
BIO X Standard 3-mL Pneumatic Printhead User Manual



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01

Package contents

01 Package contents

Item	Part number	Quantity
Standard 3-mL Pneumatic Printhead	000000020340	1
Locking screw	000000020106	1

02

Technical specifications

02 Technical specifications

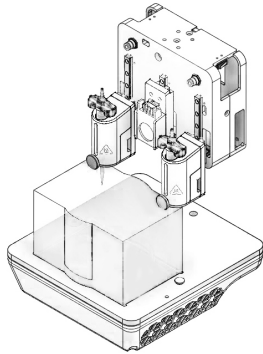
- Compatible with 3-mL plastic cartridge.
- Dimensions (height x width x depth):
 - With screw: 60 x 30.5 x 42 mm.
 - Without screw: 60 x 30.5 x 39.5 mm.
- Weight: 73.8 g.
- Maximum recommended pressure: 700 kPa.
- Material composition:
 - External surfaces: Powder-coated aluminum.
 - Internal surfaces: Aluminum.
- Recommended operating temperature: 20-23 degrees Celsius.

NOTE: We recommend operating the BIO X in temperatures between 20 and 23 degrees. In normal operating conditions, the chamber temperature of the BIO X will rise 2-3 degrees above room temperature, and the Standard Pneumatic Printhead temperature will rise above the chamber temperature.

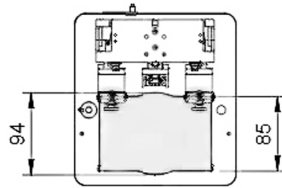
02 Technical specifications

- Running temperature (in recommended conditions): 26-32 degrees.
- Heating capacity and rate:
 - Total heating range: 30-65 degrees.
 - Heating rate: Room temperature-40 degrees (10 minutes).
 - Heating rate: 40-65 degrees (12 minutes).
- Cooling capacity: N/A.
- Build volume (X, Y, Z): 128 x 85 x (83.4-A) mm; A represents tip length.

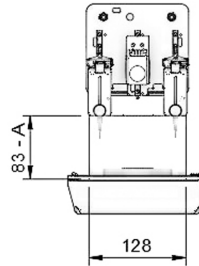
02 Technical specifications



ISOMETRIC VIEW



TOP VIEW



FRONT VIEW

*All measurements are in mm.

Figure 1: Theoretical maximum build volume as shown from the top and side view using the Pneumatic Printhead. Render assumes a nozzle length of zero. Exact build volume depends on cartridge position in printhead and nozzle/needle used. Build volume may be lower when combining the Pneumatic Printhead with a printhead that restricts printbox movement. Please consult the printhead manual for the printheads being used for specific limitations.

02 Technical specifications

Table 1: Theoretical build volume based on tips of common lengths. Actual volume can vary based on build surface thickness, size, shape and level, as well as cartridge position and tightness of tip. X, Y and Z are measured from the front left corner of the printbed.

Tip Type	Tip Length	Nozzle/Needle Length	X	Y	Z	Build Volume
Conical	3.2 cm	3.2 cm	12.7 cm	9.36 cm	5.14 cm	614 cm ³
Blunt	2.4 cm	0.635 cm	12.7 cm	9.36 cm	5.94 cm	706 cm ³
Blunt	3.0 cm	1.27 cm	12.7 cm	9.36 cm	5.34 cm	635 cm ³
Blunt	4.3 cm	2.54 cm	12.7 cm	9.36 cm	4.04 cm	480 cm ³
Micron-S	1.9 cm	1.9 cm	12.7 cm	9.36 cm	6.46 cm	768 cm ³

03

Safety information

03 Safety information

3.1 BIO X system warnings

- Please consult the BIO X manual for BIO X-specific and general warnings and safety procedures.

3.2 Standard Pneumatic Printhead warnings

- Do not clean the printhead by submerging it in liquid or using excessive spraying. Liquid inside the printhead can damage the circuitry and motor.
- The LED on the printhead will turn yellow to indicate a warm printhead and red to indicate a hot printhead. Do not touch the printhead when the LED is red. Touching a hot printhead risks serious injury.
- Do not manually move the printhead mount. Moving the printhead mount manually will damage the motor. If the mount needs to be moved, place the printhead in the loading position described in the Getting Started section.
- Use the proper method to load and unload the Standard Pneumatic Printhead onto the BIO X printhead mounts. Instructions are outlined in the Getting Started section.
- Do not use a jerking motion to remove the printhead. Using a jerking motion risks hitting and damaging the HEPA filter.
- Tighten the locking screw before printing to ensure that the cartridge remains in place during the printing process. Improper tightening can cause the cartridge to move and material to dispense improperly.
- Do not touch a moving printhead. Fingers can be crushed between the printhead base and motor arm and can result in serious injury.

04

Getting started

04 Getting started

4.1 Unpacking and installation

NOTE: The printing parameters displayed on this manual might not be accurate for your specific protocol. For recommended parameters please consult the documentation for the bioink being used.

1. Open the package. Remove the Standard Pneumatic Printhead.
2. Attach the Standard Pneumatic Printhead to the BIO X printbox. Align the printhead above the desired printhead mount (Figure 2A) and push downward using one hand. Secure the bottom of the printhead mount with your opposite hand to avoid straining the motor (Figure 2B,C,D).

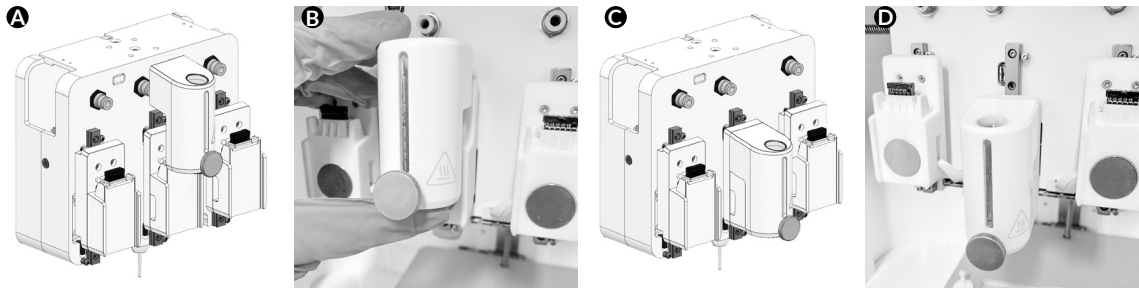


Figure 2: Attaching the Pneumatic Printhead to the BIO X. (A) Line up the printhead with a mount on the printbox. (B) Slide the printhead down with one hand and secure the bottom of the printhead mount with the other hand. Once attached, the lights on the printhead will turn from white to blue.

4.2 Raising and lowering the printhead

1. On the user interface, navigate to the Utilities menu and then go to the Tools submenu.
2. Lower the desired printhead mount into the active or loading position by pressing the down arrow (Figure 3).
3. Press the up arrow to return the printhead to the inactive position.

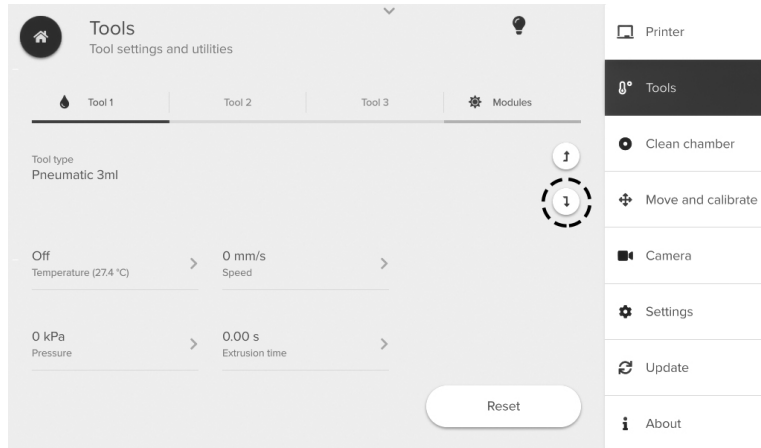


Figure 3: Lowering the printhead into the active position.

04 Getting started

4.3 Loading the bioink cartridge

1. Make sure that the printhead is installed and in the active or loading position (Section 4.1 and 4.2) (Figure 2).
2. Fill a cartridge with bioink or obtain a prefilled cartridge. Remove the bottom cap if using a prefilled cartridge (Figure 4A). Attach a nozzle (Figure 4B) and remove the end cap (Figure 4C).
3. Align the cartridge adapter to the loaded cartridge (Figure 4D). Twist until secure (Figure 4E).

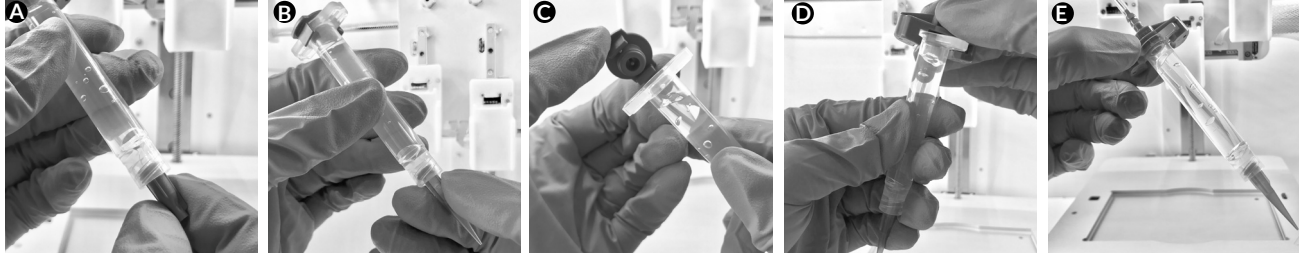


Figure 4: Preparing a prefilled bioink cartridge for bioprinting.

04 Getting started

4. Load the cartridge into the printhead from the top (Figure 5B). Tighten the locking screw. To make this step easier, we recommend placing the printhead in the active position (lowered) as explained in the Raising and lowering the printhead section (page 13) (Figure 5A).
5. Connect the air tubing to the air inlet on the printbox above the respective printhead (Figure 5C).
6. Raise the printhead to the inactive position (Section 4.2).

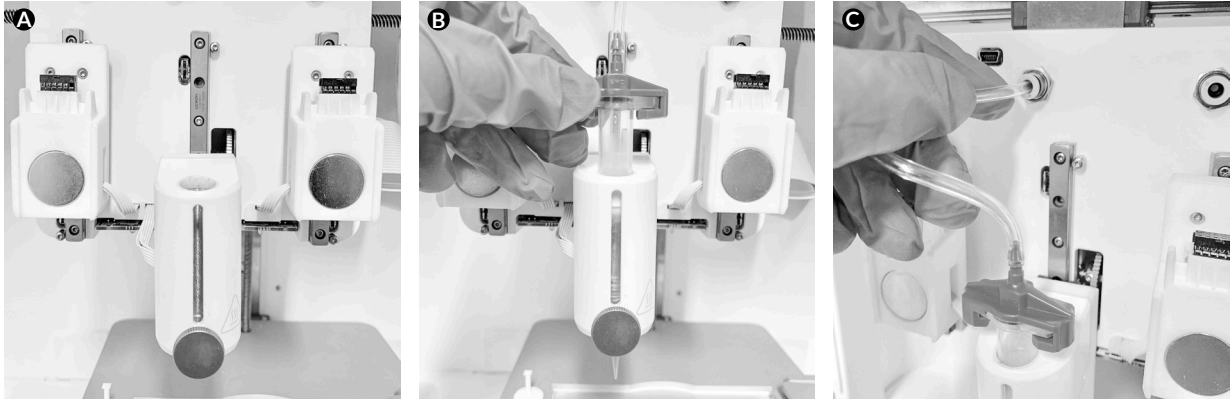


Figure 5: Attaching the air adapter to the cartridge and attaching to the printbox.

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4.4 Your first bioprint

1. Prepare the printhead and load the bioink as detailed in Section 4.3.
2. Select Bioprint from the Start menu (Figure 6).

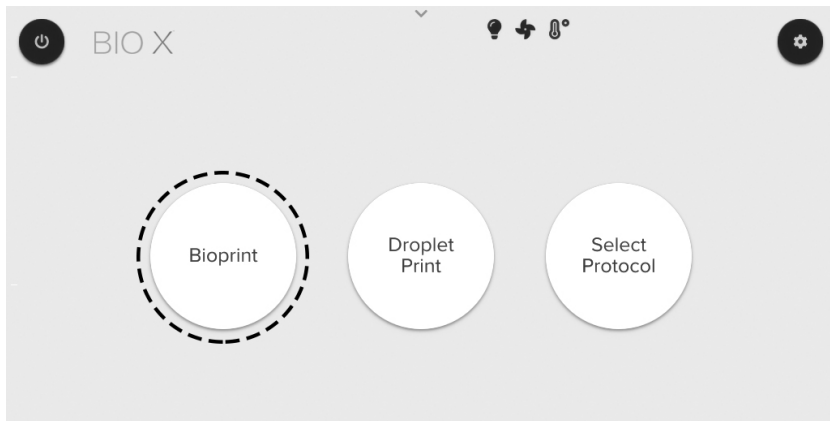


Figure 6: Select Bioprint on the Start menu and proceed to print setup.

04 Getting started

3. Select an STL file (filename.stl) or G-code file (filename.gcode) from the Model menu (Figure 7). Proceed to the next menu by selecting Surface along the bottom of the screen.

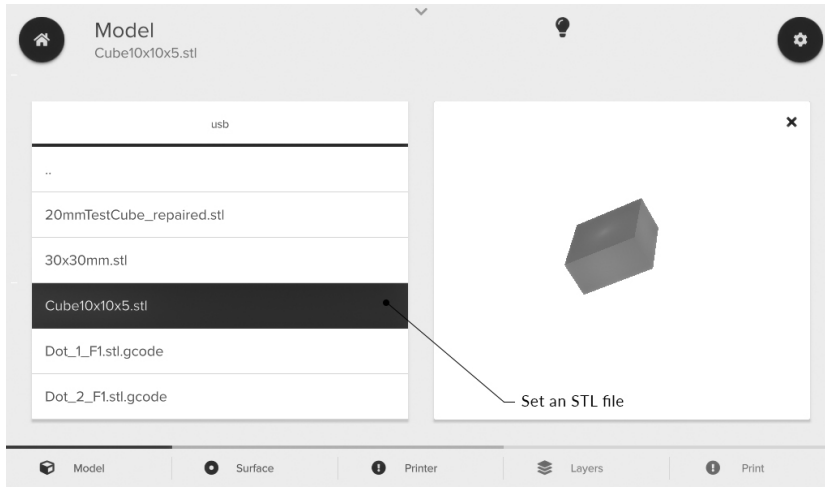


Figure 7: Selecting an STL file from the Model menu.

04 Getting started

4. Select a surface to print on (e.g., a Petri dish) and proceed to the next menu (Figure 8).

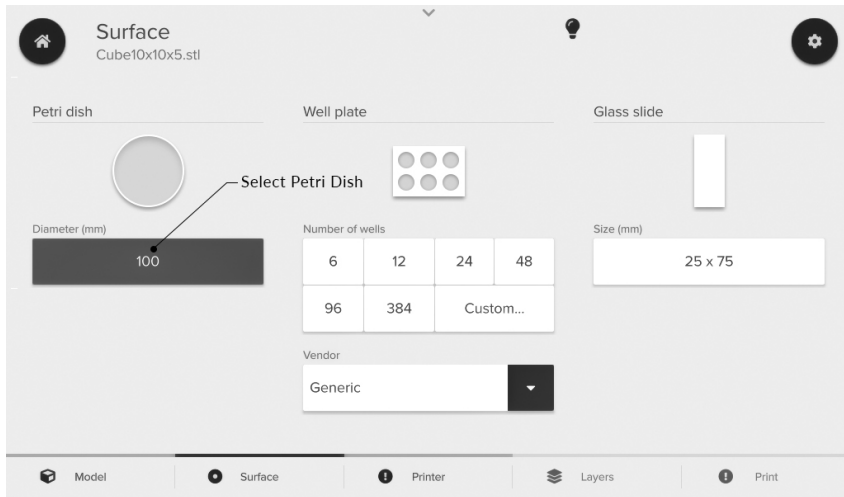


Figure 8: Select a print surface from the Surface menu.

5. Select printhead positions that have been set up. Ensure that Pneumatic 3-mL is selected under the printhead type (Figure 9).
6. Enter your printhead parameters (Figure 9) and proceed to the Layers menu. Parameters needed for the Pneumatic Printhead include:

- Nozzle diameter.
- Pressure.
- Print speed.
- Preflow delay.
- Postflow delay.

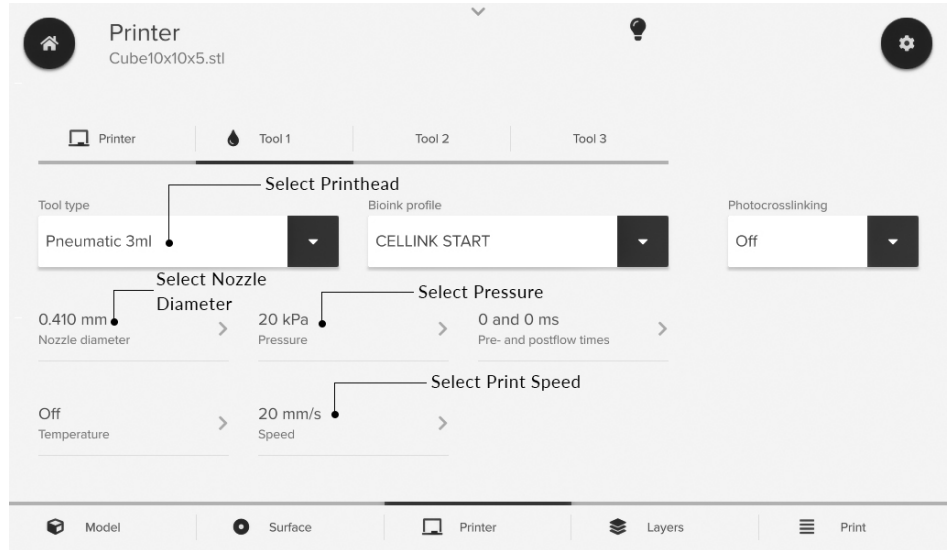


Figure 9: Printing parameters on the Printer menu.

04 Getting started

7. Assign the enabled printhead to the respective layer characteristics and proceed to the next menu (Figure 10).

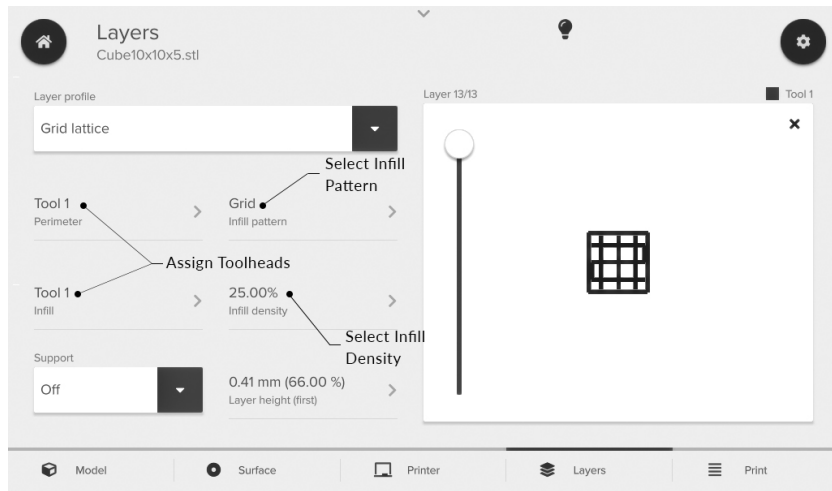


Figure 10: Layer menu. You can assign toolheads to respective print areas like perimeter, infill and support. You can select the infill pattern and density and preview the layers.

04 Getting started

8. Prime the nozzle/needle and test bioink flow by using the Drop button next to the pressure setting (Figure 11). Press Print to proceed to the calibration page.

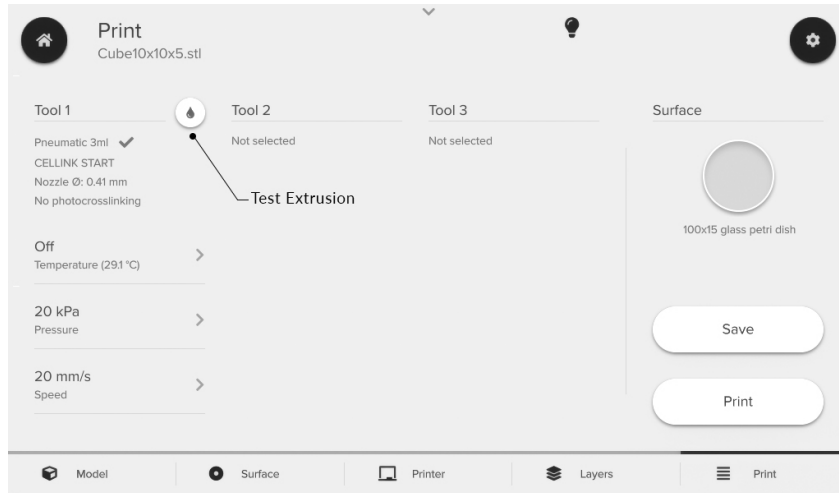


Figure 11: The Print menu is where you can preview your print parameters and test the extrusion rate. You can adjust the parameters on this menu. Press Print to proceed to the Calibration menu. Press Save to save your configuration as a protocol.

04 Getting started

9. Select Calibrate to calibrate the system to the desired start position (Figure 12). Bring the tip of the nozzle/needle to your desired starting position. We recommend making sure that the tip touches the surface of your printing substrate.

10. Start the bioprinting process (Figure 12).

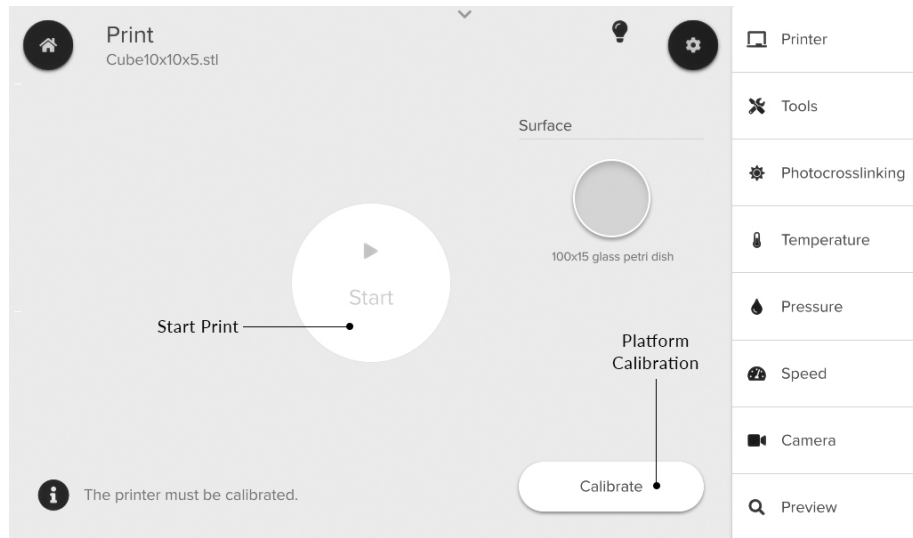


Figure 12: Use the Bioprint menu to calibrate the system and start the bioprinting process.

4.5 Removing the Pneumatic Printhead

1. Move the printhead to the active or loading position from the Tools submenu under the Utilities menu. See Section 4.2 for instructions.
2. Loosen the screw and remove the cartridge from the top.
3. Twist the cartridge pneumatic adapter and lift it off the cartridge to disconnect the cartridge from the pressure supply.
4. Cap your cartridge with the top and bottom caps and store it.
5. To remove the printhead, stabilize it using two fingers on the top surface of the printhead mount (Figure 13A). Press your thumb against the bottom side of the printhead to push the printhead slightly up (Figure 13B).

04 Getting started

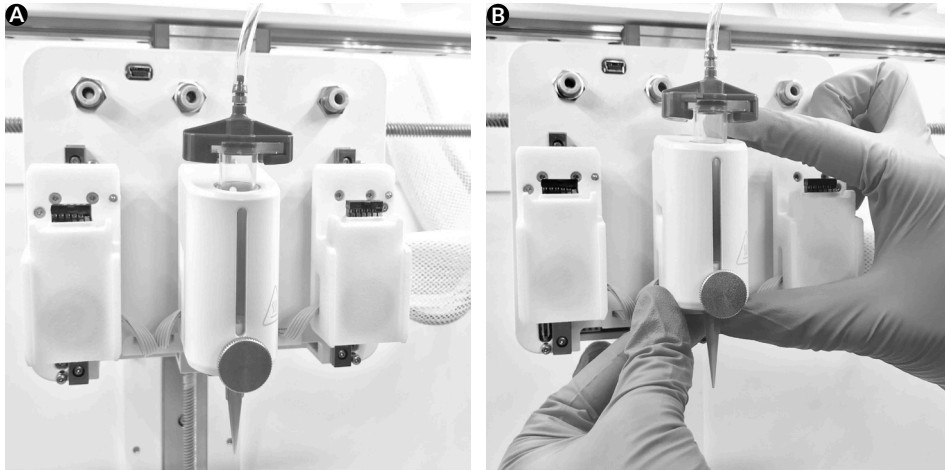


Figure 13: Removing the Pneumatic Printhead from the BIO X.

4.6 Pneumatic Printhead printing optimization

The Pneumatic Printhead uses compressed air to extrude bioink through a nozzle or needle with diameters between 50 and 600 micrometers. As a result, you can increase or decrease the air pressure to control the filament dispensing rate. Parameters that affect the deposited material's diameter include nozzle diameter, translation speed, layer height, bioink chemistry and thermal environment. Bioinks should be characterized thoroughly to determine the optimal parameters for the Pneumatic Printhead.

It is necessary to control the applied pressure to maintain viability while bioprinting. High pressure and small nozzle diameters can produce excessive shear stress that results in cell damage or rupture. Pneumatic-based extrusion systems enable users to control the pressure within the cartridge and the resulting cell viability. However, pneumatic-based extrusion systems offer less control over the flow rate and filament diameter.

The diameter of a filament extruded by the Pneumatic Printhead depends on nozzle diameter, applied pressure, temperature and translation speed of the printhead. These factors will affect the resulting filament diameter. As the applied pressure increases, the resulting filament diameter will increase. Similarly, as the translation rate is increased, the resulting filament diameter will decrease – and resulting filaments will be noncontinuous and nonuniform. In balanced conditions, the resulting diameter will approach the diameter of the nozzle or needle used. Adjusting the temperature of the printhead and applying pressure might result in changes to the bioink viscosity depending on its rheological behavior.

04 Getting started

Preflow delays can be set to ensure continuous filaments at the beginning of each layer and various starts and stops. This is due to the mechanical compression of the bioink in the cartridge, resulting in a delay in flow. The preflow delay setting will pause printhead movement after the air pressure valve is opened at the designated time. This allows the bioink to start extruding before the printhead moves, resulting in more continuous filaments at faster printing speeds.

The layer height must also be considered. A layer height that is shorter than the nozzle or needle diameter will result in filaments that are wider in diameter, while a layer height that is too large will result in poor filament attachment to the substrate. It might not be ideal to use a layer height equal to the nozzle or needle diameter because the bioink could expand after extrusion, or contract as it gels or self-assembles. This parameter is both bioink- and nozzle-dependent and must be evaluated individually for each bioink.

For depositing droplets, please follow the instructions on the droplet printing menu.

To optimize cell viability, we recommend depositing cells in droplets using increasing pressure to determine acceptable cell loss. Analyzing the resulting droplets after crosslinking and cell culture will enable quantification of survival rate under the different pressures, nozzle/needle diameters and shear forces. The optimal pressure can be used to determine the translation rate and nozzle needed to achieve the target filament diameter.

Please see the G-code section (Section 5) for coding the Pneumatic Printhead and customizing functionality.

05

Relevant G-code commands

05 Relevant G-code commands

Commands	Description
G1 Xnnn Ynnn Ennn Fnnn	When used in combination with the G90 command, which defines absolute coordinates, G1 is the absolute move command. The values for the X and Y parameters are the coordinates (in mm) directing where to move. E tells the BIO X to open the valve for extrusion. The F parameter is the speed of the printhead in mm/min.
G1 Znnn Ennn Fnnn	When used in combination with the G90 command, which defines absolute coordinates, G1 is the absolute move command. The value for the Z parameter is the coordinate (in mm) directing where to move. E tells the BIO X to open the valve for extrusion. The F parameter is the speed of the printhead in mm/min.
G4 Snnn Pnnn	The G4 command tells the system to dwell. The S command is the wait time in seconds and P is the wait time in milliseconds.
G7 Xnnn Ynnn Ennn Fnnn	When used in combination with the G90 command, which defines absolute coordinates, G7 is the relative move command. The values for the X and Y parameters are the coordinates (in mm) directing where to move relative to the current position. E tells the BIO X to open the valve for extrusion. The F parameter is the speed of the printhead in mm/min.

05 Relevant G-code commands

G7 Znnn Ennn Fnnn

When used in combination with the G90 command, which defines absolute coordinates, G7 is the relative move command. The value for the Z parameter is the coordinate (in mm) directing where to move relative to the current position. E tells the BIO X to open the valve for extrusion. The F parameter is the speed of the printhead in mm/min.

G92 Xnnn Ynnn Znnn

G92 sets the current position of the printhead to the specified X, Y and Z coordinates. If no values are given, the position is assumed to be 0, 0, 0 (this will also change the Z position).

Tx

Switch between printhead (x). Where 0, 1, 2 designate printheads 1, 2 and 3.

M771 Tx Pyy

M771 turns on the heater in printhead Tx at temperature Pyy. yy is between 30 and 65 degrees Celsius.

06

Frequently asked questions

06 Frequently asked questions

- **Can I print temperature-sensitive materials using the Pneumatic Printhead?**

The Pneumatic Printhead will warm up during normal operation due to the embedded chips and other electronic components. If precise temperature control is needed, we recommend using the Temperature-controlled Printhead in a room with an ambient temperature between 20 and 23 degrees Celsius.

- **What do the LED colors indicate?**

The LED will change color depending on its status. When the printhead is first put on the printhead mount, the light will turn white to indicate that it is initializing. Once it is recognized, the LED will turn blue. Blue light can also indicate a temperature below 35 degrees. When the temperature of the printhead is between 35 and 50 degrees, the light turns yellow to indicate that the printhead is warm. When the printhead is above 50 degrees, the light turns red to indicate that the printhead is hot.

- **What is the mechanism that enables bioprinting with this printhead?**

This printhead leverages pneumatic micro-extrusion bioprinting. Compressed air actuates a plunger valve opening and closing, causing materials with certain viscosities to be extruded through a small nozzle.

- **My bioink is not extruding.**

The needle or nozzle might clog because of the bioink used, or because of a pause in use. Replace the needle or nozzle. If your bioink contains aggregates, ensure that they are small enough to pass through the needle or nozzle without clogging it. If using a thermosensitive

06 Frequently asked questions

bioink, check to see if it has gelled at the tip. If it has, consider using the Temperature-controlled Printhead to prevent thermal gelation.

- **My filament is not attaching to the surface while printing.**

Your layer height may be too high, or your print speed may be too fast. These factors can prevent the filament from attaching to the surface.

- **My needle is dragging through my printed structure while moving.**

Your layer height may be too small, or your print speed may be too slow. These factors can cause thicker filaments that build a print that is taller than expected, resulting in the nozzle contacting the previous layer.

07

Maintenance

07 Maintenance

- Regularly clean the printhead with a damp cloth to remove any dust or debris.

Appendix A: Consumables— needles and nozzles

Appendix A: Consumables- needles and nozzles

Table 2: Compatible needles

Part Number	Type	Color	Gauge	Length (inch)
NZ5180505001	Needle	Green	18	0.5
NZ6200255001	Needle	Pink	20	0.25
NZ6200505001	Needle	Pink	20	0.50
NZ5201005001	Needle	Pink	20	1.00
NZ6210255001	Needle	Purple	21	0.25
NZ6210505001	Needle	Purple	21	0.50
NZ5211005001	Needle	Purple	21	1.00
NZ6220255001	Needle	Blue	22	0.25
NZ6220505001	Needle	Blue	22	0.50
NZ5221005001	Needle	Blue	22	1.00

Appendix A: Consumables- needles and nozzles

NZ7220505001	Needle	Steel	22	0.50
NZ6230255001	Needle	Orange	23	0.25
NZ6230505001	Needle	Orange	23	0.50
NZ6231005001	Needle	Orange	23	1.00
NZ6250255001	Needle	Red	25	0.25
NZ6250505001	Needle	Red	25	0.50
NZ5251005001	Needle	Red	25	1.00
NZ6270255001	Needle	Clear	27	0.25
NZ6270505001	Needle	Clear	27	0.50
NZ5271005001	Needle	Clear	27	1.00
NZ6300255001	Needle	Lavender	30	0.25
NZ6300505001	Needle	Lavender	30	0.50

Appendix A: Consumables- needles and nozzles

NZ5320255001	Needle	Yellow	32	0.25
NZ5320505001	Needle	Yellow	32	0.50
NZ5340255001	Needle	Green	34	0.25
NZ5340505001	Needle	Green	34	0.50

Table 3: Compatible conical nozzles

Part Number	Type	Color	Gauge	Length (inch)
NZ4180005001	Conical	Green	18	1.25
NZ4200005001	Conical	Pink	20	1.25
NZ3220005002	Conical	Blue	22	1.25
NZ3250005002	Conical	Red	25	1.25
NZ3270005002	Conical	White	27	1.25

Appendix A: Consumables- needles and nozzles

Table 4: Compatible precision conical nozzle

Part Number	Type	Color	Gauge	Length (inch)
NZ2210000801	Micron-s Conical	Light Blue	21	0.75
NZ2230000801	Micron-s Conical	Purple	23	0.75
NZ2250000801	Micron-s Conical	White	25	0.75
NZ2270000801	Micron-s Conical	Red	27	0.75
NZ2300000801	Micron-s Conical	Black	30	0.75
NZ1150000501	Micron-s Conical	Blue	150 μm	0.75
NZ1100000501	Micron-s Conical	Orange	100 μm	0.75
NZ1050000501	Micron-s Conical	Yellow	50 μm	0.75

Appendix A: Consumables- needles and nozzles

Table 5: Compatible cartridges and cartridge accessories

Part Number	Description	Quantity
CSC010300502	3-mL empty cartridges	50
CSC010300102	3-mL empty cartridges with end and tip caps	10
CSC010311502	3-mL empty cartridges with end and tip caps	50
CSO010311102	3-mL UV-shielding cartridges with end and tip caps	10
CSO010311502	3-mL UV-shielding cartridges with end and tip caps	50
OH000000010	Female/female Luer lock adapter	10
OH000000050	Female/female Luer lock adapter	50

Support information

- Official site: www.cellink.com
- Contact: support@cellink.com
- Contact: sales@cellink.com
- Web store: www.cellink.com/store



Store



Sales



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Support



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