The Developments of a Helium Acoustic Levitation Environment for Time Resolved XFEL Experiments

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ABSTRACT

X-ray Free Electron Lasers (XFELs) provide the opportunity to investigate the structures of proteins at room temperature. An ideal sample delivery system would present the protein crystal of choice to the XFEL beam without any support structures, just in the 'mother' liquor. Such a system should be able to operate at room temperature and within a helium filled environment to reduce the beam attenuation and scatter of diffraction data, and to limit evaporation effects. Acoustic levitation has recently become a popular technique with the availability of low cost transducers and open source methods being provided, allowing for the levitation of particles and droplets within air. In this article we highlight the challenges faced to provide a system that can operate in a helium environment.

Keywords: XFEL, acoustic levitation, helium, droplets, protein

1 INTRODUCTION

With recent developments of X-ray Free Electron Lasers (XFEL) in recent years, there are arising opportunities to investigate the structures of proteins at room temperature, owed to its high flux and photon delivery rate. In addition, the delivery rate of the x-rays makes an XFEL an ideal method to perform time resolved experiments. These entail investigating the structure of a protein and observing its change as it is exposed to external stimuli, such as light or additional biochemistry. As often there is no observation of interstitial states of proteins as their structures change [1]. As this application matures, nature's biochemistry will be better understood.

The XFEL beam has a flux density which is orders of magnitude greater than that of a synchrotron, and when delivered at a frequency of 100Hz (as it is at the LCLs XFEL), data determining structural changes can be aquired before the protein crystal is destroyed due to radiation damage. During a typical data set collected at a synchrotron, over 90% of the X-rays used for the acquistion of diffraction data lead to the sample's destruction. This is the case despite the sample being presented at cryogenic ground states [2].

The ideal sample delivery system would present the protein crystal of choice to the beam without support structures, only within it's mother liquor. This is because any sample support would instigate the attenuation of the beam and loss of diffraction data, it would also likely be destroyed due to continuous exposure to X-rays.

From these requirements of a sample delivery system for XFEL, we determined that acoustic levitition is an excellent candidate system, as this would facilitate sample support without a frame or an enclosed sample environment. Such acoustic levititation systems are capable of levitating a single droplet with the ability to simultaneously rotate it in the position in which it is being suspended.

The system we propose will operate at room temperature within a helium filled environment, to reduce the beam attenuation, so as to lend itself to being further developed for time resolved studies.

Acoustic levitation has previously been explored to suspend liquid samples within a synchrotron beam for x-ray diffraction experiments at the Swiss Light Source, however such a system was used within ambient air [3].

The concept of acoustic sample manipulation was first devised by Kundt, leading to the development of the Kundt's tube, to initially observe the speed of sound within various gases [4]. The tube was a transparent horizontal pipe, containing a small amount of fine powder. A source of sound emitting a single frequency was positioned at one end of the pipe. The other end had a movable piston which was used to adjust the length of the tube. The sound waves generated were reflected by the movable piston, allowing for a resonant condition to be met. This meant that the distance that the sound waves travelled was a multiple of the wavelength λ of the sound waves within the gas. Therefore, the length of the tube was a multiple of half a wavelength. At this point, the sound waves are in the form of standing waves, and the amplitude of the vibrating gas is zero at equally spaced intervals along the tube, called nodes. The powder settled in the positions of these nodes, as the gas is comparitively stationary there. The distance between each pile of powder is one half wavelength $\lambda/2$ of the sound within the gas. And this, along with the driving frequency f used, could be used to determine the speed of sound c within the gas, as in equation 1.

$$c = \lambda f \qquad (1)$$

The motion of the powder was due to an effect called acoustic streaming caused by the interaction of the sound wave with the boundary layer of the gas at the surface of the tube.

A by-product of such a system is the ability to manipulate and place particles in the nodes of an acoustic wave which has formed a standing wave. The interest in this technology for XFEL applications is to position samples without introducing any additional material into the path of the beam. Depending upon the frequency and spacing between the sound emitter and reflector, there can be a single node or multiple nodes where there is zero pressure for the suspension of droplets or particles.

In recent years, other workers have explored using acoustics in this way [5], including multiple transducers (which are effectively single frequency emitting speakers) , and various mirrors so that the location and motion of the particles can be specifically manipulated in multiple dimension by controlling the relative phase of the acoustic signals emitted. The basic concept of a reflector system is shown in figure 1.

A variant to this technology was reported by Andrade [6] which allows for the distance between the reflector and the transducer to be varied into non-resonant conditions offering additional flexibility.

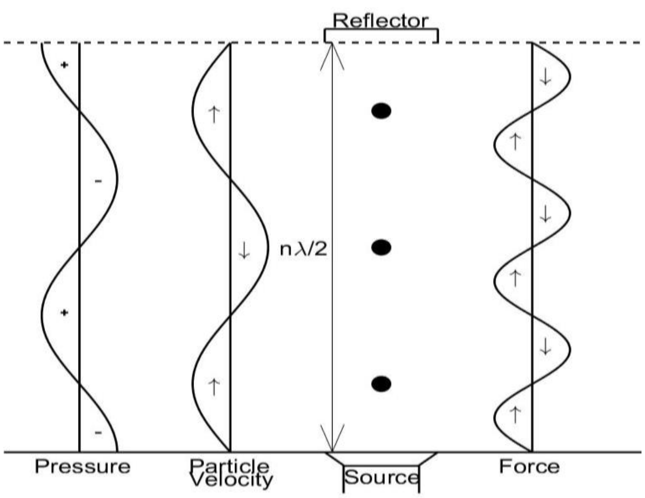

Figure 1- The schematic of acoustic levitation discerning the pressures and forces present due to a standing wave, in addition to the levitating particles velocities and positions.

Acoustic levitation has recently become more popular amongst the public, owing to the work by Marzo showcasing both the one-sided tractor beam design [7] and the TinyLev system [8]. This research showed a cheap alternative to the existing technologies, for the levitation of particles and droplets within air. The tractor beam system, although producing stable nodes to levitate particles, is unable to support droplets due to its uneven pressure gradient on either side of the node, causing droplets to become unstable and fall. Therefore, this system will not be discussed further in this work. However, the TinyLev system shows alternative geometry to the traditional single transducer and reflector

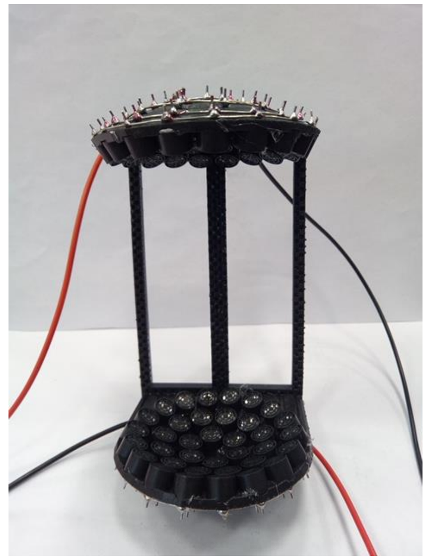

Figure 2- An inexpensive acoustic levitator developed by Marzo et al., constructed for preliminary acoustic levitation experiments within air. This system utilises two domes of transducers which are operating at 40kHz, each dome of transducers is 180° out of phase with the other.

system, as shown in figure 2. It utilizes 72 transducers operating at 40kHz with a radius of 10mm, mounted within two seperated domes. Either dome in this case is 180° out of phase with the other.

2 SAMPLE INTRODUCTION

The task of launching the drop into the trap with precision and when the drops themselves are so small is not trivial. PolyPico Technologies Ltd (Galway, Ireland) produce a drop-on-demand liquid handling system which uses acoustic pressure waves for the ejection of micro-droplets of liquid from a hole at the base of their dispensing cartridge. The system can be used to dispense various liquid substances.

Figure 3- The PolyPico dispensing head is imaged on the left and the dispensing cartridge which can dispense between 10-120pL is imaged on the right.

Unique to this technology is the use of disposable dispensing cartridges, which avoid cross-contamination risks and also support quick fluid replacement. The volume of the micro-drops can be controlled to be in the range of approximately 10pL to 120pL (1pL = 10^{-12} Litres) depending on the properties of the fluid and dispensing can take place up to a frequency of 10kHz. Figure 3 shows a PolyPico dispensing head on the left and a dispensing cartridge on the right.

Depending on the characteristics of the fluid being dispensed, and the application, dispensing cartridges can be selected a in range of orifice sizes (e.g. 30um, 50um, 70um and 100um).

The technology works by introducing an acoustic pressure wave into the fluid in the dispensing cartridge. This pressure wave propagates in the fluid and focuses at the orifice of the dispensing cartridge. This focused pressure wave is sufficient to break the surface tension at the orifice and eject a precisely controlled volume of fluid from the hole. Each time an acoustic impulse is introduced into the fluid column a micro-drop is ejected. This system is the only one currently available to actually offer the potential to dispense the pico-litre slurries of crystals required for the XFEL application.

3 CHALLENGES

Existing acoustic levitation systems typically use transducers which operate at ~40kHz, like the MCUST10P40B07RO multicomp transducer in the Marzo

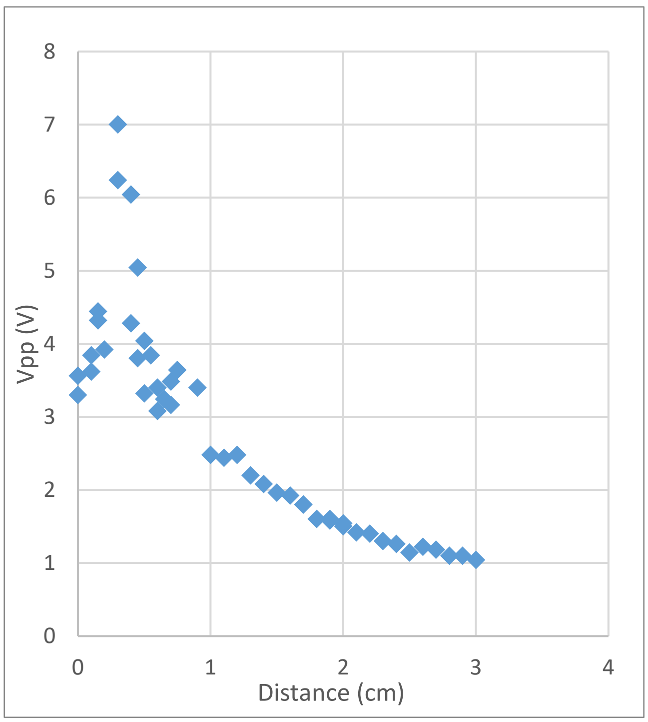

Figure 2- The peak to peak voltage of the recieve transducer as a function of distance from a 40kHz transmitting transducer within air. The voltage decreases as the the distance is increased, with peaks where the resonant condition is met, at ~4mm seperations.

systems. This allows for a wavelength of 8.5 mm in air, in which particles and droplets may be levitated.

The sound pressure level of such transducers may be measured by connecting an oscilloscope to a transducer with the same operating frequency and measuring the peak to peak voltage received, the distance between the transducers may be varied to observe the sound pressure level as a function of distance. Such an experiment has been performed within air and the results are shown in figure 4. The transmitting transducer was powered by a 40kHz sine wave, with 10V peak to peak. This graph showed a decreasing receive voltage, with peaks in the positions in which the resonant condition is met.

This experiment was repeated within ambient helium, expecting peaks to be seperated by ~12mm, owing to the increased speed of sound within helium to 972ms^{-1} compared to that within air of 340ms^{-1}. However, these experiments showed a negligible receive voltage at reasonable separations so results are not shown here. This is owing to the much lower density of helium when compared with air.

In addition, the wavelength of these transducers is much longer when in helium than in air, which creates a much

Figure 1- A MCUSD40A100B17RS-70C Multicomp ultrasonic transducer, which operates at 200V, driving at a frequency of 100kHz.

lower pressure gradient, unable to trap particles or liquid droplets.

In order to increase the pressure gradient within the acoustic levitation system, a MCUSD40A100B17RS-70C transducer was instead selected, as shown in figure 5.

This transducer operates at 200V_{pp} with a driving frequency of 100kHz. Within air this amounts to a wavelength of ~ 3.4mm, but within helium it provides a wavelength approximately equal to that of a 40kHz transducer in air. In addition, the output power of these transducers is significantly higher and should more effectively vibrate the helium molecules, despite the reduced sound pressure level within the gas. This will allow for a sufficient pressure gradient to confine particles or droplet at the positions of the nodes.

4 CONCLUSIONS

This methodology for handling small volumes of microcrystals in mother liquor for presentation to X-ray beams at both synchrotron and XFEL light sources is showing great promise. With the inclusion of the ability to unite multiple droplets and therefore instigate room temperature mixing, we believe we have an excellent sample environment for time resolved experiments.

As the reaction takes place in free space, there is no contamination of the unreleased sample meaning that further experiments may be performed without needing to alter the set up or create additional samples. It also offers the potential to reduce handling quantities down to pico-litre slurries of crystals.

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