

INSTRUCTION MANUAL

NANOLITER2020 Injector With MICRO2T SMARTouch™ Controller

Serial No._____

www.wpiinc.com

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ABOUT THIS MANUAL

The following symbols are used in this guide:



This symbol indicates a CAUTION. Cautions warn against actions that can cause damage to equipment. Please read these carefully.

This symbol indicates a WARNING. Warnings alert you to actions that can cause personal injury or pose a physical threat. Please read these carefully.

NOTES and TIPS contain helpful information.



Fig. 1—Nanoliter 2020 setup. Micromanipulator is not included. The 300704 Injector head can be purchased separately or with the MICRO2T controller.

INTRODUCTION

The **NANOLITER2020** Injector works on the principle of positive displacement. An internal micro-step motor precisely moves the metal (stainless steel) plunger (also referred to as the *piston*) which pushes the oil inside the micropipette, and the oil layer pushes the aqueous sample, dispensing specific nanoliter volumes ranging from 6-4200 nL at selectable delivery rates. Target injection volumes and rates can be easily adjusted using the **MICRO2T** SMARTouch[™] controller. Fine control of plunger displacement, along with proper sealing in the gasket, glass micropipette and oil, ensures precision and accuracy.

Notes and Warnings

CAUTION: Do NOT apply solvents or oils to any part of the **NANOLITER2020**. Do NOT wash or lubricate any parts of the system.

CAUTION: The **NANOLITER2020** Injector Head (**3004704**), **MICRO2T** controller, and other part are not autoclavable. Do not autoclave.

CAUTION: Sterilize the **NANOLITER2020** Injector Head with EtO or by wiping the exterior of the pumps and controller with alcohol or Cidex (WPI# **7364**).

CAUTION: Do not disassemble unless it is discussed in the manual. There are no serviceable parts inside either the **NANOLITER2020** Injector Head (**300704**) or the **MICRO2T** controller.

CAUTION: Always hold the **NANOLITER2020** Injector Head (**300704**) by the main body or the mounting bar. Do not swing it or carry it by its cable.

Parts List

After unpacking, verify that there is no visible damage to the instrument. Verify that all items are included:

(1) **300704 NANOLITER2020** Injector head. (The injector comes with gaskets to use with 1.1-1.15 mm glass installed.)

Items listed below are mandatory accessories already included with the **NANOLITER2020** system that may be purchased separately. The **300704** is just the injector head and does not include any controller or accessories.

(1) MICRO2T SMARTouch[™] Controller with 12V Power Supply and Power Cable for 110V/220V

- (1) 501981 Tweezers to scoop out the gaskets
- (1) 504949 Glass Capillaries (ID: 0.530 mm, OD: 1.14 mm) pkg. of 300
- (2) **TIP10XV119** Micropipettes (ID: 0.530 mm, OD: 1.14 mm, Tip ID: 10 µm)
- (1) 300746 Spare Parts Kit which includes:
 - (1) 14456 Allen wrench (0.035") hex tool for replacing wire plunger
 - (1) MF34G MicroFil non-metallic filling needle, 34 gauge (used for backfilling)
 - (5) 300733 O-ring Kit for the NANOLITER2020
 - (1) 3563 1 CC Syringe
 - (2) 300514 Replacement Pistons for the NANOLITER2020

This Instruction Manual for the **NANOLITER2020** can be downloaded at <u>https://www.wpiinc.com/manuals</u>.

Unpacking

Upon receipt of this instrument, make a thorough inspection of the contents and check for possible damage. Missing cartons or obvious damage to cartons should be noted on the delivery receipt before signing. Concealed damage should be reported at once to the carrier and an inspection requested. Please read the section entitled "Claims and Returns" on page 39 of this manual. Please contact WPI Customer Service if any parts are missing at (941) 371-1003 or customerservice@wpiinc.com.

Returns: Do not return any goods to WPI without obtaining prior approval (RMA # required) and instructions from WPI's Returns Department. Goods returned (unauthorized) by collect freight may be refused. If a return shipment is necessary, use the original container, if possible. If the original container is not available, use a suitable substitute that is rigid and of adequate size. Wrap the instrument in paper or plastic surrounded with at least 100 mm (four inches) of shock absorbing material. For further details, please read the section entitled "Claims and Returns" on page 39 of this manual.

INSTRUMENT DESCRIPTION

NANOLITER2020 Description

The NANOLITER2020 is the system which includes NANOLITER2020 Injector Head (300704), MICRO2T SmartTouch[™] Controller and the required accessories. Two sample micropipettes are included with the NANOLITER2020 (WPI #TIP10XV119) and a pack (300) glass capillaries (WPI #504949).

- Additional 1.14 mm OD fire-polished glass micropipettes can be purchased from WPI or glass capillaries can be pulled using a puller (e.g., **PUL-1000**).
- 1.14 mm OD fire polished glass is to be used with green color gasket.

CAUTION: Never attempt to use micropipettes pulled from glass containing a filament. Injection volumes will not be accurate.

- NANOLITER2020 has compatibility with 1.3-1.35 mm and 1.5 mm OD fire-polished glass capillaries by using black colored and red colored front gasket, respectively.
 NOTE: Always use fire-polished glass. Make sure to select glass capillary or pipettes matching the front gasket.
- Colorless and odorless mineral oil (to be used for back-filling glass micropipettes) is not included.
- The injector head can be mounted on a micromanipulator (commonly used) or on a stereotaxic frame. WPI's M3301 micromanipulator (not included with a NANOLITER2020 purchase) is a popular among NANOLITER users.



Fig. 2—NANOLITER 2020 System can be used on a micromanipulator which is NOT included with the NANOLITER2020.

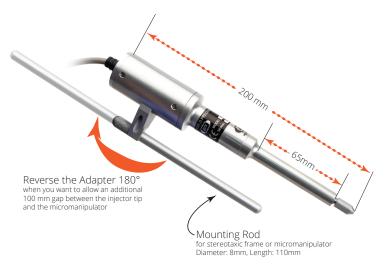


Fig. 3—NANOLITER2020 Injector Head (300704) dimensions.

Power/Run LED Indicator on Nanoliter Injector Head

When the **NANOLITER2020** is connected to the **MICRO2T** controller, a dim red light illuminates. The light turns bright red when the *RUN* button is pressed and the **NANOLITER2020** pump is running.







Off - Unit is unplugged.

Dim - Unit has power.

Bright - Unit is running.

Fig. 4—The LED indicator on the NANOLITER2020 Injector Head indicates proper communication with the MICRO2T Controller.

MICRO2T Description

The MICRO2T is used to control one or two NANOLITER2020 or UMP3 pumps.



Fig. 5—The MICRO2T touch screen controller shows the command screen.

Pump Information Display Showing Two Pumps



Pump Information Display

Fig. 6—*The Command screen provides pump information and has buttons to control the pumps.*

Selected Pump-Touch one of the pump data displays in the Pump Information Display area to select a pump. The selected pump may be controlled using the command buttons or setup using the *Configure* button.

Command Buttons–The three command buttons are used to control the selected pump (or pumps if they are grouped).

Run starts the programmed sequence for the selected pump. The syringe delivery sequence is established through the Configuration screen. While the pump is running, the border of the pump's information display flashes to indicate which pump is running. As soon as you press *Run*, the button changes to *Stop* and the *Direction* button changes to *Pause* (Fig. 7). To pause a running delivery sequence, press *Pause*. The button changes to *Resume*, and the *Stop* button changes to *Resume* button and the delivery sequence will finish the programmed volume delivery. The ongoing delivery can be paused as many times as needed with no limitation until the delivery is complete. To stop a delivery and reset the program, press *Reset*. The volume will be reset to zero and the unit will be ready for a new run.



Fig. 7—*When the Run button is pressed, it changes to Stop and the Direction button changes to Pause.*

- **Direction** toggles the direction of the selected pump between infuse and withdraw. When the direction button is pressed, the selected pump's information display updates with the new direction of travel. A yellow arrow pointing toward the needle end of the syringe indicates infusion. A red arrow in the opposite direction indicates withdrawal.
- Manual gives you direct control of the selected pump. Press Manual to command the selected pump to travel in the direction and rate configured. When you release the button, the pump stops.

Configuration Menu–Press *Configure* to access the Configuration screen for the selected pump. Parameters which may be set include the volume target, counter mode, delivery rate, selected syringe, motor drive mode, units of delivery and grouping status. (See "Defining Pump Parameters" on page 14.)

Pump Information Display–This area shows information about each pump connected to the controller.



Fig. 8—The pump display area shows vital information for each pump attached to the controller.

Command screen (Fig. 8) shows:

- *Pump Number* is located in the upper left corner.
- *Grouped Status*. When grouped, a G appears next to the target volume. If it is not grouped, the G disappears.
- Target Volume for dispensing (nL)
- Counter shows the volume dispensed or volume remaining in the syringe (nL).
- Rate (nL/sec or nL/min)
- Infuse or Withdraw Mode
- *Counter Mode* is the delivered volume or volume remaining to be delivered.
- *Volume Counter* (indicator) shows the live occurrence of volume changes as the pump is withdraws or infuses.
- Delivered Volume (nL)
- *Mode.* Grouped, Non-grouped, Disabled is spelled out more clearly on the 2-channel display.
- *Time* (hr:mm:sec) indicates the elapsed time the pump has run.
- A diagram of the syringe shows the percentage of volume in the syringe.

NOTE: When pumps are not grouped, they can be operated independently. While one pump is running, the remaining pump can be made active and operated. The controls change to reflect the state of the active pump.

Back Panel of MICRO2T

The back of the **MICRO2T** has all the electrical connection ports and the power switch.



Fig. 9—The rear panel of the MICRO2T controller has the power switch and the connection ports for power, pumps, foot switch and computer control via USB.

Setting Up the System

Cleaning the NANOLITER2020

NOTE: The **NANOLITER2020** injector Head can be cleaned by removing the collet, O-rings and spacers and wiping them with alcohol.

CAUTION: Do NOT soak the NANOLITER2020 parts in liquid. Do NOT autoclave the NANOLITER2020 injector or controller.

The control box may be cleaned by wiping it with a clean, damp cloth.

Setting up the Injector

The **NANOLITER2020** Injector Head by default is configured with a green front gasket and two o-rings. Follow steps 1 and 2 to replace the gaskets.

1. **Select the appropriate O-rings** – The Sealing O-ring and the Plastic Sleeve (spacer) are always used. The O-ring used depends on the outer diameter of your glass micropipette.



Fig. 10—Choose appropriate gaskets based on the size of the glass used.

NOTE: **TIP10XV119** (1.14 mm OD micropipette) or 504949 (1.14 mm OD glass capillary) is recommended for use with the Green Front Gasket. **TIP10XV119** and 504949 are included with the **NANOLITER2020** system. Fire-polished glass needs to be used for any Front Gasket.

2. **Install the O-rings on the plunger** – First, slide the sealing O-ring (black disc shape gasket) on the plunger with the large hole facing towards the barrel of the pump and the smaller hole facing toward the micropipette. Then, slide the white plastic sleeve (spacer) on the plunger with the flat surface side facing towards the barrel of the pump and the and concave surface facing towards the micropipette.



Fig. 11—Install the O-rings on the plunger.

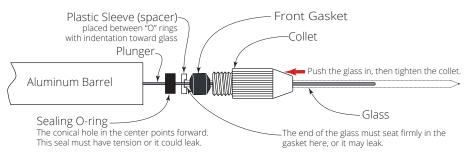


Fig. 12—The front gasket can be installed in either direction.

NOTE: It is essential that these components (O-ring/gaskets) be installed in the correct orientation to hold the glass micropipette in place and proper functionality of **NANOLITER2020** Injector.

Replacing the Gaskets

To change the gaskets, unscrew the collet and slide the collet assembly off of the wire plunger. Unscrew the aluminum barrel from the Nanoliter injector. The O-ring should slide easily from the collet (Fig. 13).



Fig. 13—(Left)The injector is disassembled to replace the gaskets. Fig. 14— (Right) Use the tweezers to remove the gasket from the aluminum barrel.

When you slide the barrel over the plunger without the collet assembly, the sealing O-ring (black) and the plastic sleeve (white) should come off. If not, use a small pair of curved forceps to scoop them out of the end of the barrel (Fig. 14). To reassemble the unit, slide the barrel over the wire plunger and screw it in place on the injector. DO NOT bend the plunger. Position the Sealing O-ring and the Plastic Sleeve as described above. Install the appropriate O-ring on a fresh micropipette and screw the collet in place. See "Setting up the Injector" on page 9.

Connecting the NANOLITER2020 Injector with the Controller

1. Plug the **NANOLITER2020** cable into one of the *Output* connection ports on the rear panel of the SMARTouch[™] controller (Fig. 15). Make sure the marked arrow on the connector is facing upwards for proper pin alignment. The controller allows one or two pumps to be controlled independently or simultaneously.



- Fig. 15—Plug the NANOLITER2020 cable into one of the Output connection ports.
- 2. Connect the AC power supply unit to power port on the rear panel of the **MICRO2T** controller (Fig. 16).



Fig. 16—*Connect the power supply to the power port on the controller.*

NOTE: The switchable power supply included with the controller automatically senses input line voltage between 100 and 240 V and converts it to 12 V.

3. Connect the power cord to the power supply, and plug the AC power cord into a wall outlet (Fig. 17).



- *Fig.* 17—*Connect the power cord to the power supply.*
- 4. If the foot switch (WPI# **13142**, not included) is needed, plug it into the foot switch port on the rear panel of the **MICRO2T** controller (Fig. 9).
- 5. Press the power button on the rear panel of the **MICRO2T** SMARTouch[™] controller to power on the unit. Verify that the LCD screen is illuminated.

IMPORTANT NOTE: Before operating the **NANOLITER20202** pump (**300704**), you must set the end of travel limits of the pump. See "Calibrating the MICRO2T System Travel Limits" on page 15.

OPERATING INSTRUCTIONS

Setting Parameters

NOTE: All parameters are stored in memory, even when the controller is powered off.

Accessing the Configuration Screen

1. Select the appropriate pump channel on the *Command* screen by tapping the desired channel on the display.



Fig. 18—*Channel* 1 *is selected, and PUMP1 is shown with a blue field behind it in the upper right corner of the display screen.*

2. Press *Configure* to access the Configuration screen for your pump.



Fig. 19—The Configuration screen allows you to set the limits of travel and define parameters for a selected pump.

Defining Syringe Parameters

The syringe parameters are set on the Configuration screen for your selected pump.

1. To access the *Configuration* screen, select a pump by tapping on the display from the *Command* screen. Then, click *Configure* for your selected pump. See "Accessing the Configuration Screen" on page 12.

| Volume Target | Actual Delivery | Delivery Rate |
|---------------|-----------------|---------------|
| 0.0 nL | 0.0 nL | 10.0 nL/Sec |

Fig. 20—The syringe parameters are shown across the top of the Configuration screen.

- Volume Target shows the volume in nanoliters that the pump is set to deliver. Whenever a parameter is changed, the controller calculates the actual volume that can be delivered. It is not always possible to deliver the exact desired volume because of the limits of discrete motor steps.
- Actual Delivery is read only and not directly editable. It is not always possible to deliver the exact target volume. The unit calculates the actual delivery volume to be as close as possible to the target volume based on the selected step microstepping and syringe size. This value changes to reflect the effects of changes to the target volume, syringe type and motor drive selection.
- **Delivery Rate** shows the rate at which the volume is dispensed by the pump. The unit for this parameter can be set to nL/sec or nL/min. The units are set by pressing the corresponding radio button on the *Configuration* screen. The units are updated on the text box as the unit is selected.

Four white navigation buttons in the center of the *Syringe Stop Definition* screen (Withdraw, Rapid Withdraw, Rapid Infuse and Infuse) let you inject or withdraw the pump at two different rates, fast or slow.



Fig. 21—The Syringe Stop Definition screen buttons let you inject or withdraw at different speeds.

2. To update the volume target or the delivery rate, tap the text field. A keypad appears with 0.0 in the text field. Use the keypad to enter the desired value.



Fig. 22—Use the keypad to change the Volume Target. A similar keypad appears when you press the Volume Counter or the Delivery Rate fields.

3. Press *Enter* to store the new value or press *Back* to return to the *Configuration* screen without saving any changes.

TIP: If you enter an erroneous value, click *Delete* to clear it. Then, use the keypad to enter a new value.

Defining Pump Parameters

The pump parameters define how the selected pump responds. These are set on the *Configuration* screen for the selected pump.

1. To access the *Configuration* screen, select a pump by tapping on the display from the *Command* screen. Then, click *Configure* for your selected pump. See "Accessing the Configuration Screen" on page 12.



Fig. 23—The bottom left corner of the Configuration window shows pump parameters that may be set.

 Touch the control to select the Motor Drive. The control toggle between Smooth and Max Load. In order to simplify and to guarantee that the desired rate is achieved, the controller has two options to determine the amount of microstepping needed. The actual microstepping ratio is dependent on the selected syringe and the desired delivery rate.

Motor Drive lets you set the pump to deliver the aliquot volume using the minimum number of microsteps or the maximum number of microsteps that are possible for the selected delivery rate.

- *Smooth* yields the smoothest delivery for the selected rate. It sets the pump to deliver the desired rate with the maximum number of microsteps. Depending on the rate and the syringe geometry, this ratio can change between 256 microsteps per step to full stepping
- Max Load yields a more pulsatile flow with a better force delivery. It allows the pump to deliver at the desired rate with the fewest number of microsteps per step. Depending on the rate and the syringe geometry, this ratio can change from full stepping to 256 microsteps per step.

| Smooth Delivery | Max Load |
|---|--|
| Smooth out undesirable pressure pulses when injecting very small volumes or for injections over extended periods of time. This option is quieter and has reduced vibration. | More forceful delivery, as much as 15– 30% increase over <i>Smooth</i> delivery. Improved precision. |
| When undesirable pressure pulsations are observed during injects at a low rate, use a smooth delivery. | |

- 3. **Volume Counter** determines how the volume counter is affected when volume is delivered. Tap the control to toggle between *Delivered* and *Remaining*.
 - When the *Volume Counter* slider is set to *Delivered*, the volume counter increments when injecting and decrements when withdrawing. The volume count shown is the amount delivered.
 - When *Run* is pressed the counter resets to zero and increments. When the slider is set to *Remaining*, the volume counter decrements when injecting and increments when withdrawing. When *Run* is pressed the counter is set to the volume target and decrements. Negative values signify a withdrawal and positive indicate an infusion.
- 4. Tap on a round button to select the desire units. Choose between nL/sec and nL/min for the delivery rate.
- 5. Tap on a round button to select your mode. Mode buttons determine the pump operating mode.
 - *Disabled*–The selected pump is disabled and will not operate. When you return to the Command screen, DISABLED appears over the selected channel.
 - Grouped–You can start and stop all grouped pumps simultaneously. See "Grouped Operations" on page 31. When Grouped is selected, both pumps (if present) are set to grouped mode.
 - Non-Grouped–Non-grouped pumps operate independently.

Calibrating the MICRO2T System Travel Limits

The **MICRO2T** monitors the location of the **NANOLITER2020** plunger. It is important to define the limits of travel before you begin using the pump. This keeps the pump from driving past the mechanical limits of the pump and avoids any jamming issues. When you install a pump on a new channel, you must set the stop positions so the controller will properly monitor the pump. Usually this only needs to be done once. But, if the pump is moved to another channel or the pump, you will need to reset the end stops.

1. **End Stop**: The *End Stop* calibration procedure sets the limit for the maximum distance that the pump's plunger carrier can travel in the withdrawn direction. This setting prevents the motor from forcing the plunger carrier to the mechanical limit of the drive mechanism for an extended period of time, in the event the pump is left running unattended. This prevents the possibility of binding the lead screw which can cause the pump to lock up. The ideal location for the *End Stop* limit is just a fraction of a millimeter before the maximum mechanical limit is reacted.

The calibration procedure is simple and involves using the controller to manually position the **NANOLITER2020** into the fully withdrawn position by pressing a button. When the *END STOP* button is released, the controller automatically performs the calibration. The calibration should be checked:

- Every time a pump is newly connected to a MICRO2T controller
- Any time a pump is switched to a new channel that was not previously calibrated.
- Any time a pump is stalled due to excessive load or other accidental misuse.

Once the End Stop Calibration is performed, it is not necessary to do it again, even if the pump is disconnected from the controller as long as it is re-connected to the same channel on the same controller.



CAUTION: IF YOU SUSPECT THAT THE PUMP HAS STALLED, CHECK THE END STOP CALIBRATION AGAIN.

2. Set Syringe: This button is not used for the NANOLITER2020 Injector.

Setting End Stop in the Nanoliter Injector

1. Power on the **MICRO2T** unit. The splash screen appears. Touch the screen anywhere. The *MAIN MENU* appears.



Fig. 24—The Splash Screen appears on power up.

2. On the Main menu, touch the *DISPLAY ALL CHANNELS* button. The *Syringe Status* screen appears.

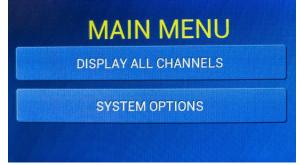


Fig. 25—The Main menu allows access to the pump information and system setup options.

Press anywhere in the PUMP1 display area to make PUMP1 the active pump.
NOTE: Make sure the NANOLITER2020 is plugged into Port 1 of the MICRO2T controller.



Fig. 26—To make a pump active, press anywhere in the PUMP Display Area on the Syringe Status screen.

4. Press the *Configure* button to open the *Pump Configuration* screen.



Fig. 27—Press Configure.

5. Press the Selected Syringe (Type) box to bring up the Syringe Selection screen.



Fig. 28—Press the Selected Syringe area to open the Syringe Selection screen.

6. On the *Syringe Selection* screen, press the *DWN* key until you see the Type NL, 4.5 uL, 25.0 mm option. Tap the NL line to select it. It is then highlighted.



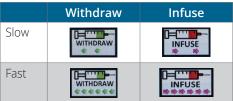
Fig. 29—Select the NL option to highlight it.

- 7. Press the BACK button to return to the Pump Configuration screen..
- 8. Press the RESET POS button to open the Syringe Stop Definition screen.



Fig. 30—*The Syringe Stop Definition screen lets you define the limits of travel for the pump.*

Four, white navigation buttons in the center of the Syringe Stop Definition screen (Withdraw, Rapid Withdraw, Rapid Infuse and Infuse) let you Inject or withdraw the pump at two different rates. (Fast and Slow).



 Use the navigation buttons to set the position of the plunger at the base of the NANOLITER2020 collet. The plunger should extend 1-2 mm outside of the collet opening.

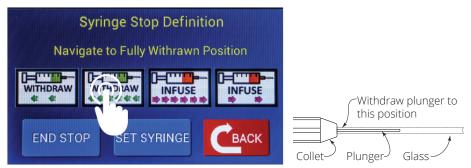


Fig. 31—(Left) Use the four white navigation buttons to set the position of the plunger. Fig. 32—(Right) Using the navigation buttons, withdraw the plunger until you see it reach its withdrawn position at the base of the collet where the arrow is pointing in this image.

10. When you see that the plunger is properly positioned at the base of the collet (1-2 mm outside the opening of the collet), press *END STOP*. The travel limits are stored.



Fig. 33—Press the End Stop button.

11. Press *Back* to return to the *Configuration* screen. Press *Back* again to return to the *Command* screen.



Fig. 34—The Command screen displays.

Micropipette Filling Techniques Back Filling

"Back filling" refers to the process of filling the pipette from the large, non-pulled end. Back filling offers a great advantage when performing a bulk fill, because the large opening allows you to use a syringe and **MicroFil** flexible needles.

- 1. Immerse a Luer syringe into the filling liquid.
- 2. Pull the liquid into the syringe. Attach a MicroFil needle onto the syringe.
- 3. Using the 34G **MicroFil** included, place the **MicroFil** needle into the micropipette from the back side so that the needle's tip is pushed all the way into the front part of the pipette near the pulled end.
- 4. Eject the liquid into the micropipette and slowly withdraw the **MicroFil** needle as the liquid fills the micropipette. Back fill the entire length of a glass micropipette. Be careful to prevent bubbles. Wipe any excess filling liquid with a Kimwipe.

NOTE: This is how mineral oil is back-filled into glass micropipettes.

Front Filling

"Front filling" describes the filling of a micropipette through the small, pulled front end of the pipette. Front filling is achieved by creating a vacuum at the back end of the pipette that pulls the filling liquid into the micropipette. Once the micropipette is installed on the **NANOLITER2020** Injector Head, it is possible to front fill. Front filling is advantageous for small volumes of expensive samples.

Typical Filling Technique

The technique described below is a typical process for filling a micropipette with a small volume of liquid for microinjection applications.

- 1. Back fill the micropipette with mineral oil.
- Use the MICRO2T controller to eject the plunger from the home position. This is necessary in order to allow the plunger to move backwards to create a vacuum for front filling.
- 3. Install the micropipette in the **NANOLITER2020** Injector. Then, submerge the micropipette tip into the sample.
- 4. Pull the sample through the tip of the micropipette by moving the plunger of the **NANOLITER2020** Injector in the withdraw mode.

Installing Oil Back Filled Micropipettes in NANOLITER2020 Injector Head

1. **Back fill the micropipette** – Using a 34G **MicroFil**, back fill the entire length of a glass micropipette with mineral oil. Be careful to prevent bubbles. Wipe any excess oil with a Kimwipe.



Fig. 35—MicroFil is a thin flexible needle that lets you quickly back fill a micropipette.

2. Install the back filled micropipette on to the NANOLITER2020 Injector – First unscrew the collet. Slide the blunt end of the micropipette through the tip of the collet. Then, push the blunt end of the micropipette through the appropriate O-ring (Fig. 36). Line up the wire plunger with the micropette and slide the micropipette over the plunger (Fig. 37). As you push the micropette in, feel it go through the large O-ring and seat in the white spacer. The glass end must be firmly seated in the plastic sleeve (spacer), or it could leak. It is absolutely essential that these components be properly assembled.



Fig. 36—(Left) Slide the blunt end of the micropipette through the tip of the collet and push it through the appropriate sized O-ring.

Fig. 37—(*Right*) *Line up the micropipette with the wire plunger and slide it into place.*

Select the front gasket based on the outer diameter of the glass micropipette.

| Front Gasket Color | Green | Black | Red |
|-----------------------|----------|----------|-----|
| Pipette Diameter (mm) | 1.1-1.15 | 1.3-1.35 | 1.5 |

NOTE: First, the white spacer has one flat side and one side with a concave surface. This concave surface receives the back end of the pipette and protects the plunger sealing O-ring from damage. Second, the black sealing O-ring has a small hole on one side and a larger hole on the other. The larger hole must be positioned facing the injector, and the smaller hole faces towards the white spacer disc. The cylindrical shaped front gasket goes on top of the glass (end of the glass micropipette) in any orientation. Once positioned, tighten the collet securely "finger tight." Pull on the micropipette (with your fingers) to verify that it is securely seated. Micropipette does not come out when you pull gently.



Fig. 38—Micropipette does not come out when you pull gently

NOTE: THE NANOLITER2020 WILL NOT OPERATE PROPERLY WITHOUT BACKFILLING THE MICROPIPETTE.

Front Filling Sample and Injecting Sample

1. With the oil back filled micropipette installed onto the **NANOLITER** Injector Head, press the *CONFIGURE* button to access the *Pump Configuration* screen. Set the *Volume Target* and the *Delivery Rate* by touching each field and entering the desired value.

| Volume Target | Actual Delivery | Delivery Rate |
|---------------|-----------------|---------------|
| 0.0 nL | 0.0 nL | 10.0 nL/Sec |

Fig. 39—Touch the appropriate field the change the value.

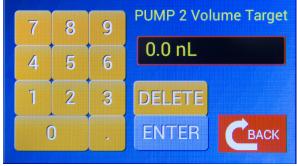


Fig. 40—When you press a target field, a keypad appears. Enter the value and press BACK. (Example Parameters might be: Delivery Rate = 200 nL/sec, Volume Target = 3000 nL, Motor Drive = MAX LOAD)

2. Press the *BACK* button. The image of the micropipette on the controller screen should have a red arrow pointing towards the tip, indicating that the pump is infusing. If not, press the *DIRECTION* button to set the pump to infuse.



Fig. 41—The arrow indicates the direction of the pump.

NOTE: The forward arrow shows the injector running in infusion mode to eject oil and create space inside the micropipette.

- 3. Press the *RUN* button to eject a volume of oil.
- 4. The micropipette tip can be touched very gently and carefully with a Kimwipe to absorb any remaining oil from the tip.

CAUTION: Tip of the micropipette is extremely delicate and can get damaged easily.

5. Next place the micropipette tip into the fluid to be loaded. Make sure the tip is immersed in the liquid, or gas bubbles will collect in the sample.

NOTE: The presence of bubbles inside the micropipettes can result in an inaccurate volume dispensed.

6. Press the *DIRECTION* button to set the pump to withdraw. The image of the micropipette on the controller screen should have a green arrow pointing away from the tip.



Fig. 42—The arrow indicates the pump is withdrawing. (Example Parameters: Delivery Rate = 200 nL/sec, Volume Target = 3000 nL, Motor Drive= MAX LOAD)

7. Press the *RUN* button. This will draw fluid (front fill) into the micropipette.

NOTE: The backward arrow shows the injector is running in withdrawal mode to fill up micropipettes through the tip (front fill).

- 8. The micropipette is ready to inject into target cell or tissue.
- 9. Press the *DIRECTION* button to set the pump to infuse. Now the micropipette can be placed at the desired injection location and after setting the desired Volume Target and Delivery Rate, make an injection by pressing the *RUN* button.

NOTE: The first injection after front filling may be less accurate due to the internal pump's mechanical configuration change from reverse to forward motion. We recommend performing a blank injection right after front filling. (For example, inject >15 nL and discard the volume.) All subsequent injections will be accurate while the injector continues to run in the same direction (infuse/inject).

NOTE: The **MICRO2T** *Motor Drive Mode* of Max Load is recommended for use with the **NANOLITER2020** Injector.

MAINTENANCE

Replacing O-Rings

After a period of time, you may observe some leakage around the seals. To correct this, replace the O-rings. Replacement O-rings are included with your **NANOLITER2020**. Additional O-rings (WPI **#300746**) may also be ordered. Fig. 44 shows the proper installation configurations. Correct orientation of the O-rings is critical for proper operation.

- 1. Unscrew the collet and slide the collet assembly off of the wire plunger. Unscrew the aluminum barrel from the Nanoliter injector. The O-ring should slide easily from the collet.
- 2. When you slide the barrel over the plunger without the collet assembly, the sealing O-ring (black) and the plastic sleeve (white) should come off. If not, use a small pair of curved forceps to scoop them out of the end of the barrel. DO NOT bend the plunger.
- 3. Discard the old O-rings. Slide the sealing O-ring (black disc shape gasket) on the plunger first with the large hole facing towards the barrel of the pump and the smaller hole facing toward the micropipette.
- 4. Then, slide the white plastic sleeve (spacer) on the plunger with the flat surface side facing towards the barrel of the pump and the and concave surface facing towards the micropipette.



Fig. 43—Install the O-rings on the plunger. To reassemble the unit, slide the barrel over the wire plunger and screw it in place on the injector.

5. Install the appropriate O-ring on a fresh micropipette and screw the collet in place. See "Setting up the Injector" on page 9.

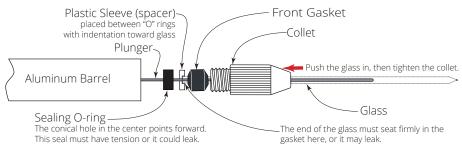


Fig. 44—The front gasket can be installed in either direction.

NOTE: It is essential that these components (O-ring/gaskets) be installed in the correct orientation to hold the glass micropipette in place and proper functionality of **NANOLITER2020** Injector.

Replacing the Wire Plunger

- 1. Verify that the plunger is in the home (retracted) position, just inside the collet. Do not try to retract it too much.
- 2. Hold the motor housing with one hand while loosening the aluminum barrel. To remove it, unscrew it in a counter-clockwise direction with your other hand.



Fig. 45—Unscrew the barrel from the Injector Head.



Fig. 46—The barrel is removed to expose the plunger.

CAUTION: Do NOT disassemble the Motor Housing. Doing so may damage the plunger drive assembly.

3. Using the 0.035" hex wrench provided with the replacement plunger, loosen the two set screws.



Fig. 47—Loosen the set screws with the hex wrench.

- 4. Remove the plunger.
- 5. Insert the new plunger and tighten the set screws snugly. Do NOT overtighten. The set screws must be flush with the edge, otherwise the barrel will not seat properly.

NOTE: The pointed end of the plunger is normally oriented out.

6. Slide the barrel over the tip of the plunger. Hold the handle so the plunger is point straight up. Gently shake the unit until the barrel hole lines up with the plunger and slides down over it. Carefully position the O-rings and collet on the tip of the plunger.

CAUTION: DO NOT FORCE THE ALUMINUM BARREL OVER THE PLUNGER. The plunger is easily bent.

7. Simply turn the cylinder (barrel) clockwise with gentle pressure to re-tighten it. Do NOT over-tighten it!



Fig. 48—The NANOLITER2020 is reassembled.

Cleaning Recommendations

The injector can be cleaned by removing the collet, O-rings and spacers and wiping them with alcohol.

CAUTION: Do NOT soak the **Nanoliter 2020** parts in liquid. Do NOT autoclave the **Nanoliter 2020** injector or controller.

The control box may be cleaned by wiping it with a clean, damp cloth.

ACCESSORIES

| Part Numb | per Description |
|------------|---|
| 300746 | Spare Parts Kit (includes MicroFil™ MF34G , displacement plunger, five |
| | O-ring sets, tools) |
| 504949 | Replacement 3.5-in. glass capillaries (300) |
| 504950 | Replacement 7-in. glass capillaries (300) |
| TIP10XV119 | Pre-pulled Micropipettes for Nanoliter Injector (10) |
| 13142 | Foot switch for MICRO2T controller |
| MF34G-5 | MicroFil™, 34g (pkg. of 5) |
| MICRO2T | SMARTouch [™] Programmable, Touch Screen Controller, 2-Channel |
| 300704 | NANOLITER2020 Injector Head only |

TROUBLESHOOTING

| Issue | Possible Cause | Solution |
|--|---|---|
| Plunger does not travel | The channel is improperly pro- grammed. | Test the pump in another channel, with the same program parameters. |
| Pump display is blank | There is a loose connection on a pump that is plugged in. | Look for a loose connector at the rear of the MICRO2T , make sure the NANOLI- TER Injector Head plug is firmly seated. The gray plastic plug should fit flush with the connector on the controller. Verify that the pins in the connector are not damaged. |
| Pump does not move when commanded | The selected pump is disabled or absent. | Make sure that the intended syringe is selected on the <i>Command</i> screen by tapping the appropriate pump display area. |
| Pump c move comm | The end of travel in the selected direction will exceed the stops. | Set the direction away from the end of travel if the syringe is at the stop. |
| Plunger fails to retract completely | The home position is set incorrectly. | Reset the End Stop. See "Setting End Stop in the Nanoliter Injector" on page 16. |
| s off the ctor head) | The front O-ring seal is worn out. | Replace the O-ring seals. Follow the correct configuration. See "Replacing O-Rings" on page 24. |
| Glass pipette pushes off the ump (Nanoliter Injector heac | The wire plunger is bent enough to add friction to the glass, which pushes it off. | Examine the plunger. If it is bent, replace the wire plunger. See "Replacing the Wire Plunger" on page 25. |
| Glass pipette pushes off the pump (Nanoliter Injector head) | The O-ring seal is soaked in oil and tightening the collet is not enough to secure the glass. | Wash the front O-ring in alcohol and clean it with a Kimwipe or paper towel before placing it on the glass pipette shank. |

NOTE: If you have a problem/issue with that falls outside the definitions of this troubleshooting section, contact the WPI Technical Support team at (941) 371-1003 or technicalsupport@wpiinc.com.

SPECIFICATIONS

NANOLITER2020 Injector

This unit conforms to the following specifications:

| NANOLITER 2020 Plunger Outer Diameter | 482 μm |
|---|----------------|
| Plunger Movement for 100 nL Volume Dispense | 550 µm ± 55 µm |
| Piston Movement per dispensed volume (nL) | 5.5 μm/nL |
| Linear Travel Per Full Step | 12.7 µm/step |
| Maximum Possible Volume | |
| Maximum Rate | 644 nL/sec |

Minimum Recommended Volume Injection

25 nL with 1.14 OD glass (fire polished glass and green front gasket)50 nL with 1.5 mm OD glass (fire polished glass and red front gasket)

Recommended Glass

1.14 mm OD Fire polished glass capillaries: **504949** and **504950**1.14 mm OD Fire polished Pre-pulled micropipettes: **TIP10XV119**

Glass Use Capabilities

1.10-1.15 mm OD Fire Polished Glass with Green Front gasket 1.30-1.35 mm OD Fire-polished Glass with Black Front Gasket 1.5 mm OD Fire-polished Glass with Red Front Gasket

MICRO2T Controller

APPENDIX A: MICRO2T CONFIGURATIONS

Selecting System Options

The *System Options* screen gives you immediate access to basic parameters for your operations. To access the *System Options* screen:

- Select System Options from the main menu, or
- From the Pump Configuration screen, press Options.



Fig. 49—The System Options screen allows you to set basic system parameters for operations. It also lets you revert to the factory default setup.

NOTE: Most parameters are stored in memory, even when the controller is powered off. However, when the unit is powered on, *Remote Access* is always disabled, and the pump drive motors are always enabled.

Disabling Sound Feedback

To disable all audio feedback from the **MICRO2T**, unselect the *Sound Enabled* check box. To re-enable the controller sounds, select the *Sound Enabled* check box. Press *Back* to save the setting and bring up the *Command* screen.

Enabling Remote (Computer) Access

Remote control of the **MICRO2T** is available through the USB port on the back of the controller using a terminal access program on a computer. Before the controller can receive commands from a computer, remote access must be enabled. By default, this is disabled. To enable remote access:

- 1. Select the *Remote Access* check box.
- 2. Press Back to save the setting and bring up the Main Menu.
- 3. Press *Display All Channels* to bring the *Channel Data Display*. Remote commands are only accepted while the unit is on this command screen.

Remote access is always disabled when the **MICRO2T** is powered up. If you power the unit off, you will need to reset this parameter in order to operate with remote access. See "Appendix B: MICRO2T Computer Control" on page 33.

Disabling Motors for Low Noise Recording

For applications where a low electrical noise environment is necessary, you may turn off the drive electronics to the motors until you need to use them. When you return to any other screen, the motors automatically re-enable themselves.

To disable the drive motors on the **NANOLITER2020** pumps, select the *Low Noise Recording* check box. When you press the Back button to return to the previous screen, a warning appears.



Fig. 50—Warning message appears to let you know that the motors will be re-engaged and the Low Noise Recording period will end.

Press *Yes* to re-enable the motors and return to the *Command* screen. Press *No* to remain on the System Options page without enabling the motors.

The drive motors are always enabled when the **MICRO2T** is powered up. The only time the motors are disabled is when the *Low Noise Recording* check box is selected and the *System Options* screen is displayed.

Setting Screen Brightness

You may adjust the brightness intensity of the unit's display by using the *Brightness* slider. The unit will always power up with the brightness set to maximum.

Resetting System Defaults

To reset the factory default settings, press *Defaults*. A message appears to confirm that you want to load the default settings.



Fig. 51—Warning message appears indicating that you are about to reload the factory default settings.

Click *OK* to reload the factory defaults or *Cancel* to exit without loading the defaults. Press *Back* to bring up the *Main Menu*.

Displaying Firmware Revision

Press *About* to display the current firmware version running on the instrument. The *About* button appears in the upper right corner of the System Options screen. See Fig. 49.

Resetting Limits after Power Failure

The **MICRO2T** records the location of all syringes when they stop. The information is recalled the next time the **MICRO2T** powers up. This allows the instrument to resume operation from the previous state without need to re-enter the syringe's position. If the **MICRO2T** is powered down while a volume delivery is taking place, the location at the time where power is removed is not recorded. However, the **MICRO2T** detects that this occurred and reminds you the next time the instrument is turned on. When you tap the startup screen to begin operation, the warning appears.



Fig. 52—This warning displays if the controller loses power while a pump is running.

Press *OK* to continue operation. The message indicates which pump or pumps require attention. Reset the end stop and syringe limits for the displayed pump or pumps. Until the limits are reset, the message continues to display on power up.

To reset the end stop, see "Calibrating the MICRO2T System Travel Limits" on page 15.

Grouped Operations

Pumps may be grouped so that they start and stop together. Commanding any grouped pump to *Run* initiates all the grouped pumps. However, the pump that is highlighted when the run command is issued becomes the master pump. The other pump is a slave.

- If the master pump reaches a mechanical limit before the delivery sequence completes, then the master and the slave pump stop. A short, high pitch tone is emitted when a mechanical limit is reached.
- If a slave pump reaches a mechanical limit, that slave pump stops, and the master pump continues to run until it completes its sequence or until the master pump reaches a mechanical limit.
- 1. Complete the operational preparations for each pump that will be grouped. Be sure to set one of the pumps to *Grouped* so that they can run simultaneously. Grouped pumps are marked on the *Command* screen.



Grouped pumps designation

Fig. 53—Grouped pumps are indicated on the Command screen.

2. When you finish setting up the pump and syringe parameters, press *Back* to return to the *Command* screen.



Fig. 54—Pump 2 is the Master pump, and Pump 1 is the slave in this example.

3. Press *Run* to infuse the required volume of fluid for the injection or for multiple injections. Both pumps will begin delivery of their sequences. Each pump will complete delivery of its sequence unless it reaches a mechanical limit or the master pump reaches a mechanical limit. If the master pump reaches a limit, all grouped pumps still infusing will stop.

NOTE: When pumps are not grouped, they can be operated independently. While one pump is running, the remaining pump can be made active and operated. The controls change to reflect the state of the active pump.

APPENDIX B: MICRO2T COMPUTER CONTROL

Serial commands are used to control the **MICRO2T** via the serial port of a computer using a free USB port.

Serial Commands

All commands are case sensitive. The settings for the serial port are 9600 baud rate, 8 data bits, 1 start bit and 1 stop bit. Numbers and decimal points are indicated below by the "#" symbol. Enter a carriage return (*Enter* key on the keyboard) after each command.

| Command | Syntax | Notes |
|---|--|--|
| Set target volume | V ######## | ######## - Desired Volume Target in nL. Value can be entered with or without decimal point (Range: 0 to 999999.9) |
| Set delivery rate | R##################################### | ######## - Desired Delivery Rate in nL/ sec or nL/min. Units are set with S and M commands. Value can be entered with or without decimal point (Range: 0 to 999999.9) |
| Set active pump to infuse direction | 1 | |
| Set active pump to withdraw direction | W | |
| Start delivery from stopped condition or resume from paused | G | |
| Halt volume delivery (STOP) | Н | Cancels delivery in process |
| Pause delivery | U | Pauses delivery in process |
| Set delivery units to nL/sec | S | |
| Set delivery units to nL/min | M | |
| Set active pump | L# | # is between 1 and 2 |
| Set mode to non-grouped | N | |
| Set mode to grouped | Р | |
| Set mode to disabled | D | |
| Select syringe | T# | # Is between 1 and 12. 1–9 for types 1–9, 10–12 for A, B and C. |
| Set motor drive to max load | BT | |
| Set motor drive to smooth | BS | |
| Set volume counter mode to Delivered | EN | |
| Set volume counter mode to Remaining | EI | |
| Display target volume | ?V | Response: Target Volume = 10000.0nL OK |
| Display volume counter | ?C | Response: Volume Counter = 1000.0µL OK |
| Display motor drive option | ?В | Response: Smooth Drive (Max Load Drive) OK |
| Display motor counter mode | ?E | Response: Delivered Volume (Remaining Volume) OK |
| Display pump mode | ?M | Response: Mode: Non-Grouped (Mode: Grouped, Mode: Disabled) OK |

| Command | Syntax | Notes |
|-------------------------|--------|--|
| Display syringe type | ?S | Response: Type 8, 100.0uL, 60.0 OK |
| Display direction type | ?D | Response: Direction: Infuse (Direction: Withdraw) OK |
| Display rate units | ?∪ | Response: Rate Units: nL/min (Rate Units: nL/sec) OK |
| Display run mode | ?G | Response: Motor State: Stopped (Motor State: Running, Motor State: Paused) OK |
| Pause script execution | A#### | #### is length of pause in seconds/100 Response: PAUSING (while in pause) OK (when finished pause) |
| Веер | F#### | #### is length of beep in seconds/100 Response: BEEP (while beeping) OK (when finished beeping) |
| Kill command | Z | Ends current command and cancels any pending commands |
| Blocking start delivery | *G | Same as G command, but it stops accepting any new commands until the commanded delivery is finishd at which point it responds with OK. This is useful when host computer wants to ensure delivery is finished before commanding another action. |

APPENDIX C: FREQUENTLY ASKED QUESTIONS

What is the advantage of using oil back-filled glass micropipettes?

"Back filling" refers to the process of filling the pipette from the large, non-pulled end. "Front filling" describes filling a micropipette through the small, pulled front end of the pipette. The glass micropipettes are first backfilled completely with mineral oil and secured to the **NANOLITER** Injector Head. Then, some mineral oil is dispensed through the tip. This creates the space and generates the pressure to front fill samples through the tip. Front filling sample prevents the spillage or loss of costly or scarce sample involved with back filling samples. When the sample volume is low, first back filling glass micropipettes with oil and then front filling the sample may be the only option.

Can I use WPI's old standard controller to run the 300704 Injector head?

WPI **NANOLITER2020** Injector Head (**300704**) needs the **MICRO2T** controller (recommended). WPI's **MICRO4** controller may be used to control the pump. The **300704** Injector Head cannot be used with WPI's old standard controller.

Can I use same MICRO2T controller that I use with UMP3 pump? Do I need an additional adapter to use the 300704 pump with my existing MICRO2T controller?

Yes, the same **MICRO2T** controller can be used to control the **UMP3** pump and the **NANOLITER2020**. No additional adapter is needed. The **300704** connects directly to **MICRO2T** controller.

Can I run the pump using a foot switch?

Yes, you can use the **13142** Foot switch. This is not included in the **NANOLITER2020** system and is sold separately.

What do I do when glass micropipettes come off after attaching or I am unable to front fill a sample?

Odorless and colorless mineral oil is used for back filling glass micropipettes. While the glass micropipette is installed and with gradual spillage over multiple uses, the gaskets become too slippery to hold glass micropipette in place. Use Kimwipes to soak up oil spillage inside the injector head, outside the glass micropipettes and to clean the gaskets. If needed, install a new set of gaskets to resolve the issue.

I followed the instructions manual and I still have questions using the NANOLITER2020 system. What should I do?

Please contact WPI Technical Support team at (941) 371-1003 or email technicalsupport@ wpiinc.com.

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DECLARATION OF CONFORMITY



WORLD PRECISION INSTRUMENTS, LLC. Telephone: (941) 371-1003 Fax: (941) 377-5428 e-mail <u>wpi@wpiinc.com</u>

DECLARATION OF CONFORMITY CE

We:

World Precision Instruments, Inc. 175 Sarasota Center Boulevard Sarasota, FL 34240-9258, USA

as the manufacturer/distributor of the apparatus listed, declare under sole responsibility that the product(s):

NANOLITER 2020

To which this declaration relates is/are in conformity with the following standards or other normative documents:

Low Voltage Directive (Safety) 2014/35/EU:

• EN 61010-1:2010+A1:2019

EMC Directive 2014/30/EU:

- EN IEC 61326-1:2021
- EN IEC 61326-2-3:2021
- EN IEC 61000-3-2:2019+A1:2021
- EN IEC 61000-3-3:2013+A2:2021

Issued On: December 13, 2022

Cory Boyes / Director of Design and Development Europe Representative Mr Andrew Waldes Managing Director World Precision Instruments Germany GmbH, Pfingstweide 16, 61169 Friedberg, Germany

WARRANTY

WPI (World Precision Instruments) warrants to the original purchaser that this equipment, including its components and parts, shall be free from defects in material and workmanship for a period of one year* from the date of receipt. WPI's obligation under this warranty shall be limited to repair or replacement, at WPI's option, of the equipment or defective components or parts upon receipt thereof f.o.b. WPI, Sarasota, Florida U.S.A. Return of a repaired instrument shall be f.o.b. Sarasota.

The above warranty is contingent upon normal usage and does not cover products which have been modified without WPI's approval or which have been subjected to unusual physical or electrical stress or on which the original identification marks have been removed or altered. The above warranty will not apply if adjustment, repair or parts replacement is required because of accident, neglect, misuse, failure of electric power, air conditioning, humidity control, or causes other than normal and ordinary usage.

To the extent that any of its equipment is furnished by a manufacturer other than WPI, the foregoing warranty shall be applicable only to the extent of the warranty furnished by such other manufacturer. This warranty will not apply to appearance terms, such as knobs, handles, dials or the like.

WPI makes no warranty of any kind, express or implied or statutory, including without limitation any warranties of merchantability and/or fitness for a particular purpose. WPI shall not be liable for any damages, whether direct, indirect, special or consequential arising from a failure of this product to operate in the manner desired by the user. WPI shall not be liable for any damage to data or property that may be caused directly or indirectly by use of this product.

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Inspect all shipments upon receipt. Missing cartons or obvious damage to cartons should be noted on the delivery receipt before signing. Concealed loss or damage should be reported at once to the carrier and an inspection requested. All claims for shortage or damage must be made within ten (10) days after receipt of shipment. Claims for lost shipments must be made within thirty (30) days of receipt of invoice or other notification of shipment. Please save damaged or pilfered cartons until claim is settled. In some instances, photographic documentation may be required. Some items are time-sensitive; WPI assumes no extended warranty or any liability for use beyond the date specified on the container

Do not return any goods to us without obtaining prior approval and instructions from our Returns Department. Goods returned (unauthorized) by collect freight may be refused. Goods accepted for restocking will be exchanged or credited to your WPI account. Goods returned which were ordered by customers in error are subject to a 25% restocking charge. Equipment which was built as a special order cannot be returned.

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Contact our Customer Service Department for assistance in the repair of apparatus. Do not return goods until instructions have been received. Returned items must be securely packed to prevent further damage in transit. The Customer is responsible for paying shipping expenses, including adequate insurance on all items returned for repairs. Identification of the item(s) by model number, name, as well as complete description of the difficulties experienced should be written on the repair purchase order and on a tag attached to the item.

* Electrodes, batteries and other consumable parts are warranted for 30 days only from the date on which the customer receives these items.

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