A Piezoelectric Interfacial Phenomena Biosensor

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# Table of Contents

List of Tables ...................................................................................................................... vi
List of Figures ................................................................................................................... vii
Abstract ............................................................................................................................... x

1. Introduction ..................................................................................................................... 1
   1.1. Background .............................................................................................................. 1
   1.2. Previous Work ........................................................................................................ 3
   1.3. Purpose and Objectives ........................................................................................ 7
   1.4. Organization .......................................................................................................... 7

2. Theory ............................................................................................................................. 8
   2.1. Visco-elastic Properties of Colloidal Systems ......................................................... 9
   2.2. Principles of Operation of the Sensor .................................................................... 11
       2.2.1. Physical Model of Piezoelectric Sensor .................................................. 12
       2.2.2. Equivalent Electric Circuit Representation ............................................. 16
       2.2.3. Foundations of the TSM Electronic Measurement Techniques .............. 20

3. Design ............................................................................................................................ 22
   3.1. Sensing Element ..................................................................................................... 22
   3.2. Sensor Enclosure ................................................................................................... 25
   3.3. Electronic Measurement System .......................................................................... 26

4. Experimental Results and Discussion ........................................................................... 29
   4.1. Target System Studied ........................................................................................... 29
   4.2. Laboratory Testing Procedure .............................................................................. 29
   4.3. Calibration with Reference Medium - DMEM ...................................................... 30
   4.4. Colloidal System Behavior .................................................................................... 33
   4.5. Sample Handling ................................................................................................... 40
   4.6. Results .................................................................................................................... 40
       4.6.1. Influence of The Sphere Size On The Transient Frequency Response of the
              Sensor ................................................................................................................ 41
           4.6.1.1. Influence of 1 µm Polystyrene Spheres ............................................. 41
           4.6.1.2. Influence of 10 µm Polystyrene Spheres in Solution ...................... 44
4.6.1.3. Influence of 90 µm Polystyrene Spheres in Solution............................... 47
4.6.1.4. Comparison Between Theoretical And Experimental Data ..................... 49
4.6.2. Influence of Sphere Size As A Function Of Time on Sensor Response ....... 51
  4.6.2.1. Influence of The Sphere Size on Resonant Amplitude as A Function of
          Time ...................................................................................................................... 51
  4.6.2.2. Influence of The Sphere Size on Resonant Frequency as A Function of
          Time ...................................................................................................................... 54
4.6.3. Change In The Resonant Frequency and Amplitude as a Function of Sphere
       Size ................................................................................................................................ 57
4.6.4. Influence of Concentration of Spheres on Sensor Response ...................... 58
  4.6.4.1. Influence of The Concentration on Resonant Amplitude as a Function of
           Time ...................................................................................................................... 59
4.6.5. Influence of Power on Sensor Response ......................................................... 64
  4.6.5.1. Influence of Power on Sensor Amplitude ................................................ 65
  4.6.5.2. Influence of Power on Sensor Frequency ................................................ 68
4.6.6. Influence of Cells Sedimentation on Sensor Response ............................... 70
4.6.7. Influence of Cells Culture and Fixation on Sensor Response .................... 73
4.7. Discussion .............................................................................................................. 74
4.8. Applications ........................................................................................................... 75
5. Summary, Conclusion and Future Work................................................................. 76
  5.1. Summary and Conclusions ............................................................................... 76
  5.2. Future Work ....................................................................................................... 76
References ......................................................................................................................... 78
List of Tables

Table 1: Schematic representation of wave motion ......................................................... 15
Table 2: Sensor operational parameters ................................................................. 25
Table 3: Shear wave parameters at different frequencies in water. .................. 37
Table 4: Experimental sensor data. ................................................................. 50
Table 5: Theoretical 5 MHz sensor response as volume fraction increases........ 50
List of Figures

Figure 1: Electrostatic adsorption of polystyrene particles with different surface charges
QCM............................................................................................................................ 5

Figure 2: Simple shear motion generated in fluid by a moving solid plate. ....................... 9

Figure 3: Geometrical arrangement of a piezoelectric interfacial sensor.......................... 12

Figure 4: Shear wave parameters in liquid........................................................................ 15

Figure 5: Sensor electric equivalent circuit....................................................................... 17

Figure 6: Sensor electric equivalent circuit in liquid. ........................................................ 18

Figure 7: Sensor electric equivalent circuit for fundamental frequency and third and fifth
harmonic.................................................................................................................... 19

Figure 8: Influence of operation frequency on shear wave penetration............................ 19

Figure 9: Piezoelectric sensing element geometry............................................................ 22

Figure 10: Shear motion of the surface of the sensor......................................................... 23

Figure 11: Temperature stability of quartz crystal sensor................................................ 24

Figure 12: Measurement Chamber.................................................................................... 25

Figure 13: Electrical Measurement System. ..................................................................... 26

Figure 14: TSM sensing resonator connected to the Network Analyzer ......................... 27

Figure 15: Typical Sensor response in air and water. ....................................................... 28

Figure 16: Sensor magnitude response in air, water and DMEM. .................................... 31

Figure 17: Sensor frequency stability in DMEM............................................................... 32

Figure 18: Sensor magnitude stability in DMEM............................................................... 33

Figure 19: Uniform colloidal system arrangement. .......................................................... 34

Figure 20: Colloidal system exhibiting sedimentation..................................................... 34

Figure 21: Colloidal system after sedimentation and packing is complete....................... 35

Figure 22: Changes in density and viscosity as volume fraction of solid particles
increases. .................................................................................................................... 36

Figure 23: Penetration depth as a function of volume fraction for 5 MHz shear wave in
water and DMEM...................................................................................................... 38

Figure 24: Visual representation of surface contact of the spheres with the sensor as size
increases. .................................................................................................................... 39
Figure 25: Sensor response to 1 µm spheres at 5 MHz .................................................. 42
Figure 26: Sensor response to 1 µm spheres at 15 MHz ........................................... 43
Figure 27: Sensor response to 1 µm spheres at 25 MHz ........................................... 43
Figure 28: Sensor response to 10 µm spheres at 5 MHz ............................................. 44
Figure 29: Sensor response to 10 µm spheres at 15 MHz ......................................... 45
Figure 30: Sensor response to 10 µm spheres at 25 MHz ......................................... 46
Figure 31: Sensor response to 90 µm spheres at 5 MHz ............................................. 47
Figure 32: Sensor response to 90 µm spheres at 15 MHz ........................................... 48
Figure 33: Sensor response to 90 µm spheres at 25 MHz ........................................... 49
Figure 34: Influence of sphere size on sensor amplitude at 5 MHz ............................... 51
Figure 35: Influence of sphere size on sensor amplitude at 15 MHz ............................. 52
Figure 36: Influence of sphere size on sensor amplitude at 25 MHz ............................. 53
Figure 37: Influence of sphere size on frequency at 5 MHz ........................................ 54
Figure 38: Influence of sphere size on frequency at 15 MHz ...................................... 55
Figure 39: Influence of sphere size on frequency at 25 MHz ...................................... 56
Figure 40: Influence of sphere size on sensor frequency shift after 60 minutes ............. 57
Figure 41: Influence of sphere size on sensor amplitude after 60 minutes ................. 58
Figure 42: Influence of concentration on sensor amplitude at 5 MHz ......................... 59
Figure 43: Influence of concentration on sensor amplitude at 15 MHz ......................... 60
Figure 44: Influence of concentration on sensor amplitude for 25 MHz ....................... 61
Figure 45: Influence of concentration on frequency shift at 5 MHz ............................. 62
Figure 46: Influence of concentration on frequency shift at 15 MHz ............................. 63
Figure 47: Influence of concentration on frequency shift for 25 MHz .......................... 64
Figure 48: Influence of power on sensor amplitude at 5 MHz ..................................... 66
Figure 49: Influence of power on sensor amplitude at 15 MHz .................................... 67
Figure 50: Influence of power on sensor amplitude at 25 MHz .................................... 68
Figure 51: Influence of power on frequency shift at 5 MHz ........................................ 69
Figure 52: Influence of power on frequency shift at 15 MHz ........................................ 69
Figure 53: Influence of power on frequency shift at 25 MHz ...................................... 70
Figure 54: Influence of cells on sensor amplitude at 5 MHz ...................................... 71
Figure 55: Influence of cells on sensor amplitude at 15 MHz ...................................... 72
Figure 56: Influence of cells on sensor amplitude at 25 MHz ........................................ 73
Figure 57: Influence of cells cultured and fixed on sensor amplitude ................................. 74
In numerous biomedical applications, it is vital to observe the activity of various biological systems such as cells, proteins or bacteria in solution, and their interaction with different natural and artificial interfaces. Monitoring sedimentation of cells or antibodies, their adhesion to the surfaces of different levels of biocompability, or the study of the impact of the drugs on cells or bacteria is of vital importance for biomedical and pharmaceutical applications. Currently, there is no instrumentation capable of quantitatively determining the kinetics of these processes in real time. In this work, we study the use of a Thickness Shear Mode (TSM) piezoelectric resonator as a biosensor for rapid and sensitive detection of a broad range of biological interfacial phenomena. The TSM utilizes shear mechanical waves, which penetrate liquids at a distance of tens of nanometers, and hence effectively monitor interfacial processes. The sensor has been tested and calibrated with a colloidal solution based on polystyrene spheres, which simulates the cells in solution. This well-defined medium allows identification of several important processes and the calibration the proposed sensor system that would be extremely difficult to identify with a complex, biological system. The work is concluded by measurements of endothelial cell properties such as sedimentation, adhesion and fixation.
1. Introduction

It is vital to observe the activity of various biological systems and their interaction with natural and artificial interfaces. Currently, there is no instrumentation capable of quantitatively determining the mechanical properties of cells, the different stages of cell adhesion and the consequences of varying environmental conditions in real time. A thickness shear mode (TSM) piezoelectric sensor, which is the focus of this thesis, utilizes shear motion generated by a vibrating resonator and has the ability to observe mechanical changes due to various cellular processes. Shear motion penetrates a very short distance, on the order of hundreds of nanometers, into the liquid, allowing the sensor to monitor changes occurring at the liquid/sensor interface. The properties of the sensor are influenced by phenomena at the interface between the surface of the sensor and the liquid under test. The principle of the proposed sensor is based on the measurements of variation of both the resonant frequency and amplitude of a piezoelectric quartz resonator caused by the presence of biological entities. However, monitoring cell behavior is complex and the interpretation of the sensor response needs to be understood before a correlation between cell processes and specific sensor responses is reached. In order to understand these sensor responses, a model colloidal system, which simulates cells in solution, is studied in depth. It leads to sensor calibration and identification of various processes in a controllable environment. This work is concluded with monitoring endothelial cell sedimentation, adhesion and proliferation.

1.1. Background

New developments in microelectronic and micro-mechanical manufacturing technologies have unlocked exciting potential for the development of a novel group of devices for chemical (Kwoun, et. al., 2000) and biological study. These microchip size devices can sense gases (Lec, et al., 1988, Lucklum, et al., 1997, Wegener, 1999), study biological fluids and solid materials with high accuracy and reproducibility. The majority of these
devices function on voltage, current, resistance, acoustic, magnetic, optical and thermal effects. These devices can be manufactured using silicon, fiber optic and piezoelectric technologies (Moussy, et al.).

Piezoelectric transducer technology offers a platform for realizing an assortment of highly sensitive, small devices with rapid response times, high accuracy, compatibility with Integrated Circuit (IC) technology, outstanding aging characteristics and the ability to measure numerous quantities in one sensor enclosure (Schwyer, et al., 1997). Sensors based on this technology can be created using typical photolithography techniques and are therefore reasonably priced.

Typically, piezoelectric sensors transmit acoustic waves into a medium under examination. The waves interact with the medium, and the measured parameters of the acoustic waves are correlated with medium properties. Shear waves only penetrate a small distance in liquids. They monitor neighboring properties of a liquid in the surrounding area of the sensor at the liquid(sensor) interface; therefore, they supply a technique to study interfacial processes in a liquid environment. Finally, shear waves can be generated by a variety of piezoelectric devices including bulk and surface acoustic devices. Shear wave sensors have been used to measure many biological systems such as antigen-antibody reactions and enzyme kinetics (Andle, et al., 1990).

Studying cell properties is important in many biological and bioengineering applications such as tissue engineering and drug development. One of the most significant areas is cell adhesion.

Several techniques have been devised for assessing cell adhesion. Perhaps the most common is the simple application of a suspension of cells to the adhesive surface followed by “rinsing.” Then, some measure of the number of cells left adhering to the surface is used to characterize the adhesion strength. In this technique, the mechanical force used to dislodge non-adherent or weakly adherent cells is ill defined and poorly controlled.
The parallel-plate flow chamber and variants apply a nominally uniform shear stress to the culture surface. The wall shear stress is reproducible and quantitative. However, drag force on the cell and the force transmitted to the adhesion receptors is strongly dependent on the surface morphology and cytoskeletal structure (Burmeister, et al. (1998). Centrifugal force techniques apply a body force to the cell rather than a surface traction (Moussy, et al., 1990). Thus, the force on the cell will be less dependent on surface topography, but the degree of spreading and focal adhesion distribution will still influence the detachment behavior. Both of these techniques allow a single loading condition to be applied in each experiment. Furthermore, the adhesion strength can be determined for only one time point per experiment. The radial flow detachment assay (Crozen-Roberts, et al. 1990) and spinning disk techniques (Garci, et al., 1997) allow a range of shear stresses to be applied to a population of cells allowing direct determination of the critical shear stress for detachment. Unfortunately, as with the techniques mentioned above, only one time point per experiment can be evaluated. None of the currently used techniques for assessing cell adhesion allow continuous, nondestructive monitoring of the cell-substrate interaction. Therefore, piezoelectric sensor technology has potential to become a very effective approach for development of a very sensitive technique capable of monitoring interfacial phenomena in real time.

1.2. Previous Work

Piezoelectric sensor technology has been used for the development of a wide range of devices allowing for the study of a broad range of biological phenomena such as antigen/antibody reactions, and enzymatic glucose reactions (Moussy et al., 1990, Cozen-Roberts, et al., 1990, Garcia et al., 1997). In recent years, there has been growing interest in using this type of technology for monitoring biological processes. One of the very promising approaches is based on using TSM shear wave technology.
A broad assortment of methods has been developed to describe cell organization, mechanical properties and adhesion strength. Among the objective of providing a nondestructive technique for constant observation of cell adhesion and structure on a broad selection of substrate materials, it is evident that present methods have severe restrictions. Also, many of these methods are time and labor intensive and require especially skilled personnel and expensive equipment to execute.

TSM technology offers sensors that are capable of real time measurements and are quite inexpensive. TSM sensors have been used extensively to monitor properties of non-uniform liquids, mixtures, polymer solutions as well as biological systems, such as antigen/antibody reactions (Dybwad, 1985, Lucklum, 1997, Ballantine, 1997)

Recently, there have been promising advances in the use of TSM sensors for monitoring cell properties under different physiological conditions. The ASW techniques have been used for the study of cell sedimentation and adhesion (Wergener et al., 2000) and to the evaluation of cell structural properties. Another sequence of studies (Wergener et al., 2000) shows the viability of determining the mechanical properties of cellular structures. By using impedance analysis of the ASW and by removing components from the cell layer, changes in the viscosity and mass were related to the presence of cytoplasm, cytoskeletal filaments, and extracellular protein.

One study uses a flexural-plate-wave sensor, which is an ultrasonic sensor that can sense variations in E. coli W3110 concentration in solution as cells settle on top of the membrane of the sensor under the influence of gravity. A model of the sensor’s response to cell settling has been developed and is in excellent agreement with the experimental data. The flexural-plate-wave sensor consists of a thin micro-machined silicon nitride membrane created by semiconductor manufacturing methods. The thickness of the membrane is smaller than the wavelength of the ultrasonic waves that pass through the membrane. As a consequence, the ultrasonic waves travel at velocities smaller than the velocity of sound in water. The flexural-plate-wave method develops on conventional techniques for determining cell concentrations because this method permit on-line data
gathering, is nondestructive, and needs small sample volumes. The flexural-plate-wave sensor has application as a device to measure cell concentrations and growth rates in manufacturing fermentors, biofilms, and wastewater management facilities (Cown et al., 1999)

There have been very little studies conducted on the target colloidal system, which provides a well-defined calibration medium. The adsorption of polystyrene particles with different surface charges from aqueous dispersions on the surfaces of cationic ultra-thin polymer films was studied (Serizawa et al., 2000) using the TSM method. Particle adsorption, as a consequence of electrostatic interaction, on the outermost surface was observed by quartz crystal microbalance. The TSM was used to compute the accumulation of the adsorbed particles based on the sensor frequency change.

![Ultrathin polymer film on QCM](image)

**Figure 1: Electrostatic adsorption of polystyrene particles with different surface charges QCM.**

The adsorption behavior of two differently charged particles was observed and revealed better adsorption of particles with larger surface charge. The dependence of adsorption of all the particles against an outermost thickness in a polymer precursor film was described by a sigmoid curve, demonstrating that a critical charge on the film is necessary for adsorption.

In another study (Dybwad et al., 1985), a TSM quartz resonator with gold electrodes was used to monitor the behavior of solid particles placed on the surface of the sensor. When a particle was placed on the electrode, the resonant frequency increased, contrary to mass
loading theory, which states that frequency should decrease as mass is deposited on a resonator. If the particle/resonator system is modeled mechanically as a coupled oscillator system the resonant frequency should increase. The increase is determined by the bonding force constant between particle and substrate, which can be calculated. The particle/resonator system was modeled as a pair of coupled oscillators. Although the resonator is usually modeled electrically, a one-dimensional mechanical model provides physical insight into the attachment properties and behavior. In this model, the resonator is represented by a mass attached to a spring with a spring constant, which is determined from the elastic constants of quartz. The mass is determined from the resonant frequency and the spring constant. The particle mass is assumed to be attached to a spring with an unknown spring constant. The coupled oscillator now resonates at a new frequency determined by the additional mass and value of the additional spring constant. For weak particle binding, the system frequency should increase. Maximum frequency is reached when the resonant frequency of the particle is the same as the resonant frequency of the resonator. For stiffer values of particle the spring constant, the frequency approaches a value lower than the resonator resonant frequency. This value is the frequency decrease the resonator would display if the was a part of the crystal and not loosely bound.

Another study (Razavian et al., 1997) attempts to measure the sedimentation rate of non-aggregated sphere-shaped particles in solution by an ultrasonic interferometry method or an “Echo-Cell”. The “Echo Cell” is a vertical temperature controlled tube, which is open at the top. The suspension of particles is enclosed inside the vertical cylinder. A transmit/receive ultrasonic transducer makes contact with the base of the vertical cylinder, while a pulse generator excites the transducer and produces wave bursts, which are transmitted inside the cylinder and are echoed by acoustic impedance mismatches. This technique is based on A-mode echography and measures the speed of sedimentation on a solid plate throughout settling. The accumulation of particles on the bottom of the cylinder induces a fixed solid/sediment interface. There is also a solid/suspension interface, which moves up as the layer grows. The amplitude of ultrasonic wave bursts reflected by a horizontal solid plate is monitored during particle sedimentation. The particle buildup rate, which is related to sedimentation rate, is found from the interfering
two waves echoed by two boundaries. Studies were conducted with latex spheres of different diameters and floating in distilled water at different volume fractions. As predicted by the Stokes model, linear relationships were found between sedimentation rate and both particle density and the square of particle radius. Experimental sedimentation rate values decrease with increasing suspension particle concentration.

1.3. Purpose and Objectives

The purpose of this research is to design and test an interfacial phenomena sensor based on piezoelectric technology. The designed sensor utilizes shear acoustic waves, which penetrate a liquid media at the order of tens of nanometers and are very sensitive to subtle changes at the interface. The target media studied is a polystyrene-based colloidal system, which simulates a typical biological media, such as cells in suspension. The measured properties include sedimentation rate, dynamics of interfacial accumulations, the influence of different sensor operating conditions such as frequency and power and the influence of different colloidal parameters such as concentration and size. Finally, the sensor is tested with a biological system. The ultimate goal of this project is to develop reliable, multipurpose and low cost, miniature instrumentation for characterization of the interaction of cells and proteins with solid surfaces including biomaterials for the analysis of interfacial phenomena. The sensor should find a broad number of applications in the medical, biochemical and environmental industries.

1.4. Organization

The following chapters describe the methods used in the development of the piezoelectric interfacial biosensor. Chapter 2 introduces the theory behind the system studied and the operation of the sensor. Chapter 3 explains sensor design and the measurement system. Chapter 4 describes the measurements technique, experimental setup and results. Chapter 5 presents the conclusions and proposed future work.
2. Theory

In this research, an interfacial phenomena sensor for the monitoring of interfacial phenomena is proposed. The sensor uses a piezoelectric quartz crystal resonator operating in shear mode in the MHz frequency region. Changes in the resonator’s resonant frequency and amplitude indicate different phenomena such as mass loading or changes in viscosity as particles sediment and come in contact with the surface of the sensor. A colloidal suspension of polystyrene spheres was chosen as a target media. Colloidal systems are complex solutions whose behavior depends on the properties of both the medium and the particles in suspension. The behavior of the particles in the suspension also depends on the viscosity of the medium, the size and concentration of the particles and the difference in the density of the particles and the medium. The behavior of the colloidal system simulates several important phenomena in real biological systems, such as cells or proteins in suspension. In order to develop the sensor, the theoretical model for the sensor and the target media needs to be developed.

Sedimentation rate of particles is one of the key parameters most affected by the parameters mentioned above. By knowing a few of the above parameters, unknown mechanical properties can be calculated if the sedimentation rates are accurately measured. The implications of this type of sensor have far reaching effects both in industry and in medical applications. For example, the “health” of a cell can be measured by the sedimentation rates exhibited in solution. While sedimentation rates alone are not the most useful parameters in practical applications, the sensor has the ability to extract other information from the medium such as densities and viscosity.

Currently, there are no commercial sensors available to monitor the behavior of colloidal systems. The main challenge is creating measurement systems that offer high sensitivity to small changes in frequency and amplitude. This sensor has tremendous applications in industry as well as medical applications where sensitivity, accuracy and fast response time are very important.
2.1. Visco-elastic Properties of Colloidal Systems

The behavior of colloidal systems exposed to shear forces is an important part of rheology. Rheology is the study of flow and deformation of matter under the influence of a mechanical shear force. Two types of behaviors are possible. Elastic behavior, exhibited by rigid solids, is observed when the deformation is reversed spontaneously or relaxed when the force is removed. Viscous behavior, exhibited by simple liquids, is observed when the deformation is not reversed and the material stops flowing when the force is removed. In this case, the liquid’s viscosity governs its behavior. Real world systems fall somewhere between the two extremes and are called visco-elastic (Everett, 1988). Colloidal systems typically behave as visco-elastic systems, whose behavior depends on the interaction between the suspended solid particle and the suspending liquid.

The transmission of the shear mechanical motion into the fluid is determined by the visco-elastic properties of the liquid, in particular shear viscosity. Consider a case were fluid is contained above a plate, which generates a shear motion penetrating the liquid by depth \( h \). The plate moves at a velocity \( V \) by a force \( F \) as shown in the figure 2.

![Figure 2: Simple shear motion generated in fluid by a moving solid plate.](image)

Assuming there is no slip at the plate/fluid interface, the velocity, \( v \), of fluid relative to the plate is approaching zero at depths much higher than \( h \) and increases to \( V \) at the plate (Everett, 1988). The mechanical deformation of the liquid is represented by:
\[ \frac{dS}{dt} = \frac{V}{h} \quad (1) \]

and is called the shear rate. The force required to maintain the plate velocity, \( V \), is proportional to \( A \), the area of the plate. Therefore, the following ratio represented by \( T \):

\[ T = \frac{F}{A} \quad (2) \]

is called the shear stress. The ratio of the force to the rate of shear strain characterizes the viscous properties of the liquid:

\[ \eta = \frac{T}{\frac{dS}{dt}} \quad (3) \]

and is referred to as the viscosity coefficient (Everett, 1988).

There are two types of behaviors observed in viscous systems. Newtonian systems exhibit a constant viscosity that is independent of shear rate. Simple liquids and dilute colloidal systems are Newtonian systems. Non-Newtonian systems exhibit a shear dependant viscosity or an apparent viscosity. The apparent viscosity may increase with shear rate when the fluid exhibits shear thickening. In the case of shear thinning, the apparent viscosity decreases (Everett, 1988). Biological fluids, which consist of a suspension of biological particles such as cells, antibodies and proteins, can be modeled as colloidal systems.

The Newtonian viscosity of a liquid is modified and may become non-Newtonian if a high concentration of colloidal particles is introduced. This outcome is due to hydrodynamic interactions between the liquid and solid particles, attractive and repulsive forces between the particles and particle/particle interactions (Everett, 1988).

For spherical particles at very low concentrations, the colloid viscosity, \( \eta/\eta_s \), is related to the volume fraction, \( \varphi \), of particles by the following equation (Everett, 1988):

\[ \frac{\eta}{\eta_s} = 1 + 2.5\varphi \quad (4) \]

where \( \eta_s \) is the viscosity of the pure liquid medium.
These relations are valid at infinite dilutions, where there is no interaction between particles in solution and only particle/liquid interactions are significant. In practice, the relations are accurate for volume fractions up to 0.01. At higher concentrations, the effects of particles in close proximity have to be considered (Everett, 1988).

2.2. Principles of Operation of the Sensor

The main component of the interfacial phenomena sensor is a piezoelectric quartz crystal resonator. Piezoelectric materials have the distinctive capability to induce a mechanical strain when an electrical voltage is applied, and vice versa. By stimulating a sensor with an alternating voltage, standing acoustic waves are created within the sensor, and the sensor acts as an extremely responsive resonator. If the sensor is weighed down with a medium, it transmits a shear wave through the medium. The sensor and medium at the surface can be characterized as a coupled resonant system whose properties depend on the properties of the sensor, the medium, and the interface of the two at the boundary. The generated shear wave penetrates a very short distance into the liquid, and the influence of the boundary conditions on the behavior of the sensor is extremely dominant. The penetration depth factor, $\delta$, is proportional to the viscosity of the liquid and is inversely proportional to the density of the liquid and the frequency of the wave. Consequently, by varying the frequency, the distance at which a wave probes the medium can be controlled. At 5 MHz, the depth of penetration is about 250 nanometers in water (Lec, 1999).

The theory of operation of a piezoelectric shear wave sensor loaded a viscous and viscoelastic media is well developed (Lec, 1999). The key points of the model are summarized below, both in physical interpretation and electric equivalent circuit representation.
2.2.1. Physical Model of Piezoelectric Sensor

Mechanical forces and electrical fields are coupled in piezoelectric materials. As a result, the equations describing the vibration of the piezoelectric resonant elements require a simultaneous solution of Maxwell equations and Newton equations with appropriate boundary conditions, which describe the geometry of the piezoelectric device. The speed of acoustic waves is five orders of magnitude smaller than the speed of electromagnetic waves, therefore Maxwell equations, coupled to mechanical motion, are reduced to electrostatic charge equations.

Usually piezoelectric resonators can be well approximated as a one-dimensional vibrating element (see figure 3), and the equations can be considered in one-dimensional space. In this case, consider a piezoelectric resonator, or sensing element, loaded on one side with a liquid and with air on the other. The piezoelectric sensor can be disk, plate or bar shaped.

Maxwell and Newton’s equations in a piezoelectric element take the following form. For the Maxwell charge equation:

\[
\frac{dD_z}{dz} = 0
\]

(5)

where \(D_z\) is the z-component of the electric displacement vector \(D\) (Lec, 1999)

![Figure 3: Geometrical arrangement of a piezoelectric interfacial sensor](image-url)

and the relationship between the electric field and the applied potential has the form:
\[ E_z = -\frac{d\phi}{dz} \]  \hfill (6)

where \( E_x \) is an electric field and \( \phi \) is the potential (Lec, 1999).

Newton’s equations of motion follow:

\[ \frac{d^2 T_{xz}}{d z^2} = \rho \frac{d^2 u_x}{d t^2} \]  \hfill (7)

Electrical and mechanical properties of materials are described by constitutive equations. In piezoelectric materials, the constitutive equations couple the electrical and mechanical variables:

\[ T_{xz} = c \frac{d u_x}{d z} - e E_z \]  \hfill (8)

\[ D_z = e \frac{d u_x}{d z} + \varepsilon E_z \]  \hfill (9)

where \( e \) is the piezoelectric constant and \( \varepsilon \) is the permittivity. Therefore, the wave equation is as follows:

\[ \frac{-d^2 u_x}{c d z^2} = \rho \frac{d u_x}{d t} \]  \hfill (10)

and \( T_{xz} \) is the shear stress, \( u_x \) is the shear displacement, \( c \) is the stiffened elastic constant and \( \rho \) is the density (Lec, 1999):

\[ c = c + \frac{e^2}{\varepsilon} \]  \hfill (11)

The solution of the mechanical motion in piezoelectric material represents the displacement wave motion of the form:

\[ u_x = [A_1 e^{jkp z} + A_2 e^{-jkp z}] e^{j\alpha} \]  \hfill (12)

where

\[ k_p = (\omega^2 \rho/c)^{1/2} = \omega' V_q \]  \hfill (13)

and \( k_p \) is the wave vector, \( A_1 \) and \( A_2 \) are the partial amplitudes of the wave and \( V_q = (c/\rho)^{1/2} \) is the velocity of the shear wave. The potential distribution across the piezoelectric material is:
Equations for the liquid media are follows. For simple or Newtonian liquids, which do not support the propagation of shear waves, the constitutive equation has the form:

\[ T_{zx} = \eta \frac{du_x}{dt} \]  (15)

and the shear mechanical motion is described by the Navier-Stokes equation expressed in terms of a particle velocity, \( v_x \), takes the form:

\[ \eta \frac{d^2v_x}{dz^2} = \rho \frac{dv_x}{dt} \]  (16)

In liquid, the solution to the (Newton) Navier-Stokes equation has a form of an exponentially decaying wave:

\[ v_x = A_x e^{-k_L z} e^{j\alpha} \]  (17)

where \( v_x \) is the particle velocity and the wave vector \( k_L \) is given by:

\[ k_L = \left( \frac{\omega \rho}{2\eta} \right)^{1/2}(1+j) \]  (18)

or

\[ v_x = A_x e^{-k_L z} e^{j(\alpha-k_L z)} \]  (19)

where the real part of \( k_L \) is the attenuation factor represented by:

\[ \text{Re}[k_L] = \alpha = \left( \frac{\omega \rho}{2\eta} \right)^{1/2} \]  (20)

and \( v_s \) represents the velocity of the shear wave represented by:

\[ v_s = \text{Im}(k_L)/\omega = (2\eta\omega/\rho)^{1/2} \]  (21)

and the penetration depth of the wave is

\[ \delta = 1/\alpha = (2\eta\omega/\rho)^{1/2} \]  (22)

The mechanical motion transmitted from the piezoelectric sensor to the liquid is coupled through the boundary conditions. The description of wave motion can be schematically summarized in the table below:
Table 1: Schematic representation of wave motion

<table>
<thead>
<tr>
<th></th>
<th>Liquid Navier-Stokes - $\eta \frac{d^2 u_x}{dz^2} = \rho \frac{du_x}{dt}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mechanical Motion (Particle Motion) – $V_x = A_3 e^{-k_{z}z} e^{i\omega t}$</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Boundary Conditions (Liquid/Solid)</th>
<th>$u_x(h^-) = u_x(h^+) - \text{Non-Slip Condition}$</th>
<th>$T_{xz}^L - T_{xz}^D = \rho \frac{d^2 u_x}{dt^2}$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$\phi(h) = -\phi_0 e^{i\omega t}$</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Z=h Piezoelectric Sensor</th>
<th>Wave Equation – $c_p \frac{d^2 u_x}{dz^2} = \rho \frac{d^2 u_x}{dt^2}$</th>
<th>Constitutive Equation – $T_{xz} = c_p \frac{d u_x}{dz} - c_p E_z$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$u_x(z) = [A_1 e^{jkpz} + A_2 e^{-jkpz}] e^{i\omega t}$</td>
<td>$\phi(z) = [(e/\varepsilon)u_x(z) + A_2 z + A_3] e^{i\omega t}$</td>
<td></td>
</tr>
</tbody>
</table>

| Boundary Conditions (Air/Solid) | $T_{xz} = 0$ for $h = 0$ | $\phi(z = 0) = \phi_0 e^{i\omega t}$ |

The wave motion in the sensing element and the liquid can be represented in the conceptual form given in figure 4 (Lec, 1999).

**Figure 4:** Shear wave parameters in liquid
The TSM piezoelectric sensor can be considered a source of a distributed stress, $T_{xz}$ or force, $F_x = T_{xz} \times \text{Area}$, which generates a displacement $u_x$ and a particle velocity $v_x$ at the interface. The magnitude of $u_x$ and $v_x$ depend on the boundary conditions and the type of media in contact with the sensor. The ratio of stress to the particle velocity is the mechanical shear interfacial impedance $Z_s$, which for viscous liquids equals:

$$Z_s = \frac{T_{xz}}{v_x} \bigg|_{z=h} = \frac{\eta}{\delta} (1+j) \quad (23)$$

where $\eta$ is the viscosity and $\delta$ is the penetration depth.

The concept of interfacial impedance is important in relation to the electrical impedance of the TSM sensor. From the electrical point of view, the TSM being excited by voltage, where a current flows through the sensor, represents electrical impedance. The form and the magnitude of the electrical impedance depend on the electrical and mechanical properties of the TSM sensor, the boundary conditions and the loading liquid. Therefore, the sensor electric impedance is directly related to the sensor mechanical shear interfacial impedance, $Z_s$, and the sensor can be described as an electromechanical transformer, which transforms $Z_s$ to $Z_e$ (Lec, 1999).

### 2.2.2. Equivalent Electric Circuit Representation

The equivalent electric circuit representation of the TSM sensor provides a very important tool for the analysis of the interfacial phenomena because it directly relates the measured electrical parameters of the sensor with the boundary conditions and the mechanical and visco-elastic properties of the liquid media (Lec, 1999).

The electrical characteristics of the TSM sensor can be expressed in terms of its electrical parameters. For an unloaded sensor (in air), the circuit parameters are the capacitance $C_0$, which is coupled in parallel to inductance $L_1$, capacitance $C_1$ and resistance $R_1$. 
Figure 5: Sensor electric equivalent circuit.

The capacitance $C_0$ is described by:

$$C_0 = \frac{\varepsilon \varepsilon_0 A}{h} \quad (24)$$

where $\varepsilon$ is the permittivity, $A$ is the area of the electrode and $h$ is the thickness of the sensor, and represents the static capacitance sandwiched between the electrodes occurring from the insulating quartz. The additional three elements form the motional branch that describes the piezoelectric dynamics of the resonator. The electric motional impedance, $Z_m$, is given by:

$$Z_m = j\omega L_1 + \frac{1}{j\omega C_1} + R_1 \quad (25)$$

and corresponds to the inertia ($L_1$), compliance ($C_1$) and losses ($R_1$) of the resonant branch (Lec, 1999) and the electrical impedance:

$$\frac{1}{Z_e} = Y_e = j\omega C_0 + \frac{1}{Z_m}$$

$$R_1 = \frac{\eta}{c_{66} C_1} \left( \frac{\omega}{\omega_s} \right)^2$$

$$L_1 = \frac{1}{\omega^2 C_1}$$

$$C_1 = \frac{8K^2C_0}{(N\pi)^2}$$

Note that $L_1$ is directly proportional to the total mass of sensing element and $C_1$ is directly proportional to the elasticity and $R1$ is the viscous losses in the resonator (Lec, 1999).

When a quartz crystal resonator functions under a liquid load, a fraction of its kinetic energy escapes, which alters the motional impedance and the liquid load can be expressed
in terms of additional electrical impedances to the motional branch. It can be shown that a Newtonian liquid at the boundary can be represented by an inductive \((L_2)\) and resistive element \((R_2)\).

\[
\begin{align*}
R_2 &= \frac{N\pi}{4K^2C_0Z_q}\left(\frac{\rho\eta}{2\omega_s}\right)^{1/2} \\
L_2 &= \frac{N\pi}{4K^2C_0Z_q\omega_s}\left(\frac{\rho\eta}{2\omega_s}\right)^{1/2}
\end{align*}
\]  

(27)  

(28)

The resistance \(R_2\) corresponds to power dissipation and the inductance to the kinetic energy accumulated in the liquid shear wave. When additional mass is accumulated at the interface, it can be represented as an additional inductance \((L_3)\).

\[
L_3 = \frac{N\pi}{4K^2C_0Z_q\omega_s}\rho h_{eff}
\]  

(29)

The parameter \(L_3\) describes the kinetic energy that is accumulated in the synchronous movement of the trapped liquid layer (Lec, 1999, Lucklum et al. 1997, Martin et al., 1994).

It can be shown that the TSM sensor can operate at odd harmonics. This important because the same sensor can generate a shear wave at the resonant frequency, the third harmonic, the fifth harmonic, etc. and can probe the interface at different depth as shown in Figure 8.
If the frequency increases, then the depth of penetration is reduced. By make use of the fact that piezoelectric quartz crystal resonators function also at odd harmonic frequencies.
in addition to the fundamental frequency, observing the behavior of a medium at different distances at the same time with the same sensor is achievable (Lec, 1999).

2.2.3. Foundations of the TSM Electronic Measurement Techniques

One more significant characteristic of a piezoelectric quartz crystal resonator is its ability to directly monitor mechanical properties of a surrounding medium by electronic means. In piezoelectric materials, the transformation of electrical energy to acoustic energy takes place inside the material almost immediately. Consequently, the sensor perceives variations in the liquid rapidly. By understanding the sensing process, explanation of important sensor response is feasible. When a voltage is applied to the sensor, the sensor generates a shear deformation at the solid/liquid boundary. This deformation is applied to the liquid media and the strain is developed at the border. Mechanical impedance describes the process at the solid/liquid interface, and the piezoelectric quartz crystal resonator can be represented as a tool that converts mechanical impedance to electrical impedance. The impedance has a resonant nature and is characterized in the form of an electric circuit. The components of that circuit are a function of the material and the properties of media at the interface as shown in the previous section (Lec, 1999).

By linking mechanical parameters and the equivalent electric components, a circuit representation offers a great tool for direct explanation of interfacial phenomena. An initially bare sensor only has a resonant frequency given by:

$$\omega_{s1} = 1/(L_1C_1)^{0.5} \quad (30)$$

and the losses are determined by $R_1$. As soon as the sensor is immersed into the liquid, then $L_2$ and $R_2$ become active, and the resonant frequency and the losses experience the variations as a result. The resonant frequency given by:

$$\omega_{s2} = [(L_1+L_2)C_1]^{0.5} \quad (31)$$

decreases and the losses given by:

$$R_{\text{total}} = R_1 + R_2 \quad (32)$$
increase. If mass builds up at the boundary, the frequency will decrease once more since the sum of the inductance increases by:

$$L_{\text{total}} = L_1 + L_2 + L_3$$  \hspace{1cm} (33)

On the other hand, the losses will remain stable (Lec, 1999).

If the mass becomes bigger and the solid film becomes thicker, then viscous losses will begin to appear, and the addition of $R_3$ introduces more losses. If structural rigidity increases in the layer, then the viscous losses will be decrease, and the frequency will increase and losses will decrease. Consequently, monitoring the progression of the sensor characteristics as a function of time provides insight into interfacial processes and allows their interpretation (Lec, 1999).

Electrical impedance can be measured by an array of methods. One of the techniques makes use of the Network Analyzer based system. This technique allows for fast determination of the behavior of the resonator over a broad frequency range and for the recognition of a wide variety of contributions to the sensor response (Lec, 1999).

By monitoring the changes in resonant frequency and the amplitude, a correlation can be drawn about liquid properties at the interface (Lec, 1999). Using the equivalent circuit representation, it can be shown that:

$$\frac{f_s(Liquid) - f_s(air)}{f_s} = \frac{\Delta f_s}{f_s} = \frac{\Delta L}{2L} = -\frac{2f_s}{N\sqrt{c q \rho q}} \left[ \rho_s + \left( \frac{\rho_\eta}{4\pi f_s} \right)^{1/2} \right]$$  \hspace{1cm} (34)

$$\alpha[\text{dB}] = 20\log \left[ \frac{R}{Z_o} + 1 \right]$$

where

$$R = \frac{\eta_l c_q C_1}{c_q C_1} + \frac{1}{N\pi C_1} \left[ \frac{\rho_\eta}{4\pi f_s c_q \rho_q} \right]^{1/2}$$  \hspace{1cm} (35)

and $Z_o$ is the reference impedance of the electronic measurement system, $\eta_l$ is quartz viscosity and $c_q$ is quartz elasticity.

Specific details of the electronic measurement system are presented in section 3.3.
3. Design

A laboratory model of an interfacial piezoelectric sensor consists of a sensing element enclosed in a chamber with the accompanying measurement system.

3.1. Sensing Element

The sensing element is piezoelectric AT-cut Quartz resonator. In piezoelectric sensors, an applied electrical voltage induces a mechanical strain, and vice versa. By exciting a sensor with an alternating voltage, standing acoustic waves are produce within the sensor, and the sensor behaves as a highly sensitive resonator. If the sensor is loaded with a medium, it transmits a shear wave into the medium. Piezoelectric sensing elements can be fabricated in various shapes, such as thin disks (Figure 9), plates or bars, and covered with a system of metal electrodes (Lec, 1996).

Figure 9: Piezoelectric sensing element geometry.
Figure 9 shows a schematic drawing of the piezoelectric sensing element used. The shaded region is the gold electrode deposited on the quartz to provide electrical contact. An applied voltage generates shear motion at the surface of the sensor as shown in the figure below:

![Shear motion of the surface of the sensor](image)

**Figure 10: Shear motion of the surface of the sensor**

At a specific frequency determined by the thickness of the sensor, $h$, and the velocity of shear acoustic waves in the piezoelectric material, $v$, the sensing element exhibits a resonant frequency as indicated by the equation below:

$$f_r = \frac{v}{2h} \quad (36)$$

There are several piezoelectric materials that can be used as shear wave generating sensing elements. Some piezoelectric materials include: quartz ($\text{SiO}_2$), lithium niobate ($\text{LiNbO}_3$), and lithium tantalite ($\text{LiTaO}_3$) (Lec, 1996). These materials have stable piezoelectric, mechanical and temperature properties and are chemically inert.
Figure 11: Temperature stability of quartz crystal sensor

Certain SiO$_2$ cuts exhibit the unique feature of being temperature compensated over a wide range of temperatures around room temperature, which is important for using the same sensor in many different applications in different conditions without having to compensate for temperature effects on the sensor (Lec, 1996). In this design, an AT-cut quartz is used and the temperature coefficient of the resonant frequency is zero over a very wide temperature range around room temperature.

Table 2 lists the parameters of the sensor used in this study.
Table 2: Sensor operational parameters

<table>
<thead>
<tr>
<th>Property</th>
<th>Value</th>
<th>Units</th>
</tr>
</thead>
<tbody>
<tr>
<td>Density ($\rho$)</td>
<td>2651</td>
<td>kg/m$^3$</td>
</tr>
<tr>
<td>Elasticity ($c$)</td>
<td>$2.947 \times 10^{10}$</td>
<td>N/m$^2$</td>
</tr>
<tr>
<td>Permittivity ($\varepsilon$)</td>
<td>$3.982 \times 10^{-11}$</td>
<td>A$^2$s$^4$/kg m$^3$</td>
</tr>
<tr>
<td>Velocity</td>
<td>7920</td>
<td>m/s</td>
</tr>
<tr>
<td>Electrode Diameter</td>
<td>5</td>
<td>mm</td>
</tr>
<tr>
<td>Sensor Diameter</td>
<td>12</td>
<td>mm</td>
</tr>
<tr>
<td>Thickness</td>
<td>.5</td>
<td>mm</td>
</tr>
<tr>
<td>Resonant Frequency</td>
<td>$\approx 5$</td>
<td>MHz</td>
</tr>
</tbody>
</table>

3.2. Sensor Enclosure

The chamber provides mechanical integrity, proper electrical connections and is depicted in figure 12. It consists of a compartment for the liquid, a housing for piezoelectric sensors and a compartment for electric connectivity. A cross-section view of the measurement chamber is shown below. The liquid compartment is of a cylindrical shape and at its bottom houses a piezoelectric sensor. The piezoelectric sensor is sealed from the liquid with silicon o-rings, and its electrodes are mounted with wires to SMA electrical connectors.

Figure 12: Measurement Chamber
The measurement chamber provides a stable and versatile interface for handling biological liquids and for integration with electronic measurement system.

### 3.3. Electronic Measurement System

The measurement system provides an accurate and versatile technique capable of measuring the parameters of the piezoelectric interfacial sensor, especially the resonant frequency and amplitude.

A schematic block diagram of the measurement system is presented in Figure 13. It consists of four measurement systems based on the Network Analyzer, Vector Voltmeter, Time Domain Analyzer, and Impedance Meter measurement techniques and a PC computer system for data acquisition and signal processing (Lec, 1999). These four measurement systems can be connected sequentially to the sensing chamber for performing the measurements.

![Figure 13: Electrical Measurement System.](image-url)
The common feature of these techniques is that their principle of operation involves the measurement of the impedance of the sensor. They differ in how the specific method implements the impedance measurement principle and their technical specifications. For example, these methods operate in either wide or narrow frequency bands, with varying sensitivities, response times and high accuracy. The Network Analyzer based method provides the most versatile measurement system. It allows for a rapid and wide band scanning of the transmission characteristic of the sensor. The kinetics and the time and frequency domain signatures of the sensor response to media can be obtained easily. The Vector Voltmeter method implements a single frequency measurement technique, which allows for rapid and very sensitive monitoring of processes, but only at one specific frequency. The Time Domain Analyzer measurements are narrow band and sensitive. This method provides the data based on a free decay of the vibrations of the sensor. The impedance meter technique directly supplies the values of the elements of the sensor equivalent electric circuit, which are directly related to the mechanical parameters of media. However, this method is relatively slow and does not allow presentation of data in real time. For this study, the Network Analyzer method was used because of its versatility, rapid response time and ability to monitor wide band characteristic of the sensor. Consequently, the frequency domain signatures of the sensor response to media are obtained easily.

A typical measurement procedure involves measuring of the scattering parameters or $S_{21}$, which is related to the sensor transfer function. The sensing element is connected to the network analyzers as shown in the figure below (Andle et al., 1994).

![Figure 14: TSM sensing resonator connected to the Network Analyzer](image)
A scattering matrix defined for the two-port network for the TSM sensing element and is presented below.

\[
\begin{bmatrix}
V_1^- \\
V_2^-
\end{bmatrix} =
\begin{bmatrix}
S_{11} & S_{12} \\
S_{21} & S_{22}
\end{bmatrix}
\begin{bmatrix}
V_1^+ \\
V_2^+
\end{bmatrix}
\]

(37)

where,

\[
S_{11} = \frac{V_1^-}{V_1^+} \bigg|_{V_2^- = 0} \quad S_{22} = \frac{V_2^-}{V_2^+} \bigg|_{V_1^- = 0} \quad S_{12} = \frac{V_1^-}{V_2^+} \bigg|_{V_1^- = 0} \quad S_{21} = \frac{V_2^-}{V_1^+} \bigg|_{V_2^- = 0}
\]

(38)

The TSM sensing element used in this project is electrically symmetric, so \(S_{11} = S_{22}\) and \(S_{12} = S_{21}\). In this study, \(S_{12}\) is used to characterize the sensor response in air using the Hewlett-Packard 4395A Network/Impedance/Spectrum analyzer. As will be explained in subsequent chapters, liquid loading results in a resonant frequency shift and changes in viscosity result in a decrease in amplitude (Andle et al., 1994). Figure 15 shows the typical sensor response in air and water.

![Figure 15: Typical Sensor response in air and water.](image-url)
4. Experimental Results and Discussion

4.1. Target System Studied

The goal of this work is to explore biomedical applications of a piezoelectric sensor for monitoring interfacial phenomena. Unfortunately, biological systems are very complex. They usually consist of biological entities, such as cells and proteins, suspended in liquid. Such systems belong to a group of liquid media known as colloidal systems. However, proper handling of biological samples is difficult and prone to a variety of ambient factors that make characterizing a new technique more complex. Therefore, for in-depth testing of the proposed sensor, a colloidal system was studied. The target artificial medium was a polystyrene sphere colloidal solution, simulating the cells in suspension. This well-defined medium allows identification of several important processes and makes the calibration the proposed sensor system possible. Measuring endothelial cell properties such as sedimentation, adhesion and fixation is used as a case study to conclude the work.

Two types of mechanical behaviors are possible with colloidal systems. Elastic behavior, exhibited by rigid solids, is observed when the deformation is reversed spontaneously or relaxed when the force is removed. Viscous behavior is observed when a material flows under shear stress, the deformation is not reversed and the material stops flowing when the force is removed. In practice, systems exhibit a combination of elastic and viscous behavior. Colloidal systems under shear motion exhibit complex visco-elastic behavior.

4.2. Laboratory Testing Procedure

To assure the reliability and reproducibility of the testing results, the sensors parameters were measured before each experiment. Characterization of the sensor is important
because the response of the device to an outside stimulation can be better understood if the device’s characteristics are known prior to the application of the stimuli.

Following the pre-measurement characterization procedure, a large number of measurements were performed on the interfacial phenomena sensor to determine if the system was operating within a sensitive region given the size of spheres introduced to the medium.

The parameters of interest are the magnitude of the peak and the frequency, at which the peak occurs. In the figure above, the plots show the sensor response in air and water. The peak amplitude shows a decrease in amplitude and the frequency at which the peak is observed also decreases. If a mass accumulates at the interface, the frequency will change, however, the losses will remain constant. If the mass gets larger and the solid layer thicker, then viscous losses will start to emerge. If structural stiffness builds up or a resonant structure develops, the frequency will increase. If viscous losses decrease, losses will decrease. Observing the evolution of the sensor characteristics as a function of the time gives insight to subtle interfacial processes and allows for their interpretation.

4.3. Calibration with Reference Medium - DMEM

The reference medium, namely Dulbecco's Minimum Essential Medium (DMEM) was used for calibration. DMEM is a buffered saline solution used for cell cultures and cell suspensions. The response of the sensor obtained from the Network Analyzer includes the amplitude and the phase as a function of the frequency. These measurements were also collected as a function of time to determine the stability of the medium alone over time.
Figure 16 represents the amplitude of the sensor response as a function of frequency obtained for three cases: when the sensor is exposed to air, water and DMEM. The reference sensor characterization was always in air. Figure 16 shows when water is added to the measurement chamber the sensor responds in two ways. First, the frequency decreases indicating the addition of an inductive component. As mentioned earlier, an inductive component indicates a mass loading effect coupled through the viscous effect. Second, the peak magnitude decreases indicating the addition of a resistive component. A resistive component indicates the water introduced mechanical losses attributed to an increase in viscosity. Next water was removed from the measurement chamber and replaced with DMEM. Comparing the difference between the sensor response from water versus sensor response from water shows that only the amplitude changed. There was no change in frequency because of the fact that the density of water and DMEM are almost identical. However, the dramatic decrease in amplitude can be attributed to the presence of ions in DMEM that are not present in water. Since the walls on the
measurement chamber are made of brass, the introduction of DMEM creates an electrical path to ground.

The next experiment was to determine the stability of DMEM in the sensor. For this experiment, the measurement chamber was filled with DMEM. Measurements were recorded using a LabView program to record data every minute for 20 minutes.

Figure 17: Sensor frequency stability in DMEM.

The figure above shows the resonant frequency does not vary with time. This measurement is very important because it verifies the fact that there are no time dependant frequency contributions from DMEM.
The figure above shows the resonant amplitude varies slightly with time. The variation is very small, the order of ± .15 dB. The measurement is very important because it verifies the fact that there are no time dependant amplitude contributions from DMEM.

4.4. Colloidal System Behavior

Colloidal systems are very complex systems. This complexity is transferred to the mechanical properties of the system at the sensor/liquid interface. The figure below shows a uniformly mixed colloidal system.
At the interface, the system is more complex than a pure liquid. If the colloids are denser than the liquid, the density at the interface begins to increase because of the presence of the particles. The viscosity also increases due to liquid/particle interaction and particle/particle interaction. The volume occupied by the particles leads to additional viscous dissipation.

As the particles begin to sediment, the density continues in increase. The viscosity also increases due to the increased particle/particle interaction.
Eventually, all the particles will sediment to the bottom and the density will reach a maximum and stop increasing. The viscosity also will reach a maximum when the particles are packed very close together. The equations below show how the average density and viscosity vary with volume fraction (Everett, 1988).

\[
\rho_{\text{col}} = VF_{\text{sphere}} \rho_{\text{sphere}} + (1 - VF_{\text{sphere}}) \rho_{\text{liquid}}
\]

\[
\eta_{\text{col}} = \eta_{\text{liquid}} \left(2.5VF + 6.2VF^2 + \ldots\right)
\]

where \(VF\) is the volume fraction given by \(VF = \text{Volume (Spheres)}/\text{Volume (Liquid)}\).

The figures below show how density and viscosity change as a function of volume fraction.
Figure 22: Changes in density and viscosity as volume fraction of solid particles increases.
Volume fraction and density exhibit a linear relationship. Volume fraction and volume exhibit a parabolic relationship because the higher the volume fraction, the higher the particle/particle interaction and the higher the viscosity.

Penetration depth, shear wave velocity and wavelength of the shear wave penetrating the colloidal system are greatly influenced by the changes in mechanical properties mentioned above.

\[
\delta = \left(\frac{2\eta}{\rho \omega}\right)^\frac{1}{2},
\]

\[
\lambda = \frac{1}{\delta},
\]

\[
v = \omega \lambda
\]

The table below shows how a shear wave behaves in a viscous liquid.

<table>
<thead>
<tr>
<th>Frequency f (MHz)</th>
<th>Penetration Depth (\delta) ((\mu)m)</th>
<th>Velocity (v) ((\mu)/s)</th>
<th>Wavelength (\lambda) ((\mu)m)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>0.25</td>
<td>7.93</td>
<td>1.59</td>
</tr>
<tr>
<td>15</td>
<td>0.15</td>
<td>13.73</td>
<td>0.92</td>
</tr>
<tr>
<td>25</td>
<td>0.11</td>
<td>17.72</td>
<td>0.71</td>
</tr>
</tbody>
</table>

The table above shows that the penetration depth of the wave decreases as the frequency increases. Therefore, varying the frequency and observing the characteristics of the fundamental frequency or any of the crystals odd harmonics, one can measure the mechanical properties of various layers on the medium. As the mechanical properties of at the interface change, so do wave parameters such as penetration depth as shown in the figure below.
Figure 23: Penetration depth as a function of volume fraction for 5 MHz shear wave in water and DMEM.

The figure above shows that the penetration depth increases as the volume fraction of particles increases. The increase is from about .25 µm for water to over .6 µm for a closely packed colloidal system, assuming that the arrangement of spheres is face centered cubic with a 74% packing density.

Depending on the size of the particles, the amount of contact area the spheres make with the surface of the sensor changes greatly. The series of figures below gives a visual picture of the difference in amount of contact spheres of different sizes make with the surface.
Figure 24: Visual representation of surface contact of the spheres with the sensor as size increases.

From the figures above it is very clear that a shear wave penetrating only .25 μm is more likely to interact with a 1 μm polystyrene particle than a 90 μm particle, with 10 μm somewhere in the middle. Therefore, the 1 μm spheres are more likely to have a stronger sensor response.
4.5. Sample Handling

Consistent sample handling is very important for the reproducibility of results when testing such complex and sensitive systems. All liquids were removed from the refrigerator approximately half an hour before each experiment to ensure that temperature stabilizes around room temperature. First, .4 ml of DMEM solution was placed into the liquid chamber and the magnitude and phase response curves were measured and saved using the network analyzer. This measurement served as the baseline for the subsequent measurements after the spheres were added. Then .1 ml of the spheres solution was gently shaken, added and mixed by pipeting and re-pipeting the solution approximately ten times to ensure a uniform mixture. Finally, the automated data acquisition system was started and the magnitude and response curves were collected every minute for 60 minutes.

After the experiment is completed, the sensor is rinsed using tap water. Then the sensor is gently dried with paper towel. Finally, acetone is added to the sensor and the sensor is gently cleaned again with a q-tip and left to dry.

4.6. Results

The sensor was tested with a colloidal suspension of polystyrene spheres with diameters of 1 µm, 10 µm and 90 µm. The concentration of the suspension defined as a volume sphere solution of a given diameter over the total volume of the suspension. The volume fraction of the sphere solution was 2.7 % solid. The measurements were performed at the piezoelectric sensor’s resonant frequency of 5 MHz and the odd harmonics of 15 MHz and 25 MHz. All measurements were performed using a network analyzer based measurement system, which measure the resonant frequency and amplitude of the sensor as a function of time. Typically the measurements were performed over a 1-hour period, with measurements recorded every minute. The following measurements were performed:
4.6.1. Influence of The Sphere Size On The Transient Frequency Response of the Sensor

4.6.1.1. Influence of 1 µm Polystyrene Spheres

Several experiments were conducted using the 1 µm polystyrene spheres. These experiments were conducted at 5, 15 and 25 MHz each for an hour. The figures below show the changes in sensor frequency and amplitude response time at the beginning and at the end experiment when a dilute solution becomes more and more concentrated at the interface and the particles begin to arrange themselves into a stable structure.
Figure 25: Sensor response to 1 µm spheres at 5 MHz.

Figure 25 shows the effects of the 1 µm polystyrene spheres sedimentation and packing has a 5 MHz piezoelectric quartz crystal operating at its fundamental resonant frequency. The frequency decreases as time increases indicating an increase in the inductive element of the equivalent electric circuit. An increase in inductance indicates an increase in mass loading or an increase in the density of the medium surrounding the crystal. Figure 25 also shows the effects of the 1 µm polystyrene spheres sedimentation and packing has on the resonant amplitude. The magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit. An increase in resistance indicates an increase in density and viscosity of the medium surrounding the crystal.
Figure 26: Sensor response to 1 µm spheres at 15 MHz

Figure 27: Sensor response to 1 µm spheres at 25 MHz
Similar behavior is observed at 15 MHz and 25 MHz. However the change in magnitude increases with the frequency indicating the stronger interaction the sensor and the spheres at those frequencies.

### 4.6.1.2. Influence of 10 μm Polystyrene Spheres in Solution

Similar experiments to the 1 μm spheres were conducted using the 10 μm polystyrene spheres. These experiments were conducted at 5, 15 and 25 MHz each for an hour. The figures below show the changes in resonant frequency and amplitude in time. In this case, very different behavior was observed in the sensor response at 15 MHz and 25 MHz.

![Graph](image)

**Figure 28: Sensor response to 10 μm spheres at 5 MHz**

Figure 28 shows the effects of the 10 μm polystyrene spheres sedimentation and packing has a 5 MHz piezoelectric quartz crystal operating at its fundamental resonant frequency.
Again, the frequency decreases indicating an increase in the inductive element of the equivalent electric circuit. This increase in inductance indicates an increase in mass loading of the medium surrounding the crystal. Figure 28 also shows the effects of the 10 µm polystyrene spheres sedimentation on the resonant amplitude. Again, the magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit. This increase in resistance indicates an increase in viscosity of the medium surrounding the crystal.

Figure 29: Sensor response to 10 µm spheres at 15 MHz

Figure 29 shows the effects of the 10 µm polystyrene spheres sedimentation and packing has on a 5 MHz piezoelectric quartz crystal operating at its third harmonic, or 15 MHz. Unlike results obtained with the 1 µm polystyrene spheres, the resonant frequency actually increases. The frequency increases as time increases indicating a decrease in the inductive element of the equivalent electric circuit, which is not possible. Since a decrease in inductance indicates a decrease in mass loading or a decrease in the density
and viscosity of the medium surrounding the crystal, a different model has to be applied to this system. In this model, the polystyrene particle mass is assumed to be attached to another spring with a spring constant that is proportional to the bonding energy between the surface of the sensor and the polystyrene sphere. The coupled oscillator now resonates at a new frequency determined by the additional mass and value of the additional spring constant. For weak particle binding, the system frequency increases, which contradicts normal mass loading theory that states that resonant frequency should decrease as mass is deposited on a resonator. Figure 29 also shows the effects of the 10 µm polystyrene spheres sedimentation and packing has on the resonant amplitude. The magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit. An increase in resistance indicates an increase in viscosity of the medium surrounding the crystal.

![Graph showing sensor response to 10 µm spheres at 25 MHz](image)

**Figure 30:** Sensor response to 10 µm spheres at 25 MHz
Figure 30 shows the effects of the 10 µm polystyrene spheres sedimentation and packing has on a 5 MHz piezoelectric quartz crystal operating at its fifth harmonic, or 25 MHz. Both the frequency shift and the change in magnitude behave similarly as in the case of the 15 MHz experiments.

4.6.1.3. Influence of 90 µm Polystyrene Spheres in Solution

Similar experiments were conducted using the 90 µm polystyrene spheres. These experiments were conducted as previously at 5, 15 and 25 MHz each for an hour. Figures 31-33 show the changes in frequency and amplitude in time. Contrary to the previous experiments, very small changes were observed.

Figure 31: Sensor response to 90 µm spheres at 5 MHz
Figure 31 shows the effects of the 90 µm polystyrene spheres sedimentation and packing has on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The frequency is constant with time due to the small surface contact as depicted in figure 24. Due to the large diameter of the 90 µm spheres, minimal contact is achieved between the surface of the crystal and the spheres. Therefore, the contributions from all the spheres are not large enough to elicit a shift in resonant frequency or peak amplitude.

Figure 32: Sensor response to 90 µm spheres at 15 MHz
Figure 33: Sensor response to 90 µm spheres at 25 MHz

Phenomena similar to those observed at 5 MHz were observed at 15 MHz. At 25 MHz, the penetration depth is very small and the relatively small contact area of the spheres is actually detected by the sensor, in comparison, with the 5 MHz and 15 MHz experiments where no change was detected.

4.6.1.4. Comparison Between Theoretical And Experimental Data

As outlined before, a single model of viscous fluid is used to describe the behavior of the polystyrene colloidal system under shear stress. Initially, the spheres are uniformly distributed in the solution. However, at the end of the experiment when the spheres have accumulated at the sensor surface the model becomes more complex. In table 4, the relative frequency change and the change in amplitude are compared.
Table 4: Experimental sensor data.

<table>
<thead>
<tr>
<th>Frequency (MHz)</th>
<th>1 µm delta f/f</th>
<th>1 µm delta α (dB)</th>
<th>10 µm delta f/f</th>
<th>10 µm delta α (dB)</th>
<th>90 µm delta f/f</th>
<th>90 µm delta α (dB)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5</td>
<td>-4.97E-05</td>
<td>-4.824</td>
<td>-2.49E-04</td>
<td>-4.534</td>
<td>0</td>
<td>0.06</td>
</tr>
<tr>
<td>10</td>
<td>-9.96E-05</td>
<td>-3.572</td>
<td>2.32E-04</td>
<td>-3.839</td>
<td>1.66E-05</td>
<td>-0.327</td>
</tr>
<tr>
<td>25</td>
<td>-8.96E-05</td>
<td>-1.953</td>
<td>4.98E-05</td>
<td>-0.648</td>
<td>0</td>
<td>-0.431</td>
</tr>
</tbody>
</table>

The theoretical sensor response was calculated to determine frequency shift and changes in amplitude (equation 33 and 34). This analysis is based on a very simplified model, where changes in density are calculated with no regard to sphere size (equation 39). The final solid/liquid volume fraction is 74%. This value is based on packing structure of a face center cubic crystal lattice. The results are shown in table 5 as sphere volume fraction increases for a sensor operating at 5 MHz.

Table 5: Theoretical 5 MHz sensor response as volume fraction increases.

<table>
<thead>
<tr>
<th>Volume Fraction</th>
<th>delta f/f (Hz)</th>
<th>Amplitude (dB)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.00E+00</td>
<td>0</td>
</tr>
<tr>
<td>0.1</td>
<td>2.12E-05</td>
<td>-1.26</td>
</tr>
<tr>
<td>0.2</td>
<td>4.69E-05</td>
<td>-2.58</td>
</tr>
<tr>
<td>0.3</td>
<td>7.57E-05</td>
<td>-3.86</td>
</tr>
<tr>
<td>0.4</td>
<td>1.07E-04</td>
<td>-5.04</td>
</tr>
<tr>
<td>0.5</td>
<td>1.39E-04</td>
<td>-6.13</td>
</tr>
<tr>
<td>0.6</td>
<td>1.72E-04</td>
<td>-7.13</td>
</tr>
<tr>
<td>0.7</td>
<td>2.07E-04</td>
<td>-8.05</td>
</tr>
<tr>
<td>0.74</td>
<td>2.20E-04</td>
<td>-8.40</td>
</tr>
</tbody>
</table>

The experimental results seem to be in excellent with the theoretical calculations especially for the 1 µm spheres. Frequency shift and changes in amplitude are about the same order of magnitude as theory suggests for both the 1 µm and 10 µm spheres at 5 MHz. Variation in theoretical and actual numbers result from variations in volume fraction and density the penetration depths of .25 µm. Another problem is, as sphere size increases, the volume fraction approximation becomes less accurate because the volume fraction of solid is much smaller than for larger spheres than smaller spheres at the shallow layer penetrated by the wave. This model is very simple, is very qualitative and
requires further development to account the penetration depth of the wave as particle size varies.

4.6.2. Influence of Sphere Size As A Function Of Time on Sensor Response

In these experiments, the influence of spheres size on the resonant amplitude and frequency as a function of time for a given frequency at 5, 15 and 25 MHz was examined.

4.6.2.1. Influence of The Sphere Size on Resonant Amplitude as A Function of Time

![Figure 34: Influence of sphere size on sensor amplitude at 5 MHz](image)

Figure 34 shows the effects of the 1 µm, 10 µm and 90 µm polystyrene spheres sedimentation and packing has on the resonant magnitude of a 5 MHz piezoelectric
quartz crystal operating at its fundamental frequency. The magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit for both the 1 µm and 10 µm spheres. An increase in resistance indicates an increase in viscosity and density of the medium surrounding the crystal. The amplitude of the 10 µm spheres stabilizes quicker than the 1 µm spheres. This is due to the fact that the 1 µm spheres travel slower and take longer to reach the sensor surface than the 10 µm spheres. The plot also seems to be noisier for the 10 µm spheres than for the 1 µm spheres. One of the possible explanations for the “noise” is the 10 µm spheres are less packed than the 1 µm spheres and tend to move around the gaps. These motions results in fluctuation in viscosity and are detected by the sensor. The 90 µm spheres are virtually undetected by the sensor due to the relatively little contact they make with the sensor (See figure 22).

Figure 35: Influence of sphere size on sensor amplitude at 15 MHz

Figure 35 shows the effects of the 1 µm, 10 µm and 90 µm polystyrene spheres sedimentation and packing has on the resonant magnitude of a 5 MHz piezoelectric
quartz crystal operating at 15 MHz. The magnitude decreases as time increases as it did in the previous case at 5 MHz. The plot also seems to be less noisy than the plot for 5 MHz. The reason for the decreased noise is sensor is penetrating less into the medium and is less prone to detecting fluctuations as the spheres move with in their structure. In Addition, the plots do not stabilize in the measure time period as they did in the 5 MHz experiment. This lack of stabilization is even more prominent in the 25 MHz experiments, which suggests that the sensor is more sensitive to interfacial changes as the penetration depth decreases.

Figure 36: Influence of sphere size on sensor amplitude at 25 MHz

The figure above shows the effects of the 1 µm, 10 µm and 90 µm polystyrene spheres sedimentation and packing has on the resonant magnitude of a 5 MHz piezoelectric quartz crystal operating at 25 MHz. Like the previous cases, the magnitude decreases as time. However, the amplitude of the 10 µm spheres does not shift as it did in the case of 5 and 15 MHz because the shear wave is penetrating less distance into the 10 µm spheres
and sensing more liquid than solid. The 90 µm spheres are detected by the sensor due to the relatively small penetration depth of the wave generated by the sensor.

4.6.2.2. Influence of The Sphere Size on Resonant Frequency as A Function of Time

The resonant frequency changes exhibit a similar behavior to the resonant amplitude changes. For 5 MHz, the behavior is typical of a Newtonian liquid and a decrease in frequency occurs. For 15 MHz and 25 MHz, an increase in frequency is observed for the 10 µm spheres, but a decrease is observed to the 1 µm spheres.

Figure 37: Influence of sphere size on frequency at 5 MHz

Figure 37 shows the effects of the 1 µm, 10 µm and 90 µm polystyrene spheres sedimentation and packing has on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The frequency decreases as time...
increases indicating an increase in density and viscosity of the medium surrounding the crystal. The frequency shift of the 10 µm spheres stabilizes quicker than the 1 µm spheres. This is probably due to the fact that the 1 µm spheres travel slower and take longer to reach the sensor than the 10 µm spheres. The plot for the 10 µm spheres seems noisier for the reason mentioned above related to the motion of the spheres. Another interesting observation is that the frequency shift is smaller for 1 µm than for 10 µm. This may be due to and increase in the elasticity of the medium that forces a frequency increase. The 90 µm spheres are virtually undetected by the sensor due to the relatively little contact they make with the sensor.

![Figure 38: Influence of sphere size on frequency at 15 MHz](image)

The figure above shows the effects of the 1 µm, 10 µm and 90 µm polystyrene spheres sedimentation and packing has on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at 15 MHz. The frequency decreases as time increases indicating an increase in the density and viscosity of the medium surrounding the crystal. Unlike
results obtained with the 1 µm polystyrene spheres, the resonant frequency actually increases for the 10 µm spheres. The frequency increases as time increases indicating spring constant as outlined earlier. For weak particle binding, the system frequency should increase, which contradicts normal mass loading theory that states that resonant frequency should decrease as mass is deposited on a resonator. The 90 µm spheres are virtually undetected by the sensor due to the relatively little contact they make with the sensor.

Figure 39: Influence of sphere size on frequency at 25 MHz

The figure above shows the effects of the 1 µm, 10 µm and 90 µm polystyrene spheres sedimentation and packing has on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at 25 MHz. The behavior is the same as that observed at 15 MHz. The model of the coupled resonator can qualitatively explain observed phenomena. This means that the strength of the interaction between the sphere and substrate can be calculated.
4.6.3. Change In The Resonant Frequency and Amplitude as a Function of Sphere Size

The influence of sphere size on frequency and amplitude after the 60-minute experiments was determined to be linear at 5 and 15 MHz. At 25 MHz, the sensor seems less sensitive due to the very shallow penetration depth. For smaller spheres, the relationship is no longer linear due to their slow sedimentation and increased contact area. The figures below show plots of sphere size versus frequency shift and change in amplitude.

The figures below show that a linear relationship exists between sphere size and sensor response, especially in the amplitude plot. Therefore, using this technique an unknown sphere size can be determined using piezoelectric interfacial biosensor.

Figure 40: Influence of sphere size on sensor frequency shift after 60 minutes
4.6.4. Influence of Concentration of Spheres on Sensor Response

Several experiments were conducted using the 10 µm polystyrene spheres to determine if concentration influences the sensor response. These experiments were conducted at 5, 15 and 25 MHz each for an hour. The figures below show the changes in amplitude and frequency in time.
4.6.4.1. Influence of The Concentration on Resonant Amplitude as a Function of Time

Figure 42 shows the effects of varying the concentration of 10 µm-polystyrene spheres on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The amplitude decreases as time increases as usual, however they stabilize at different amplitudes. The reason may be due to changes in viscosity, but are more likely due to variations from making measurements on different days.
Figure 43: Influence of concentration on sensor amplitude at 15 MHz

Figure 43 shows the effects of varying the concentration of 10 µm-polystyrene spheres on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at 15 MHz. Same behavior is exhibited as in the 5 MHz case. However, the amplitudes do not stabilize during the time of the experiment. A possible explanation is that the spheres could be coming closer and closer to the sensor as time goes by and these changes are not observed at lower frequencies.
Figure 44 shows the effects of varying the concentration of 10 µm-polystyrene spheres on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at 25 MHz. The amplitude shift has about the same slope for all concentrations, but does not stabilize during the time of the experiment. This effect may be caused by higher sensitivity of the 25 MHz wave to interfacial effects, which are not stationary.
Figure 45 shows the effects of varying the concentration of 10 µm-polystyrene spheres on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The resonant frequency decreases as time increases, which is similar for both power experiments, but they stabilize at different frequencies.

**Figure 45: Influence of concentration on frequency shift at 5 MHz**
Figure 46 shows the effects of varying the concentration of 10 µm-polystyrene spheres on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. Unlike results obtained with at 5 MHz, the resonant frequency actually increases at 15 MHz, however they stabilize at different frequencies.
The figure above shows the effects of varying the concentration of 10 µm-polystyrene spheres on the resonant frequency of a 5 MHz piezoelectric quartz crystal operating at 25 MHz. The results are similar to those obtained at 15 MHz, however there may be a trend where the frequency increase results in a larger frequency shift. More experiments are required to determine if there is a correlation.

### 4.6.5. Influence of Power on Sensor Response

A piezoelectric shear wave sensor operates as an active device since it transmits mechanical motion into the surrounding medium. In this case, the medium is a colloidal suspension of polystyrene spheres and a suspension of endothelial cells. The transmitted mechanical energy may have an impact on the properties of the tested medium and...
biological processes or start new processes. Therefore, it is important to evaluate the impact of the acoustic energy on the measurements.

Although the amplitude of the mechanical motion is very small, on the order of 10 nanometers, the frequency of the motion high and on the order of MHz with a penetration depth on the order of 100 nanometers where all the acoustic energy is dissipated. So far, existing literature points out that the acoustic affect is negligible. However, the studied colloidal system because of its suspended particles and their arrangement are very sensitive to acoustic motion.

The measurements were performed at two levels of power excitation. The first is –10 dBm, which corresponds to .1 mW and the second is +10 dBm, which corresponds to .01 W.

### 4.6.5.1. Influence of Power on Sensor Amplitude

Several experiments were conducted using the 1 μm-polystyrene spheres to determine if concentration influences the sensor response. These experiments were conducted at 5, 15 and 25 MHz each for an hour. The figures below show the changes in frequency and amplitude in time.
The figure above shows the effects of varying the sensor power on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The amplitude decreases for all powers used.

Figure 48: Influence of power on sensor amplitude at 5 MHz

The figure above shows the effects of varying the sensor power on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The amplitude decreases for all powers used.
The figure above shows the effects of varying the sensor power on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at 15 MHz. The amplitude decreases for all powers used. There is very small variation in the final amplitude between the two powers used. Further investigation is required to determine the source of the variations.

Figure 49: Influence of power on sensor amplitude at 15 MHz
The figure above shows the effects of varying the sensor power on the resonant amplitude of a 5 MHz piezoelectric quartz crystal operating at 25 MHz. Similar results were observed as shown in figure 48. Further investigation is required to determine the source of the variations.

4.6.5.2. Influence of Power on Sensor Frequency

Similarly, the change of the resonant frequency of the 1 um sphere colloidal system was measured and the changes are presented in figures 51-53. Much like the amplitude data, the frequency data does not vary as power changes, except for 25 MHz where there is some small variations. However, this issue needs to be studied further with more experiments in the future.
Figure 51: Influence of power on frequency shift at 5 MHz

Figure 52: Influence of power on frequency shift at 15 MHz
4.6.6. Influence of Cells Sedimentation on Sensor Response

Several experiments were conducted using endothelial cells to determine their influence on sensor response. These experiments were conducted at 5, 15 and 25 MHz each for an hour. The concentration of cells used was about 50,000 cells/ml and about .2 ml of solution was used. These concentrations ensured that the surface of the sensor would be completely covered after sedimentation. The figures below show the changes in frequency and amplitude in time.
The figure above shows the effects of the endothelial sedimentation and adhesion has on the resonant magnitude of a 5 MHz piezoelectric quartz crystal operating at its fundamental frequency. The magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit. An increase in resistance indicates an increase in viscosity and density of the medium surrounding the crystal. The amplitude stabilizes quickly as it did with the polystyrene spheres.

**Figure 54: Influence of cells on sensor amplitude at 5 MHz**
The figure above shows the effects of the endothelial sedimentation and adhesion has on the resonant magnitude of a 5 MHz piezoelectric quartz crystal operating at 15 MHz. The magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit. An increase in resistance indicates an increase in viscosity of the medium surrounding the crystal. The amplitude never stabilizes as it did with the polystyrene spheres, indicating that the more process that are increasing the viscosity at the interface are taking place.

Figure 55: Influence of cells on sensor amplitude at 15 MHz
The figure above shows the effects of the endothelial sedimentation and adhesion has on the resonant magnitude of a 5 MHz piezoelectric quartz crystal operating at 25 MHz. The magnitude decreases as time increases indicating an increase in the resistive element of the equivalent electric circuit. An increase in resistance indicates an increase in viscosity of the medium surrounding the crystal. The amplitude appears ready to stabilize around 15 minutes, but is followed by a large drop. This drop may be the beginning of the adhesion process. Then it continues to drop and never stabilizes as it did with the polystyrene spheres, indicating that the more process that are increasing the viscosity at the interface are taking place.

4.6.7. Influence of Cells Culture and Fixation on Sensor Response

An experiment was conducted where endothelial cells were cultured on the surface of the sensor using to determine their influence on the sensor’s response. These experiments
were conducted at 5, 15 and 25 MHz each for an hour. The figures below show the changes in frequency and amplitude in time.

![Graph showing changes in frequency and amplitude](image)

**Figure 57: Influence of cells cultured and fixed on sensor amplitude**

For the 5 MHz, 15 MHz and 25 MHz The amplitudes steadily decreased indicating the presence of the cells and the proliferation of new ones. At the end of the second day, the medium was removed and replaced with formaldehyde, which is a fixative that causes the cells to become stiffer. The sensor shows these changes with a dramatic increase in amplitude.

### 4.7. Discussion

The studied sensor consists of a thickness shear mode piezoelectric quartz crystal resonator and measurement system, based on a Network Analyzer System. Preliminary results show that the interfacial phenomena sensor can be used to monitor phenomena
taking place at the interface of the liquid and the sensor. The piezoelectric interfacial phenomena sensor was tested with the polystyrene colloidal system and proved with a biological system. The sensor can monitor the sedimentation process, changes in interfacial density, changes in interfacial stiffness, changes in interfacial viscosity. The piezoelectric interfacial phenomena sensor can differentiate between different sizes of spheres in suspension. The sensor, operating at different frequencies, is capable of “slicing” the media and revealing different phenomena. The sensor is sensitive to interfacial structural arrangement. The sensor can monitor sedimentation, adhesion and fixation of cells. The sensor exhibits high sensitivity, reproducibility and stability. The sensor offers a promising technique for monitoring a variety of interfacial phenomena in real-time.

4.8. Applications

The piezoelectric interfacial biosensor has tremendous potential in areas such as:

- Tissue engineering, where the mechanical properties of cells and tissue is very important
- Biocompatibility, where cell adhesion and proliferation plays a vital role in success of implants
- Pharmacology, where influence of drugs on biological processes is crucial before trials begin
- Other application where the interaction between and medium and a solid surface can yield important information about the behavior of the system

Piezoelectric sensors are highly sensitive, small devices with rapid response, high accuracy, compatibility with Integrated Circuit (IC) technology, outstanding aging characteristics and the ability to measure numerous quantities in one sensor enclosure. They provide an excellent platform for developing sensors for all application mentioned above.
5. Summary, Conclusion and Future Work

5.1. Summary and Conclusions

Preliminary results show that the interfacial phenomena sensor can be used for the monitoring of colloidal systems. The sensor consists of a thickness shear mode piezoelectric quartz crystal resonator and measurement system, in this case a Network Analyzer. The interfacial phenomena sensor is capable of differentiating between different sizes of spheres in suspension by monitoring changes of the mechanical properties at the interface as solid sediments to the bottom of the chamber and comes in contact with the crystal.

The most important feature of the interfacial phenomena sensor is its sensitivity to small changes in viscosity and large dynamic range. By automating the measurement equipment, remote operation can be easily implemented. The accompanying electronics can also be miniaturized and made of small, inexpensive, common components. The knowledge gained from this project can be used in other applications, such as monitoring industrial processes where colloidal systems play an important role and medical applications where sedimentation rates of solids in biological fluids can determine the degree of health or disease of an individual.

5.2. Future Work

The interfacial phenomena sensor presented in this study has proven to be sensitive enough to warrant further investigations of possible applications. However, some basic optimizations should be performed before further study of the sensor. First, a redesigned measurement chamber must be constructed of non-conductive material to prevent the leakage current that decreases the amplitude sensitivity. Second, several sensors must be
constructed to accommodate different volumes of liquids under investigation for different investigation. Third, the systems would benefit from comparative studies of different colloidal systems, where the behavior can be simply modeled and compared to sensor response. Fourth, an advanced theoretical model explaining quantitatively the operation of the sensor needs to be developed. Finally, the sensor should be tested with different biological systems, such as cells and proteins.

An interfacial phenomena sensor has tremendous potential in many different applications. The sensor and the measurement chamber can be customized for a variety of functions. The signal processing and data analysis components can also be miniaturized for real-time measurements.
References


Lec RM, "Introduction to Biosensors, BMES-401", Lecture Notes, Drexel University, 1999


