Two-photon Lithography with the Nanoscribe GT2

Quick Start Guide

1. Enable the tool in Coral.
2. Determine the correct objective(s) (10X, 25X, and 63X available) for your application.
3. Select the objective in the microscope control console near the keyboard.
4. Ensure the Process light on the tool (green) is not lit. If it is currently lit, ensure that the Holder Exchange dialog is open. If not, click Exchange Holder.
5. Press the Open button, it should flash and the lid can now be opened.
6. Open lid and sample holder lid.
7. Remove black cap from center-most microscope position, install the objective
   a. Objective should be inserted only until rotation stops. Do not overtighten.
   b. For the 25X objective, ensure the correct Adjustment Ring setting is selected. For most cases, Glyc with the 3 longest bars is used.
8. Install resin stop on the objective.
   a. For 10 and 25X, a silicone stop is used. For the 63X, a fiber ring is used.
9. Select an appropriate sample holder and place on the white and black support with the Resist side facing up.
10. Place your substrate in the holder and tape down with at least two strips of tape from the dispenser.
11. Place a drop of resin on the substrate.
    a. For 63X oil-based printing, please be sure to place the oil on the proper orientation - the oil should be in contact with the lens, not the resin
12. Ensure the sample holder lid in the printer is placed in the partially opened position, supported by the latch.
13. Bring sample holder to printer. Immediately before inserting, flip sample holder upside-down and insert in the printer.
    a. For all resin-based prints, the resin side of your substrate should be facing the objective.

b. For oil-based printing, the oil side should be facing the objective
14. Close sample holder lid and secure down with both clamps.
15. Ensure all lens holders, tools, etc. are clear of the tool and close the main lid.
16. Select the correct sample holder and sample position in the Exchange Holder dialog box.
17. For standard prints, click Approach Sample. For manual interface finding, please see the information provided in Manual Interface Finding.
18. Click Load Job and select your *_job.gwl file.
19. Click Start Job.
20. When printing has finished, click Exchange Holder. Leave this dialog box up for the next user.
21. Partially open sample holder lid and remove print, inverting it so that it is resin-side up.
22. Open sample holder lid and remove the objective; you may leave the resin stop on if there is resin on it during this process, otherwise you may remove the resin stop first.
23. Clean objective as outlined in objective cleaning
24. Log out from tool
25. Develop your samples as outlined in the sample development section

Introduction

Direct laser lithography or direct laser writing using two-photon femtosecond near-IR laser for additive manufacturing using conventional resin sensitive to two-photon absorption resulting in two-photon polymerization to create volumetric prints from CAD models.

Exposure System

The NanoScribe GT2 Laser Lithography System is an ultramodern two-photon polymerization volumetric maskless printer. It is one of our newest acquisitions here at the Cornell NanoScale Facility (CNF). The tool was purchased in late 2019 and delivered and installed during the second week of January 2020. This was accomplished with the help of a committee of faculty and researchers, and a grant from the NSF. The NanoScribe GT2 can create three-dimensional nanostructures using a NIR femtosecond laser via direct-write onto a photosensitive resin, which is subjected to a non-linear two-photon absorption process. This process involves cross-linking the resin via UV absorption. The laser sets a focal light cone where a concentration of the light intensity defines the exposure focal spot volume or a "3D Pixel." Using this technique, a CAD design can be broken into an X-Y-Z coordinate system to define the structure pixel by pixel and layer by layer. The exposure side of the system sits in a fine-tuned vibration isolation table and a high-speed ultra-precise piezoelectric stage for movement in X-Y-Z and uses a galvanic mirror defection system for focusing and beam rasterization. Models for printing can be designed using the stand-alone software DeScribe—which comes with the tool—or with any CAD software capable of outputting file formats DXF or STL. The DeScribe software can import these formats.
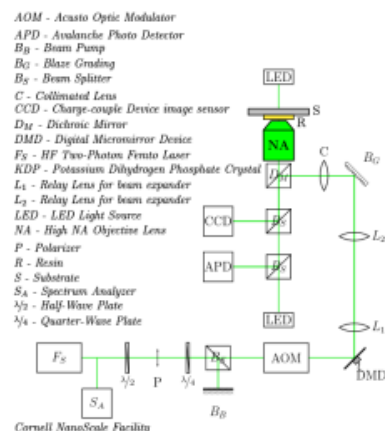
3D Print Technology & Direct Write

Laser-based direct writing has been around for a few years especially in the semiconductor industry known as maskless lithography (MPL). Typically, the two-dimensional CAD design is transferred onto the substrate that is pre-coated with a photosensitive resin via reduction laser projection. The projected pattern was reduced given the tool's numerical aperture and scan across the surface of the substrate using either a raster or vector format. These techniques are the same ones utilized in 3D direct-write. The only difference is that now you are working with a thickness degree of freedom that can be scanned across the Z-axis. The NanoScribe GT2 has a Piezo Nanopositioning stage and a high-speed galvo scanner. In the case of two-photon lithography, the light source is a pulsed femtosecond fiber laser with the center wavelength being around 780 nm.

Hardware Overview

Diagram of a simplified optical path of a Two-Photon Lithography System.

Simplified Optical path of a Two-Photon Lithography System

AOM - Acusto Optic Modulator
APD - Avalanche Photo Detector
B_B - Beam Pump
B_G - Blaze Grading
B_S - Beam Splitter
C - Collimated Lens
CCD - Charge-couple Device image sensor
D_M - Dichroic Mirror
DMD - Digital Micromirror Device
F_S - HF Two-Photon Femto Laser
KDP - Potassium Dihydrogen Phosphate Crystal
L_1 - Relay Lens for beam expander
L_2 - Relay Lens for beam expander
LED - LED Light Source
NA - High NA Objective Lens
P - Polarizer
R - Resin
S - Substrate
S_A - Spectrum Analyzer
$\lambda/2$ - Half-Wave Plate
$\lambda/4$ - Quarter-Wave Plate

Cornell NanoScale Facility

Utilized Objectives [NA: 10x, 25x, 63x]
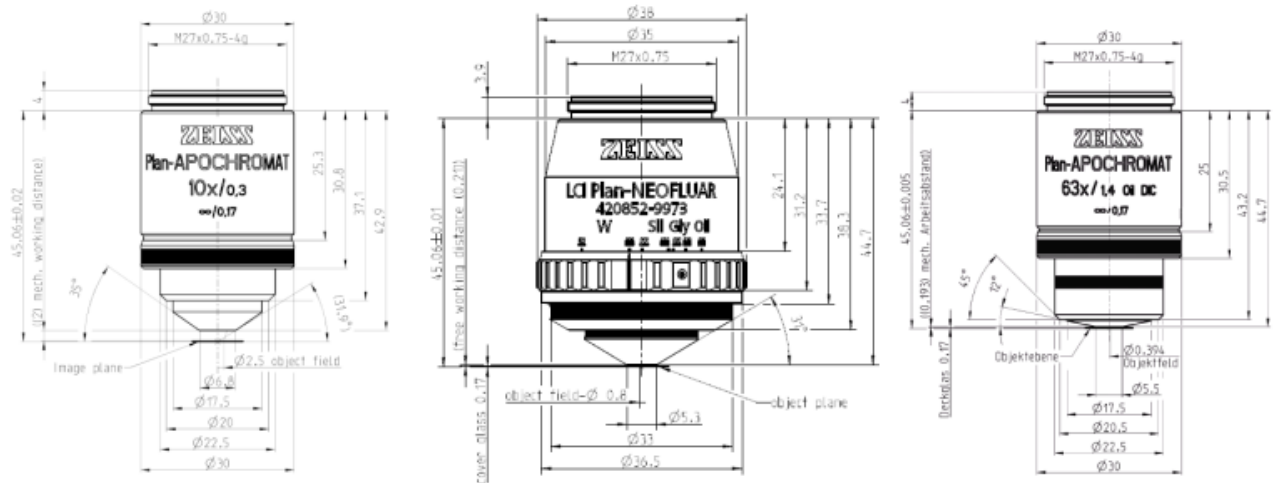
The 25X has an adjustment ring for printing with different liquids. For most cases, the Glyc setting with the three longest bars is used, as shown below:

Two-photon Light Source

- Hight Performance NIR femtosecond laser [FemtoFiber Pro]

Training

Cornell NanoScale's Training Workflow

- Send the tool manager a request for online access to NanoScribe's NanoGuide Webpage.
- Meet with the tool manager to discuss your project.
- Complete the CULearn training for the tool.
  - CULearn Training links:
    - RSRCH - CNF - NanoScribe GT2 (NetID Link)
    - RSRCH - CNF - NanoScribe GT2 (GuestID Link)
  - Once you completed the CULearn video and quiz, email the tool manager to schedule a hands-on training session.
- Users are required and responsible for reviewing the services and procedures provided by NanoScribe on the following:
  - You will need to log in to NanoGuide to view these materials.
    - Model Creation (CAD)and Import
    - Printing
    - Print with Solution Sets
    - Print Applications
    - Print Strategies
    - Programming Structures
    - Post Print Processes
    - Further REading
- Check your STL model for common mesh errors. You can use CAD software capable of mesh verification such as Autodesk Meshmixer or MeshLab

- Once your file has been checked for mesh errors, run it through DeScribe to do the conversion. Be sure to select the desired solution set (SF, MF, or LF) for your process.
- Once the CULearn

NanoScribe's Subsystems

- Electronics Cabinet
- Optics Cabinet [contains the laser, optics, and Galvo]
- Emergency Off (EMO) switch
- Printing Cabinet [Microscope, Piezo, and Stage]
- Power Supply 232
- System Controller
- System Computer
- Laser System Controller
- Galvo Stage Power Supply
- Monitor, Keyboard, and Mouse
- Manual Microscope Controller
- Joystick

CAD & Software

CAD Model

Since NanoScribe only handles the fracturing and slicing of the imported CAD design utilizing its own proprietary General Writing Language (GWL) complex models need to be designed and verified outside of the system and when ready exported as a Standard Triangle Language or Standard Tessellation Language (STL). STLs describe the raw unstructured triangular surface geometry of three-dimensional using normalized units and vertices objects without any color or texture attributes. STLs can be written in both ASCII and binary.

3D CAD Software Available at the CNF

Note: Software usage is limited and or restricted depending on the type of license the CNF currently is under contract.

- Autodesk
  - AutoCAD
  - Inventor
  - Meshmixer
- DeScribe
- FreeCAD
- MeshLab
- LinkCAD

You may opt to use the CNF 3D GPU Conversion Computer which has a number of software tools above as well as DeScribe available. Compile time on DeScribe is typically faster on this system than a standard Windows desktop PC by 2-3 times for complex 3D object. Instructions for its use are available here.

STL Conversion From GDSII Files

GDSII files are common for traditional photolithography processes and it is possible to convert them to a 3D model suitable for use with the DeScribe slicer. Three primary applications are used in this process:

- KLayout
- OpenSCAD
- gds3xtrude

Installation is easiest on a Linux distribution. General installation instructions are available on the gds3xtrude link.

DeScribe

- Options for running DeScribe Software
- The most current version is v2.7 as of 12/7/22
  - It is highly recommended that you use the most current version of DeScribe to have access to the latest recipes and print modes.
  - In v 2.7, most recipes are included in DeScribe and do not need to be downloaded separately
  - Users can download their copy here (requires software license and Nanoguide access)

Recipes: DeScribe Tabulated Solution Set based Parameters:

Solution Set	Objective	Resin	Substrate	Sample Holder	Slicing	Hatching	Dev/Rinse
SF	63x	IP-L / IP-DIP	Fused Silica / Borosilicate Substrates	DILL / DiLL Oil	0.3 um	0.2 um	12 min
MF	25x	IP-S	ITO-Coated	DiLL	1 um	0.5 um	20 min
LF	10x	IP-Q	Silicon	DiLL	5 um	1 um	20 min

Printing Modes

The Nanoscribe uses solution sets which are paired to an objective in order to slice a CAD model. In general, each objective has a Solid, Shell and Scaffold, and Swift printing mode.

| Mode	Use Case
Solid	Slowest printing method, generates parts which do not require post-process UV curing generally. High resolution printing. Used for delicate or intricate parts where strength is a priority or long development times are required.
Shell and Scaffold	Faster than solid, speed dependent on part complexity and infill percentage. Parts have a high resolution shell with coarse infill to balance speed and high resolution. Will require UV cure. Used for intricate parts where development times are short (<<1hr).
Swift	Fastest printing method, prints coarsely in a layer-wise fashion in half the time of most shell and scaffold prints. Parts usually do not require UV cure. Balanced Swift is higher resolution than Swift, but longer print times. Used for bases or support structures where dimensional accuracy is not critical.
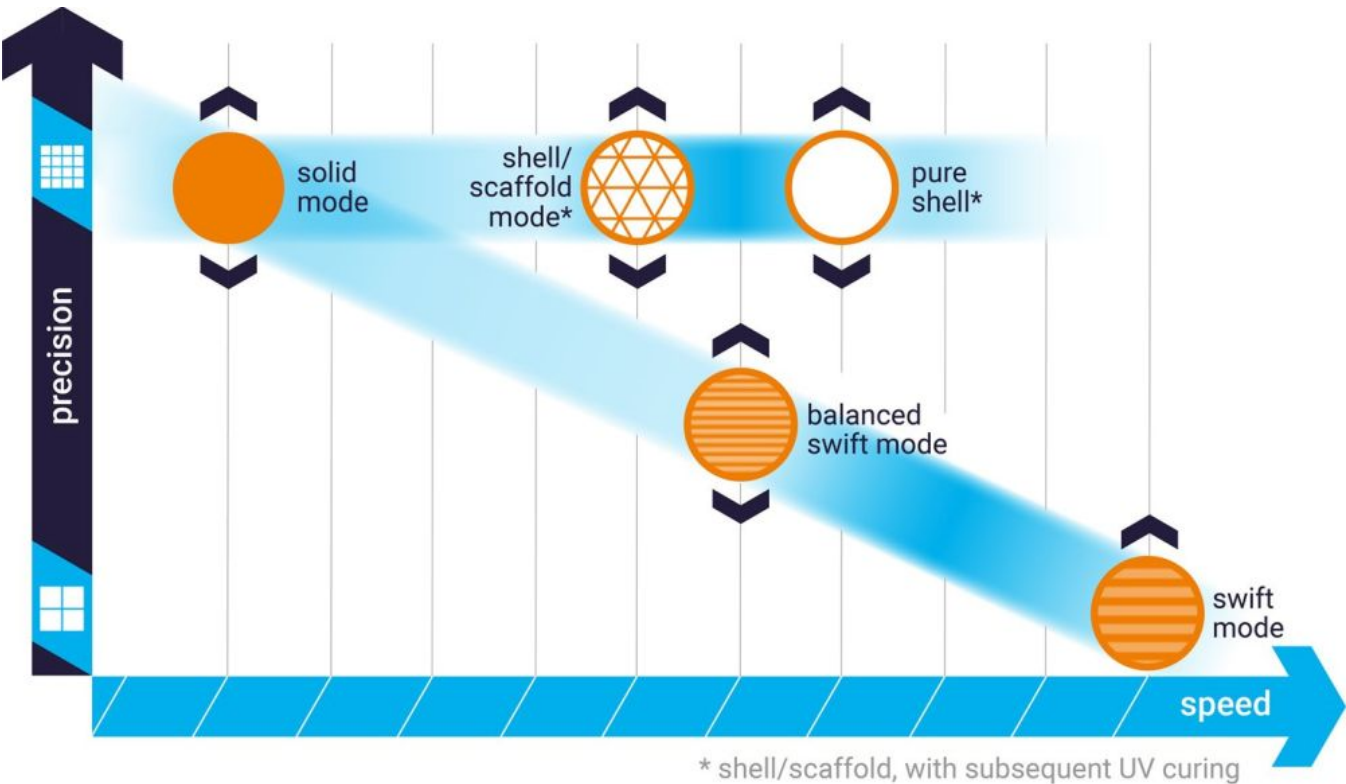
Comparison of printing solutions by part resolution and print speed. Image from Nanoguide

NanoWrite

Sequence Of Operations
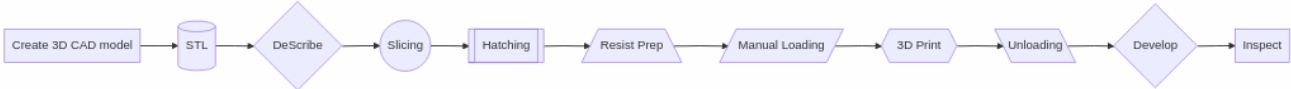
General Printing: Operational Workflow (SOP) as stated in the NanoGuide

The basic standard operation procedure (SOP) is summarized below. The titles link to pages with more detailed information.
The paragraph headers of this page link to important safety measures. Carefully read through each article to avoid harm to the operator and damage to hardware components. Additionally read through printing considerations, which provides general information for a successful print along with best practice advice.

Check the status of the printer

- Make sure the printer components are switched on. If the laser was switched off wait for 30 min before starting a print job to ensure a stable laser output.

Import a CAD model file

- Drag and drop an STL file (CAD model file) into *DeScribe* to start the import wizard.
- Center the model and adjust the orientation as well as size. Ensure that it fits within the substrate dimensions and consider the working distance of the objective for the desired print set.
- Choose a recipe matching the desired print set and follow the steps given in the import wizard.
- Save the GWL print job file.
- Preview the print job (shortcut: F5 key).
- Alternatively, the print job can be programmed manually.
- If the import was performed on a personal computer, copy all GWL files and the subfolder "..._files" to the control computer.

Prepare a sample and change the objective

- Prepare the required materials for the intended print set and print job.
- Fix the substrate to the sample holder and apply resin.
- Check that the resin is free from bubbles.
- Select an objective and inspect it for cleanliness. Clean it if necessary.
- Use a clean and dry felt ring or resin stop with all objectives except for air objectives.
    - Wash felt rings with isopropanol before the first use to remove loose fibers.
- Mount objective in the correct position. Use the touch screen to check/select the position if necessary.

Start *NanoWrite* and insert sample holder

- Start *NanoWrite*.
- Wait for the stage calibration warning message.
- Ensure sure the stage will not be impeded by obstacles.
- Ensure the microscope objective is at the lower z-drive limit.
- When it is safe to do so, confirm the warning message by clicking *Calibrate*.
- Wait for the *NanoWrite* initialization to finish.
- When the Exchange Holder dialog appears, carefully insert the sample holder.
- Select the correct sample holder and sample position from the dialog box.
- Confirm the selection by clicking *OK*.
- Switch on an illumination. Choose reflection illumination for non-transparent substrates.
- Start AxioVision software and position it next to *NanoWrite* to view the print process in real-time.

Perform a sample approach

- Ensure that the selected sample position is correct. If it is not, double click the respective position on the sample holder icon in *NanoWrite*.
- Click *Approach sample* to move the objective towards the interface. The interface will be automatically found and the working distance set such that the interface is in focus. The result of the interface approach is displayed in the *NanoWrite* message log window.
- Adjust the brightness and contrast in the AxioVision software.
- After each subsequent movement of the stage, the interface position must be verified. Either click *Find Interface* to autofocus the interface again, or issue FindInterfaceAt commands in the GWL code of the print job. Imported models utilizing block splitting and stitching have the commands included automatically.

Load a print job

- Click *Load Job* and choose a print job (*name_job.gwl* file).
- Wait for loading to finish (indicated by *name_job.gwl loaded* in the message log window).
- If compilation takes a long time, use DeScribe to perform this step (debug -> compile, or shortcut: F5)

Print your structure

- For high-precision applications allow ~30 min for the resin and holder to reach thermal equilibrium.
- Click *Start Job.*
- Wait until the print job is finished or press *Abort* to immediately halt the job.

Unload the sample

- Click *Exchange Holder*.
- Wait for the Exchange Holder dialog to pop up.
- Carefully remove the sample holder.
- Confirm the dialog by clicking *OK*.

Develop the sample

- Carefully remove the substrate from the sample holder.

- Place the substrate in a developer bath (PGMEA for most resins, methanol for GP-Silica) to wash away excess liquid 2PP resin.
- Leave substrate in developer at least 15 minutes, high aspect ratio structures require additional development time.
- Wash away the developer with IPA.
- Transfer to IPA for at least 15 minutes.
- Cure remaining liquid 2PP resin if a shell and scaffold structure was printed.

Clean the objective lens

NEVER USE ACETONE WHEN CLEANING THE OBJECTIVES

- **The objective lenses must be thoroughly cleaned after each user has finished using the tool.** Multiple prints may be conducted during the same user session without the need to clean the objective, assuming the same 2PP resin will be used for each print.
- In most cases, IPA alone is appropriate for lens cleaning.
    - Specific lenses have different cleaning requirements, as do different resins and immersion oils.
    - If you are unsure of the proper cleaning procedure for your use case, please contact the staff before printing.

10X Objective

1. Remove the objective and screw it into its holder
2. Carefully wipe the metal portion of the objective with a lab wipe or swab, taking care not to touch the glass.
3. Cover the lens with the clear top cover and move the lens to the development room.
4. Place the lens on clean wipes where you will be cleaning - do no assume wipes left in the hood are clean.
5. Carefully fold a wipe to about 1 to 1.5" wide and wrap around the body of the objective. Take note of the location of the vent hole by placing your thumb directly over it.
6. Invert the lens over the waste container and funnel.
7. Gently flow IPA over the objective using a wash bottle - be careful to dispense the IPA slowly and keep the tip of the bottle away from the nosepiece.
8. With the lens still inverted, use the N2 blowgun and dry off the IPA with a low pressure stream of dry nitrogen
9. Return the lens to an upright position and using a wipe or swab remove any resin which has flown onto the metal body of the nosepiece
10. Repeat, typically 2-3 additional times, using IPA sparingly.
11. *Note, for use with GP-Silica, alternate cleaning methods must be used, as outlined in the section on GP-Silica*

25X Objective

1. Remove the objective and screw it into its holder
2. Carefully wipe the metal portion of the objective with a lab wipe or swab, taking care not to touch the glass.
3. Check the objective body, and in particular, the adjustment ring for any residual resin and clean this with a lab wipe dampened with IPA.
4. Cover the lens with the clear top cover and move the lens to the development room.
5. Place the lens on clean wipes where you will be cleaning - do no assume wipes left in the hood are clean.
6. Carefully fold a wipe to about 1 to 1.5" wide and wrap around the body of the objective.
7. Follow steps 6-10 as above, being careful to prevent any IPA from coming into contact with the rubber gaskets and the adjustment ring in particular.

63X Objective

Follow cleaning for the 10X objective, omitting the need to cover a vent hole in step 5.

Please consult Nanoguide for any other questions regarding lens cleaning.

Failure to properly maintain the objectives can and will result in significant tool downtime.

Leaving the printer

- Clean the workspace with a wipe and 70% IPA spray to ensure there is no residual resin on the desk or the printer.
- Ensure the sample holders are clean and free of resin and wipe with 70% IPA is any residue is left on them.
- Close *NanoWrite*. This makes sure that used print commands are set to the default values for the next user.
- Only switch off the laser and other printer components if the printer is not in use for a longer period of time.

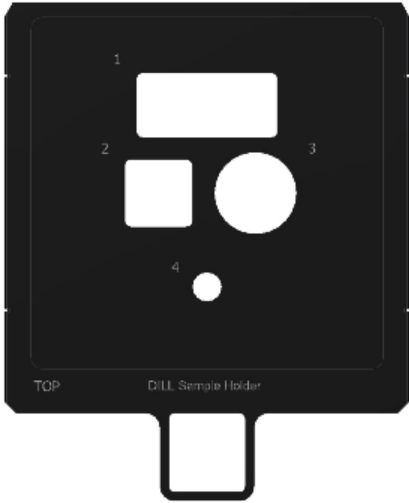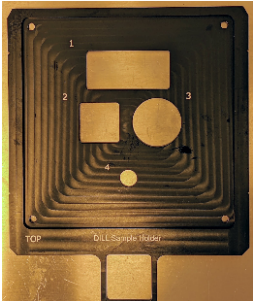# Material Specifications for standard printing at CNF

Substrates Specifications

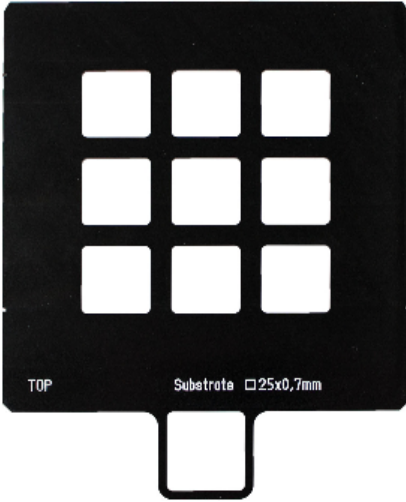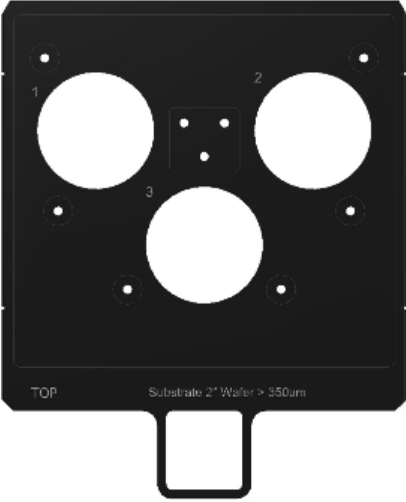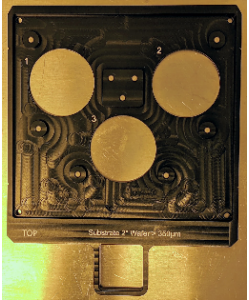| @ 780 nm	Fused Silica Substrates	ITO-Coated Substrates	Silicon Substrates	Borosilicate Substrates	Microscope Slides
Material	Fused Silica	Soda Lime w/ ITO	Si Substrates	Borosilicate Sub	Microscope Slides
Sample Holder	DiLL	DiLL	DiLL	10 × Ø 30 mm	DiLL
Refractive index	1.454	1.624 [ITO] 1.518 [soda lime]	3.710	1.517	1.518
Dimension	[25, 25, 0.7] mm	[25, 25, 0.7] mm	[25, 25, 0.725] mm	30 mm Ø, 170 µm	[22, 75, 1] mm
Thickness variation	± 25 µm	± 60 µm	± 25 µm	± 10 µm	± 100 µm
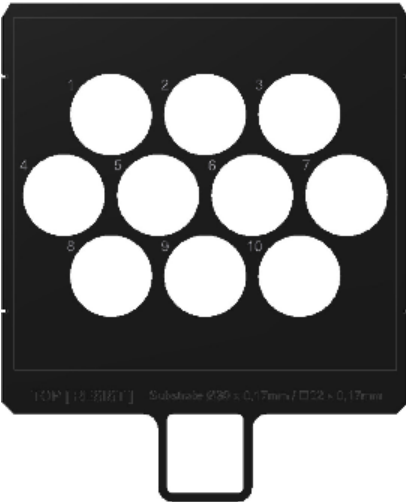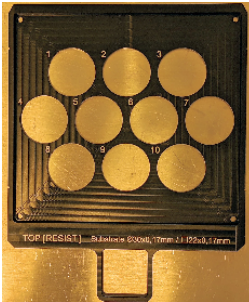
Surface finish	DSP	DSP	Polished	DSP	DSP
Density	2.2 g/cm3	2.5 g/cm3	2.3290 g/cm3		
Mohs hardness	5.3-6.5	5-6	9-10		
Melting point	1400°C	1000°C	3265°C		
Thermal expansion coefficient	0.54x10-6 K-1	0.937 W/m·K	2.6 μm/(m·K)		
Heat conductivity	1.38 W/(m·K)	0.937 W/(m·K)	149 W/(m·K)		
Compatible w/ Solution Set	3D SF	3D MF (3D SF, 2D ML)	3D LF (3D MF, 3D SF, 2D ML)	3D SF Oil	3D SF
Compatible w/ 2PP resin	IP-DIP	IP-S	IP-Q (IP-S, IP-Dip, AZ resin)	IP-L 780 or IP-G 780	None, unless coated
Compatible w/ Objective	63X	20x, 25x , 63x	10x, (20x, 25x, 63x)	63x	63x
Combination & Ref index	-0.058 @ 20°C IP-Dip / -0.025 @ 20°C IP-S	-0.145 @ 20°C			
Contrast					
Cleaning & Prep	O2-plasma & silanization	O2-plasma & silanization	RC1		

Indium-Tin Oxide (ITO) Coating Specifications

ITO	Value
Film Thickness	18 nm ± 5 nm
Film Surface Resistance	100 - 300
Film Transmittance	89%

Sample Holders available at the CNF

Sample Holder	Substrate Type	Substrate Thickness	Holder Image	CAD Files
DiLL	1. Microscope Slide: 24-26 mm x 50-76 mm 2. Fused Silica Substrate, ITO-Coated substrate, Silicon Substrate: 25 x 25 mm² 3. Borosilicate Substrate: Ø 30 mm 4. Tiny Substrates: 12 mm > Ø 30 mm > 25.4 mm	1.00 mm 0.70 mm, 0.70 mm, 0.725 mm 0.17 mm 0.30 mm		

Multi-DiLL	Fused Silica Substrate, ITO-Coated substrate, Silicon Substrate: 25 x 25 mm²	0.70 mm, 0.70 mm, 0.725 mm		multidill.stl multidill.stp *Note: some dimensions are approximated
2" Wafer	Ø 2 inch Wafer	0.35 – 0.55 mm		
10 × Ø 30 mm	Ø 30 mm	0.17 mm		

Resins Available at the CNF
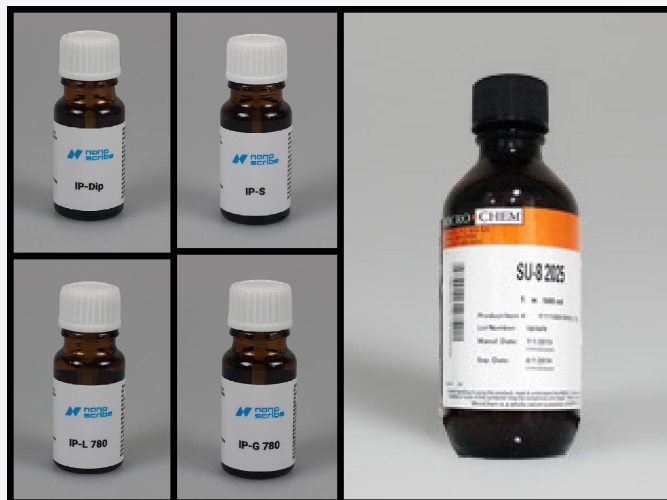
- **IP-DIP:** IP-Dip is a liquid negative-tone 2PP resin formulation by NanoScribe designed for printing the finest possible features. IP-Dip is a component of the 3D SF Solution Set. Tall structures exceeding the working distance of the objective can be printed using the DiLL configuration.
- **IP-S:** IP-S is a highly viscous liquid negative-tone 2PP resin formulation by NanoScribe designed for printing medium-sized features in DiLL. IP-S combines low shrinkage and high stability with a smooth structural surface finish perfect for micro-optics. The photosensitivity is tailored for two-photon polymerization and UV-curing is also possible. As a component of the 3D MF solution set, IP-S is suitable for printing structures with dimensions of several cubic millimeters.
- **IP-L 780:** IP-L 780 is a liquid negative-tone 2PP resin formulation designed for printing small features in oil immersion configuration. IP-L 780 combines high resolution with low shrinkage and high stability.
- **IP-Q:** IP-Q is a highly viscous liquid negative-tone 2PP resin formulation by *NanoScribe*. The 3D LF Microfabrication Solution Set, including *IP-Q*, extends 2PP 3D printing towards rapid printing of mesoscale structures with millimeter sizes, micrometer precision, and maximum throughput up to the macroscale.
- **SU-8:** SU-8 is a UV-curable negative-tone 2PP resin by Kayaku Advanced Materials. This resin is well-known and widely used in the lithography community. [The following article applies only to SU-8 100 determined to be the most suitable resin for 2PP as it balances quality and printing speed.]

GP-Silica Resin

A new resin will soon be available for use. GP-Silica contains silica particles which can be sintered into solid fused silica parts. Using the LF solution set, users can develop robust parts with mechanical and optical properties very similar to bulk fused silica.

Some substantial differences exist compared to IP-Q and similar acrylate resins:

- Maximum part printing time of 15 hours - exceeding this time may cause severe lens damage, as the resin will slowly cure and solidify over time.
- Parts must be developed in methanol and should develop horizontally to prevent premature part-substrate separation
- After development, parts must be sintered to come to full strength. Currently, CCMR's controlled atmosphere furnaces are recommended for thermal processing. Sintering curves are available on Nanoguide.

Developer

- PGMEA (CAS NO. 108-65-6) (For IP-Dip, IP-S, IP-Q, IP-L, SU-8)
- Methanol (CAS No. 67-56-1) (For GP-Silica only)
- Isopropanol (CAS No. 67-63-0) (For secondary wash, all resins)

Metrology & Post Processing

Model UV Curing

- Parts, particularly shell and scaffold components, may require a UV cure after development.
  - Shell and scaffold parts contain uncured resin within solid walls. Failure to UV cure will result in weak parts and may leak resin.
- The resin's highest sensitivity is to 405 nm UV, however broad spectrum UV curing stations may also work.
  - NOTE: The resin may burn or become distorted with UV power densities above 7 W/m^2
  - Typical maximum cure depth is 2.5 mm
  - Recommended exposure time is 1-15 minutes, dependent on part geometry
- Alternately, parts may be heat cured. IP-S cures in 5 minutes at 190°C and 17 h at 140°C. Care must be taken to slowly heat samples; thermal shock may break your part off the substrate prematurely. IP-DIP and IP-L cannot be heat cured.

UV Cure Station

A 405 nm UV cure station is available in Room 121 in the cleanroom.

Technical Specifications:

- Fourteen 405 nm LED in two arrays (2x7)
- Maximum 40W total LED power
- Fluence: 21 mW/cm^2
- Solar powered rotating stand
- Maximum running time of 40 minutes. Exceeding this may cause damage to the equipment.
- Typical exposure time of 0.5 - 10 minutes.*

* Values are for typical Nanoscribe GT2 prints. Actual application may vary by resist or resin.

Note – the LED lighting is diffuse, making characterization of the exact energy density your sample will receive difficult to estimate, as sample location and height can have a significant impact on energy received. Overexposure of samples to 405 nm light may result in yellowing of the samples and embrittlement of the resin.

Use

Can be used for curing 405 nm wavelength resists/resins, particularly parts printed on the Nanoscribe Photonic Professional GT2. Users should only place **clean, dry** parts in the cure station. Users are responsible for cleanup after use, including wiping the stand. This cure station will run for a user-selected time and automatically shut off after the time has elapsed. Time is entered in 30 second increments up to 40 minutes. Generally, curing times will be well under 10 minutes. Running for 40 minutes or longer may result in overheating of the machine. Samples are visible through a yellow filter in the front of the cure station – you may notice intense fluorescence of your samples while the curing is ongoing.

Safety

This curing system uses near-UV 405 nm wavelength light, which is not inherently harmful to vision or cells. The lighting system is, however, very bright posing a risk of vision damage if the system is operated without being properly closed. Filtered goggles are available and encouraged for users. Additionally, the bright light may cause unwanted curing of other samples when open. As such, the station should only be run when in the properly seated closed position and only opened when the LED array is off.

Model Removal

After printing and development, parts that are still on the substrate may need to be removed if they have no detached already. For robust parts on substrates that have poor adhesion, particularly silicon wafers, parts may be detached through simple mechanical means. However for more delicate parts or substrates with superior adhesion, a few methods can be used to remove parts:

Thermal shocking of the substrate via hot plate is an effective method for some parts, typically temperatures of ~90°C are adequate, higher temperatures may risk damaging or overcuring parts.

In some cases, sonication in IPA may work to dislodge a part from its substrate, using short durations (<1 min) and checking your part often.

Silicon Wafer Substrate Production

Standard Type P silicon wafers can be cut to produce 25mm square substrates for use with the 10X and other processes using the CNF DISCO Dicing Saw. Users must provider their own wafer for slicing; for best results, wafers should be 700µm thick +/-25µm. After receiving training on this tool, a program has been saved that is available to dice 6" wafers for use in the Nanoscribe. When using the DISCO Dicing Saw, open the USER directory and select the Nanoscribe6in profile. Please note: this profile is currently set for a 675µm thick wafer; you MUST adjust this parameter for thicker or thinner wafers.

Once you have completed the alignment process (typically aligning only along the flat edge of the wafer is adequate), a total of at least 18 substrates will be produced. Take care to ensure that substrates are free of debris from cutting before using in the NanoScribe. Additional cleaning of substrates via sonication with consecutive baths acetone, IPA, and DI water is recommended.
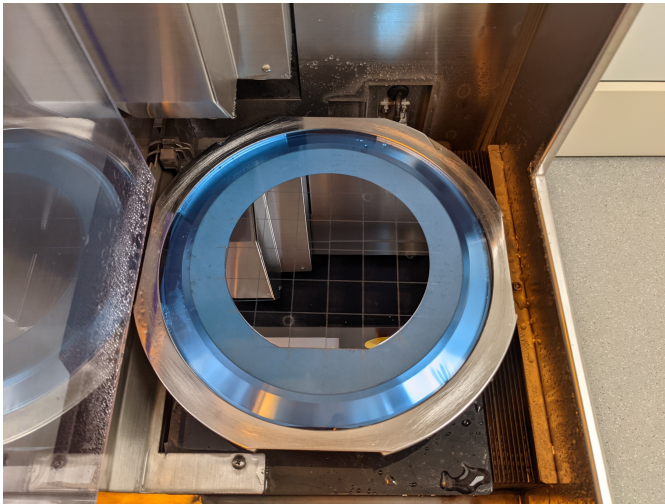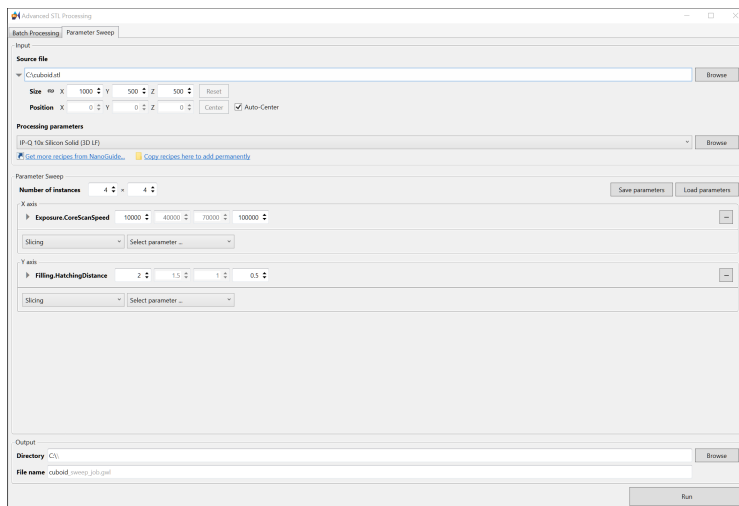

Advanced Printing Procedures

Parameter Sweeps

While the default DeScribe settings work in many cases, high precision parts may require careful tailoring of the printing parameters, such as laser scan speed, laser power, hatching distance, among others. The quickest way to test a variety of these parameters is with the Advanced STL Processing tool in DeScribe. Using a test STL, usually a small part or feature, one can generate a 1D or 2D array of parts with parameters varied between a start and end point. The image below illustrates a 4x4 array which varies scan speed and hatching distance, which impacts effective laser dose and can highlight issues due to proximity effect:

File Generation and Hierarchy

Advanced printing procedures often require modification to the basic files generated by the DeScribe slicer. In addition to offering much greater control over your print, a user can stitch together multiple jobs into a single part with differing print modes, or even objectives. Many of the most useful commands are highlighted in the sections below. A full reference list is available on Nanoguide and in the DeScribe editor under Window Command Reference.

Upon slicing an STL file, **sample.stl** for example, DeScribe generates a number of files and folders, all but the recipe files are necessary to print:

sample.recipe - Contains basic information on your part import, such as orientation, scaling, and translation. This file will let users maintain changes made to an imported part's orientation on subsequent imports.

sample_job.recipe - Contains information related to the print settings, such as block size, hatching distance, and scan speed. This can be used when importing a new part to pull the previous settings forward in DeScribe.

sample_job.gwl - File which is called by NanoWrite to start a print job. This file contains basic parameters used by the printer such as InvertZAxis, Galvo or Piezo Mode, Scan Speed, Power Scaling, Laser Power among others. This file is also used to call the print data, usually sample_data.gwl but can also call other job files.

sample_data.gwl - Contains instructions to move the stage relative to current position in microns as well as tells the printer which specific block to print in the form of gwlb files.

gwlc - This folder contains GWLC files which are a form of compiled GWLs. The GWLC files are created when generating a 3D preview and will be subsequently loaded by NanoWrite; compiling in DeScribe before loading in NanoWrite significantly speeds up the import process. These files are binary and not easily readable or editable.

sample_files - This folder holds the GWLB, or binary GWL files which are ultimately read by the printer and dictate the laser paths and stage movements. These files are binary and not easily readable or editable.

Multi-DiLL Printing

Multiple samples can be printed sequentially without the need to open the printer and switch substrates. This is useful for jobs where a large number of samples are needed, particularly if print times are short. The procedure for setting up a Multi-DiLL print will reference this build folder which is available for download: Sample01_02_03.zip The builds are based on these simple STLs: sampleSTLs.zip.

- Generate STL files as with any standard print job
- Import these files into DeScribe individually and generate a build using your selected solution set. It is important that all files use the same objective, however you may vary between solid and the shell and scaffold solutions.
    - Keep the generated files in their own respective folders - organizing these files is very important to the process
    - In this case, all files related to Sample1 are in a Sample1 folder
- Combine all sample folders to one master folder
    - An example of the file structure is included below:

> This PC > Secondary (D:) > Sample01_02_03

Name	Type	Size
📁 gwlc	File folder	
📁 Sample1	File folder	
📁 Sample2	File folder	
📁 Sample3	File folder	
📄 1_2_3_job.gwl	GWL document	1 KB

> This PC > Secondary (D:) > Sample01_02_03 > Sample1

Name	Type	Size
📁 gwlc	File folder	
📁 sample1_files	File folder	
📄 sample1.recipe	RECIPE File	1 KB
📄 sample1_data.gwl	GWL document	2 KB
📄 sample1_job.gwl	GWL document	1 KB
📄 sample1_job.recipe	RECIPE File	2 KB

- Generate a *job.gwl file - this will tell NanoWrite the order of operations and which files to reference
  - The example uses 1_2_3_job.gwl and is stored in the master folder. The gwl file can be created in a text editor (by changing .txt to .gwl or similar) and the content should be formatted as follows:

```
NewStructure
SamplePosition 4
Wait 20
MessageOut "Start printing the 1st sample"
include Sample1\sample1_job.gwl

NewStructure
SamplePosition 5
Wait 20
MessageOut "Start printing the 2nd sample"
include Sample2\sample2_job.gwl

NewStructure
SamplePosition 6
Wait 20
MessageOut "Start printing the 3rd sample"
include Sample3\sample3_job.gwl
```

- In this example, we are printing in positions 4, 5, and 6 on the Multi-DiLL holder.
  - **NewStructure**: Tells NanoWrite to begin a new print
  - **SamplePosition**: Tells NanoWrite which position in the sample holder to print
  - **Wait**: Waits for an integer number of seconds to allow the resin to settle after the lens has approached
  - **MessageOut**: Displays text to indicate a new job has started
  - **include**: Opens the build files from the included path
- You may test your gwl file by opening it in DeScribe to verify the paths are correct. Generate a 3D preview (F5) and ensure there are no errors.
  - Note: DeScribe will render all models on top of one another. This is normal behavior, as seen below:
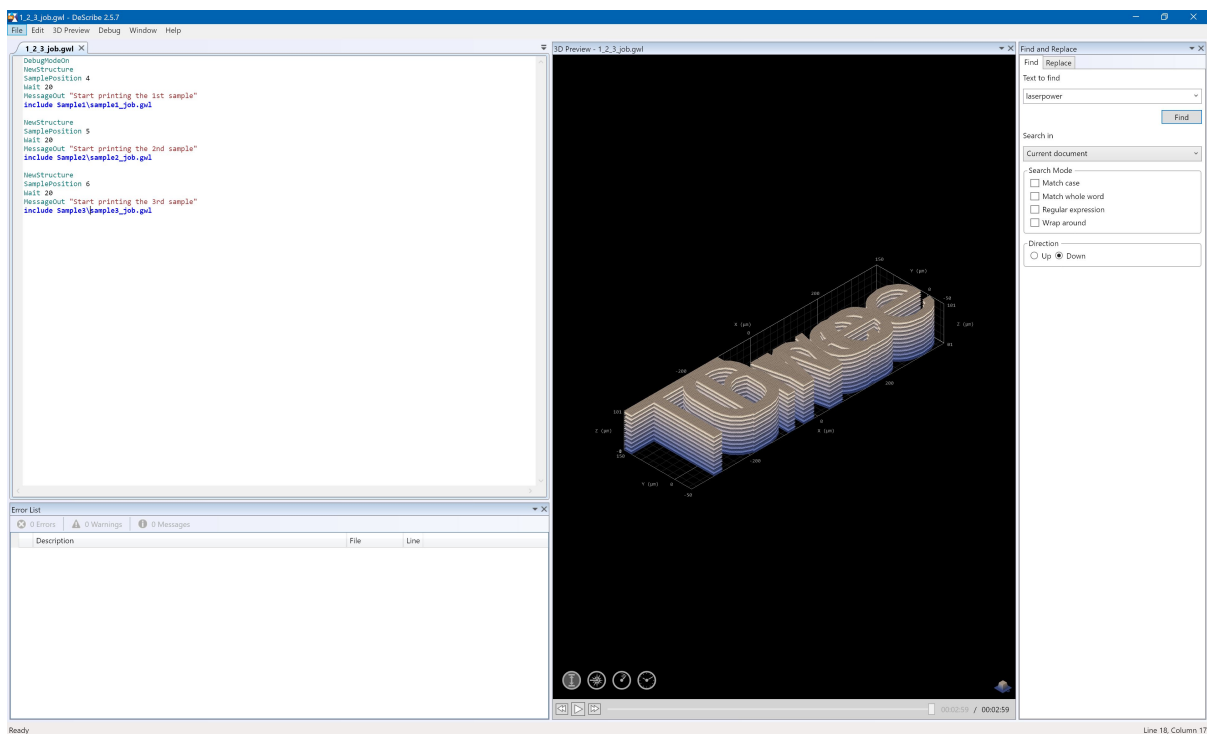
- Copy the folder to the Nanoscribe computer
- Place your substrates in the positions indicated in your gwl file and add resin to them, using an appropriate amount for the sample printed
  - Large deposits of resin may drip in the machine, please ensure a minimal amount is applied
- Insert the correct objective and the Multi-DiLL holder as with standard prints
- Select the position of your first print in NanoWrite. By default, the Multi-DiLL is set to position 5, as seen below. To reduce bubble formation in your print, it is best to change this to your first print position. Note that when clicking the new position on the Multi-DiLL graphic in the NanoWrite "Choose sample holder" dialog box, the view is from the bottom of the holder, so positions are flipped, e.g. position 1 is the top right. Click OK and then approach the sample.
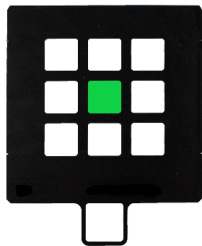
- Load the *job.gwl file in your top folder in NanoWrite, in the case of the example load 1_2_3_job.gwl
- Start print job, the total print time scales linearly with the number of samples in the job

Manual Interface Finding

WARNING: MANUAL INTERFACE FINDING MAY RESULT IN A COLLISION BETWEEN THE SUBSTRATE AND LENS IF DONE IMPROPERLY. IF YOU HAVE CONCERNS, PLEASE CONTACT A STAFF MEMBER BEFORE ATTEMPTING.

Procedure for All Objectives:

1. Load your substrate in a sample holder. If the substrate is thicker or thinner than the holder depth (i.e. 700 microns thick for the Multi-DiLL holder), it is important to measure how much taller or shorter your substrate is.
2. Add resin to your substrate and the objective.
3. Load your sample as you would normally, select the correct holder and click OK.
   a. **Do not** click Approach Sample at any point.
4. In the advanced camera settings, ensure that the light source is correct for your substrate - for opaque substrates use a Reflective Illumination, but for transparent substrates you will be better served with Transmission Illumination with all objectives.
5. Approach the sample with the manual Z control. You may use coarse control initially until you reach a Z-height on the order of 9000um. For thicker substrates, it is recommended to stop earlier, substract your substrate height above the holder from 9000.
   a. During this step you should see the lens contact the resin, it will be very obvious, looking like a bubble moving across the microscope. Keeping Auto Contrast and Auto Exposure.
6. Using the fine control, move the microscope slowly; you should slowly begin to resolve the substrate.
   a. For a 700um thick substrate this is typically a Z-value greater than 9800um.

b. Turning off auto-contrast and auto exposure can be helpful; ensuring short exposure times but also smaller Gain (<15) will result in a clearer image.
7. Once the substrate appears in view, turn off the microscope in NanoWrite and open AxioVision.
8. While AxioVision loads, in the NanoWrite Advanced Settings console type *ManualControl* and click Submit.
9. On the left hand side of the Manual Control interface is a section with Shutter Control. Set the laser power to a low value, typically 5%
10. With AxioVision loaded, open the shutter. If you see a small white dot, this is the laser. If you cannot see it, you are either too close to the substrate and its focal length is past the surface of your substrate. Alternately, you may be too far from the substrate, however this is typically not the case if you have your substrate in focus.
11. Using the fine Z control, slowly adjust the Z location towards your substrate until the white dot just disappears. Note this Z location - switching the microscope control unit to XYZ mode is helpful for this process as the Manual Control interface is not ideal for this step.
12. Close the Shutter in Manual Control.
13. Close AxioVision and Manual Control. Enable the NanoWrite microscope.
    a. Note: closing AxioVision and renabling the microscope will automatically turn on Auto-Contrast, you may wish to disable this again.
14. Your substrate should eventually come into sharp focus, move the substrate on X and Y until you have identified where you would like to print.
15. Modify your _data.gwl file:
    a. It will contain the line **FindInterfaceAt $interfacePos**, comment this out by adding "**%**" before the line
    b. If this line is not removed or commented out, the microscope will lose the position and attempt to automatically find the interface
    c. **DeScribe v2.7 and higher allows you to disable automatic interface finding in the Output section of the Import STL window. Under "Interface finder", select "Skip." Doing so should obviate this step.**
16. Load your print job and begin printing
    a. It may be valuable to have a small object (e.g. a cube) to test the print before starting a longer one. This can help validate your starting point and ensure that your print isn't floating or the laser is focused past your substrate.
17. When your print is complete, click Exchange Sample Holder. This will back the objective from your print as normal.

See this short video below which follows the same process:

*Note - the Z values in this video are not necessarily reflective of other prints and should not be used as a reference.

Printing on Porous Substrates

For substrates with holes or other features, it is necessary to manually align the microscope. First, users should follow the Manual Interface Finding procedure. This can be done on any location on your substrate, however an area away from your desired print area is suggested. Once an interface has been determined, manual alignment can begin.

In the following steps, we are assuming that we are looking to find the center of a 1000 um hole in the center of a substrate.

1. Move around the microscope until the feature is visible. Ensuring the features are near center of the substrate as possible will prevent bubbles from forming and being moved around the lens.
2. Make sure Center Cursor is on, there should be a dashed blue line on the microscope display. If it is not there, click Center Cursor.
3. Move to one edge of the feature, roughly center, in this case the lowest point of the hole, keeping this in the center of the crosshairs.
4. Move up 1000 um on Y by inputting 1000 um in the step size box and clicking the up arrow for stage adjustment. Note that in this case picking the Y axis is arbitrary and the X may be used just as easily as a starting point.
5. Realign to the top of the hole, generally 10um increments are sufficient. Record the total distance added or subtracted from the initial 1000 um.
6. Move to the new Y center point by inputting half the net distance travelled in Step Size then click the down arrow once e.g. ((1000um-distance correction)/2).
7. Move right on the X axis by current value in Step Size.
8. Align the edge of the hole as with the Y axis. If the center appears to be misaligned with the Y axis, adjustments can be made now to align on this axis.
9. Continue to move side to side and up and down in order to verify that the sample is centered.
10. Record the X and Y coordinates of this center point.
11. For multi-part prints, it is necessary to comment out any instance of **FindInterfaceAt** or **CenterStage** in the _job.gwl and _data.gwl files
    a. In lieu of **CenterStage**, a new center point can be added manually via the **StageGoToX** and **StageGoToY** commands, typically in the _job.gwl file.
        i. Formatting is **StageGoToX** *number* where **number** is the X coordinate of your new center point.

The CAD models for this substrate are available for download in STL and STP format, as well as a drawing of the substrate.

| substrate.stp | substrate.stl | porous_substrate.pdf |

The short video below demonstrates the process for a 25x25 mm substrate with a 1mm center hole:

These substrates were printed via an SLA process at 50um layer height and subsequently polished to a 3um finish.

Multi-Part Printing

The Nanoscribe is capable of printing multiple parts in a single job, assuming they use the same objective.* In general, each individual part is sliced with its own parameters and then a master job.gwl file calls these individual parts.

Some editing of _job.gwl files may be required, particularly to generate the master file. The commands which are very useful in generating this file are:

- **include *_job.gwl** : Always include the job file and NOT the data file. The job file will automatically call the data.
- **Pause**: Introduces a pause before proceeding to the next step. The user must click "Continue" or "Abort" before proceeding.
- **var $i** : This syntax declares the variable *i*. Any other string can be used as long as it is not a command and follows the $ symbol every time it is called.
- **CenterStage**: Brings the stage to the center position, the same coordinates where the automatic interface finder determined the center.

- **StageGoToX #/ StageGoToY #**: These commands can move the stage to a desired X or Y position where # is an X or Y coordinate.
- **AddZDrivePosition #**: Adds some value, #, to the current Z height in microns.
- **ShowVar $i**: displays the value of the variable *i*. Useful for outputting things like loop number, or other hardcoded variables to ensure they are still correct.
- **MessageOut "text"**: Displays the text within the quotes in the Message Center. Using this one can output current status (i.e. MessageOut "Printing job 2 of 3")
- **for i = 0 to 1**: Basic For loop syntax. The lines between the for statement and "end" will run while i increments by 1 (by default).
- **end** : Ends the For Loop.

An example folder is included which demonstrates this printing method. It can be downloaded here: NS_multi_part.zip

The code after the standard header text in the job file is below:

```
var $centerX = 0
var $centerY = 0
var $i = 0

% Include slicer output
include pedestal_job.gwl % Prints the base part

MessageOut "Pedestal printed" %Displays a message in the NanoWrite info window

MessageOut "X center:"
ShowVar $centerX  %Displays the current set value for X center
MessageOut "Y center:"
ShowVar $centerY  %Displays the current set value for Y center

MessageOut "Printing statue"
Pause %Waits for user input to continue or abort print

for $i = 0 to 1 % This loop prints two sets of 'statues', adding 200um in Z height each time to ensure they print atop the pedestal
    StageGotoX $centerX % Ensures the stage moves to a known center. Users can set other values if the subsequent prints should be offset
    StageGotoY $centerY
    AddZDrivePosition 200
    include statue_job.gwl % Make sure the interface finding is OFF in statue_data.gwl as the stage will reset to the interface location
otherwise

end
```
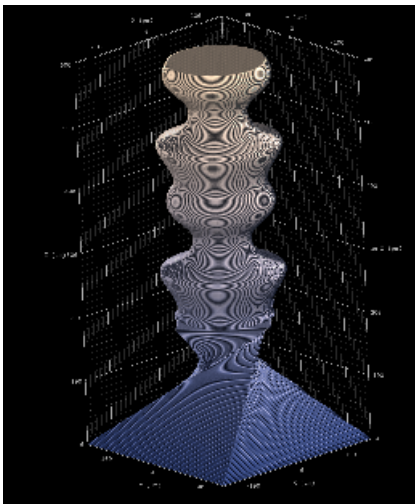
The folder contains two sliced parts: pedestal.stl via 25X Swift Mode and statue.stl via 25X Balanced Swift Mode. Master_job.gwl contains the commands to first print the pedestal and then two copies of the 'statue' atop. When generating a 3D preview, the final object should look like this:


Total estimated print time is 2.5 minutes.

*Multi-objective printing is also possible, but not covered in this example.

Marker Alignment

Printing on wafers prepared by other systems or prints that require multiple objectives may require special alignment procedures to ensure the print occurs in the exact orientation desired. A detailed description of the exact alignment procedure is available on Nanoguide. To aid with this, three job files have been prepared to aid in aligning the 10X, 25X, and 63X objectives. All files by default print a single voxel width X for orientation purposes using the *Write* command. An example of this is below:
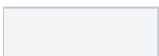
```
-10  -10   0
 10   10   0
write
-10   10   0
 10  -10   0
write
```

This will produce an **X** that is 20 um wide by 20 um tall. This command can also be run in the Advanced Commands window directly without a special job file.

Basic job files have been prepared for the 25X and 63X objectives for marker alignment:

25x_marker_job.gwl

63x_marker_job.gwl

Applications

- Microfluidics
- Micromechanics
- Biomedical Engineering
- Micro-electro-mechanical systems
- Mechanical metamaterials
- Micro-optics
- Photonic metamaterials and Plasmonics

Publications & Gallery

- **Study highlights promise of 3D printing for electrochemical reactors** [04/2021]
- **Nano-sized McGraw Tower features 161 steps, chimes** [05/2021]
- **Biologically inspired micro-robotic swimmers remotely controlled by ultrasound waves** [09/2021]
- **Building the Nano-3D McGraw Clock Tower** [12/2021]

Other Files

Users may wish to store their new and used 25mm square substrates. A set of STL files is provided for users to download and 3D print; each holder can fit 20 substrates.

substrateholder_top.stl   substrateholder_base.stl
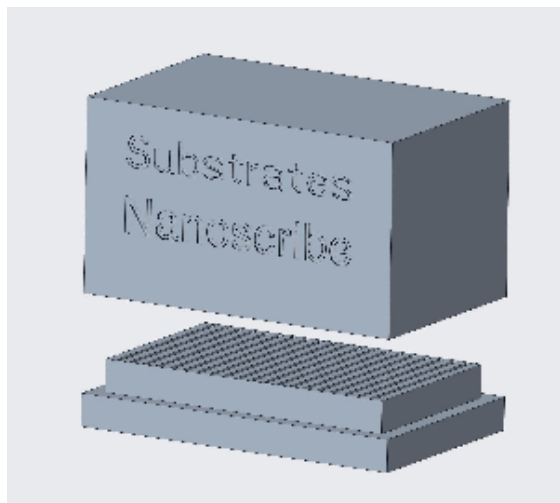
It is recommended that these be printed with an SLA or similar process, as tolerances are tighter than allowable by FDM printing.

Other Sites in the NNCI Network with a NanoScribe

- **Stanford University: Stanford Nanofabrication Facility**
- **Harvard University: Center for Nanoscale Systems**
- **Georgia Tech: Institute for Electronics and Nanotechnology Micro/Nano Fabrication Facility**

Power UP and Power DOWN Procedures [STAFF ONLY]

Power UP Procedure

Warning!

"Disregard of the guidelines concerning power-up of the Photonic Professional GT2 may cause damage to the system!"

Apply the following sequence in order to power up the Photonic Professional GT2 (See Fig)

1. Turn the mains power switch to position 1.
2. Wait some seconds until the *reset* and open *buttons* blink in alternation and the *attention* indicator is lit up.
3. Press the *reset* button, the *attention* indicator switches off.
4. Switch on the Photonic Professional GT2 Controller by activating the key switch.
5. Switch on the Control PC: Let it boot completely before proceeding, wait until the long on-screen shows up.
6. Switch on the laser controller by activating the key switch. The button `start/stop` does not have to be pushed (the backlight of the button should be off). The laser emission is controlled via the Photonic Professional GT2 control software NanoWrite.
7. Switch on the GT power supply.
8. Switch on the z-drive power supply.
9. Switch on the computer monitor.
10. Press the *open* button in order to release the safety lock and to open the printing cabinet. As long as the safety lock is released the *open* button is blinking white.

   Note: The laser gives acoustic feedback, as soon as its start-up is finished. This can take up to 30 s. After that, the control software NanoWrite may be launched. Make sure that the laser is operational when starting NanoWrite. The laser emission is switched on automatically when NanoWrite starts. The status of laser emission is indicated by the backlight of the button `start/stop` on the front panel of the laser controller. If the light is activated, the emission is on. You can manually switch on/off the emission by pressing this button. A power calibration of the laser is performed automatically before the first print starts or when NanoWrite receives the first script command.

Power Down Procedure

Warning!

"Disregard of the guidelines concerning power-up of the Photonic Professional GT2 may cause damage to the system!"

Apply the following sequence in order to shut down the Photonic Professional GT2 (See Fig)

1. Exit NanoWrite. Wait until NanoWrite is shut down. Otherwise, error messages may occur. In general, the laser emission is switched off automatically when exiting NanoWrite.
2. Shut down the computer.
3. Make sure the objective lens is in the lowermost position. Check for the message `lower z-limit reached` on the touch panel.
4. Switch off the z-drive power supply.
5. Switch off the GT power supply.
6. Check the status of the laser emission; if the backlight of the button `start/stop` is activated, press it to switch off the emission.
7. Switch off the laser controller by deactivating the key switch.
8. Switch off the Photonic Professional GT2 controller by deactivating the key switch.
9. Switch off the Photonic Professional GT2 by turning the mains power switch to position 0.

Warning!

Never switch off the Photonic Professional GT2 Controller or the mains power switch prior to any of the other components. This might cause damage to the system. The emergency shutdown of the system should only be used under dangerous situations. Do not use it as a standard shut down procedure.

Note: A large part of the background information comes from NanoScribe's NanoGuide, Wikipedia, and ZEISS .