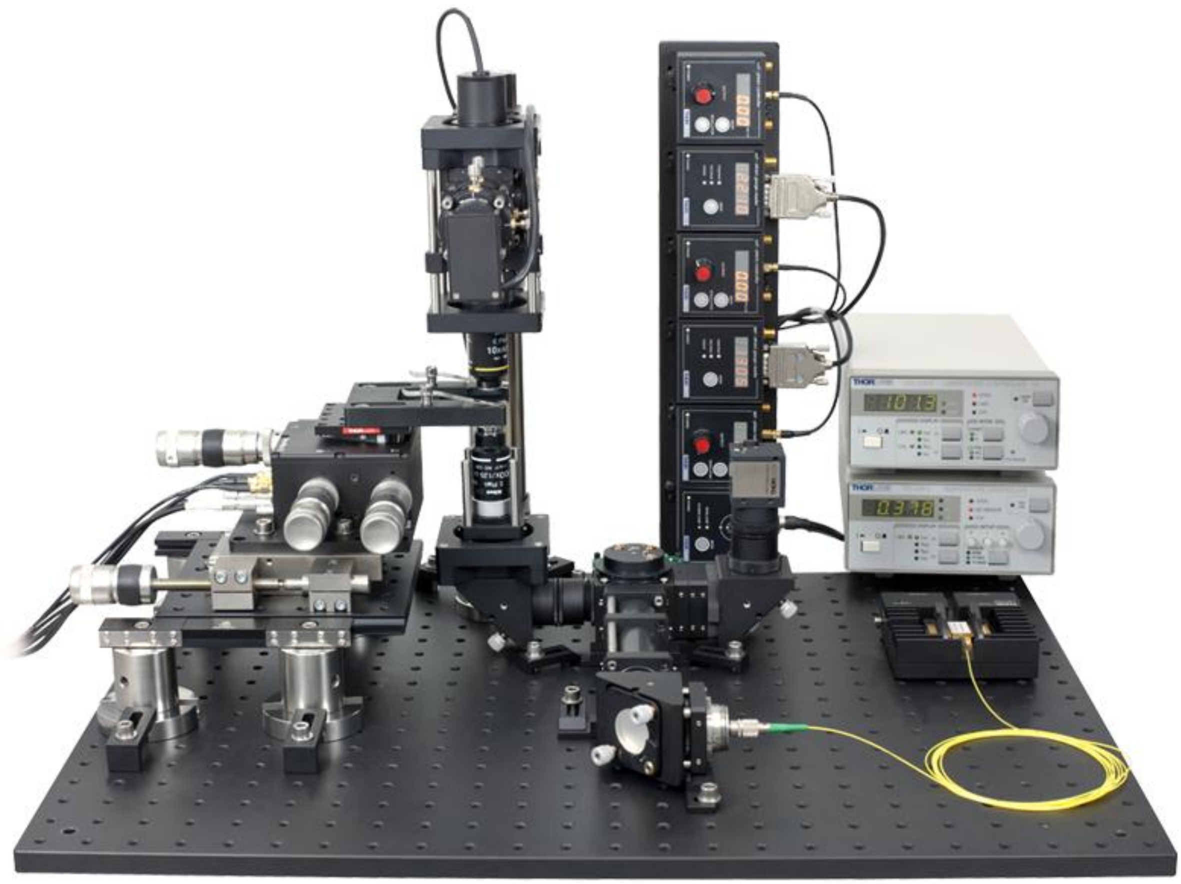
# Optical Trap Kit

## Manual



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# Part 1. Safety

All statements regarding safety of operation and technical data in this instruction manual will only apply when the unit is operated correctly.

**WARNING**

**This unit must not be operated in explosive environments**

**WARNING**

**Make sure the fuse setting at the back of the laser diode driver and temperature controller are appropriate for your region. Failure to do this will result in damage to the controllers. Set TEC and max LD current parameters according to the specifications of the trapping laser diode before operating the Optical Trapping Kit. Failure to do so may result in damage of trapping laser diode. Also refer to the manual for the white light source current driver. Failure to do this may result in damage to the LED.**

**WARNING**

**Latex gloves should be worn to prevent oil from fingers from reaching all optical surfaces. Make sure you use appropriate laser safety glasses during operation.**

**WARNING**

**Avoid Exposure – ASE and laser radiation emitted from apertures.**
**Never look directly in to beam.**

# Part 2. Introduction

In 2007, three researchers in the Department of Biological Engineering at MIT – Steve Wasserman, David Appleyard, and Matthew Lang – built an optical trapping setup for use in teaching labs. Their results were published in the American Journal of Physics [S. Wasserman, D. Appleyard, and M. Lang, *Optical Trapping for Undergraduates*, Am. J. Phys. **75**, (2007)]. Based on their design, Thorlabs has collaborated with the aforementioned authors to design an OTKB optical trapping kit that includes all necessary components and provides the same capabilities. Moreover, since Thorlabs' components are designed to be compatible with each other, the OTKB optical trapping kit is easily modified to provide additional functionalities as your research needs evolve. For example, adding the ability to steer / scan the optical trap position. Alternatively, an excitation light source can be easily added by incorporating a beam splitting cube into the beam path allowing e.g fluorescence measurements. This simple and modular nature of the system allows you to adapt it for a wide variety of applications.

All of the components needed to construct a fully functional optical trapping system are included in this cost effective kit (OTKB contains imperial components while OTKB/M contains their metric counterparts).

# Part 3. Setup Description

## *Optical Trapping Kit OTKB (OTKB/M)*

Like many optical trapping systems, this one is based on an inverted microscope design. The structure of the inverted light microscope is constructed using Thorlabs' 60mm cage system, which is supported by a damped ∅1.5″ post (Item # DP14). All of the parts discussed in this section are included in the OTKB kit unless otherwise noted. Some of the parts mentioned below are not universal so the metric version of the part is used in the OTKB/M kit.

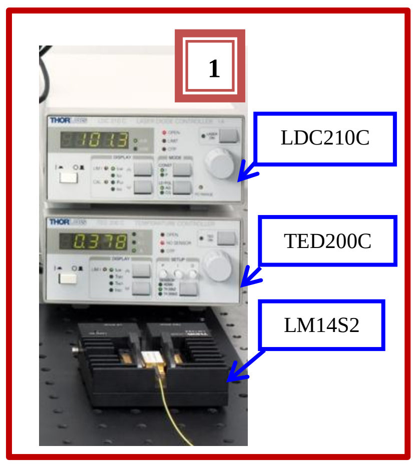



***Figure 3.1: Pigtail laser, mount and controllers***

1) The 980 nm trapping laser source (Item # PL980P330J) is a pigtailed Fiber Bragg Grating (FBG) stabilized single mode laser diode in a hermetically sealed 14-pin butterfly package. The integrated TEC element and thermistor in the butterfly package allow the temperature of the laser to be precisely controlled when mounted in the LM14S2 laser diode mount and controlled using an LDC210C laser diode current driver and a TED200C temperature controller. This laser, mount, and controller combination was chosen to ensure that the output power (330 mW max) of the laser will be extremely stable, which is important to maintaining a constant trapping force.

2) The FiberPort (Item # PAF-X-7-B) collimates the output of the trapping laser. The FiberPort is a versatile collimator since it allows the aspheric collimation lens to be precisely positioned with 5

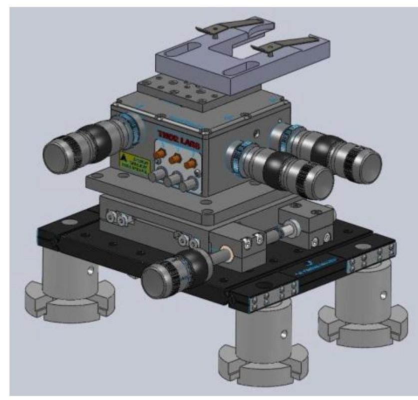
degrees of freedom (X, Y, Z, Pitch, and Yaw). For polarization sensitive applications, the keyway on the FiberPort can be rotated about the optical axis so that the orientation of a linearly polarized collimated beam can be set.

3) The two achromatic doublet lens pairs ACN254-050-B and AC254-150-B expand the collimated trapping laser beam by a factor of 3.

4) The dichroic mirror that is mounted in the central C6W reflects 980 nm light (trapping source) while passing visible light.

5) Visible light from the LED light source illuminates the sample and is then imaged on the 1280 x 1024 pixel color CCD camera (Item # DCU224C) using an achromatic doublet AC254-200-A. The dichroic mirror in the light path in combination with a short pass filter prevents backscattered light from the 980 nm laser from saturating the CCD detector.

6) A 100X oil immersion Nikon objective lens (Item # OTKOBJ) is used to focus the 980 nm laser beam down to form the optical trap. The calculated diffraction limited trap diameter is 1.1µm. A 10X Nikon objective is used as the condenser (Item # OTKCON) and collimates the beam after the optical trap.



7) The sample stage consists of a microscope slide holder mounted to a 3-axis (X, Y, and Z) translation stage that is mounted on a 1-axis long travel translation stage and to a translating breadboard which results in the following capabilities:
    A) 2" (50 mm) and 2.4" (60mm) of travel perpendicular to the beam path. This makes it easy to load the sample and coarsely position it near the trap.  The mounting configuration shown can easily be modified to suit other desired translations.
    B) 4 mm of travel in the X, Y, and Z directions using the modular nanomax differential micrometer drive with coarse knobs providing 0.5 mm/rev.
    C) 300 µm of travel in the X, Y, and Z directions using the differential knobs (50 µm/rev) on the 3-axis differential drives.
    D) 20 µm of travel in the X, Y, and Z directions using the piezo actuators (20 nm resolution without using feedback from the internal strain gauge sensors, 5 nm resolution using the internal strain gauges for positional feedback) on the 3-axis stage. Three T-Cube Piezo Drivers (Item # TPZ001) are included in the kit. (Two T-Cube Strain Gauge (Item # TSG001) are included with the OTKBFM force module.)

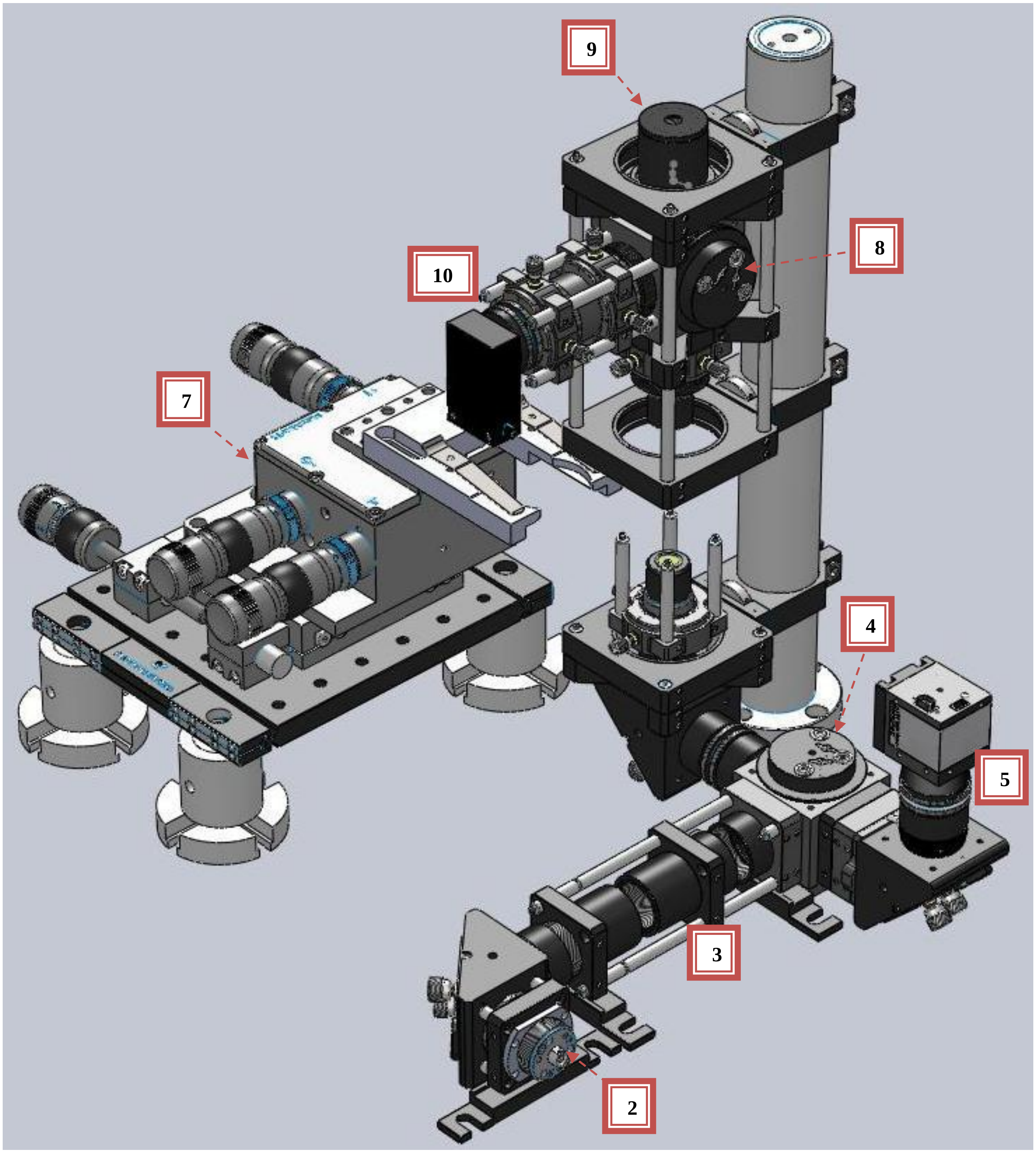



*Figure 3.2: Model of Thorlabs Optical Trapping*

8) The visible light emitted from the LED passes through the dichroic mirror and illuminates the sample while the 980 nm laser beam is reflected towards the optional OTKBFM Force Calibration Module. (If the OTKBFM is not being used, the laser is blocked by a cap, SM1CP2.

9) A single emitter white light LED (Item # LEDWE-10,10$^{o}$ half angle forward radiating) is built into the component OTKB-LS-SP. The light from the LED illuminates the sample.

10) The OTKBFM force measurement module contains the hardware needed to calibrate the trap. The iris at the back focal plane of the objective (used as the condenser) is imaged on the PDQ80A Quadrant Position Detector (QPD) using a 40 mm focal length biconvex BK7 lens (Item # LB1027-B). The detector is silicon based segmented quadrant position-sensing detector with a rise time of 40 ns and a bandwidth of 150 kHz. The signal generated by the QPD is sensitive to the relative displacement of the trapped particle from the laser beam axis. As a result the output of the detector can be used to calibrate the position, stiffness, and force of the optical trap. A TQD001 T-Cube Quadrant Detector Reader is included with this optional force module.

# Part 4. Optical Force Measurement Module OTKBFM (optional)

The OTKBFM module contains the components that can be used to calibrate the trap using positional detection of the back-focal plane of the condenser. By placing the Quadrant Position Detector (QPD) in a plane conjugate to the back focal plane of the condenser, the signal generated by the QPD is sensitive to the relative displacement of the trapped particle from the laser beam axis. As a result the output of the detector can be used to calibrate the position, stiffness, and force of the optical trap. The detector is connected to the cage cube above the condenser as in Figure 3.2. A TQD001 T-Cube Quadrant Detector Reader and two T-Cube Strain Gauge Readers are the main components included in this module. For high bandwidth measurements the QPD signal can be read out from the controller cube directly via a DAQ card (not included).

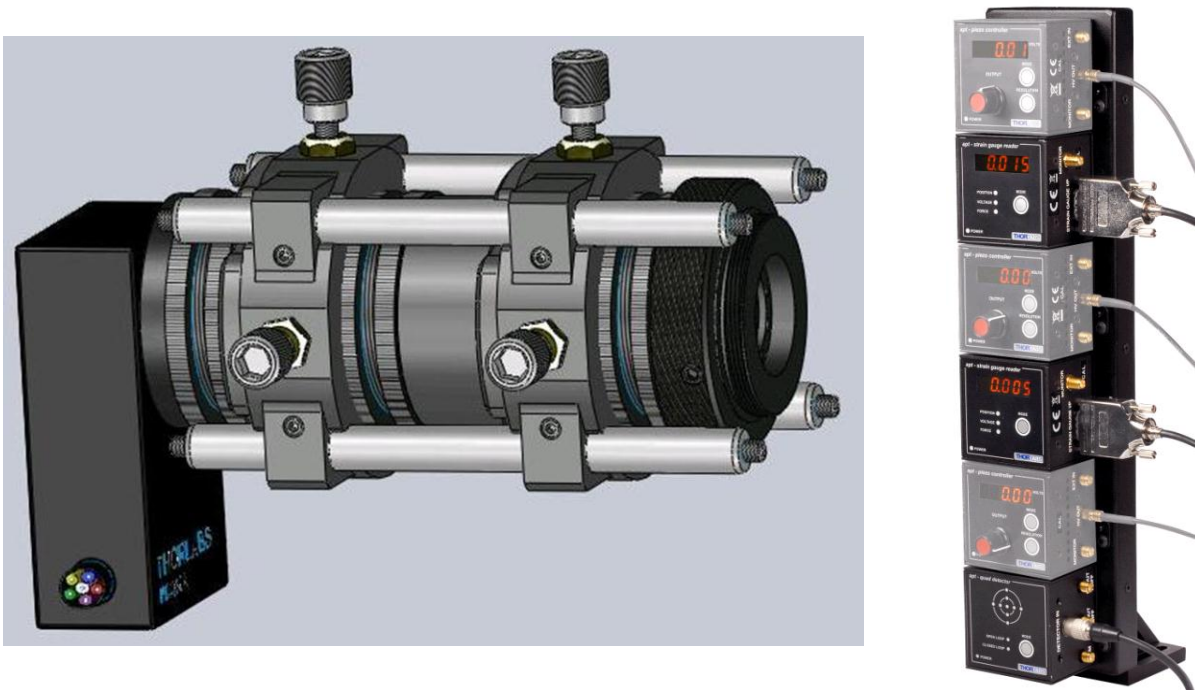


***Figure 4.1: Force Measurement Module OTKBFM consisting of two TGS001, TQD001, PDQ80A and the cage assembly for connecting the detector and optics to the base kit. The shaded controllers and the USB hub TCH002 are included with the base kit OTKB***

# Part 5. Initial Setup and Alignment

Unless otherwise noted, all of the parts mentioned in this section are included. **Please observe proper laser safety procedures. IR laser beams are particularly dangerous because they cannot be seen. Always wear the appropriate type of laser glasses (not included) when working with laser beams. To protect the diode, make sure you have an ESD guard product, like the WS01 (not included).**

The following steps provide a detailed step by step set of instructions on how to assemble the optical trapping kit OTKB. The system can also be ordered preassembled and pre-aligned on a breadboard. Please contact our technical support department (techsupport@thorlabs.com) regarding this option.

1) Select the correct laser type on the laser diode mount (LM14S2). When used with the fiber laser that is part of the kit (PL980P330J) you will have to put the 'Type 1' PCB card into the LM14S2 mount, please refer to the LM14S2 manual for details.

2) Mount the laser diode in the butterfly controller mount and connect to the laser diode controllers LDC210C and TED200C. The temperature sensor type on the TED200C should be "TH 20kOhm" and the Laser Diode polarity should be set to 'AG Ground'. Set the current limit to 700mA. Please check individual manuals for controller, mount and diode for additional information on how to set the temperature of the mount and drive current.

### 5.1 Collimating Trap beam from FiberPort

1) Connect a Cage Plate Mounting Base (CPB1) to the FiberPort Cage Adapter (CP02FP) and attach the FiberPort (PAF-7-X-B) to the adapter. You may want to look at the setup arrangement to know where to mount components on optical table. Secure this assembly to an optical breadboard or table (not included) using two CL6 mounting cleats (if the mounting slots on the CPB1 align with the hole pattern on the optical table they may be used instead of the CL6 cleats).

2) Temporarily attach an iris (SM1D12D) to the CP02FP FiberPort cage Adapter.

3) Temporarily attach two ER6 or ER8 cage rods to the CP02FP (use the top two cage rod holes).

4) Create an eye safe environment prior to turning on the laser and collimating it. In addition, use a low power beam for this alignment step. Turn on the laser and aim the beam at an IR card (not included) on a wall about 5m away. You may need to follow the beam from the fiberport to locate its direction.

5) Adjusts the FiberPort to collimate the beam. (Two people may be required.)

   A) Attach a connectorized fiber source to the bulkhead of the FiberPort and examine the output.

   B) Adjust the X-Y screws to center the output beam in the tilt plate aperture.

   C) Trace the beam away from the FiberPort to check for collimation.

a) For a converging beam (beam comes to a focus): The lens is too far away from the fiber. Alternately turn the Socket Head Cap Screws (SHCS) clockwise in small, equal increments. **Be sure to adjust all screws in equal increments.**

b) For a diverging beam (beam diameter continually increases): The lens is too close to the fiber. Alternately turn the SHCS counter clockwise in small, equal increments. **Be sure to adjust all screws in equal increments.**

D) Check the beam path and adjust the X-Y screws as needed to re-center the beam in the output aperture.

E) Use progressively smaller adjustments until collimation is achieved and the desired beam centration is obtained. Do not force the screws past their normal operating range, if collimation is not easily achieved please contact Tech Support for assistance.

F) Ensure that the beam is parallel to the cage structure by sliding the alignment plate (Item # VRC4CPT) along the ER6 cage (temporarily attached) rods as shown on Figure 5.1.1 (left) with the iris adjusted to its smallest aperture. The intensity of the beam transmitted through the hole in the alignment plate should be constant as the distance the alignment plate is from the iris is changed.

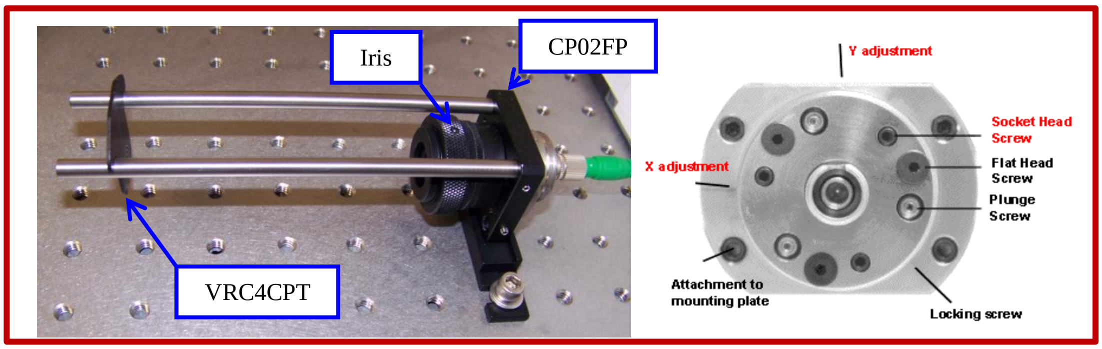



***Figure 5.1.1 (Left) FiberPort Alignment Configuration, (Right) FiberPort showing adjustments***

6) Turn off the laser and remove the ER6 or ER8 cage rods and ring-activated iris.
7) Attach an SM1T2 to the FiberPort Cage Adapter (CP02FP) and remove the retaining ring from the CP02FP. Thread in the SM1T2 and after a few revolutions, lock this to the CP02FP using one of the locking rings on the SM1T2.
8) Attach four ER05 cage rods to a KCB1 right angle mirror mount and secure a mirror, PF10-03-P01 to the KCB1.
9) Connect the KCB1 assembly to the FiberPort assembly through the ER05 rods. The ER05 cage rods should be secured in the CP02FP adapter and the other end of the SM1T2 lens tube coupler should slide into the KCB1 right angle mirror mount. You need not necessarily thread this end inside the KCB1 as the cage rods will hold this combination tightly in place. The beam path from the FiberPort to the KCB1 should be closed. See the ensuing model Figure 5.1.2.
10) Attach the SM1D12D ring iris used previously to the output port of the KCB1.
11) Temporarily attach two ER6 cage rods to the top two tapped holes on the output face of the KCB1.

12) Turn the laser on to ensure that beam is aligned through the center of the iris on the output port of the KCB1 and along the cage structure using the VRC4CPT as previously illustrated in Figure 5.1.1. Remove this KCB1 with mirror and put it aside as it will later be used for the coupling to the vertical path of the microscope.
13) Use a second KCB1 with a second mirror, PF10-03-P01 and align in the same way as in steps 10-12 above. The completed model is shown in Figure 5.1.2.

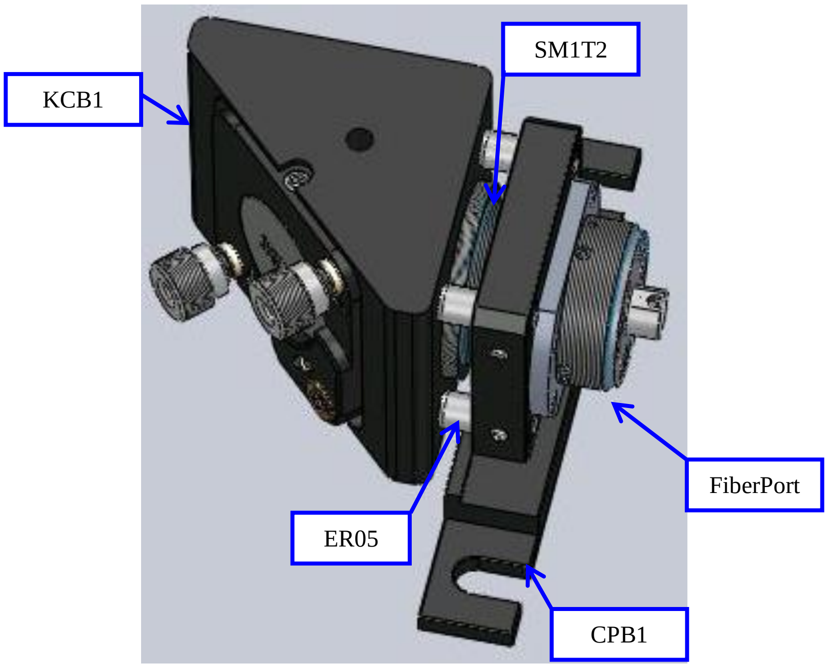



***Figure 5.1.2 Adjusting the Right Angle Mirror Mounts KCB1 and connection to Fiberport.***

## 5.2 Beam Expander Section

1. Mount a CP02 onto a CPB1.
2. Mount the achromatic doublet ACN254-050-B into an SM1V10 lens tube and thread it into the CP02 (from the previous step), such that external retaining rings on the SM1V10 are on both sides of CP02. Make sure that the lens is mounted in the lens tube so that the collimated beam from the FiberPort is incident on the curved surface of the doublet.
3. Connect an SM1T2 to the KCB1 from the previous section. Get a couple of threads into the KCB1 and lock to the KCB1 using one of the locking rings on the SM1T2.
4. Attach the SM1V10 from step 2 to the SM1T2 and lock these together using the other locking ring on the SM1T2.
5. Thread an SM1L10 on the other side of the CP02 holding the SM1V10 and first lens. The lens tube will thread into the SM1V10 also. The SM1L10 should touch the other face of the CP02 as shown in Figure 5.2.1.
6. Clamp the CPB1 attached to the CP02 to table using the miniature rail clamp CL6.
7. Attach another SM1L10 to a new CP02.

8. Mount the second lens, AC254-150-B to a half inch long lens tube, SM1L05.Thread this lens tube holding the lens unto the other side of the CP02 above. Note that the lens should be positioned in lens tube such that the trap beam goes through lens from the concave side.
9. Mount a CP02T thick Cage Plate on a CPB1 Cage Plate Mounting Base. Attach a new SM1L05 to one side of the CP02T.
10. Connect an ER4 cage rod to an ER1 cage rod to produce a 5″ long cage rod. Repeat so that you have two 5″ cage rods. You may remove the threaded studs from the ends of the assembled 5″ cage rods.
11. Connect the assembly from steps 7 to 9 together using the rods created in step 10 as shown in Figure 5.2.1. The cage rods are located diagonally from each other. In this arrangement, you have the flexibility of manually adjusting the position of the second lens along the optical axis over a range of approximately 12mm. If you want to enclose the beam completely (which we recommend), then you can put segments of the lens cover SC1L24 over the two open sections in Figure 5.2.1. You will need a two inch long cut (for the first section) and a one inch long cut (for the second section).

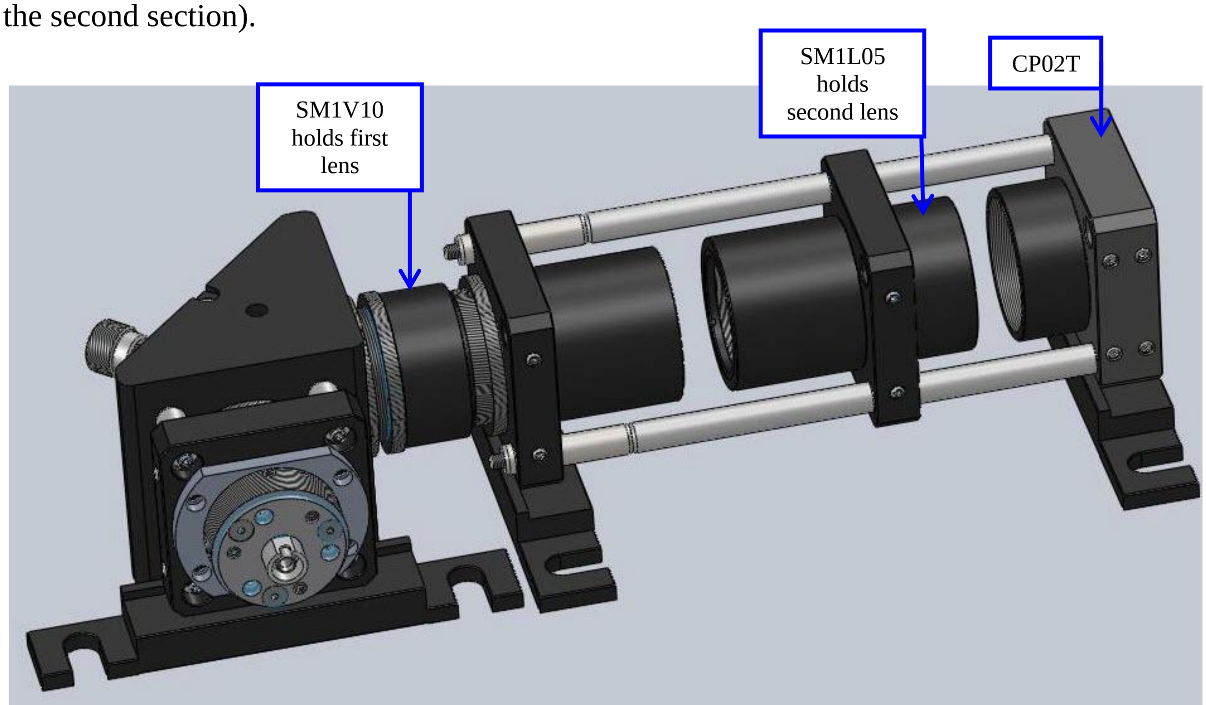
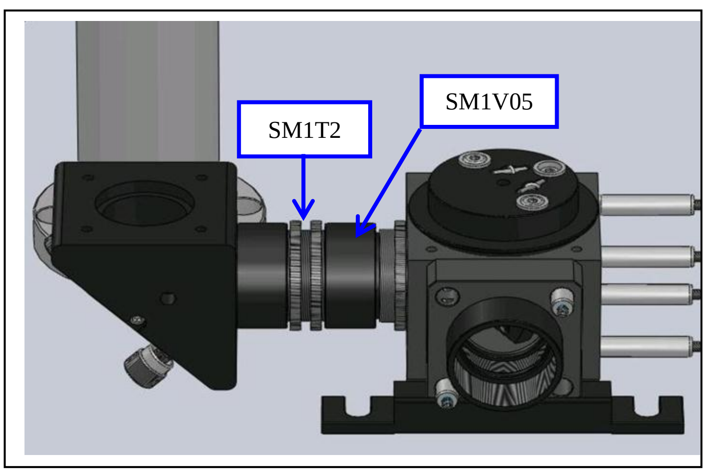



*Figure 5.2.1: Building the beam expander segment*

12. Close the bottom of a C6W cube using a B1C.
13. Remove the studs from one end of two ER1 rods and fully thread the ends with studs onto the diagonal (as shown in Figure 5.2.2) of the C6W from previous step. Connect this C6W to the CP02T from Step 10 using the two ER1 cage rods. The cage rods should be located diagonally across the cage from each other. Do not use the same orientation that was used in Step 11.
14. Temporarily attach two ER6 cage rods to the C6W on the opposite (rear) side of the cube. Use the upper mounting holes.
15. Use an iris on the cube and a VRC4CPT alignment guide resting on the ER6 cage rods to ensure that the beam is still aligned to the center of the cage system. If the beam is off, you may need to correct this by small adjustments on the KCB1 mirror.
16. Remove the temporary ER6 cage rods and close the port on that face of the C6W cube using an SM1CP2.

17. Mount the FM01 hot mirror into a B5C. To ensure that the FM01 is mounted correctly, hold the FM01 to white light and observe both surfaces. The surface that shows more red reflections should be mounted such that it reflects the trap beam.
18. Mount the B5C onto a B4C. The B5C should be mounted such that the hot mirror is positioned at the center of the B4C.
19. Insert the B4C into the top of the C6W and coarsely align it so that the beam is reflected 90˚ to the left. If the B5C is mounted at the appropriate hole on the B4C, and the coarse adjustment is correct, the kinematic adjusters on the B4C should be aligned along the diagonal of the C6W as shown in Figure 5.2.2. Use the four screws that came with the B4C to connect it to the C6W such that it can still rotate on top of it.
20. Thread an SM1V05 completely into the C6W on the face at which the trap beam exits.
21. Temporarily attach two ER6 or ER8 cage rods to this side of the C6W and place the cage system VRC4CPT alignment guide on the rods.
22. Adjust the B4C so that the reflected beam is aligned to the center of the cage system. Do this in two steps. First connect an iris, SM1D12D to the SM1V05. Then rotate the B4C on the C6W until the beam is going through the iris when it is almost closed, and through the center hole of the VRC4CPT. Then use the kinematic adjusters if necessary on the B4C to optimize the alignment and ensure that you have the maximum possible intensity going through both pinholes. Remove the ER6 cage rods and SM1D12D.
23. Attach an SM1L05 to the previously adjusted KCB1 from step 12 in section 5.1.
24. Attach an SM1T2 to the SM1L05, getting a couple of threads inside and lock to the SM1L05 using one of the locking rings.
25. Connect four ER3 rods to the upper face of the KCB1. Slide an LCP02 into these ER3 rods and tighten the set screws once the faces of the KCB1 and LCP02 are in contact as shown in Figure 5.2.2 (right). Note that the 4 ER03 rods will slide though LCP02 when the set screws have been slightly released.



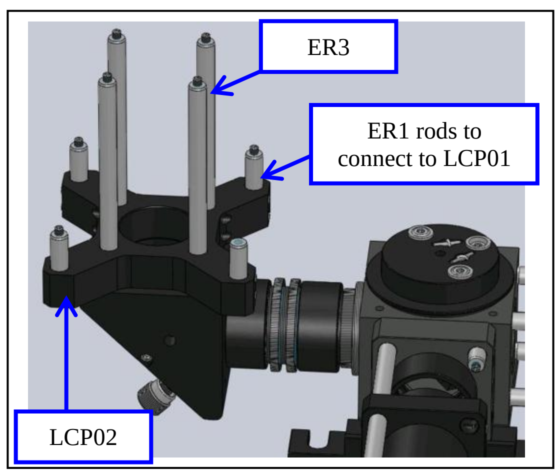




***Figure 5.2.2: (Left) Assembly between the cage cube and the KCB1 for the vertical segment. (Right) ER3 cage rods and LCP02 for vertical segment.***

### 5.3 Vertical segment

1. Attach a C1500 clamp to an LCP01 (ensure that the C1500 face is parallel to the LCP01 before locking) and slide-hold to the Dynamically Damped 14” post, DP14.
2. Stack the LCP01 on top of the LCP02 and connect them using four ER1 cage rods as in Figure 5.3.1 (you may remove the threaded studs from both ends of the cage rods).
3. Connect the SM1T2 to the SM1V05 from Step 24 in section 5.2. The SM1V05 will unthread from the C6W as it threads onto the SM1T2. Lock it in place using the locking ring on the SM1V05 to the C6W. Also lock the SM1T2 to the SM1V05 using the other locking ring on the SM1T2. The resulting arrangement is shown in Figure 5.3.1.
4. Lock the C1500 to the DP14.
5. The DP14 Post can now be locked down to the table (using CL5 clamps) along with the CPB1 bases (using CL6 clamps) when the holes in the bases do not align.
6. Temporary attach an SM1D12D to the LCP02.
7. Temporarily attach four ER8 cage rods to the ends of the four ER1 cage rods used to hold the LCP01 and LCP02 plates together.
8. On the other end of the ER8 cage rods attach an LCP02.
9. Attach an additional SM1D12D iris to the upper LCP02 as shown in Figure 5.3.1.
10. Use this arrangement to align the beam to the center of the vertical cage segment. Make sure you use protective glasses when doing this. The B4C is used to align the beam through the lower diaphragm while the KCB1 is used to align the beam through the upper diaphragm. Neither should require much adjustment. First the lower iris is wide open. Then you gradually close this and if necessary, use the kinematic adjusters to ensure the beam goes through the center. In most cases, just small adjustments on the adjusters on the B4C will center the beam. When the lower iris has its smallest aperture, you should still see the beam going through after your adjustments. Make sure the beam through the iris is optimal. Then fully open this iris and align the beam through the upper iris. Start when the upper iris is fully open, and gradually decrease the aperture while adjusting the beam position with the knobs on the KCB1 until you have the most transmission through smallest aperture size of the upper diaphragm. Now close the lower diaphragm also and ensure that you have optimal transmission through both irises, when they are at the smallest apertures. It may be necessary to iterate this alignment before you achieve the maximum transmission through both smallest apertures.

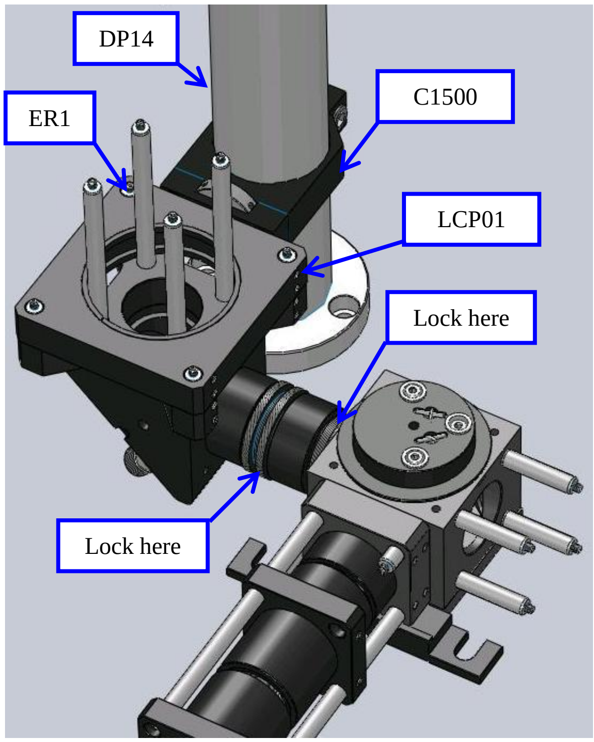



**Figure 5.3.1: The vertical segment during alignment.**

***Centering of the beam along this path is very critical. A Thorlabs power meter (not included) with an SM1 threaded measurement head (e.g., PM100D with S121C) will allow you to verify the alignment by maximizing the transmitted power when both apertures are nearly closed. As a guideline, using the 330mW laser PL980P330J, at a drive current of 100 mA and the diaphragms 8″ apart, both SM1D12D closed, the measured power should be around 0.7 mW.***

11. Thread the SM1A10 C-Mount to SM1 adapter onto the Nikon objective.
12. Slide in a 30mm cage assembly XY translating lens mount, HPT1 unto the ER3 rods.
13. Now mount the objective on to the HPT1.

***As a guideline, with the S121C sensor placed about 0.5″ above objective (see Figure 4.3.3), with diaphragm below objective fully open, the measured power should be around 6mW or more when driving the PL980P330J with 100 mA of current (14 mW at 130 mA drive current).***

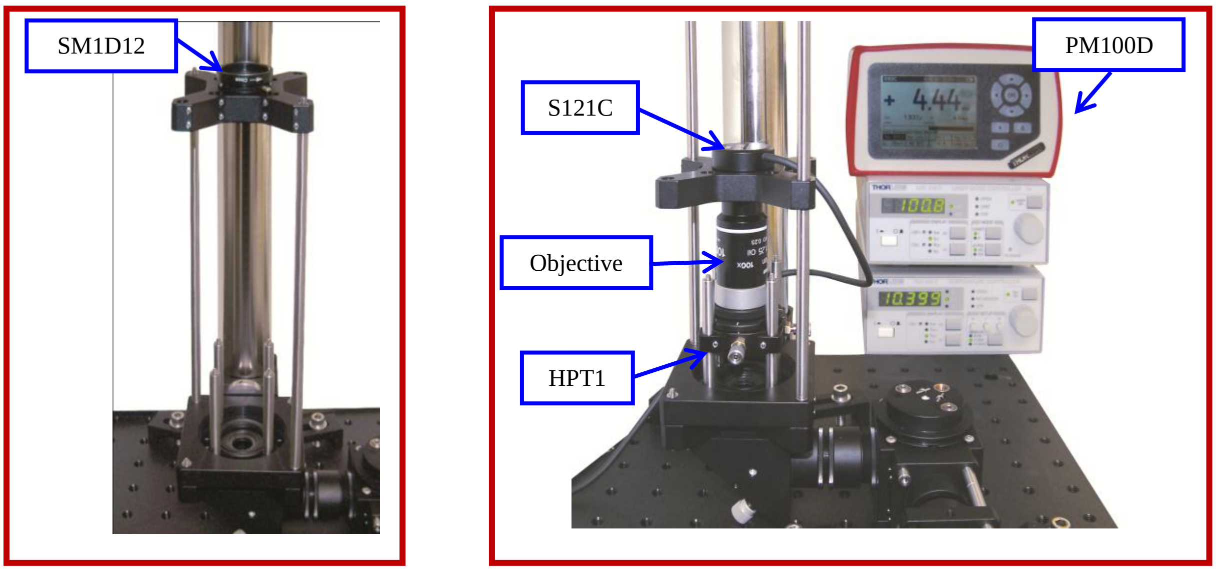



***Figure 5.3.2: (Left) Vertical bull's-eye alignment of beam using ER8 rods, a lower and upper iris. (Right) Example alignment optimization through power meter measurements.***

### 5.4 Mounting the Condenser

1. Mount another FM01 hot mirror to a B5C. To ensure that the FM01 is mounted correctly, hold the FM01 to white light and observe both surfaces. The surface that shows more red reflections should be mounted such that it reflects the trap beam.
2. Mount the B5C onto a B4C. The FM01 should be mounted such that the mirror face is along the center of the B4C.
3. Remove the threaded studs from four ER05 cage rods.
4. Place an LCP02 on a flat surface and insert the four ER05 cage rods in the 30 mm cage rod holes and secure them at such a level that only one of the two set screws in each hole of the LCP02 is used. This will give you enough rod length to connect to a C6W.
5. Connect a C6W cube to the LCP02 from Step 4. Lock the cube onto the ER05 cage rods after ensuring that the faces of the C6W and LCP02 are in contact.
6. Attach four ER1 rods to the top of the C6W above and lock cube onto ER1 rods. Make sure that the ER1 rods don't extend into the C6W such that they may interfere with the B4C rotation.
7. Connect another LCP02 to the four ER1 rods as shown in Figure 5.4.1 and lock these using the set screws on the LCP02.
8. Insert the ensuing assembly from step 2 above on one side of the C6W and coarsely align it so that the beam is reflected 90˚ to the left (if you look at Figure 5.4.1, reflection will be out of

paper as indicated by red arrow). If the B5C is mounted at the appropriate hole on the B4C, and the coarse adjustment is correct, the kinematic adjusters on the B4C should be aligned along the diagonal of the C6W as can be seen in Figure 5.4.2. Use the four screws that came with the B4C to connect this to the C6W.

9. Close off the other side of the C6W cube using a B1C, and the back with an SM1CP2.

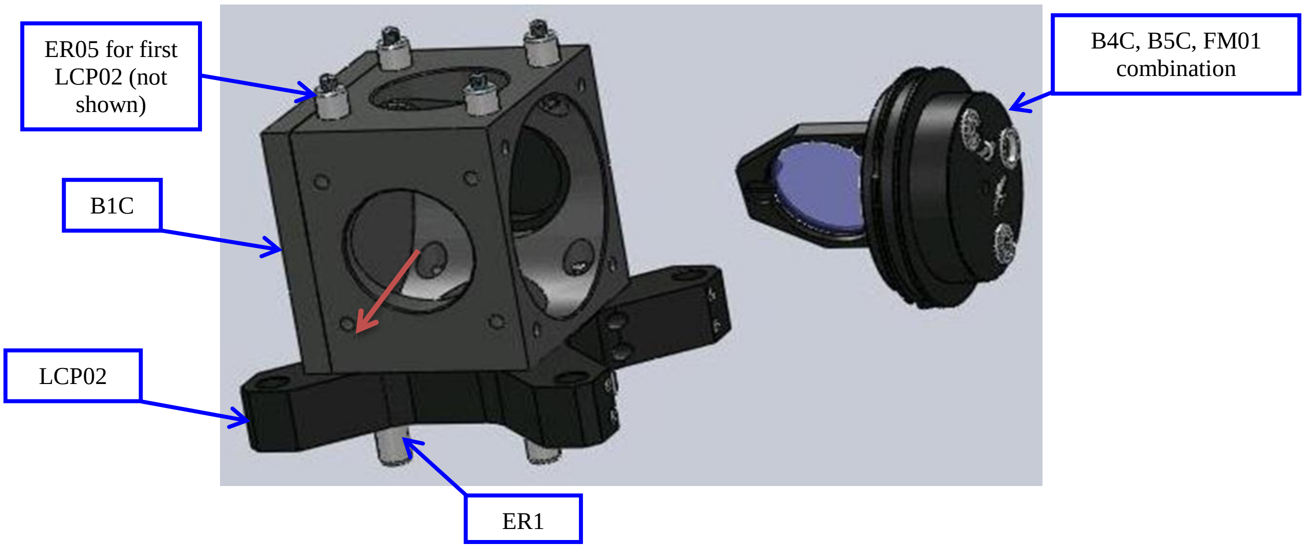



*Figure 5.4.1: Building the condenser cube section*

10. Connect an HPT1 to the four ER1 rods at the bottom of the assembly above, and lock HPT1 to assembly through the set screws. Make sure the adjusters on the HPT1 are oriented as shown in Figure 5.4.2.
11. Attach an SM1D12D to the XY translating face of the HPT1. This face has a line marker for each adjuster. The condenser will be connected to this iris.
12. Attach a C1500 clamp to an LCP01 (ensure that the C1500 face is parallel to the LCP01 before locking).
13. Repeat step 12 above with another C1500 and LCP01.
14. Using two of the four ER6 cage rods, connect the two assemblies from steps 12 and 13 to the C6W assembly as shown in Figure 5.4.2. Use the holes on the side with the C1500. The other holes will be used for alignment with the lower vertical segment.

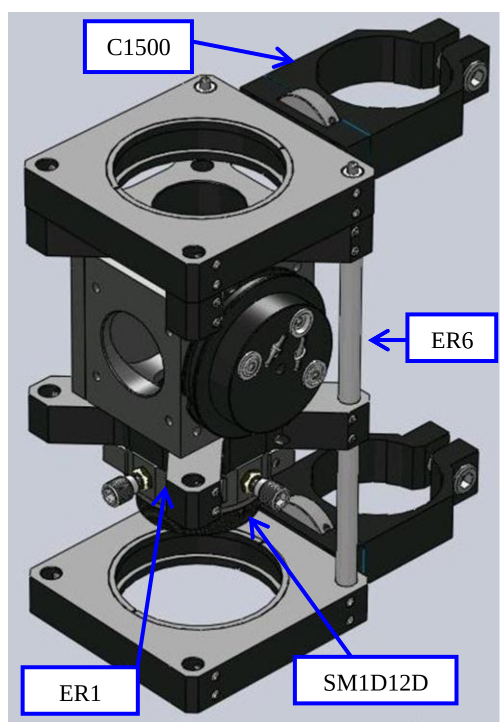



**Figure 5.4.2: The complete condenser segment**

### 5.5 Connecting to the vertical post DP14

15. Slide the assembly from step 14 in section 5.4 onto the DP14.
16. Connect two ER10 rods through the upper vertical segment to the lower vertical segment as shown in Figure 5.5.1 (Left). Use the two other holes that don't have ER6 rods. Then lower the upper assembly to the appropriate height depending on your stage and sample. If you already mounted the condenser (Item # OTKCON and SM1A10), lower this assembly such that that the tip of the condenser is about 6-8 mm above the upper surface of the sample slide.
17. Lock both C1500 to the DP14 as shown in Figure 5.5.1.
18. Use the other two ER6 rods to complete the upper vertical segment as shown in Figure 5.5.1 (right).
19. If you don't have to install the force module, then close the open end of the C6W with an SM1CP2.

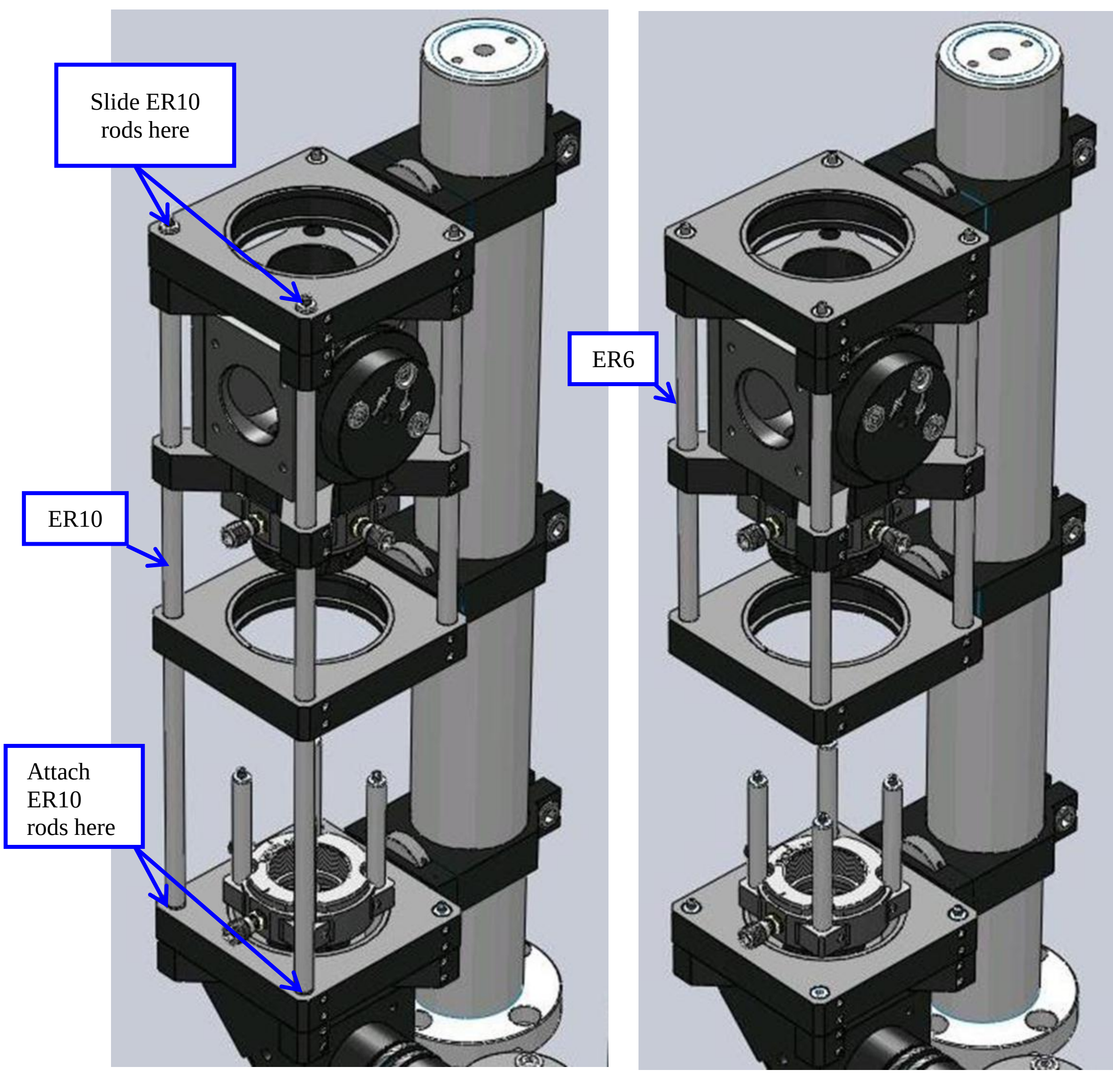



***Figure 5.5.1 (Left) Aligning the upper vertical segment to the lower vertical segment using ER10 rods. (Right) Completing the upper vertical segment using two ER6 rods.***

20. Remove one half of the SM1Q quick release adapter from the OTKB-LS-SP light source and attach it to the upper LCP02 cage plate.
21. Mount the OTKB-LS-SP LED source onto the LCP02.

### 5.6 The Camera Segment

1. Connect four ER1.5 rods to the KCB1.
2. Slide in a CPG3, then a CP02T (holding the imaging lens AC254-200-A) and a CP02.
3. Connect this to the central C6W by using the set screws in the C6W to tightly hold the four ER1 cage rods. Only a small part of the cage rods should extend into the C6W, otherwise the rods may not allow you to freely rotate the FM01 mirror after it has been installed inside the C6W cube The ensuing assembly is shown in Figure 5.6.1.
4. Mount the IR filter, FES0750 in an SM1L05.
5. Thread in an SM1D12 to the vertical face of the KCB1, then the SM1L05 from step 4, then an SM1T2, and the adapter SM1A9 (which comes with the camera).
6. Attach the DCU224C CCD camera to the SM1A9 as shown in Figure 5.6.1.

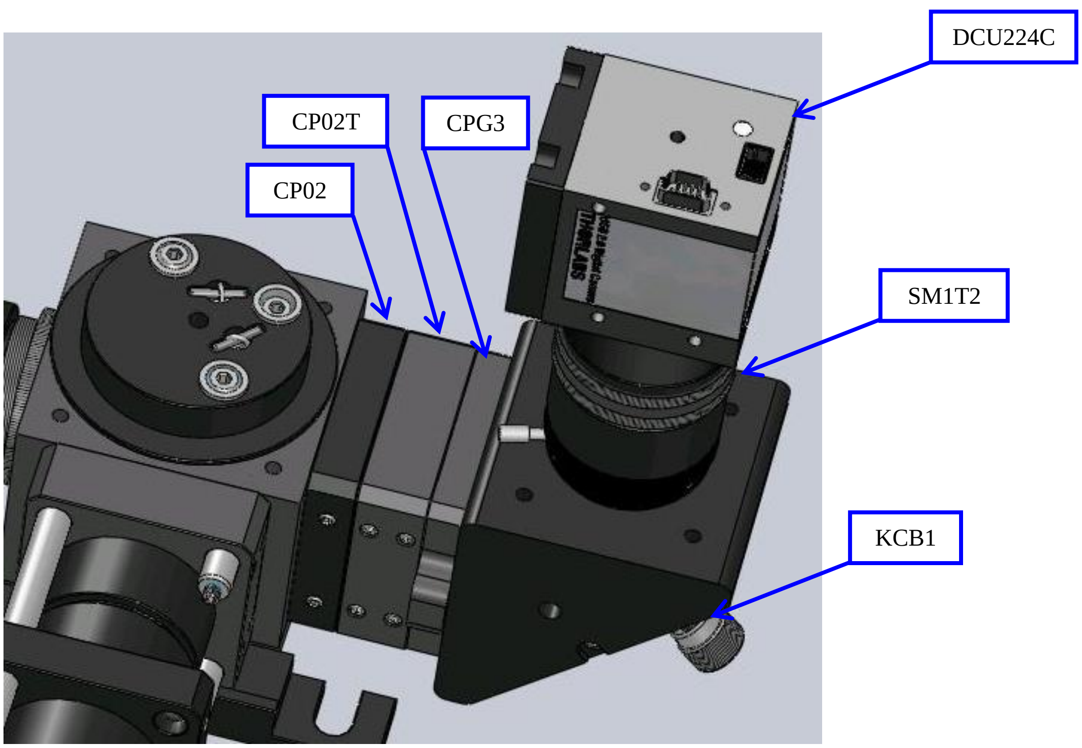



***Figure 5.6.1: Building the camera segment.***

Your trapping system is now complete and you can proceed to build the sample stage. If you use the force measurement module, see the integration instructions which were provided with the OTKBFM.

## 5.8 Sample Stage and Holder

1. Mount the TBB0606 translating breadboard onto the post assemblies P1.5 and base PB2.
2. Mount the LNR50D to the translating breadboard as shown in Figure 5.8.1.
3. Mount the MAX301 stage to the LNR50D. Three holes will line up and 2 can be conveniently used.
4. Remove the top plate (the plate has an array of mounting holes and two groves that split the plate into four quadrants) from the MAX301 stage (four screws, use a 2 mm hex key)
5. Attach the RB13P1 to the MAX301 and connect the slide holder (OTKSH) to the RB13P1. Attach the three DRV002 drives.
6. Since the oil immersion objective might push the slide from the bottom during the imaging process, the clips are used to hold the slide in place.

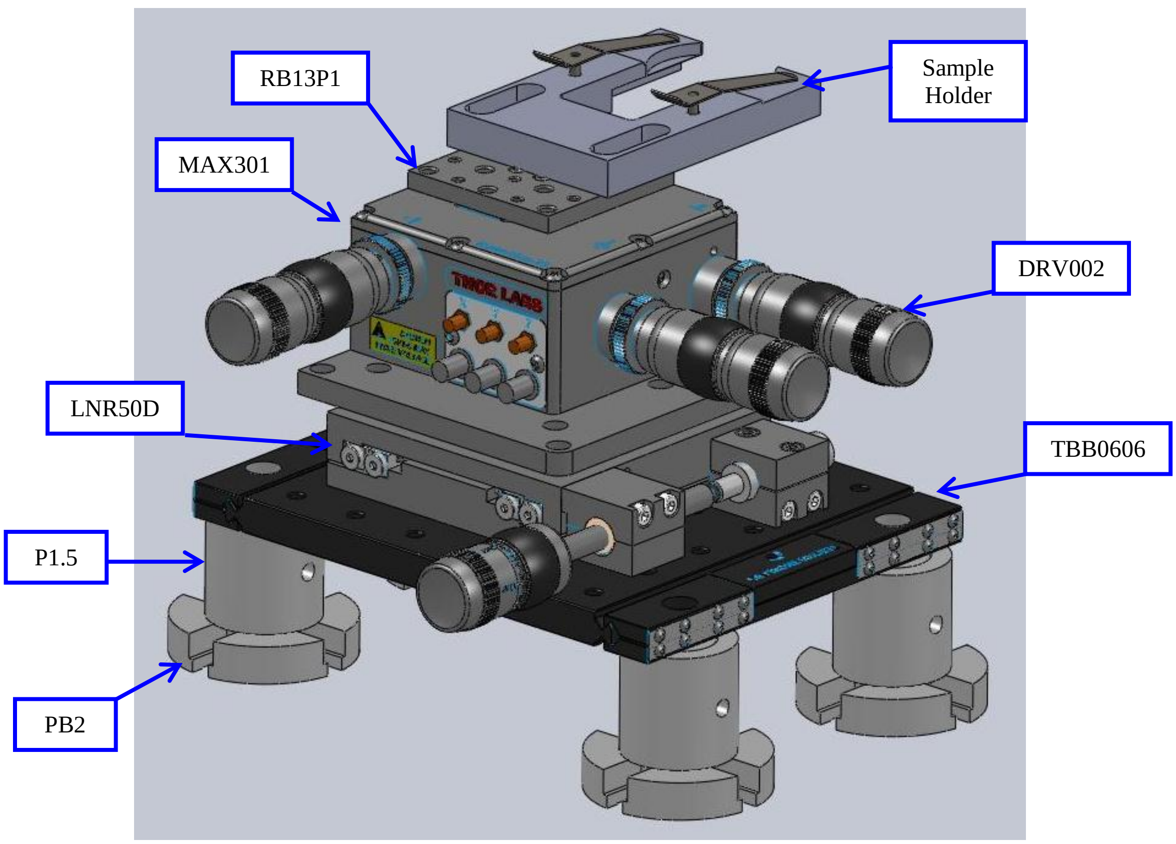



***Figure 5.8.1: The sample stage and holder***

## 5.9 The Light Source

1. Remove one half of the SM1Q quick release adapter from the OTKB-LS-SP light source and attach it to the upper LCP02 cage plate. Then connect the other half of the light source and lock in place by rotation.
2. Connect the 250mA precision constant current laser driver, LD1255R to the optical table mounting plate LD1255P, and hold this in place using the screws.
3. Mount the assembly from step 2 to the optical table using cap screws and the provided slots on the LD1255P.

4. At this step, it is important to refer to the manual provided for the LD1255R (http://www.thorlabs.com/Thorcat/0000/0009-D01.pdf). Failure to follow the instructions there may damage the LED light source.
5. Make sure you have the correct power supply line voltage for the LD1255R, i.e. PS-12DC-US for 110V (or PS-12DC-EU for 230V).
6. Turn the Current Control Potentiometer (see LD1255R manual) a full 12 turns counter clockwise to ensure the LED current is at a minimum. As a quick guide, the LD1255-CAB power cable connects to the power supply at one end and to pins 1, 2 and 3 on the LD1255R (please see LD1255R manual). The power cable from the LED light source connects to pins 6 and 7 on the LD1255R with correct polarity, i.e. the side of the connector on the cable which is marked with a '1' will connect to pin 6 (please see LD1255R manual).
7. Now gradually turn the Current Control Potentiometer and the LED should light up after a few turns. Otherwise, you may have to check the polarity of the light source connector on the driver board.

### 5.10 Thorlabs Optical Trapping Test Accessories

Thorlabs provides a set of accessories that allow the user to get started using the system and verify proper operation. This can be ordered as the item OTKBTK. This set includes the following items:

1. A sample of non-drying immersion oil for microscopy, Cargille Type B, Cat. No. 16484.
2. One plastic slide, with build in channel with height of 400 microns and volume of 100 micro liters. The bottom matches coverslip number 1.5.
3. A mini pipette with a 50 micro liter volume.
4. Some non-functionalized fused silica beads in deionized water, 1 micron in diameter, Bangs Laboratories, Inc., catalog item: SS04N/9857. Recommended storage temperature is 2 - 8$^{o}$C.

# Part 6. Software Package

The optical trapping kit comes with the standard software which is included with the electronic and nano positioning parts, i.e. it includes the APT software package to control the sample positioning stage and to read out the quadrant detector signal as well as the application software for the DCU camera. The kit does not include any routines that will analyze the data and calculate force/stiffness values. The ActiveX based software modules can be used to develop custom applications (e.g. using LabWindows CVI, Visual C++, Matlab, Visual Basic, HPVEE, any programming software that supports ActiveX). A procedure of how to approach this data analysis can be found in 'Calibration of optical tweezers with positional detection in the back-focal-plane, Review of Scientific Instruments 77, 103101, 2006'. The screen shots on the right hand side show the QPD and Strain Gauge Controller software on top (included with OTKBFM) and the piezo controller software and CCD camera software at the bottom (included with OTKB (OTKB/M)

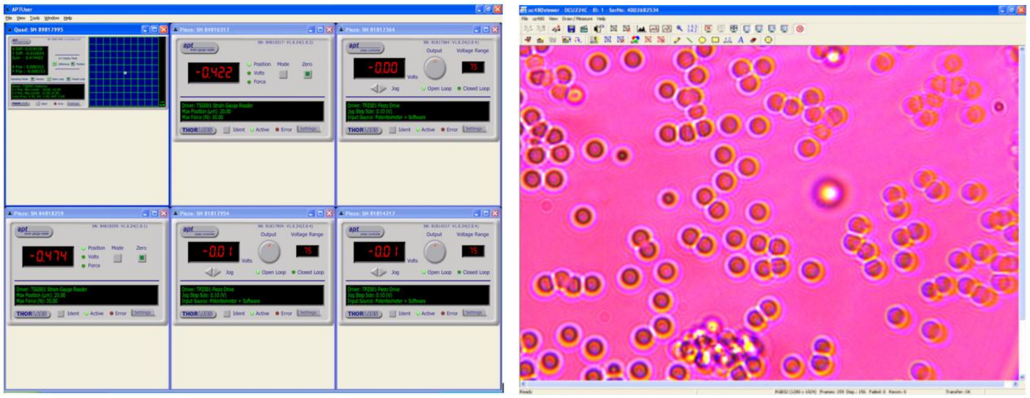


***Figure 5.1 (Left) Application Screen (Right) CCD camera application, 1 μm Silica beads. The bright spot near the center of the picture is the trapped bead.***

## Part 7. Sample preparation and measurement

For initial testing we recommend 1um or 2um silica beads. The sample solution loaded into the channel using a microscopy slide with build-in channel (offered via our optical trapping accessories kit) or you can build a simple channel by placing double sided tape on a standard slide, put liquid in-between and add a cover glass on the top. Place the slide onto the sample holder and carefully place the slide between objective and condenser. Make sure to either use immersion oil on the bottom of the slide or to apply it to the objective before trying to image the sample.

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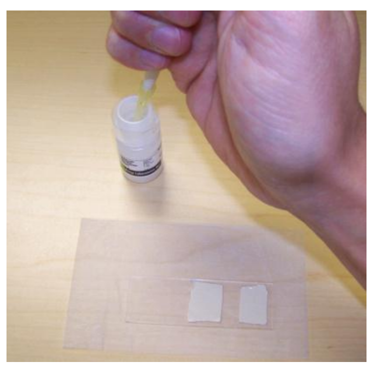

***Figure 6.1 Beads are diluted using a pipette and placed between strips of double sided tape***

Now raise the HPT1 holding the Nikon objective upwards. As soon as you see that the immersion oil gets in contact with the cover glass you will need to use the smallest possible adjustments up and down until you see a picture of the beads. Using the set screws, tighten the HPT1 to the ER3 cage rods as close to the focus position as possible and then use the height adjustment on the sample stage to get the beads into focus.

# Part 8. Additional Accessories

Following parts will be helpful to setup and operate the trap. They are not part of the ‘Optical Trap Kit’:

a) Immersion Oil, e.g. Cargille non-drying immersion oil for microscopy, Type B, Cat. No. 16484. **Immersion oil is required to able to use the Nikon Oil Immersion objective.**

b) Power meter **PM100D** with **S121C** measurement head.

c) Appropriate laser goggle, like the **LG1** – (Laser Safety Glasses, Light Green Lenses, 59% Visible Light Transmission)

d) Fiber coupled laser source emitting in visible range, like the **S1FC635**.

e) Metal Grounding Wrist Strap like the **WS01**

f) Precision Right Angle Plate to mount T-cube hub TCH002, **AP90**

g) To build this on a breadboard, you would need the **MB1824** – (Aluminum Breadboard, 18" x 24" x 1/2", 1/4"-20 Threaded) for imperial or the **MB4560/M** – (Aluminum Breadboard, 450 mm x 600 mm x 12.7 mm, M6 Threaded) for metric.

# Part 9. Summary of Key Features and Specifications

## Features

- Trap Laser Source
  - SM Fiber Coupled DFB Laser, 14-Pin Butterfly Package
  - 975 nm, 330 mW (Max)
  - Integrated TEC Element for Temperature Stabilized Output
  - LD Controller and Mount Included
- Nikon 100X Oil Immersion Objective
- Inverted Light Microscope Design
- 3-Axis Sample Positioning Stage
- CCD Camera with USB Interface for Video Imaging
- Position-Sensing Detector Module Available

## Specifications

- Trap Force: ~1 pN (with Trap Laser Output Power: 16 mW)
- Spot Size: 1.1 µm
- Depth of Focus: 1 µm (Estimate)
- Power at Optical Trap: ~42% of Trap Laser Output Power
- Max Trap Laser Output Power: 330 mW (at fiber end).
- Nikon 100X Oil Immersion Objective
  - 1.25 NA
  - Ø5 mm Back Aperture
  - Working Distance: 0.23mm
  - Transmission: 380-1100nm
  - Recommended Cover Glass thickness: 0.17mm
- Beam Diameter at Back Aperture of Nikon Objective: Ø4.74 mm
- Nikon 10X Air Condenser
  - 0.25 NA
  - Working Distance: 7mm
  - Transmission: 380-1100nm

# Part 10. Thorlabs Worldwide Contacts

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