Optical alignment: a beginner's guide for microscopy homebuilders

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Abstract

Precise optical alignment is critical for high-quality microscopy imaging, especially in custom-built systems. This guide details essential alignment steps, from beam path setup to lens collimation and centring, introduces key tools like pinholes and shearing interferometers, and advocates for a modular construction approach. This article aims to empower microscopy enthusiasts with practical alignment techniques and encourages ongoing learning and knowledge-sharing via an easy-access online resource on YouTube.

Introduction

Aligning optical components in custom-built microscopy systems is challenging due to the mix of non-standardized parts and the absence of commercial system guidelines. Loose mechanical tolerances and mismatched components often lead to complex, error-prone alignment processes without the aid of built-in tools. Misalignments can significantly degrade image quality, affecting high-resolution techniques like super-resolution microscopy. This article aims to simplify optical alignment for homebuilders by sharing fundamental principles and practical tips gathered from experience across different labs.

Beam path setup

The optical axis is crucial in an optical system, serving as the guideline for aligning all components to ensure high-quality imaging. In practical terms, it's defined by a well-collimated laser passing through strategically placed pinholes along the beam's path. Aim to maintain a constant and relatively low beam height throughout, which can be verified using a template pinhole on a fixed-height post that is movable.

Keep the beam path as linear as possible to reduce alignment errors and system complexity. To accommodate the finite optical table length, use fold mirrors or double-purpose use other reflective elements like dichroic beam splitters. Right-angle turns are usually best to avoid polarization issues. Also, employ pairs of mirrors mounted on kinematic mounts for each change in direction.

Alignment of the beam path from one segment to the next is then an iterative process with two simple steps (Figure 1). The first mirror aligns the beam through the first pinhole, the second mirror through the subsequent one (first vertical then horizontal). Iterating between these adjustments converges to an aligned beam path, making it straightforward for beginners.

To help with other alignments, adjustable irises instead of fixed pinholes are great; open irises allow visibility when the beam is off-centre and permit quick adjustments to the beam diameter. A final tip: position fold mirrors at lens focal points, which greatly simplifies daily alignment - a boon for maintaining consistent performance.

Pre-alignment procedures

Begin optical alignment by thoroughly cleaning all components to remove dust and contaminants, using lens tissue and isopropyl alcohol to prevent surface damage. Handle elements with gloves to avoid oil transfer and protect against cleaning agents. For delicate items like gratings, consult the manufacturer's instructions for proper care.

Next, mount and secure optical elements using systems that ensure both stability and adjustability. Cage systems offer a rigid structure for alignment and component protection, with adjustments mainly along the beam path. Note that this also means that you are left at the mercy of the manufacturer's precision regarding the centration of optical elements in their housings and the straightness of the optical cage. Post-mounted systems provide more flexibility and ease of reconfiguration, suitable for setups requiring frequent changes. They allow for both fixed-height positioning and kinematic adjusters for precise alignment. The choice between shimming for stability and adjustable mounts for adaptability depends on the specific needs and preferences of the user.

The last step before actual alignment is to roughly place components along the optical path, checking their sequence and orientation rather than seeking precise alignment. The actual alignment begins with a clear beam path. Sequentially add components, starting from the light source or the objective mount.

Alignment of lenses

In this article, let us exemplarily look at the practical alignment of lenses. The two essential tasks hereby are centring and collimation. Here's how both can be performed when using a post-mounted system. Collimation is the process of aligning a lens so that the light exiting it is parallel, a critical property for beam propagation over long distances or when inserting additional optical elements downstream. It generally goes hand in hand with centring, which ensures that light passes straight through the optical centre of the lens. It is vital for maintaining the quality of the beam as rotational misalignment causes astigmatic aberrations, while decentred lenses cause considerable coma. Let us begin by axially positioning the lens such that the outgoing light is collimated (a practical example would be the collimation of a fibre laser output or the second lens of a beam expander telescope). On the far side of the lens, place a shearing interferometer.

Now, get into a comfortable position, ensure good access to the post, and check that you have a proper view of the interferogram on the shear plate's diffuser window. Put pressure on the post's foot such that you have it clamped well against the optical table. By carefully rolling your thumb and fingers against both post and table, you will be able to accurately adjust the lens position with sub-millimetre precision. Start by moving the lens along the beam path until you see the interference fringes on the shearing interferometer straighten up – this indicates collimation. If the fringes are curved, that's an indicator that the lens is quite decentred. So, let's tackle that next by adjusting the lateral position and rotation of the post. Here, the pinholes along the beam path come in handy. We will make use of the pinhole after as well as the one before the lens. Reduce the beam's diameter with an iris (ideally close to the laser's output) and move the post laterally until the transmitted beam is centred on the next pinhole. Now, rotate the post until the *back-reflections* from the lens go through the preceding pinhole (a strip of paper helps to increase visibility). Iterate between the two pinholes to get the lens into position. Once done, you might have lost proper collimation though. The solution, you might have guessed it, is iterating again between collimation and centration (Figure 2). Luckily, a more beginner-friendly procedure exists that involves an "alignment crane" (see Figure 3). Use it to tightly hold a post and position it with micrometre precision – and largely without losing alignment in orthogonal axes. If you additionally put kinematic adjusters on your lens, angular adjustments are a breeze as well. Once aligned, "custody" of the post is then simply given back from the crane to post's own clamp, and you are ready to move on to the next lens.

Handy tools and tips

Access to a range of tools such as pinholes, shearing interferometers, and power meters is great for efficient optical system alignment. For instance, pinholes help define and clean the beam path, while shearing interferometers ensure accurate collimation, particularly in short paths. Power meters measure light intensity, aiding in tasks like fibre coupling and wave plate alignment for proper polarization.

More generally, effective design and a modular construction approach simplify alignment and maintenance of optical systems. By segmenting the system into functional units like laser combination, beam-shaping, delay line, and detection blocks, each can be individually optimized and aligned. This modularity is particularly beneficial for complex setups, allowing for independent adjustments on a common beam height before integrating into the overall system. When you construct and align each block on a breadboard, adjusting the blocks themselves can be accomplished with adjustable kinematic positioners ("nudgers") with micrometre precision.

Finally, when aligning optical systems, prioritize safety by using low-power lasers and removing reflective jewellery. Employ enclosures to shield against accidental exposure and contain stray light. Always terminate laser beams in beam dumps and use protective eyewear for any intensities above the eye-safe threshold.

Summary

This article has covered key principles and steps for aligning optical components in custom microscopes. We established the importance of the beam path, detailed pre-alignment tasks like cleaning and mounting, and the precise alignment of lenses. We also introduced tools like pinholes and shearing interferometers, and the modular construction approach to simplify alignment and improve system performance.

Apart from beam path setup and basic lens alignment, other equally important things like focus finding were left out for brevity. Adjusting the distance between a lens and a target is used in scenarios such camera alignment, fibre coupling, or fine-tuning TIRF illumination. Such adjustments are fundamental and deserve a separate detailed discussion. To further support your endeavours in optical alignment, I have thus curated a series of short videos on YouTube[1] (Figure 4) that expands upon the principles discussed here and introduces practical tips like the clever uses of mountains or aluminium foil during alignment (really!). Whether you are a seasoned professional or just starting out, please share your experiences with the community (YouTube has a comment section). By fostering a culture of knowledge exchange and innovation, we will be able to provide the best possible starting points for the next generation of microscopy innovators.

Acknowledgement

[1] https://www.youtube.com/playlist?list=PLh5cDpn282vaCtqzjg6laUaR-CtzghltB

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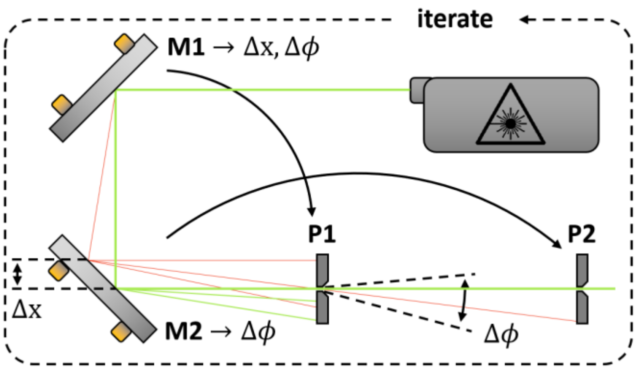

Figure 1: Two mirrors on kinematic adjusters form a great duo to align the beam path (defined by pinholes P1 and P2). The first mirror (M1) affects both lateral position Δx and angle Δϕ, the second mirror exclusively the angle. By aligning the laser through P1 with M1 and through P2 with M2 separately and iteratively, the overall light path converges onto the desired beam path.

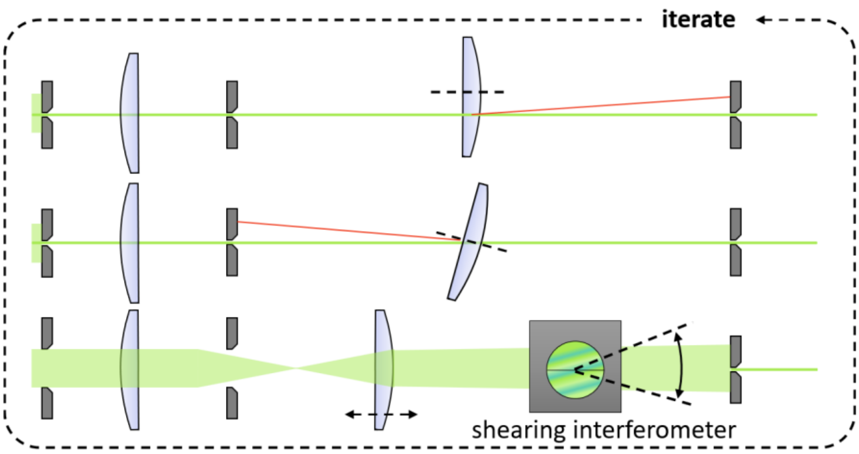

Figure 2: Aligning a lens requires adjusting five degrees of freedom (transverse and axial dimensions and orientation). Transverse positioning and orientation are adjustable with the aid of two pinholes on the beam path and a fine laser beam that lets you observe the transmitted and reflected portions of the alignment laser (note that both are coupled!). For collimation you may use a shearing interferometer, whose interference fringe orientation provides a good read-out of an expanded beams divergence. As often, iteration leads to alignment.

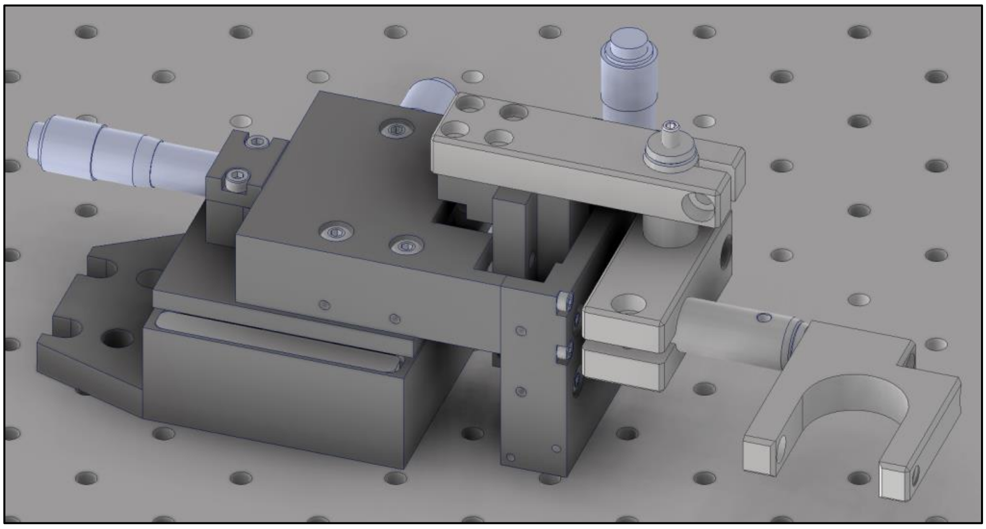

Figure 3: A simple "alignment crane" can be constructed from a multi-axis micrometre stage, a few posts as connectors, and a clamp at the end.

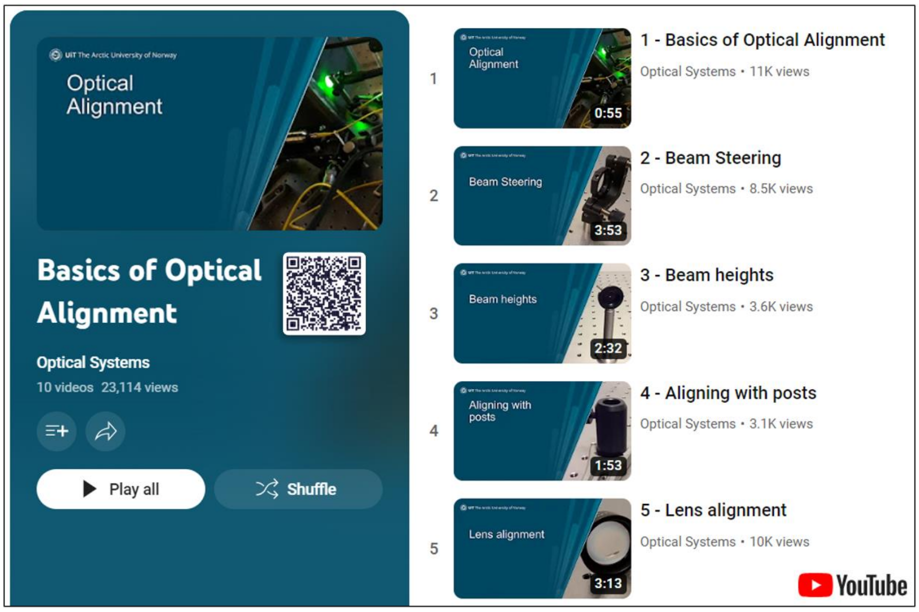

Figure 4: The "Basics of Optical Alignment" video series on YouTube. The QR code links to the entire list.