 Gently pull from the top of the disposable injector until the disposable injector just returns to the "up" position in the Injector Hub (see "Install the disposable injector" on page 18). Briefly press then release the spring-loaded top of the Injector Hub 5-10 times at the point indicated by the arrow (see illustration in "Install the disposable injector" on page 18). You should hear a click. If the Injector Hub remains in the "down" position, repeat step 2 once (up to 10 more clicks). Note: If the Injector Hub still does not move freely and click up into place, then contact
Recovery tubes filled with gel after run Note: This observation and recommended action pertain only to template preparation kits that require the use of Ion OneTouch™ Breaking Solution.	Breaking Solution was not added to Recovery Tubes prior to start of run.	 Technical Support. Add 150 μL Ion OneTouch™ Breaking Solution to each Recovery Tube. Seal the Recovery Tubes with Parafilm™ M film and vigorously invert and vortex the tubes for 10 seconds to break the emulsion. Refer to the user guide of the template preparation kit in use and follow the procedure to "Recover the template-positive ISPs". Alternatively, transfer samples to two 1.5-mL tubes and spin in an Eppendorf™ centrifuge for 10 minutes at 15,500 × g to pellet the ISPs. Refer to the user guide of the template preparation kit in use and proceed to "Wash the template-positive ISPs". Perform the ISP wash in 1 mL nuclease-free water twice.

Observation	Possible cause	Recommended action
Centrifuge lid does not open	Power failureSoftware crash	Slide a 1/8-inch L-wrench (hex wrench) or equivalent tool into the right hole and along the top edge of the centrifuge hinge:
		Push the tool into the hole until there is a slight compression of the tool against the instrument and the centrifuge lid unlocks and opens.
		3. Remove the tool from the hole, then open the lid.
		4. If necessary, troubleshoot the lid lock, then use the instrument normally. Do <i>not</i> force the lid open.
Precipitate in the Reagent Mix	Storage of thawed Reagent	1. Ensure that the solution is fully thawed.
after vortexing	Mix <2°C	2. Vortex the solution for 30 seconds, then leave the tube at room temperature for 15 minutes.
		3. Vortex the solution again at maximum speed for 1 minute.
		4. Centrifuge the tube for 30 seconds.
		5. Visually inspect the solution at the bottom of the tube and verify that there is no residual precipitate. If precipitate is visible, then repeat steps 1–4.
		Note: If precipitate is still visible, then contact Technical Support.
		Keep the Reagent Mix at room temperature during use.
		7. After use, store the solution at 2°C to 8°C.
Excessive oil in the waste tray	Various	Remove the oil waste tray, then appropriately dispose of the oil.
		Reinsert the tray into the slot, then push the tray back fully into the instrument.
		To further troubleshoot excessive oil from the instrument, contact Technical Support.

Ion OneTouch[™] ES

For Ion OneTouchTM ES vertical and horizontal axis calibration and residual volume test procedures, refer to Chapter 3 of the *Ion OneTouch*TM 2 *System User Guide* (Pub. no. MAN0014388).

Observation	Possible cause	Recommended action
Excessive foaming occurs.	Instrument is improperly calibrated resulting in	Use the recommended volumes for all wells.
	inadequate volume in one or more wells.	Ensure that fittings are tight, especially at the elbow fitting, and the pipette tip is not
	Fitting is loose.	cracked.
	Pipette tip is cracked.	If necessary, perform the residual volume test. If the residual volume test fails, then calibrate the instrument.
Brown pellet is present in centrifuged tube of enriched ISPs.	Residual Dynabeads™ MyOne™ Streptavidin C1 Beads are present.	Pipet the suspension with the brown pellet up and down 10 times to resuspend the pellet.
		 Place the 0.2-mL PCR tube against a magnet such as a DynaMag[™]-2 magnet for 4 minutes.
		 Transfer the supernatant with the enriched ISPs to a new 0.2-mL PCR tube without disturbing the pellet of Dynabeads™ MyOne™ Streptavidin C1 Beads.
		4. Sequence or store the enriched ISPs.

Observation	Possible cause	Recommended action
E12, E22, or E23 errors display during the run or during	Calibration values are out of range.	Power OFF the instrument and wait seconds.
calibration.		 While holding down Vert. Adjust, power ON the instrument. This step restores the factory default settings.
		3. Recalibrate the vertical axis:
		Note: The default setting for the vertical axis is 310. If the setting is <310, the instrument will likely display an error, because the Tip Arm position is too high.
		 a. Press the (minus) button to lower the Tip Arm until the tip touches the shelf.
		b. Press the (minus) button 8 more times. Typical vertical axis settings are ~340–370.
		 Recalibrate the horizontal axis: Press the (plus) button to move the Tip Arm to the right until the tip touches the left tab of the strip.
		Note: The default setting for the horizontal axis is 625. Typical horizontal axis settings are ~640–670.
	AC line voltage module is installed incorrectly.	 Determine the voltage of the electrical outlet to plug in the Ion OneTouch™ ES.
		2. Align the arrow by the correct voltage on the AC line voltage module with the adjacent white arrow in the lower-right corner of the fuse socket.
		If the AC line voltage module is installed incorrectly:
		Gently remove the module with your fingernail or a small flathead screwdriver.
		2. Rotate the module so that the correct voltage on the module is aligned and adjacent to the white arrow in the lower right-hand corner of the fuse socket.
		Insert the AC line voltage module into the fuse socket.

Observation	Possible cause	Recommended action
E12 or E22 error is displayed when the unit is initializing. Either of the following: E12 or E22 errors are displayed. Tip Arm does not move or moves slightly.	 Fuse is installed incorrectly. Unit is below operating temperature. Program or calibration setting is bad, or Tip Arm is not moving. AC line voltage module is installed incorrectly. Instrument is not at the	 Ensure that the fuse module is installed correctly and that the unit is at its recommended operating temperature. Reboot the instrument: Power OFF the instrument, wait 3 seconds, then power ON the instrument. If the error persists, restore the factory defaults, then re-calibrate the instrument: Power OFF the instrument and wait 3 seconds. While holding down Vert. Adjust, power ON the instrument. This step restores the factory default settings. Repeat 3a-3b as needed to restore the factory defaults. Calibrate the vertical and horizontal axes. Determine the voltage of the electrical outlet serving the Ion OneTouch™ ES. Align the arrow by the correct voltage on the AC line voltage fuse module with the adjacent white arrow in the lower-right corner of the fuse socket. Gently remove the module with your fingernail or a small flathead screwdriver. Rotate the module so that the correct voltage on the module is aligned and adjacent to the white arrow in the lower right-hand corner of the fuse socket. Insert the AC line voltage fuse module into the fuse socket. Insert the AC line voltage fuse module into the fuse socket.
	recommended operating temperature	operating temperature of 60°F to 77°F (15°C to 25°C).
Solution overflows during run.	Reagent volumes are overloaded.	Repeat with reagent volumes described in enrichment procedure.
Tip is causing 8-well strip to lift out of tray slot during run.	Tip is not aligned vertically.	Perform the vertical calibration procedure.
Percent template-positive ISPs after enrichment is <50% as measured by flow cytometry.	Multiple causes are possible.	Contact Technical Support.

Observation	Possible cause	Recommended action
One of the following: Strip lifts up during strip push. Strip lifts up when tip is raised from well. Immediately after strip push, the strip is not in contact with the magnet.	Instrument is not calibrated properly.	 Perform horizontal calibration. Perform vertical calibration.
Tip grinds into base of instrument and Code "1999" displays.	 Unit is not calibrated properly. Vertical calibration setting is too low or out-of-range. 	 Restore the factory default settings on the instrument: Hold down the vertical adjust button while powering ON the instrument. The instrument beeps several times. Re-calibrate the instrument. Perform a residual volume test.
Tip hits the top of the tray at start of run.	Tray is not properly seated in the instrument.	Check for debris between the tray and the instrument, then reinstall the tray. Press down firmly to ensure that tray is fully seated in the instrument.
Error messages display.	Various causes are possible.	 Power the instrument OFF, then ON. If the error continues to display, restore the factory default settings on the instrument. Hold down the vertical adjust button while powering ON the instrument. The instrument beeps several times. Re-calibrate the instrument. Perform a residual volume test.
Instrument does not aspirate or dispense liquids.	Fitting(s) are loose.	 Ensure that the Luer-Lok™ connections at the elbow on the Tip Arm and at the tubing on the rear syringe pump are finger-tight. Ensure that the metal tip adapter fitting on the Tip Arm is finger-tight. IMPORTANT! After any adjustments to the metal tip adapter, recalibrate the lon OneTouch™ ES.

Safety





WARNING! GENERAL SAFETY. Using this product in a manner not specified in the user documentation may result in personal injury or damage to the instrument or device. Ensure that anyone using this product has received instructions in general safety practices for laboratories and the safety information provided in this document.

- Before using an instrument or device, read and understand the safety information provided in the user documentation provided by the manufacturer of the instrument or device.
- Before handling chemicals, read and understand all applicable Safety Data Sheets (SDSs) and use appropriate personal protective equipment (gloves, gowns, eye protection, etc). To obtain SDSs, see the "Documentation and Support" section in this document.

Symbols on this instrument

Symbols may be found on the instrument to warn against potential hazards or convey important safety information. In this document, the hazard symbol is used along with one of the following user attention words:

- CAUTION! Indicates a potentially hazardous situation that, if not avoided, may result in minor or moderate injury. It may also be used to alert against unsafe practices.
- WARNING! Indicates a potentially hazardous situation that, if not avoided, could result in death or serious injury.
- DANGER! Indicates an imminently hazardous situation that, if not avoided, will result in death or serious injury.

Symbol	English	Français
	Caution, risk of danger Consult the manual for further safety information.	Attention, risque de danger Consulter le manuel pour d'autres renseignements de sécurité.
(1)	Protective conductor terminal (main ground)	Borne de conducteur de protection (mise à la terre principale)

Symbol	English	Français
	Do not dispose of this product in unsorted municipal waste CAUTION! To minimize negative environmental impact from disposal of electronic waste, do not dispose of electronic waste in unsorted municipal waste. Follow local municipal waste ordinances for proper disposal provision and contact customer service for information about responsible disposal options.	Ne pas éliminer ce produit avec les déchets usuels non soumis au tri sélectif. CAUTION! Pour minimiser les conséquences négatives sur l'environnement à la suite de l'élimination de déchets électroniques, ne pas éliminer ce déchet électronique avec les déchets usuels non soumis au tri sélectif. Se conformer aux ordonnances locales sur les déchets municipaux pour les dispositions d'élimination et communiquer avec le service à la clientèle pour des renseignements sur les options d'élimination responsable.

Conformity symbols on this instrument

Conformity mark	English	Français
c America US	Indicates conformity with safety requirements for Canada and U.S.A.	Indique la conformité avec les normes de sécurité en vigueur au Canada et aux États-Unis.
C€	Indicates conformity with European Union requirements for safety and electromagnetic compatibility.	Indique la conformité avec les exigences de l'Union européenne en matière de sécurité et de compatibilité électromagnétique.
	Indicates conformity with Australian standards for electromagnetic compatibility.	Indique la conformité avec les normes australiennes régissant la compatibilité électromagnétique.

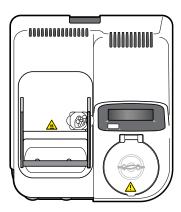
Safety alerts on this instrument

Additional text may be used with one of the symbols described above when more specific information is needed to avoid exposure to a hazard. See the following table for safety alerts found on the instrument.

English		French translation
1	CAUTION! Hazardous chemicals. Read the Safety Data Sheets (SDSs) before handling.	ATTENTION! Produits chimiques dangereux. Lire les fiches signalétiques (FS) avant de manipuler les produits.
<u></u>	CAUTION! Hazardous waste. Refer to SDS(s) and local regulations for handling and disposal.	ATTENTION! Déchets dangereux. Lire les fiches signalétiques (FS) et la réglementation locale associées à la manipulation et à l'élimination des déchets.

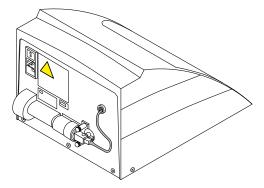
Location of safety labels on the instrument

The Ion OneTouch[™] 2 Instrument has warnings at the indicated locations:



Top view.

The Ion OneTouch[™] ES Instrument has warnings at the indicated locations:



Rear view.



Safety information for instruments not manufactured by Thermo Fisher Scientific

Some of the accessories provided as part of the instrument system are not designed or built by Thermo Fisher Scientific. Consult the manufacturer's documentation for the information needed for the safe use of these products.

Instrument safety

General



CAUTION! Do not remove instrument protective covers. If you remove the protective instrument panels or disable interlock devices, you may be exposed to serious hazards including, but not limited to, severe electrical shock, laser exposure, crushing, or chemical exposure.

Electrical



WARNING! Ensure appropriate electrical supply. For safe operation of the instrument:

- Plug the system into a properly grounded receptacle with adequate current capacity.
- Ensure the electrical supply is of suitable voltage.
- Never operate the instrument with the ground disconnected. Grounding continuity is required for safe operation of the instrument.



WARNING! Power Supply Line Cords. Use properly configured and approved line cords for the power supply in your facility.



WARNING! Disconnecting Power. To fully disconnect power either detach or unplug the power cord, positioning the instrument such that the power cord is accessible.

Cleaning and decontamination



CAUTION! Cleaning and Decontamination. Use only the cleaning and decontamination methods specified in the manufacturer's user documentation. It is the responsibility of the operator (or other responsible person) to ensure the following requirements are met:

- No decontamination or cleaning agents are used that could cause a HAZARD as a result of a reaction with parts of the equipment or with material contained in the equipment.
- The instrument is properly decontaminated a) if hazardous material is spilled onto or into the equipment, and/or b) prior to having the instrument serviced at your facility or sending the instrument for repair, maintenance, trade-in, disposal, or termination of a loan (decontamination forms may be requested from customer service).
- Before using any cleaning or decontamination methods (except those recommended by the manufacturer), users should confirm with the manufacturer that the proposed method will not damage the equipment.

Safety and electromagnetic compatibility (EMC) standards

The instrument design and manufacture of the Ion OneTouch^{$^{\text{TM}}$} 2 Instrument and Ion OneTouch^{$^{\text{TM}}$} ES Instrument complies with the standards and requirements for safety and electromagnetic compatibility as noted in the following tables:

Safety

Reference	Description			
Ion OneTouch [™] 2 Instrument				
EU Directive 2006/95/EC	European Union "Low Voltage Directive"			
IEC 61010-1	Safety requirements for electrical equipment for measurement, control, and laboratory use – Part 1: General requirements			
EN 61010-1				
UL 61010-1				
CSA C22.2 No. 61010-1				
IEC 61010-2-010	Safety requirements for electrical equipment for measurement,			
EN 61010-2-010	control and laboratory use – Part 2-010: Particular requirements for laboratory equipment for the heating of materials			
IEC/EN 61010-2-020	Safety requirements for electrical equipment for measurement, control and laboratory use – Part 2-020: Particular requirements for laboratory centrifuges			
Ion OneTouch [™] ES Instrument				
EU Directive 2006/95/EC	European Union "Low Voltage Directive"			
IEC 61010-1	Safety requirements for electrical equipment for measurement,			
EN 61010-1	control, and laboratory use – Part 1: General requirements			
UL 61010-1				
CSA C22.2 No. 61010-1				

Environmental design

Reference	Description
	European Union "WEEE Directive" – Waste electrical and electronic equipment
Directive 2011/65/EU	European Union "RoHS Directive" – Restriction of hazardous substances in electrical and electronic equipment

EMC

Reference	Description
Directive 2004/108/EC	European Union "EMC Directive"
EN 61326-1	Electrical Equipment for Measurement, Control and Laboratory Use – EMC Requirements – Part 1: General Requirements
AS/NZS CISPR 22 2009+A1 2010	Limits and Methods of Measurement of Electromagnetic Disturbance Characteristics of Industrial, Scientific, and Medical (ISM) Radiofrequency Equipment
ICES-003, Issue 5	Industrial, Scientific and Medical (ISM) Radio Frequency Generators
FCC Part 15 Subpart B (47 CFR)	U.S. Standard Radio Frequency Devices

Chemical safety



WARNING! GENERAL CHEMICAL HANDLING. To minimize hazards, ensure laboratory personnel read and practice the general safety guidelines for chemical usage, storage, and waste provided below, and consult the relevant SDS for specific precautions and instructions:

- Read and understand the Safety Data Sheets (SDSs) provided by the chemical manufacturer before you store, handle, or work with any chemicals or hazardous materials. To obtain SDSs, see the "Documentation and Support" section in this document.
- Minimize contact with chemicals. Wear appropriate personal protective equipment when handling chemicals (for example, safety glasses, gloves, or protective clothing).
- Minimize the inhalation of chemicals. Do not leave chemical containers open. Use only with adequate ventilation (for example, fume hood).
- Check regularly for chemical leaks or spills. If a leak or spill occurs, follow the manufacturer's cleanup procedures as recommended in the SDS.
- · Handle chemical wastes in a fume hood.
- Ensure use of primary and secondary waste containers. (A primary waste container holds the immediate waste. A secondary container contains spills or leaks from the primary container. Both containers must be compatible with the waste material and meet federal, state, and local requirements for container storage.)
- After emptying a waste container, seal it with the cap provided.
- Characterize (by analysis if necessary) the waste generated by the particular applications, reagents, and substrates used in your laboratory.
- Ensure that the waste is stored, transferred, transported, and disposed of according to all local, state/provincial, and/or national regulations.
- **IMPORTANT!** Radioactive or biohazardous materials may require special handling, and disposal limitations may apply.

Documentation and support

Customer and technical support

Visit **thermofisher.com/support** for the latest in services and support, including:

- Worldwide contact telephone numbers
- Product support, including:
 - Product FAQs
 - Software, patches, and updates
- Order and web support
- Product documentation, including:
 - User guides, manuals, and protocols
 - Certificates of Analysis
 - Safety Data Sheets (SDSs; also known as MSDSs)

Note: For SDSs for reagents and chemicals from other manufacturers, contact the manufacturer.

Limited product warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at www.thermofisher.com/us/en/home/global/terms-and-conditions.html. If you have any questions, please contact Life Technologies at www.thermofisher.com/support.

