
1NINA KORLINA MADZHI, 2ANUAR AHMAD, 1LEE YOOT KHUAN, 1FIRDAUS ABDULLAH
1Faculty of Electrical Engineering, Universiti Teknologi MARA,
40450 Shah Alam, Selangor
MALAYSIA
2Faculty of Engineering,
Universiti Industri Selangor,Selangor
MALAYSIA
nina6875@yahoo.com

Abstract: This paper deals with the development of Piezoresistive Microcantilever biosensor and the signal transduction to detect human stress by using salivary alpha amylase activity. A Piezoresistive Microcantilever biosensor can be used to detect saliva-amylase activity by deflecting upon interaction with a specific receptor. By measuring the amount of bending the microcantilever beam experiences in response to interactions with the molecules, and the amount of analyte in the solution can be quantified. When the Microcantilever beam deflects it caused the stress change within the microcantilever beam and applied strain to the piezoresistor material thereby causing the resistance change which can be measured with the Wheatstone Bridge circuit. The Piezoresistive Microcantilever sensor integrated with transducer components converts the biochemical signal into measurable signal when it react with salivary amylase enzyme. The enzyme concentration signal is converted to a voltage signal by the transducer. The device was designed specifically that it enables the small resistivity change due to the enzymatic reaction to be measured.

Key-Words: Biosensor, Piezoresistive, Microcantilever, Signal Transduction, Resistance change, Saliva, Alpha Amylase

1 Introduction
A biosensor is commonly defined as an analytical device that uses a biological recognition system to target molecules or macromolecules. The great development of biosensors for numerous diagnosis of infectious diseases, detection of oxidizing of free radicals in saliva[1], glucose determination[2-5] and also stress measurements[6] has lead to the technological advancement of microsensors for biological sensing.

Biosensors can be coupled to physiochemical transducers that convert this recognition into a detectable output signal. Typically biosensors are comprised of three components: the detector, the transducer and the output system which involves amplification and display the output in an appropriate format.

A microcantilever biosensor is a device that can act as a physical, chemical or biological sensor by detecting changes in microcantilever bending or vibrational frequency. Microcantilevers are simple mechanical devices. They are tiny plates or leaf springs, typically 0.2-1µm thick, 20-100µm wide, and 100-500µm long, which are connected on one end to an appropriate support for convenient handling.

2 Problem Formulation
Biosensing applications demand fast, easy-to-use, cheap, and highly sensitive methods for the recognition of biomolecules. A high degree of parallelization is also desirable because of the demands made by the pharmaceutical industry for high-throughput screening. All these points can be fulfilled by micromachined cantilever sensors, which are ideal for biosensing applications. An increasing number of reports confirm the potential of Microcantilever (MC) sensors for environmental such as gas detection, mass effect and gas sensitivity[7] and biomedical application[3].

The sensitivity of a microcantilever biosensor depends on its ability to convert biochemical
interaction into micromechanical motion of the microcantilever. The deflections of the microcantilever biosensor are usually of the order of few tens to few hundreds of a nanometer. Such extremely low deflection requires an advanced instrument for accurately measuring the deflections.

As a consequence, most of the applications of microcantilever biosensors are done in laboratories equipped with sophisticated deflection detection and readout techniques. This paper proposes and analyses a self-sensing Piezoresistive Microcantilever for electrical measurement of microcantilever deflection. Microscale cantilever beams can be used to detect biomolecules by deflecting upon interaction with a specific biomolecule as in Fig. 1[8, 9].

By measuring the amount of bending each microcantilever beam experiences in response to interactions with the molecules, the amount of analyte in the solution can be quantified.

3 Methodology

A. Piezoresistive Microcantilever Deflection Detection

Piezoresistive Microcantilever deflection method involves the embedding of a piezoresistive material such as doped polysilicon at the top surface of the microcantilever to record the stress change [8]. When the microcantilever beam deflects a stress change occurs within the beam that will apply strain to the piezoresistor. Thereby causing a change in resistance that can be measured by electronic instruments. The resistance of the piezoresistive material changes when strain is applied to it. The relative change in resistance as function of applied strain can be defined as

\[ \frac{\Delta R}{R} = K \delta \]  (1.1)

Where K is a Gauge Factor which is an important material parameter, \( \delta \) is the strain in the material and R is the piezoresistor resistance.

B. Thin film Piezoresistive Microcantilever Fabrication

The fabrication process started from patterning a 0.9µm –thick photoresist of Boron Phosphosilicate Glass(BPSG) sacrificial layer on a silicon substrate by standard photolithography. The microcantilever beam is then formed by depositing a polysilicon layer of 5000Å (0.5µm) thickness using Low Pressure Chemical Vapor Deposition (LPCVD). Next, a 500nm-thick Silicon Nitride (SiN) layer are deposited by Plasma Enhanced Chemical Vapor Deposition (PECVD) which will act as an insulator. Another polysilicon layer is then deposited with a dimension of 195µm x 75 µm u–shape resistor pattern and blanket implanted to achieve a resistor value of 1.2kΩ. Then the electrode pad was patterned and deposited with Aluminum and finally the cantilever beam is released by wet etching. The cross section SEM image of the designed piezoresistive microcantilever is as shown in Fig. 2.

By measuring the amount of bending each microcantilever beam experiences in response to interactions with the molecules, the amount of analyte in the solution can be quantified.

C. Wheatstone Bridge Circuit design

Fig. 4 shows a Piezoresistor Microcantilever which can be connected to a Wheatstone Bridge circuit as shown in Fig. 4.
For a piezoresistor embedded on the surface of the microcantilever has a length of \( l \ \mu m \), with cross-section area of \( A \ \mu m^2 \) and a resistivity of \( \rho \ \Omega \mu m \), the resistance is given by

\[
R = \frac{\rho l}{A} \ \Omega \quad (1.2)
\]

When the piezoresistor material is stressed mechanically by a load \( W \) newtons, a stress, \( \sigma \) occurs where

\[
\sigma = \frac{W}{A} \quad (1.3)
\]

By using a Taylor’s series expansion method on resistance \( R \), the resistance changes can be determined by:

\[
\Delta R = \left( -\frac{L}{A^2} \right) \Delta A + \left( \frac{L}{A} \right) \Delta \rho + \left( \frac{\rho}{A} \right) \Delta L \ \Omega \quad (1.4)
\]

Then, to obtain the fractional change in \( R \), divide eqn. 1.4 with eqn. 1.2 and we will get

\[
\frac{\Delta R}{R} = -\frac{\Delta A}{A} + \frac{\Delta \rho}{\rho} + \frac{\Delta L}{L} \quad (1.5)
\]

A differential amplifier is used to measure biomedical signals where it’s applied between the inverting and non-inverting input of the amplifier. The signal is then amplified by the differential gain of the amplifier. Fig. 5 shows the sensor integration consist of Wheatstone bridge and different op-amp circuit.

![Fig.5: Sensor Integration Circuit](image)

If the following resistor ratios equal, \( R_6/R_5 = R_6/R_5 \), the output voltage is:

\[
\Delta V = V_o \left( \frac{R_3}{R_1 + R_2} - \frac{R_4}{R_3 + R_4} \right) \quad (1.6)
\]

Where \( R_3 = R + \Delta R \)

4 Results

From testing with the actual Piezoresistive Microcantilever sensor, it is found to have a resistance value of 5.767 kilo ohms. Table 1 shows the voltage output from the bridge at and slightly off the null bridge conditions. It can be confirmed that the null bridge condition is obtained when \( R_2 \) equals 6.245 kilo ohms for actual sensor testing.

<table>
<thead>
<tr>
<th>R3 (kΩ)</th>
<th>R2 (kΩ)</th>
<th>Vout (mV) (theoretical Calculation)</th>
<th>Vout (mV) (experimental)</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.767</td>
<td>6.000</td>
<td>-49.40</td>
<td>-45.102</td>
</tr>
<tr>
<td>5.767</td>
<td>6.100</td>
<td>-28.88</td>
<td>-24.37</td>
</tr>
<tr>
<td>5.767</td>
<td>6.200</td>
<td>-8.50</td>
<td>-8.04</td>
</tr>
<tr>
<td>5.767</td>
<td>6.210</td>
<td>-6.50</td>
<td>-6.143</td>
</tr>
<tr>
<td>5.767</td>
<td>6.220</td>
<td>-4.40</td>
<td>-4.247</td>
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<td>-2.176</td>
</tr>
<tr>
<td>5.767</td>
<td>6.240</td>
<td>-0.40</td>
<td>-0.519</td>
</tr>
<tr>
<td>5.767</td>
<td>6.241</td>
<td>-0.180</td>
<td>-0.955</td>
</tr>
<tr>
<td>5.767</td>
<td>6.242</td>
<td>0.020</td>
<td>-0.481</td>
</tr>
<tr>
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<td>6.243</td>
<td>0.220</td>
<td>-0.374</td>
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<tr>
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<td>0.420</td>
<td>-0.059</td>
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<td>5.767</td>
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<td>0.620</td>
<td>0.414</td>
</tr>
<tr>
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<td>6.246</td>
<td>0.820</td>
<td>0.616</td>
</tr>
<tr>
<td>5.767</td>
<td>6.247</td>
<td>1.021</td>
<td>0.883</td>
</tr>
<tr>
<td>5.767</td>
<td>6.248</td>
<td>1.221</td>
<td>1.241</td>
</tr>
<tr>
<td>5.767</td>
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<td>1.481</td>
</tr>
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<td>5.767</td>
<td>6.250</td>
<td>1.600</td>
<td>1.623</td>
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<tr>
<td>5.767</td>
<td>6.300</td>
<td>11.50</td>
<td>10.768</td>
</tr>
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</table>

With reference to experimental outcome on the deflection of Piezoresistive Microcantilever range, a range of 6.245 to 6.25 kilo ohms is chosen as variable resistance range. The output from the differential amplifier ranges from 0.616 millivolts to 1.623 millivolts on actual experiment. A discrepancy within 13.16% (Table 4.2) on the average is detected, which could be attributed to tolerances of electronic components and wiring.

Table 4.2: Integration of Sensor and Transduction Stage

<table>
<thead>
<tr>
<th>R3 (kΩ)</th>
<th>R2 (kΩ)</th>
<th>Vo1 mV (Theoretical)</th>
<th>Vo1 mV (Experimental)</th>
<th>% Discrepancy</th>
</tr>
</thead>
<tbody>
<tr>
<td>5.767</td>
<td>6.246</td>
<td>0.820</td>
<td>0.616</td>
<td>24.88</td>
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<td>5.767</td>
<td>6.247</td>
<td>1.021</td>
<td>0.883</td>
<td>13.52</td>
</tr>
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<td>1.221</td>
<td>1.241</td>
<td>-1.64</td>
</tr>
<tr>
<td>5.767</td>
<td>6.249</td>
<td>1.420</td>
<td>1.481</td>
<td>-6.40</td>
</tr>
<tr>
<td>5.767</td>
<td>6.250</td>
<td>1.600</td>
<td>1.623</td>
<td>-1.44</td>
</tr>
</tbody>
</table>
Fig. 6 depicts the outcome from a comparative study between theoretical and experimental results with the integration of sensor and transduction stage. It can be observed that the voltage output from the differential amplifier is linearly related to the resistor, $R_2$, the variable resistor.

![Graph showing the relationship between voltage output and resistor value](image)

**Fig. 6** Comparative study between Theoretical, Simulation and Experimental results on output voltage of Integration of Sensor and Transduction Stage

### 5 Conclusion

The Piezoresistive Microcantilever biosensor can be used to detect the small biological signal in response to the proposed biosensor system. The deflection of the Microcantilever beam caused a resistance change within the beam and therefore generated signal which is converted to voltage by the Wheatstone Bridge circuit. By investigating the integration of the Piezoresistive Microcantilever sensor with the developed transducer, the result shows that the percentages different between the software simulation and the hardware developed transducer was very low and insignificant to each other. Thus, it is proven with theoretical result. The software simulation and hardware implementation have been successfully completed; this finding is useful for the future enhancement of the bioamplifier design.

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### REFERENCES


