Confocal Microscopy Imaging of Single Emitter Fluorescence and Hanbury Brown and Twiss Photon Antibunching Setup

Jacob Begis

Abstract

The purpose of this lab was to prove that a source of light can be a single-photon source and to prove they exhibit antibunching. Quantum dot fluorescence was images using an EM-CCD camera with a confocal microscope. A Hanbury Brown and Twiss setup was used with APD’s to collect antibunching histograms. We successfully achieved antibunching, and estimated the fluorescence lifetime $\tau$ to be 34.319 nanoseconds.

Theory and Background

A single photon source emits photons separated in time (antibunching). This is different from a laser source attenuated to the single photon level because in the case of the laser the photons are not antibunched; there will be cases of two or more photons grouped together. Single photons have many applications in fields such as quantum cryptography and quantum computing where a photon can be a quantum bit (qubit). Examples of single photon sources are dye molecules, quantum dots, and color centered nanodiomonds. When single photon sources are excited by an external field one photon is emitted per unit time. Once a photon is absorbed a photon of a longer wavelength is emitted. This time is called the fluorescence lifetime $\tau$. Since these sources emit photons separated by a $\tau$ they are therefore antibunched. Mandel and Kimble first experimentally observed photon antibunching in 1967. Microcavities have been used for enhancement of single emitter fluorescence using 1-D photonic band-gap materials as well as 2-D photonic crystals. Photonic band-gap materials and photonic crystals exhibit selective reflection due to their non-linear periodic structure. Liquid crystals are used to create this periodic structure. One can give liquid crystals planar alignment by placing the liquid crystals between two substrates by physically rubbing them in a single direction. When chiral liquid crystals are used a 1-D photonic band gap structure is formed that only allow one handedness of circular polarization through.

Excitonic emissions from quantum dots, excited by pulsed laser light, have become a practical way to achieve antibunched photons. Quantum dots are molecules often with semiconductor properties that fluoresce at various wavelengths depending on their geometry. Depending on this geometry photons of specific energies are absorbed and emitted. A problem with quantum dots is that they blink. This means that they stop emitting photons for brief amounts of time.
Confocal Fluorescence microscopy can be used to irradiate a sample with focused light from a single-mode laser beam and direct the sample response through a pinhole. It is important that the light originated from the laser will not be able to pass through the pinhole such that only the sample response reaches the detector. This type of imaging has a shallow depth of focus but a high numerical aperture. The numerical aperture is maximized using oil immersion at the objective lens. Confocal microscopes have the ability to collect serial optical sections from thick specimens and to reduce background information away from the focal plane that would otherwise lead to image degradation.

**Experiment**

This experiment utilized the Hanbury, Brown and Twiss interferometer setup to confirm antibunching using a confocal microscope. Below is a figure of the experimental setup.

![Experimental Setup Diagram](image)

*Figure 1: Basic experimental Setup of Hanbury Brown and Twiss setup with confocal microscopy. The laser is operating at a 76 MHz repetition rate and at 1064 nm wavelength. Note that the dichroic mirror reflects the laser light and transmits the sample’s emitted photons. This allows the fluorescent light to either enter the Hanbury Brown and Twiss interferometry or to the EM-CCD camera.*

The first filter blocks all 1064 nm laser light but still allows 532 nm light to transmit. The origin of the 532 nm light is the KTP crystal inside of the resonator which converts the fundamental frequency into its second harmonic. A spatial filter next cleans/widens the beam and shifts its shape from elliptical to circular. Next, neutral density filters diminish the intensity of the laser light illuminating the quantum dot sample to prevent bleaching which diminishes fluorescence.
intensity. The light then reflects off the dichroic mirror though the oil immersion objective to hit the sample emitting photons of a longer wavelength which return through the dichroic mirror. An interference filter now eliminates any remaining 532 nm light. Now depending on the port selection the light either travels to the cooled EM-CCD camera or the avalanche photodiode modules (APD’s).

The EM-CCD camera is used to align the system assuring that the beam is focused. It observes the single-emitter fluorescence as a function of time as the unfocused beam hits the sample. When aligning it is important to take out the interference filter as well as the filter on the dichroic mirror in order for a bright enough image to form on the EM-CCD. Once the beam is properly focused and the system properly aligned the port sector will have the light directed at the APD’s. The APD detector area serves as a functional pinhole for the confocal microscope. Each has a detector area of about 170 microns.

The beam now hits a 50/50 non-polarizing beam splitter which directs half the light toward one APD and half towards the other. We use two APD’s to compensate for the dead time on the detector. One detector is not good enough to measure two consecutive photons. One APD serves as a start signal, the other a stop signal. The difference between these two signals in time allows us to make a histogram of the time delay between two photons and the coincidence count. These APD’s are connected to a computer which contains a counter and timer card. This computer is running LabView software. This computer also controls the stage and creates images of the fluorescence. The APD’s are also connected to a TimeHarp single photon counting card. This card helps create a histogram of the time period between the detection of successive photons versus the number of occurrences. This is an antibunching histogram. After one APD detects a photon a TTL pulse tells a capacitor in the timeharp to charge. When a photon in the other APD detects a photon it tells the capacitor to stop charging. This charge is correlated to the time between the two photons being detected. A signal from one APD will be split and sent to both ports of the Timeharp card creating two electrical pulses that within a perfect system would arrive simultaneously. However there is a delay between these two pulses which is used to zero to calculate an intentional delay between the start and stop signal to the capacitor to adjust the zero time on the histogram. Now if the photons are antibunched the histogram will not show a zero time. Now there should be a dip at the zero time. If two photons are not antibunched they could arrive at both detectors at the same time.

Not pictured in the diagram is a nanodrive which uses a high voltage to very gradually move the stage as while scanning an area over the sample as large as 50 x 50 microns. This raster scan can be used to locate single quantum dots.
Procedure

1. First acquire a solution of an appropriate concentration of quantum dots. The concentration affects the quantity of fluorescing molecules. Using a pipette place this solution on a microscope cover slip. For maximum uniformity one would spin coat excess solution off the slide at 3000 RPM for 30 seconds; however we did not do so.
2. Mix a drop of cholesteric liquid crystal onto the cover slip and use a glass rod to make the solution as uniform as possible.
3. Place a second cover slip such that the solution is in between the two cover slips. Now align the liquid crystals in the solution by rubbing the top cover slip in one direction.
4. Clean the microscope objective with acetone and place a drop of immersion oil on the objective lens once cleaned. Then place the sample on the objective use magnets to attach the sample to the piezoelectric stage.
5. Attenuate the laser beam
6. Use the EM-CCD camera to align the focus on the sample. Once in focus blinking of the quantum dots will be visible.
7. Align the Hanbury Brown and Twiss interferometer in realtime while watching the raster scan in order to get a better image and better focus of the quantum dots.
8. Find a single quantum dot (opposed to a cluster).
9. Zero the Timeharp antibunching histogram and collect the antibunching histogram

Results

We wanted to detect quantum dots individually. To this we used a program to coordinate a raster scan of the sample. A nano-drive moved a piezoelectric stage allowing the laser to scan the sample area. Each pixel on the raster scan represents a photon count acquired by the APDs. The following raster scans show that the quantum dots were spaced enough to be detected individually. The scan area was 25 x 25 microns.
Figure 2: Raster scan of 25x25 microns. The crosshair is where we focused the system.

Figure 3: Raster scan of 13 by 13 microns. The crosshair is where we focused the system.

The stripes on the above figure show the presence of quantum dot blinking. In accordance with our procedure this means the system is properly aligned.

A histogram can be generated from the Hanbury Brown and Twiss setup using a computer time harp card. We determined the physical delay limit of the system to be about 60 nanoseconds. This means on the antibunching histogram the 60 ns mark is where there was no time delay between detectors. We were looking for a dip in the plot where there was no time delay
because this means there is antibunching. The following antibunching histogram shows that we successfully achieved antibunching.

Figure 4: Antibunching histogram. It shows a visible dip at 60 ns. This dip is at 60 ns because that is the physical delay. So 60 can be thought of as the 0 point.

Once we showed the presence of antibunching we next estimated fluorescence lifetime $\tau$.

Figure 5: The slope of this histogram’s Logarithmic trend line was used as an approximation of the fluorescence lifetime $\tau$. 
The fluorescence lifetime $\tau$ was estimated to be 34.319 ns. This was the slope of a logarithmic trendline generated by Microsoft excel shown in the above figure.

**Conclusion**

Single quantum dots, and quantum dot blinking were observed on raster scans. Antibunching was successfully observed using quantum dots in a Hanbury, Brown and Twiss setup using a computer time harp card. In addition fluorescence lifetime $\tau$ was estimated to be 34.319 ns.