



## Controlling acoustic streaming in an ultrasonic heptagonal tweezers with application to cell manipulation



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### ABSTRACT

Acoustic radiation force has been demonstrated as a method for manipulating micron-scale particles, but is frequently affected by unwanted streaming. In this paper the streaming in a multi-transducer quasi-standing wave acoustic particle manipulation device is assessed, and found to be dominated by a form of Eckart streaming. The experimentally observed streaming takes the form of two main vortices that have their highest velocity in the region where the standing wave is established. A finite element model is developed that agrees well with experimental results, and shows that the Reynolds stresses that give rise to the fluid motion are strongest in the high velocity region. A technical solution to reduce the streaming is explored that entails the introduction of a biocompatible agar gel layer at the bottom of the chamber so as to reduce the fluid depth and volume. By this means, we reduce the region of fluid that experiences the Reynolds stresses; the viscous drag per unit volume of fluid is also increased. Particle Image Velocimetry data is used to observe the streaming as a function of agar-modified cavity depth. It was found that, in an optimised structure, Eckart streaming could be reduced to negligible levels so that we could make a sonotweezers device with a large working area of up to 13 mm × 13 mm.

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### 1. Introduction

Techniques that allow the manipulation of cells and micro particles by non-invasive means are highly desirable so as to enable biological applications such as microarrays [1] and tissue engineering [2]. Non-invasive techniques exploiting the acoustic radiation forces have been demonstrated for trapping [3–10], separating [11–15] and rotating particles [16,17]. In a standing wave field, the radiation force drives the particles into acoustic pressure nodes or/antinodes depending on their acoustic contrast factor [18]. However, other unwanted ultrasound-induced effects may disturb the distinct and precise manipulation or aggregation of suspended particles. Acoustic streaming is one such phenomenon, characterised by time dependant flow patterns caused by the absorption of acoustic energy at both the boundaries of the device and in the bulk of the fluid. Three types of acoustic streaming [19–21] can affect the particle handling: Rayleigh streaming [22], Schlichting streaming, and Eckart streaming [23]. Rayleigh streaming creates vortices on the scale of  $\lambda/4$  [24], moving the particles away from the pressure nodes and therefore influencing particle trapping [25]. Schlichting streaming can occur together with Rayleigh

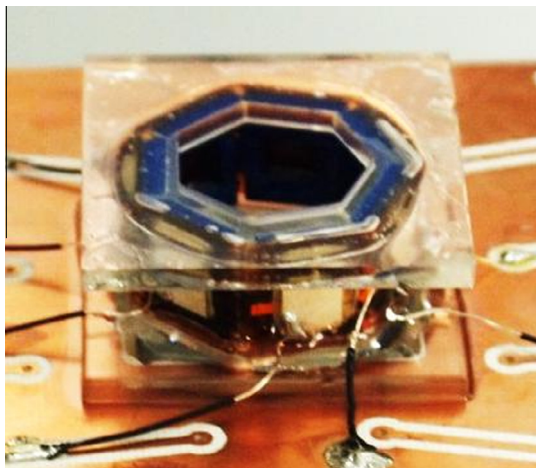
streaming, and forms a vortex with a thickness comparable to the viscous penetration depth at the boundary of the device. In contrast to Schlichting and Rayleigh streaming, which are driven by absorption in the viscous boundary layer, Eckart streaming is caused by the absorption of the acoustic energy in the bulk of the fluid [26]. It has been shown that Eckart streaming can be minimised by reducing the size of the resonator or by designing a resonator containing acoustically transparent foils [7,25].

Acoustic streaming in a microsystem is often viewed as a negative side-effect that needs to be minimised. For example, for cell patterning and engineering, acoustic streaming will affect the positioning of the cells, possibly preventing the cells from adhering to the surface upon which they are growing. In our work [27–30], standing waves were created by interference of two propagating waves. Substantial disturbance of particle manipulation within the acoustic nodes was observed. The particles were constrained to nodal lines, but were driven along those lines by apparent streaming. Under these conditions, it was difficult to pattern particles in a reliable manner.

In this paper, we show that Eckart streaming is the mechanism causing the disturbance and that it is highly dependent on the height of the liquid contained in the cavity. The streaming can be greatly diminished if the depth of the liquid in the cavity is reduced. We present a quantitative study of the Eckart streaming that occurs in a heptagonal particle-manipulating device, also

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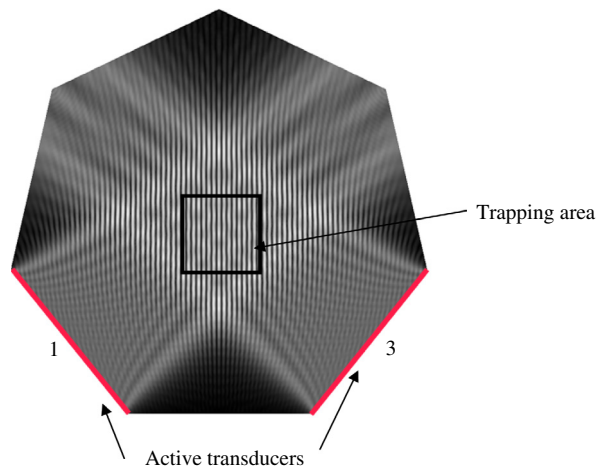
**Fig. 1.** Device shaped into a heptagon and bonded to a PCB to ease the connection of each channel (cell size  $\sim 2$  cm).

known as a “sonotweezer”, and examine the case when two transducers are excited simultaneously. In order to precisely characterise the particles’ behaviour we performed computer image analysis using Particle Image Velocimetry (PIV) of the particle movement as a function of time. We have developed a finite element model of the Eckart streaming and have used it to confirm that this is the mechanism for the generation of streaming in the device, and to identify the regions where the driving force is strongest. It is found that the acoustic streaming is dependent on the depth of the sonotweezer cavity, and this phenomenon has been studied in detail by adding an agar layer at the bottom of the heptagonal cavity to vary the liquid depth without modifying the acoustic field. Agar has acoustic properties similar to water and does not change the acoustic wave interference patterns [31]. In this manner, the amount of liquid inside the cavity of the device was modified without disturbing the acoustic properties of the device. Agar was also found to be a highly convenient filling material as it can be easily added and removed. In the newly configurable device that we present here, we are able to almost completely eliminate the effect of streaming so as to we create a large working area for a range of physical and biological experiments.

## 2. Experimental method

The sonotweezers were made by bonding  $5 \times 5 \times 0.5$  mm NCE51 Noliac Ceramic lead zirconate titanate (PZT) (*E.P. Electronic Components Limited, UK*) plates to a flexible printed circuit board ( $10 \times 72$  mm; *Flexible dynamics Ltd., UK*) and folding it into a heptagon (Fig. 1) that was stabilised by top and bottom plates with a milled heptagonal slot. Acoustic synchronisation between channels was achieved using an arbitrary waveform generator providing four output channels (*TGA12104, Aim and Thurlby Thandar Instruments, UK*) allowing independent control of the amplitude, phase and frequency. The signals from the waveform generators were amplified by the low output impedance, high-speed, BUF634T buffer (*Texas Instruments, USA*).

The characterisation and the influence of the excited transducers has been investigated and discussed elsewhere [27]. It was found that when two transducers were excited particles were trapped along lines perpendicular to the bisecting vector of the normal to the planes of the transducers (Fig. 2). The combination where the activated transducers were adjacent to each other was shown to be less effective in trapping than the combination where at least one inactive transducer separates the active transducers.



**Fig. 2.** Modelling results showing the quasi-standing waves formed in the centre of the heptagonal cavity when two transducers are excited simultaneously. The energy maxima are white, and the energy minima are black.

Adjustment of the relative phase difference between the active transducers allowed controlled movement of the position lines of particles in the direction of the bisecting vector.

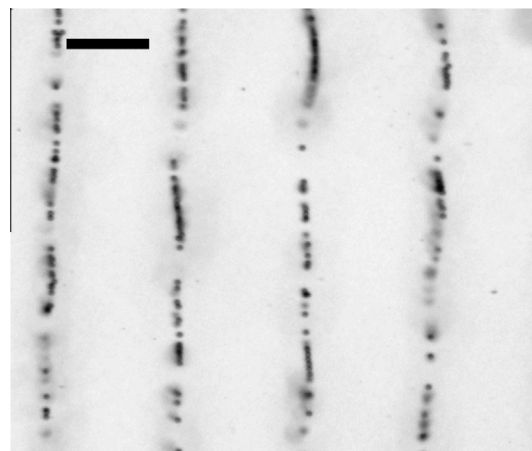
In this paper two transducers (1–3) as shown in Fig. 2 were activated. The activation of the two transducers results in a field that has an array of linear traps in the centre of the cavity. The field pattern shown in Fig. 2 was obtained using a simulation based on 2D lines of point sources that sum up to form plane waves according to Huygen’s principle [27].

Fig. 3 shows  $10 \mu\text{m}$  diameter polystyrene particles (*Polysciences Europe, Germany*) trapped at the nodes of the acoustic field in the centre of the cavity when two transducers, as shown in Fig. 2, are excited with continuous sine waves at an amplitude of  $8 V_{pp}$  and a frequency of  $4.00$  MHz. For this data the device had no agar in place, and as discussed, streaming along the lines of particles was observed.

## 3. Finite element modelling

### 3.1. Radiation force

The Huygen’s principle based method used to obtain Fig. 2 is not capable of modelling the streaming effect. In order to simulate



**Fig. 3.** Micrograph of the trapping area at the centre of the cavity showing trapping pattern when the transducers (1–3) were active. Scale bar =  $200 \mu\text{m}$ .

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