Sensors and Actuators A 188 (2012) 442–449

Contents lists available at SciVerse ScienceDirect

Sensors and Actuators A: Physical

journal homepage: www.elsevier.com/locate/sna

# Micromanipulation using cavitational microstreaming generated by acoustically oscillating twin bubbles

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ARTICLE INFO

*Article history:*
Available online 6 December 2011

*Keywords:*
Micromanipulation
Microfluidics
Microbubbles
Cavitational microstreaming

ABSTRACT

This paper describes a novel non-invasive micromanipulation technique that employs the cavitational microstreaming generated by acoustically oscillating twin bubbles. First, a single acoustically oscillating bubble was attached to the tip of a rod that was combined with a three-dimensional traverse system, and a fish egg (diameter: 1 mm) was then manipulated in an aqueous medium. Although the microstreaming generated by the single oscillating bubble was sufficiently strong to push and move the fish egg, the manipulation direction was uncontrollable. Hence, to improve the manipulation controllability, identical twin bubbles with the same size and resonant frequency were employed. The identical gas bubbles were generated on a microfabricated chip comprising sharp tip-shaped electrodes by employing an electrochemical method, electrolysis, and controlling the applied voltage and time. Subsequently, the bubbles were sequentially transferred to the tips of a U-shaped rod coated with a hydrophobic layer to improve the surface adhesion. The force generated from the acoustically oscillating bubbles and its direction were analyzed under various acoustic excitation conditions by using high-speed images. Our results showed that the generated force was proportional to the bubble oscillation amplitude, whereas the direction of the force depended on the distance between a bubble and object. A steel ball (500 $\mu$m diameter) was used for investigating the force direction. When a bubble (600 $\mu$m diameter) was acoustically excited, the steel ball was pulled toward the oscillating bubble when the distance between the bubble and ball was short (<3 mm), whereas the steel ball was pushed away from the oscillating bubble when the distance was long (>3 mm). Finally, a fish egg (diameter: 1 mm) and glass beads (diameter: 100 $\mu$m) were experimentally manipulated using acoustically oscillating twin bubbles.



## 1. Introduction

In bio-nanotechnology, micromanipulation, or, physical interactions with samples such as small tissues under microscopes using aided precision movements, is an essential technique [1–3]. Considerable efforts have been made to develop reliable and economic techniques for micro-object manipulation [4–8]. Since the late 1990s, various micro-grippers fabricated by the microelectromechanical (MEMS) technology have been developed and applied to various research fields that require precision controls of micro-objects [9–13]. However, the use of mechanical micromanipulation techniques causes an unwanted contact problem – the solid parts in the systems have to come into physical contact with the targeted objects, which thereby damages them [14–17]. In particular, the contacts induced by invasive mechanical techniques may critically damage biological objects such as cells. To prevent contact damages during micro-object manipulations, alternative techniques based on optic, electric, and magnetic forces have been developed [18–24]. Optical tweezing is one of the most popular non-invasive techniques [25–28]. In this technique, a focused laser beam is employed to attract or repel micro-objects and a three-dimensional traverse system is employed to transport the micro-objects in three-dimensional space. Despite their many advantages, optical tweezers present some problems such as the existence of insufficient force and expensive and bulky setups [29].

Chung and Cho [30] developed microbubble tweezers in which an acoustically oscillating mobile bubble can be used to manipulate micro-objects in microfluidic chips. When an air bubble is excited by an acoustic wave at its resonant frequency, it oscillates and simultaneously captures neighboring objects by means of an attractive radiation force generated by the oscillating bubble [31]. The captured objects can then be carried to the desired place by the mobile oscillating bubble via electrowetting-on-dielectric (EWOD) actuation [32–34]. However, this technique inevitably requires the operation distances between the bubble and objects to be small because the radiation force is proportional to the negative fifth power of the distance [35]. When the operation distances are small, the manipulated objects can be damaged by shear stresses induced

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doi:10.1016/j.sna.2011.11.037

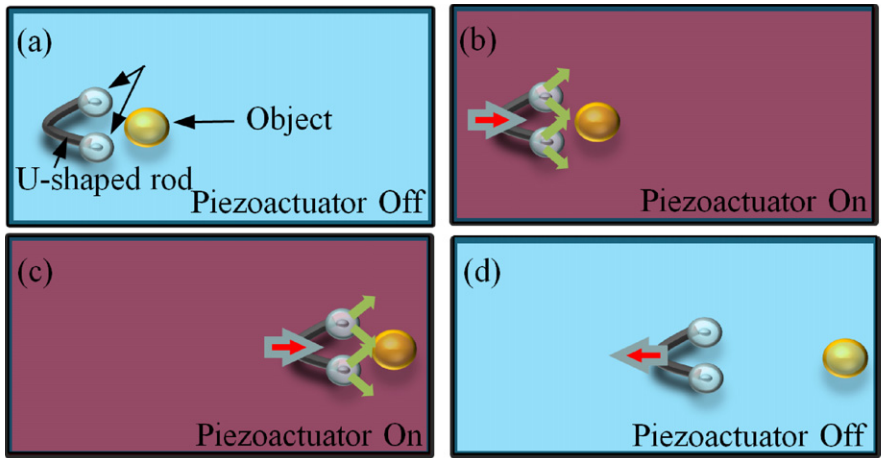


**Fig. 1.** Micro-object manipulation by acoustically exited identical twin bubbles: (a) initial state; (b) when identical bubbles on the tips of a U-shaped rod are acoustically exited, they simultaneously oscillate and generate microstreaming, which, in turn, pushes an object; (c) the rod moves to the right, and the object is carried by the microstreaming; (d) the excitation is turned off and the rod moves to the left; the object is released from the bubbles and remains at its position.

by the cavitational microstreaming generated by the oscillating bubble as well as the radiation force [36,37].

The biological effects of cavitational microstreaming have been investigated by many research groups [38]. Rooney [39] showed that the microstreaming-induced shear stresses generated by two different sources – oscillating bubbles and oscillating tungsten wires – cause the hemolysis of erythrocytes. Wu et al. [40,41] demonstrated significant microstreaming using the encapsulated microbubbles of ultrasound contrast agents and applied it to reparable sonoporation that can be used in gene delivery by temporarily opening cell membranes. They also investigated the effects of the operating distances between oscillating bubbles and cells, which play an important role in ensuring successful reparable sonoporation [42]. However, uncontrolled microstreaming often causes lethal damages to cells, and this causes their membranes to become permanently opened, leading to the so-called lethal sonoporation [41]. Recently, Marmottant and Hilgenfeldt [43] experimentally demonstrated the deformation and rapture of lipid vesicles by microstreaming, while Zhong et al. [44] also reported serious vascular injuries due to the oscillating bubble.

In this study, we have applied cavitational microstreaming from acoustically oscillating identical twin bubbles, having the same volume and resonant frequency, to manipulate micro-objects in an aqueous medium, as shown in Fig. 1. We were able to successfully manipulate the microstreaming strength by controlling the intensity of the applied acoustic signals such that the effects of the shear stresses from the microstreaming were reduced. This micromanipulation technique is easy to use and economical and can be easily applied to various applications such as cell manipulation and microassembly technologies (MAT).

## 2. Theoretical background

A steady flow generated from an acoustically oscillating object in an aqueous medium is called acoustic streaming [38,45]. In particular, cavitational microstreaming involves a gas bubble object and small-scale and boundary-associated streaming characteristics [46]. Further, when a gas bubble in a liquid is oscillated by using acoustic waves, the Reynolds stresses induced in the oscillatory boundary layer, i.e., the so-called Stokes layer, of the

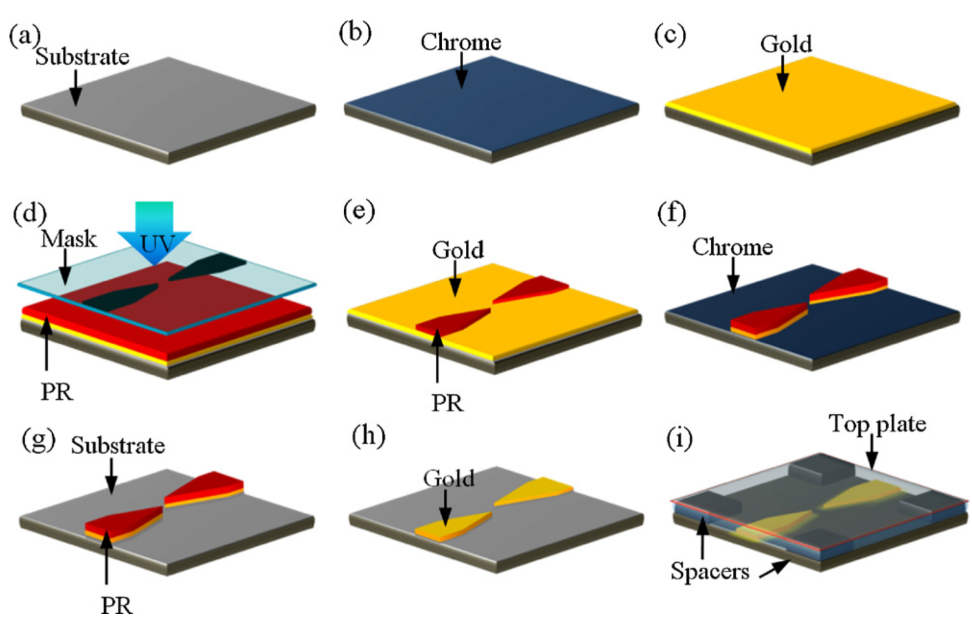


**Fig. 2.** Microfabrication steps of chips used for testing.