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Optical Tweezers: Exerting Force with Light

A Thesis Presented By

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To the Keck Science Department Of Claremont McKenna, Pitzer, and Scripps Colleges In partial fulfillment of The degree of Bachelor of Arts

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Introduction

Photons carry momentum. When a tightly-focused beam of photons hit a particle, they transfer some of their momentum to the particle. This radiation pressure exerts a tiny force on the particle; for a ~ 100 mW laser, this force is on the scale of piconewtons and unnoticeable on the scale of a human being. But for a 1.0 μ m particle, the force is significant.

In 1986, physicist Arthur Ashkin and colleagues used this phenomenon of photon momentum transfer to create optical tweezers [1] [2], an instrument that uses a converging laser beam to attract a small particle and hold it in place in three dimensions. The 2018 Nobel Prize in Physics was awarded "for groundbreaking inventions in the field of laser physics" with one half to Arthur Ashkin "for the optical tweezers and their application to biological systems," [3].

In biological systems, optical tweezers are uniquely useful because of the precise and measurable force exerted. In active microrheology, for example, optical tweezers are used to apply a force on a "tracer" particle to find the stress/strain relation of a material [4]. Scientists have also used optical tweezers to measure the force exerted by single motor proteins and the proteins that unwind DNA [5].

Outside of biological applications, the Nobel Prize in Physics 1997 was awarded "for development of methods to cool and trap atoms with laser light," [6]. Physicists used an array of laser beams finely tuned to a wavelength of light that sodium atoms can absorb to force the atoms to absorb and emit photons. As the incoming photons carry momentum only in a desired direction and emitted photons carry momentum in a random direction, this process reduces atom speed by forcing atoms into the center of the laser array [7].

This thesis will outline the fundamental principles of optical trapping, the setup built, predictions for the force exerted, and a comparison to the measured force. Chapter 1 covers the fundamental principles of optical trapping, using ray optics to follow the path of the light beam as it reflects off of and refracts through the bead, culminating in a prediction for the force exerted on the bead by the tweezers. Chapter 2 outlines the build of the optical tweezers that I constructed. And lastly, chapter 3 measures the force exerted on particles in the optical trap by tracking particle motion.

1 Fundamental principles of optical trapping

In Arthur Ashkin's original 1970 proposal of a system similar to optical tweezers [2], he analyzed the effect of a collimated laser incident on a bead (Figure 1). This is the most extensive published analysis of the forces exerted during optical trapping to date; it is very limited because it assumes the laser is collimated, and only qualitatively compares forces, not quantitatively.

The sphere of high index $n_H = 1.58$ is situated off the beam axis. in water of lower index $n_T = 1.33$. Consider a typical pair of rays symmetrically situated about the sphere axis B. The stronger ray (a) undergoes Fresnel reflection and refraction (called a deflection here) at the input and output faces. These result in radiation pressure forces $F_R^{\ i}$, $F_R^{\ o}$ (the input and output reflection forces), and $F_D{}^i$, $F_D{}^\circ$ (the input and output deflection forces), directed as shown. Although the magnitudes of the forces vary considerably with angle Φ , qualitatively the results are alike for all Φ . The radial (r) components of $F_D{}^i$, $F_D{}^o$ are much larger than F_R^{I} and F_R° (by ~10 at Φ =25°). All forces give accelerations in the +z direction. $F_R^{\ i}$ and $F_R^{\ o}$ cancel radially to first order. F_D^{i} and F_D° add radially in the -r direction, thus the net radial force for the stronger ray is inward toward higher light intensity. Similarly the symmetrical weak ray (b) gives a net force along +z and a net outward but weaker radial force. Thus the sphere as a whole is accelerated inward and forward as observed.



Figure 1: Ashkin's original physical explanation compares forces quantitatively, but only one rough calculation result is mentioned (for $\phi = 25^{\circ}$. The figure on the right shows "A dielectric sphere situated off the axis A of a TEM₀₀ mode beam and a pair of symmetric rays a and b. The forces due to a are shown for $n_H > n_L$. The sphere moves toward +z and -r." [2]

This chapter presents new mathematical analysis of a bead trapped in a diverging beam, and quantitatively analyzed force. I will follow the path of a light ray through a bead trapped in the optical tweezers, and find the angle and magnitude of the four major forces exerted: the input reflection force, input refraction force, output reflection, and output refraction. Net force from each ray is calculated from these four forces, and integrated over the upper half of the bead to find the total force exerted on the bead by the diverging laser beam.

As photons in a laser beam hit a spherical particle, they exert a small force. In the Mie regime of particle size, the diameter of the particle is much larger than the wavelength of the laser light, so the effect can be analyzed using ray optics and physics covered in introductory mechanics.



Figure 2: Two rays from a diverging laser beam hit a spherical bead. The rays refract through the bead (solid orange arrows) and reflect (dashed pink arrows). The net force is drawn in thick green. These color conventions (solid orange arrow shows refraction, dashed pink arrow shows reflection, and thick green is force) are maintained throughout all figures in this thesis.

When a ray hits the particle, it is partly reflected and partly refracted. In Figure 2, both rays refract through the bead (solid orange lines) and reflections are shown for ray a (pink dashed lines). The angle of reflection and refraction can be determined by imagining the point of contact as a flat surface, because a sphere is flat at any infinitesimally small point.



Figure 3: The incoming ray is partially reflected off the surface of the bead (pink dashed arrow) and partially refracted through (solid orange arrow).

1.1 Input Reflection



Figure 4: An incident ray (solid orange arrow) reflects off of a flat surface (dashed pink arrow). The change in photon momentum (dashed blue arrow) transfers momentum to the flat surface, exerting a force on the flat surface (thick green arrow).

By the law of reflection, the reflected ray will leave at the same angle to the normal as the incoming ray. The change in momentum of the photon points along the line normal to the bead's surface, directly out of the bead. By Newton's 3rd law, the bead experiences an equal and opposite change in momentum. This force on the bead points along the normal into the center of the bead, and is equal in magnitude to twice the momentum of the incoming photons that are reflected.

The force from the reflected photons' change in momentum is the input reflection force, here labeled FRi.



Figure 5: The first reflection (the input reflection) produces a force pointing into the center of the bead (thick green arrow).

The input reflection force points directly into the center of the bead from the point of contact, so the angle in relation to the +z axis is:



Figure 6: As light rays hit the bead, they each produce an input reflection force pointing into the center of the bead (thick green arrow). The force is at an angle $-\phi$ from the +z axis.

$$\theta_{FRi} = -\phi$$

Where ϕ is the angle between the horizontal +z axis and the point of contact of the ray, shown in Figure 6.

1.2 Input Refraction

The refracted ray will not be at the same angle to the normal as the incoming ray. Its angle is determined by Snell's law:

$$n_1 \cdot \sin\left(\theta_1\right) = n_2 \cdot \sin\left(\theta_2\right)$$

Where n_1 and n_2 are the indices of refraction of the first and second materials, respectively, and θ_1 and θ_2 are the angles to the normal of the incoming and refracted rays, respectively. This change in angle will transfer some amount of momentum to the bead, perpendicular to θ_2 .



Figure 7: An incoming ray at angle θ_1 from a line normal to the surface of the bead refracts through the bead. After refraction it is at an angle θ_2 from the line normal to the surface of the bead, transferring momentum to the bead perpendicular to the refracted ray.

This transfer of momentum to the bead is the input refraction force, labeled FDi, and its angle to the +z axis is (Figure 8):



Figure 8: The direction of the force exerted on the bead by refraction (thick green arrow), relative to the +z axis, depends on the angle between point of contact and the center of the bead, ϕ , and the angle of the light ray after refraction, θ_2

$$\theta_{FDi} = -\phi + \theta_2 + 90^\circ$$

Where θ_2 is the angle between the refracted ray and the line normal to the surface of the bead at the point of contact.

1.3 Output Reflection

As the refracted ray hits the opposite edge of the bead, a similar process of reflection and refraction occurs (Figure 9). The angles can be determined by

analyzing the bead as a circle, and once again applying the law of reflection and Snell's Law:



Figure 9: The portion of the ray that originally refracted through the bead now hits the opposite edge of the bead (labelled incoming). It now partially reflects (dashed pink arrow) and refracts (solid orange arrow) again.

The reflection of the incoming beam creates the output reflection force, FRo. The reflection force is once again equal to twice the momentum of the incoming ray, and points directly outward from the bead because here the ray bounces off the inside of the bead. The angle of this force can be determined by finding the angle of the line normal to the surface of the bead relative to the +z axis:



Figure 10: As the ray reflects off of the second surface of the bead, it produces a force directly outward from the center of the bead (thick green arrow). The three angles drawn along the equator of the bead $(\phi, \theta_3, \text{ and the angle of this reflection force})$ sum to 180° . Analysis of the triangle shown in the middle of the bead produces a relationship between θ_3 and θ_2 .

$$\theta_{FRo} = 180^\circ - \phi - \theta_3$$

= 180° - \phi - (180° - 2\theta_2)
= 2\theta_2 - \phi

1.4 Output Refraction

The direction of force exerted on the bead by the refracted ray, the output refraction force FDo, will once again be perpendicular to the exit angle of the refracted ray.



Figure 11: As the ray refracts through the second surface of the bead, it produces a force perpendicular to the refracted ray because of the photons' change in momentum.

$$\theta_{FDo} = \theta_{FRo} - \theta_1 + 90^\circ$$
$$= 2\theta_2 - \phi - \theta_1 + 90^\circ$$

1.5 Net Force Prediction

The net force exerted on the bead by the laser is the sum of the above forces. The last reflected ray will continue to bounce around inside the bead exerting forces, but since only a small portion of the ray is reflected rather than refracted (at most angles, 97% of the ray is refracted), the forces are minimal in comparison. Assuming that the initial incoming beam exerted force F, and a portion of the beam c_D was refracted and c_R reflected, and that the outgoing refracted portion was split into c_{D2} refracting and c_{R2} reflecting, the net force exerted on the bead is:

Input reflection force FRi: angle $\theta_{FRi} = -\phi$

$$FRi = F \cdot c_R \cdot (\cos(-\phi)\,\hat{z} + \sin(-\phi)\,\hat{r})$$

Input refraction force FDi: angle $\theta_{FDi} = -\phi + \theta_2 + 90^{\circ}$

$$FDi = F \cdot c_D \cdot \left(\cos\left(-\phi + \theta_2 + \frac{\pi}{2}\right) \hat{z} + \sin\left(-\phi + \theta_2 + \frac{\pi}{2}\right) \hat{r} \right)$$

Output reflection force FRo: angle $\theta_{FRo} = 2\theta_2 - \phi$

$$FRo = F \cdot c_D \cdot c_{R2} \cdot \left(\cos\left(2\theta_2 - \phi\right)\hat{z} + \sin\left(2\theta_2 - \phi\right)\hat{r}\right)$$

Output refraction force FDo: angle $\theta_{FDo} = 2\theta_2 - \phi - \theta_1 + 90^\circ$

$$FDo = F \cdot c_D \cdot c_{D2} \cdot \left(\cos\left(2\theta_2 - \phi - \theta_1 + \frac{\pi}{2}\right) \hat{z} + \sin\left(2\theta_2 - \phi - \theta_1 + \frac{\pi}{2}\right) \hat{r} \right)$$

To sum up all of these forces, use Snell's Law to write θ_2 in terms of θ_1 , and rewrite θ_1 in terms of ϕ and ω : $\theta_1 = \phi + \omega$

The c coefficients, which dictate what percent of the laser beam is reflected vs refracted, can be determined by the Fresnel equation. In this case, the laser is perpendicularly or s-polarized, so

$$c_{R} = \left| \frac{n_{1} \cdot \cos\left(\theta_{1}\right) - n_{2} \cdot \sqrt{1 - \left(\frac{n_{1}}{n_{2}} \cdot \sin\left(\theta_{1}\right)\right)^{2}}}{n_{1} \cdot \cos\left(\theta_{1}\right) + n_{2} \cdot \sqrt{1 - \left(\frac{n_{1}}{n_{2}} \cdot \sin\left(\theta_{1}\right)\right)^{2}}} \right|^{2}$$
$$c_{D} = 1 - c_{R}$$

$$c_{R2} = \left| \frac{n_2 \cdot \cos\left(\theta_2\right) - n_1 \cdot \sqrt{1 - \left(\frac{n_2}{n_1} \cdot \sin\left(\theta_2\right)\right)^2}}{n_2 \cdot \cos\left(\theta_2\right) + n_1 \cdot \sqrt{1 - \left(\frac{n_2}{n_1} \cdot \sin\left(\theta_2\right)\right)^2}} \right|^2$$
$$c_{D2} = 1 - c_{R2}$$

Combining the force angles determined by ray optics with the coefficients provided by the Fresnel equation, the net force in the r and z direction can be found, and depends only on the indices of refraction of the bead and its surrounding medium, n_1 and n_2 , as well as angles ϕ and ω , and the net force of the incoming photons F.

A quick check of these calculations: This exact math behind the forces in optical trapping has never been published, but in Arthur Ashkin's first proposal for optical tweezers in 1970, he wrote that the radial component of the force was on the scale of 10x larger than the z component at $\phi = 25^{\circ}$. In Ashkin's original proposal, the incoming laser beam did not converge and was instead parallel to the z axis at all points, so in this situation $\omega = 0^{\circ}$. He used a spherical bead with an index of refraction n = 1.58 in water (n = 1.33).

Following these specifications, the force equations produced by the ray optics analysis predict forces in the +r and +z direction with magnitudes 1.955 and 0.233 greater than the initial photon force, respectively. This result aligns with Ashkin's calculation.

But this quick calculation only produces relative force at one angle. To find the net force exerted on the bead during optical trapping, I will integrate the force over all possible ray angles in the top half of the laser beam.

In an example successful trapping scenario, the particle sits within the laser beam as such (Figure 12):



Figure 12: The collimated laser coming from the left side of the figure (solid orange arrows) passes through a positive lens, converging to a point and then diverging. The trapped bead sits in the diverging beam, just after the waist. As the laser beam reflects off of and refracts through the bead, it exerts forces (thick green arrows) holding the bead in place.



Figure 13: Close-up of Figure 12. The upper ray of the diverging beam hits the bead at an angle ϕ up from the +z axis. The incoming ray is at angle ω from the +z axis, and hits the bead at a distance y up from the +z axis. The bead sits some distance nr from the beam's convergence point, where r is the radius of the bead.

I will integrate over angle ω , so first I need ϕ as a function of ω . From

geometric analysis of the bead in the laser beam (Figure 13):

$$\sin(\phi) = \frac{y}{r}$$

$$\tan(\omega) = \frac{y}{(r+nr) - \sqrt{r^2 - y^2}}$$

$$\phi = \sin^{-1}(\tan(\omega) n + \tan(\omega))$$

$$-\frac{\tan(\omega) \left(\tan(\omega)^2 n + \tan(\omega)^2 + \sqrt{-\tan(\omega)^2 n^2 - 2\tan(\omega)^2 n + 1}\right)}{\tan(\omega)^2 + 1}$$

These relationships produce a function ϕ that depends on ω as well as the distance between the bead and the beam waist relative to the radius of the bead, n.

The upper limit of integration is the angle where the ray is just barely touching the edge of the bead. In this case, the ray is tangent to the edge of the bead, so it is at a right angle to the bead radius (Figure 14).



Figure 14: At the upper limit of integration, the ray is just barely touching the bead and tangent to its surface, so perpendicular to the bead's radius. The triangle formed establishes a relationship between ω and ϕ .

Using triangle rules,

$$\sin(\omega) = \frac{r}{nr+r}$$
$$= \frac{1}{n+1}$$

So the maximum value of ω is:

$$\omega max = \sin^{-1}\left(\frac{1}{n+1}\right)$$

Then the z and r force exerted on the top half of the bead as a function of ω are (Figure 15):



Figure 15: Force exerted by the laser beam on the top half of a trapped bead, at each angle ω up from the +z axis. The force exerted is proportional to F, the initial force exerted by the incoming photons. The z force is consistently close to 0. The r force is almost twice the incoming force F and continuous across ω , only dropping off at the very upper edge of the bead.

$$Netzforce = \int_{0}^{\omega_{\max}} \frac{\hat{z} \cdot (FRi + FDi + FRo + FDo)}{\omega max} d\omega = -0.0210F$$
$$Netrforce = \int_{0}^{\omega_{\max}} \frac{\hat{r} \cdot (FRi + FDi + FRo + FDo)}{\omega max} d\omega = 1.8795F$$

This integration over the angle omega shows the net force exerted on the top half of the bead by the laser. The bottom half of the bead experiences the same force, but with the +r and -r directions flipped. This integral doesn't take into account the higher intensity of the laser close to its center, but the graph shows that the direction of the force is unchanged, even if its magnitude is different.

So if the bead is perfectly placed in this location after the waist of the beam, the optical trap will hold it in place. If the bead starts to slide out, for example, if pulled downward, it will experience a restoring force pulling it upward. As this top half of the bead slides into the more intense center of the laser, the upward force will increase, bringing it back to the original position.

The force is balanced in only this one location, and the bead is acted upon by restoring forces if it deviates from this position. This can be thought of as a potential well, or a spring, with a spring constant that relates how far the bead is from the optical trap to how much restoring force it feels.

The magnitude of the force acting on the bead can be derived from the power level of the laser:

$$F = \frac{momentum}{second}$$

$$= \frac{momentum}{photon} \cdot \left(\frac{energy}{photon}\right)^{-1} \cdot \frac{energy}{second}$$

$$= \frac{h}{\lambda} \cdot \frac{\lambda}{hc} \cdot power$$

$$= \frac{power}{c}$$

This force depends only on the power of the beam, not the wavelength. This calculation doesn't take the fact that wavelength influences speed in a material into account.

This is the original force exerted by photons hitting the bead. But as the beam reflects off of and refracts through the bead, the force changes direction and magnitude, as many photons are bounced back. In the above example of a bead held just beyond the waist of the laser beam, the force exerted in the r direction at each point of the bead was an average of 1.8795x the original force of the photons hitting the bead. So the magnitude of the restoring force is:

$$F_{restoring} = 1.8795 \cdot \frac{power}{c}$$

For context, a bead that simply absorbed the photons would feel 1x the incoming force, while a perfect reflector would feel 2x the incoming force. So this spherical bead is a partially reflective surface, in between the two extremes.

At a current of 100mA, the measured power of the laser beam in the optical tweezers is .094 mW, in which case the magnitude of the restoring force is:

$$F_{restoring} = 1.8795 \cdot \frac{.094 \cdot 10^{-3}}{c} \\ = 5.8892 \cdot 10^{-13} N$$

This chapter set out to predict the net force exerted on the top half of the bead (the restoring force if the bead slides halfway out of the trap), and found it to be $5.8892 \cdot 10^{-13}N$. This force prediction relies on tracing the ray path through the bead and a measurement of laser power, and should be an upper bound for the measured force, which will be calculated in chapter 3.

2 Optical tweezers build

In chapter 2, I will explain how to realize the optical trapping described in chapter 1, going over the light path, beam expander segment, and other mirrors and lenses. I built the Thorlabs optical tweezer setup over the course of fall 2022 to spring 2023 (Figure 17).



Figure 16: Optical path diagram. The path of the 976 nm trapping laser is drawn in solid orange, and the path of the brightfield light in dashed light blue.



Figure 17: Photo of the optical tweezers build! The laser input line (usually present on the bottom right) was removed for repairs.

The Thorlabs optical tweezer setup [8] starts with a 976 nm collimated laser at 100 mA current, which is fed into the base of the optical tweezers. The laser beam path is drawn in solid orange in Figure 16.

The beam reflects completely off of mirror 1, which can be adjusted to correctly align the laser. Next, the beam passes through the beam expander segment.



Figure 18: Galilean beam expander. The incoming laser beam (left) passes through a negative lens and diverges. The diverging beam is then incident on a positive lens, which collimates the beam again. The lenses are lined up so that the virtual focus of the negative lens and the focus of the positive lens are at the same location. Figure source [9]

The optical tweezers use a Galilean beam expander (Figure 18) because it's compact. The beam first passes through Lens 1, which is negative.



Figure 19: Negative and positive lenses. The negative lens, with focal length f1, reshapes a collimated beam coming from the left into a diverging beam. The positive lens reshapes a diverging beam coming from the left into a collimated beam.

The negative lens produces a diverging beam, with a virtual focus a distance f1 to the left of the lens (Figure 19). The diverging beam is then incident on Lens 2, which is positive with a focus distance f2 to the left of the lens, and reorients the diverging beam into its original shape, but now expanded.

When the two lenses are placed a distance f2 - f1 apart, their two focal points align to correctly expand the beam. The beam expander provides an expansion factor of 3. After the beam expander, the beam passes through a narrowed iris and then reflects off a dichroic mirror. The long-wavelength laser beam (976 nm) completely reflects off the mirror and is diverted to the left, while shorter-wavelength light from the brightfield crosses through the mirror to be picked up by the camera.

At the very end of the horizontal segment, the beam reflects off of mirror 2, which can be adjusted to correctly align the beam. The beam then reflects off mirror 3 and through another iris, now in the vertical segment of the optical tweezers.

Finally, the laser beam passes through the objective, changing from a collimated to converging beam, and passes through the sample about 4 mm above the objective, where the optical trapping effect takes place.

Between the objective and the condenser 6-7mm above it, the laser beam converges to its narrowest point. In an optical trap, the bead sits just above (just after) this narrowest point, as explained in the previous section. The converging laser produces the focal point necessary for the optical trap.

To image the sample, an LED white light is placed at the very top of the vertical segment. This light path is drawn in blue in Figure 16. This beam passes through a condenser to collimate the beam, and then through the sample. The beam then follows the sample path as the 976 nm tweezer beam but in reverse, through the objective, reflected off mirror 3 and then mirror 2, and then at the dichroic, this short-wavelength light passes through the mirror rather than reflecting as the tweezer beam did. It then passes through a second short-pass filter and to the camera.

This chapter explains the laser path and the imaging light path through the optical tweezers, which successfully create a diverging beam described in chapter 1 capable of optical trapping.

3 Force measurements

In this chapter, I track the motion of a bead trapped in the optical tweezers, and then use the Equipartition Theorem to find the trap's spring constant. This spring constant reveals the actual force exerted on beads in the optical trap, which can be compared to the prediction from chapter 1.

Observing the behavior of 1.0 μ m fluorescent beads in the optical trap, the beads experienced a strong force pulling them toward the laser (Figure 20).



Figure 20: A 1 μm fluorescent bead is initially out of the camera's plane of focus, in the upper left corner of the image. It is pulled in toward the optical trap (the purple laser light), and in the last frame successfully trapped within the laser, along with 5 other beads.

The "stiffness" of the optical trap, the strength of the restoring force, can be calculated with the Equipartition Theorem.

$$\frac{1}{2}k_x < x^2 >= \frac{1}{2}k_BT$$

The Equipartition theorem relates each degree of freedom that the particle has to its average energy. In this case, the particle has two degrees of freedom, able to wiggle out of the optical trap in either the x or y direction.

On the right-hand side of the equation, the Boltzmann constant k_B and the temperature T are known, as the laser source has a temperature controller. On the left-hand side, the standard deviation $\langle x^2 \rangle$ is caused by Brownian motion, or random diffusion and will be suppressed by the optical trap. The spring constant k_x measures the stiffness of the optical trap.

In an identical optical tweezers build, the trapped bead drifts around its potential well (Figure 21):



Figure 21: x- and y-displacement of a bead in an optical trap, in units of video pixels. Figure source [10]

Here, displacement is measured in pixels, where 1 pixel = $.0118 \pm .0049 \mu m$. The laser current is 100mA and power is .094mW. The horizontal and vertical axes are the x- and y-direction, each a different degree of freedom for the trapped bead.

The standard deviation in the x direction is $.0744 \pm .0306 \mu m$, and the corresponding spring constant is $7.379 \cdot 10^{-7} \pm 3.038 \cdot 10^{-7} N/m$.

The beads used have a diameter of $1.0\mu m$. Assuming that a bead is half its diameter away from the center of the laser beam, so all of the force is acting solely in one radial direction, but not far away enough to leave the center of the laser beam, the force according to this measured spring constant is:

$$F = k_x \Delta x$$

= 7.379 \cdot 10^{-7} N/m \cdot 0.5 \cdot 10^{-6} m
= 3.6895 \cdot 10^{-13} N

In this chapter, I analyzed video of fluorescent beads, and found the restoring force exerted on the beads in the optical trap.

Conclusion

This thesis set out to predict the force exerted on beads trapped by optical tweezers, build the optical tweezers, and measure the restoring force. In chapter 1, analysis of the ray optics within the bead in combination with the power level of the laser predicted that the force in the radial direction would be $5.89 \cdot 10^{-13}N$. The measured force found in chapter 3 is 63% of the prediction, which is unusually high compared to the expected force. Some of the laser force was likely lost in the physical alignment described in chapter 2; if the laser enters the objective at a slight angle, it will lose some of its cohesion, or if the slide is at a height slightly off from an ideal bead position just after the waist of the beam, the force will be reduced.

Additionally, some of the energy of the photons will end up heating the sample rather than exerting force on the bead. Liquid water has absorption bands (light frequencies where the chemical absorbs more energy or heat from photons) at 970 nm and 1200 nm, as well as others. Standard optical tweezer lasers have a wavelength of 1064 nm in order to stay between bands and reduce heat absorption, but this laser has a wavelength of 976 nm, so more energy will be lost to heating the water.

This unusually high measured force is likely due to errors in tracking the trapped beads' "wiggle." The uncertainty in the force measurement is $\pm 1.52 \cdot 10^{-13}N$. With this very high uncertainty, it's more important that the measured and predicted forces are on the same scale of magnitude; little can be concluded about the actual laser efficiency.

Future experiments can increase the optical tweezer strength by using a laser with a wavelength between water absorption bands (for example 1064 nm) to reduce heating. Additionally, force uncertainty can be reduced by recording more videos to more precisely measure the trapped beads' variation in position.

Overall, in this thesis I accurately predicted the order of magnitude of the restoring force acting on beads in an optical trap, built a working optical tweezers system, and measured a force that agreed with the prediction.

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