

Acoustic Wave Sample Preparation

MICROFLUIDIC FRONT END FOR BIODETECTION

Overview

To date, most biodetection efforts have been concerned with optimization of the specificity and sensitivity of biosensors, rather than sample preparation methods in the microchip format (Fig. 1). For a majority of biosensors, the starting samples consist of tissue, blood, environmental, or food samples. Assays that are based on polymerase chain reaction (PCR); however a variety of contaminants can inhibit amplification and diminish the success of such analytical approaches. Currently, sample preparation is the rate-limiting step in any diagnostic procedure, due to the very laborious and time-consuming processing of complex biological specimens. Integration of the entire process (DNA extraction, isolation, detection) would be highly beneficial – high throughput capabilities would result in low analysis cost due to the small volume of sample and reagents required for the analysis.

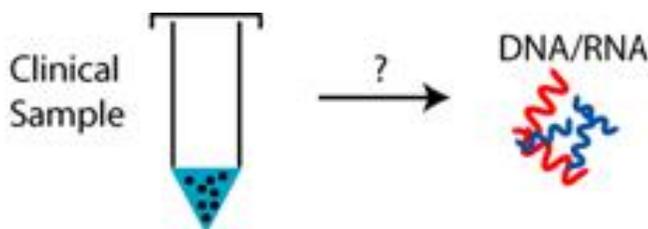


Fig. 1 A key problem in biodetection systems is achieving efficient extraction of DNA/RNA, independent of the source.

It is well known that ultrasonic waves can induce significant pressure variation within fluids and even induce cavitation – the rapid formation and collapse of bubbles. Recently, acoustic methods have proven powerful for the disruption of cell membranes and spores for subsequent DNA analysis. This is especially applicable toward sealed microsystems, where ultrasonic actuation can remotely access the sample flow, permitting continuous flow operation.

Acoustic Technology for Sample Preparation and Lysis

Analogous to an ultrasonic bath on a chip, we have developed a high frequency (54 MHz) bulk acoustic wave (BAW) actuator to perform lysis within fluidic channels. The 54 MHz actuator is based on 36° YX lithium niobate. Our BAW devices can produce large acoustic pressure fields in fluids due to their efficient coupling. Finite Element Modeling (FEM) results have shown that the acoustic pressure fields are in excess of 108 Pa for an input level of 300 mW. This is sufficient to disrupt biological membranes and the protein coat around spores. The lysing mechanism is due to the large acoustic pressure variation throughout the channel, which creates large shear forces on cells, viruses, or spores. The degree of lysing can be controlled by the input power and acoustic profile in the microchannel.

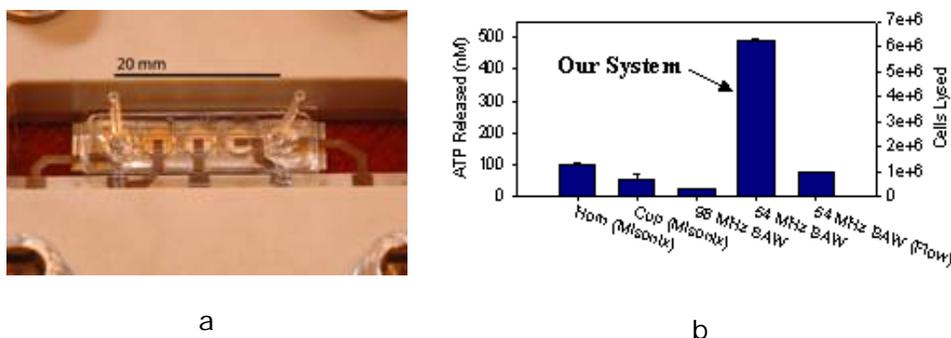


Fig. 2 Acoustic lysing system. **a)** Actual operating device with an attached microchannel **b)** Comparison of lysing systems as measured by ATP release. The ATP assay was linear from 102 – 108 cells/ml. The flow rate was 50 μ l /ml for rightmost case.

Packaging of Acoustic System and DNA Fragmentation

The system allows the microchannel system to be removed from the acoustic transducer after completion of the lysing process if required. The top section of the microchannel was fabricated from acrylic and the bottom layer was fused silica, with a thin matching layer to couple acoustic energy into the fluid filled microchannel. A key advantage with this approach is that the fluidic structure can be replaced without impacting the acoustic transducer or drive electronics (Fig. 2a). As shown in Fig. 2b the ATP release was ~ 400% higher for our 54 MHz system using only 300 mW for 10 seconds. Our system has demonstrated that cells can be lysed efficaciously with minimal power requirements and hardware, having a small overall footprint.

DNA Extraction

We have developed an integrated plastic cartridge system that lyses cellular samples and then purifies the released nucleic acids. Our method of DNA extraction uses magnetic-core beads with an electrostatic surface to first bind DNA in solution. Excess lysate is removed by confining the DNA-bead complexes to the interface. The DNA is extracted from the beads by slightly shifting the pH. This process can be repeated over and over again. Our sample preparation system is small, scalable, chemical-free and well suited for PCR applications.

Please address comments or questions to BioNano@sandia.gov