# DESIGN AND SIMULATION OF MEMS BASED MICRO HEATER FOR DNA AMPLIFICATION

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**Abstract**: PCR (polymerase chain reaction) is a recent well researched technique which can be employed to obtain an amplified DNA (Deoxyribonucleic Acid).A continuous flow polymerase chain reaction with integrated MEMS based design micro heater for DNA amplification is implemented. Temperature zone is obtained by natural heat conduction rather than making use of typical silicon thermal cycler. In this process, the DNA sample with reagents has to be heated and cooled in a controlled manner. By doing this, the two strands of the DNA are split and each strand becomes a template by itself which can be copied and replicated and thus the amplification of the DNA sample takes place.Inter connect based upon LTCC or PDMS can be employed for integration. The authors of this paper will be looking in detail about the design implementation of the two way heater, and also will be looking into details about the temperature variation obtained while using two different types of sample (saliva and blood) for DNA amplification. The simulations were carried out using COMSOL Multi physics.

Keywords*: **MEMS based micro heater, PCR chip, DNA amplification, PDMS,LTCC***

## 1. Introduction

The introduction of DNA amplification for the first time was given by researcher Kleppe and his research groups. The first introduced amplification was not recognized until 1985 by saiky and group who demonstrated the amplification and ever since it is the key component of PCR models used today [1]. The PCR (polymerase chain reaction) is well know as a molecular photocopying of single to few copies of DNA and is the fasted emerging indispensable technique that is employed for the amplification of a specific gene or region of a DNA sample across several orders of magnitude. Due to its inherent simplicity PCR is known as an ideal bio analytical procedure for miniaturization. PCR finds wide range of application ranging from diagnosis of infection disease, environmental monitoring, scientific research, DNA Isolation to amplification and quantification of DNA. In addition to the diagnosis of malignant diseases PCR is applicable to diagnosis of viral DNA. One of the most recent powerful DNA amplification technique employed is multiplex PCR which makes use of multiple primer set instead of one primer set, hence the name multiplex PCR. Also PCR reaction is conducted for a small portion of DNA sample for example, reaction of 50-1000 bases rather than entire human genome of more than 3000 bases [2]. The paper proposed makes use of Micro fluidic PCR device that is based on continuous flow. Development of many PCR devices is influenced by micro fabrication technique. There are two types of PCR devices. One is the conventional PCR device and the other being RTPCR (Real Time Polymerase Chain Reaction). In the conventional PCR, the process is carried out inside the machine within the tube. On completion of the process the agarose gel electrophoresis is used in order to analyze the amplified DNA. The major drawback of this process is the time consumption (2-3 hours); also the time that is required analyzes the amplified DNA which makes this process practically not feasible. Not only the time consumption but also the unawareness of the initial status of nucleic acid and the number of copies of the DNA obtained that is unknown makes it not applicable in the field. The use of the agarose gel electrophoresis is not required in the real time PCR process and flour meter is employed to monitor the DNA amplification. The machine emits light which is captured by the optic compound of the machine and is analyzed. The disadvantage that comes along with the RTPCR is the limitation of the light received by optic component of the machine [3]. In this paper we offer two way heater designs in order to implement the DNA amplification rather than traditional one way or three heater design. The first section briefs about the PCR methodology describing the principle behind the amplification process of DNA followed by the design and implementation. Lastly we elaborate on the simulation that we have conducted and the results obtained while using two types of sample.

## 2. PCR Methodology:

Deoxyribonucleic Acid is a molecule that is present in each and every living organism which encodes the genetic information which is used in the development and functioning of all living organisms. Polymerase Chain Reaction is currently the best technique that can be used in order to amplify any amount of DNA. The paper proposed makes use of Micro PCR device that is based on continuous flow. In this process the DNA sample flows through the three well defined temperature zones. Temperature zone is obtained by natural heat conduction rather than making use of typical silicon thermal cycler. In this process, the DNA sample with reagents has to be heated and cooled in a controlled manner.

By doing this, the two strands of the DNA are split and each strand becomes a template by itself which can be copied and replicated and thus the amplification of the DNA sample takes place [4]. When the DNA samples are made to flow through the micro channel, the samples experience three distinct temperature zones as shown in figure 1.

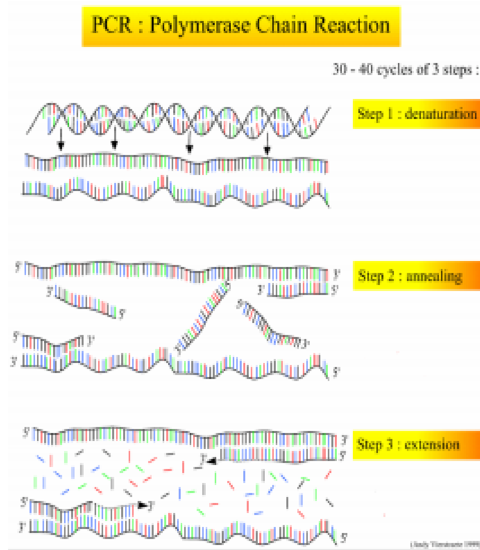


Figure 1: PCR mechanism [1]

Denaturation at a temperature variation of 90-98$^{0}$C for a duration of 1-2minutes, Renaturating (annealing) at a temperature cooled down to 45-60$^{0}$C for duration of 45 seconds, Extension (hybridization) over a temperature variation of 72$^{0}$C for a duration of 2 minutes. Until enough amount of DNA sample is established this three step process is carried out usually 20-30 times. Heating and cooling of the sample for about 20-30 times demand for the sample to be enough so that the process does not evaporate the sample completely drying it up with no DNA left for amplification. This paper proposes to introduce a new concept of obtaining three temperature zones rather than using copper block to provide the temperature zone. The major drawback of using copper blocks is the thermal cross talk between the temperature zones. The limitation of thermal cross talk is eliminated in this paper. Lateral heat conduction is used in order to create the temperature zone renaturating rather than direct micro heater heating. Lateral Heat conduction of denaturation and Extension (hybridization) is employed for renaturating zone heating [5].

## 3. Design and Implementation

In order to conduct the DNA amplification the reaction mixture of the sample has to be cycled several times through the temperature zones. The authors have proposed a design of obtaining three temperature zones using three different independent thin film heating element which is brought together in one platform using glass wafer as depicted in figure 2. In continuous flow PCR the cyclic hold time is independent of repetitive temperature variation. The DNA sample is flown through the micro channel with the help of simple mechanical injector. The input pressure and the initial velocity of the fluid highly influence the sample flow. When the sample is made to flow through the micro channel, the sample experiences three different heating at distinct temperature zone. There are various ways to design the temperature zones namely one heater, two heater and three heater design. In this paper two heater designs is employed. In three heater temperature design, the temperature is obtained distinctly. This type of design is governed with large amount of cross talk and complication in design process.

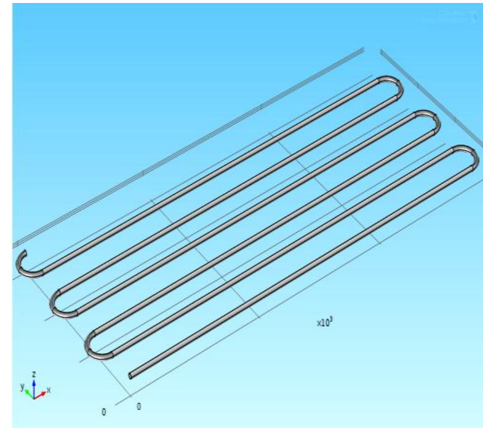

Figure 2: structure of micro heater design

In one heater design, the denaturation temperature is obtained by one micro heater and the temperature in Extension and Renaturating is established by lateral heat conduction through the glass chip structure. This type of design eliminates the cross talk and complexity in the design faced previously in three heater design, however this type of design may lead to non informality. The design used in this paper is two heater design where two separate heaters are used to obtain the uniform temperature in denaturation and replication where as the temperature zone in hybridization is obtained through lateral heat conduction through glass substrate. This type of design is obtained by using specific dimension which is listed in table 1.

| Parameter | Major radius | Minor radius | Unit |
|---|---|---|---|
| Cylinder 1 | 5.5 | 1400 | $\mu m$ |
| Cylinder 2 | 7.5 | 1400 | $\mu m$ |
| Torus 1 | 50 | 5.5 | $\mu m$ |
| Torus 2 | 50 | 7.5 | $\mu m$ |

Table 1: Dimension for micro heater

The interconnect acts as interconnect between the micro fluid and the environment. The continuous flow PCR device can be made with PDMS or LTCC interconnect. Using PDMS as an

interconnect is an emerging technology in micro fluidic devices. It enables the ease to interconnect the devices and the environment at an inexpensive process in laboratories. But due to the pressure built at interconnect which occurs due to flow rate the application of PDMS interconnects is a bottle neck. However due to the high throughput and multi inlet/outlet PDMS offers promising wide range of application [6].

## 4. Use of COMSOLE multiphysics:

The proposed structure for the micro heater is very simple and can be designed with greater ease. It consists of a pair of concentric cylinders in which the inner cylinder acts as the micro channel in which the reaction sample which in this case is blood flows through. Pairs of concentric cylinders are connected together to form a closed path through torus structures. Combination of three pairs of cylinders and torus structure will make up the three sections where the three steps in PCR i.e. Denaturaton, Annealing and Extension will take place. In the paper the authors have used in COMSOl Multiphysics conjugate heat transfer physics for simulation of micro heaters this physics provides laminar flow along with the heat transfer in solids. And in this physics one can define the solid domain outer boundary to one which one is applying temperature and the fluid domain can be defined as blood sample or DNA sample. Firstly by the help of this software the structural design of the heater is established then two domains are created (outer cylinder and inner cylinder). The inner cylinder is used to define the fluidic domain (which in our study is blood sample and saliva) where as the outer cylinder is used to define the solid domain which is usually made up of polysilicon. In the COMSOL tool poly silicon is the built in material where as the blood and the saliva sample material are user defined materials with properties defined in table 2.

| Properties | Blood Sample | Saliva | Unit |
|---|---|---|---|
| Density | 1060 | 1000 | $kg/m^3$ |
| Viscosity | 4.53e-3 | 1.3e-3 | Pa*s |
| Heat capacity at constant pressure | 3900 | $2.3*10^6$ | J/kg*K |
| Ratio of specific heat | 10000 | 1 | - |
| Thermal conductivity | 0.643 | .58 | W/m*K |

Table 2: Properties of blood and saliva

## 5. Simulation and result:

Due to good thermal conductivity property of silicon, continuous flow PCR is normally made up of silicon. Even with good conductivity silicon shows a major drawback when used for DNA amplification. Silicon if used reduces the quantity of the sample hence causing serious problem. The disadvantage does not only limit to reducing the quantity but also the high conductivity has a negative effect in which the thermal isolation adds to high complexity to micro system. Also silicon is not transparent because of which it limits the application of optical detection. These disadvantages offered by silicon makes the architecture complex and also cause difficulties in fabrication. Therefore a material that is bio compatible and also transparent (for optical detection) has to be used. Soda lime glass which has ten times the thermal conductivity is employed as a substrate material. The property of soda lime glass that is employed is listed in table 3.

| Properties | Soda lime glass | Unit |
|---|---|---|
| Density | 2400 | $kg/m^3$ |
| Viscosity | - | Pa*s |
| Heat capacity at constant pressure | 49 | J/kg*K |
| Ratio of specific heat | 753 | - |
| Thermal conductivity | 1.1 | W/m*K |

Table 3: Properties of soda lime glass

Heat transfer in the PCR device is studied to determine the required thermal input for the selection or design of the heater, to determine the controller needed to maintained the steady state temperatures for the PCR cycle and to check the stability and time constant. The simulation is carried out at an input pressure of 470kPa. Saliva and blood samples are used for DNA amplification. Three different temperature zones are maintained. The simulation result of temperature distribution is obtained for both the samples. In order to obtain the amplification the principle follows three simple steps. Firstly the original sample that is taken has to be duplicated. In order to do so sample is heated to denaturize which breaks the sample into two segments (copy of the original segment).The second step is to obtain two new strands from the previously broken strand. Therefore polymerase enzyme is used to synthesize and to build two new strands which in turn is the copy of the original sample [7]. The result molecule contains one new and old strand. The final step is to make use of newly developed strand to create additional new molecule which in turn will contain one new and one old strand. The heating and cooling process (synthesize-build) is continued for almost 30-40 times and with fully automated devices with in the interval of few hours the process results in amplification of DNA sample. The obtained simulation result for saliva sample and blood sample is shown below in figure 3 and figure 4 respectively. Also it is noticed that the velocity of the fluid (sample) is highest at the centre of the micro channel and lesser at adjoining areas of the inner wall as shown in figure 5. When we take into consideration the saliva sample there is a prominent change in the temperature levels at Denaturation where it is observed that the temperature in this region falls slighter than its original temperature. This phenomenon is depicted in figure 3; here one can clearly see the overlap in the temperature at denaturation zone. Unlike saliva sample, blood

sample in the other hand has no much change in it's either of zones. Also since the temperature sensors is difficult to fabricate, and there is no proper temperature distribution in this micro heater. The obtained result of temperature flow for blood sample is shown in figure 4.

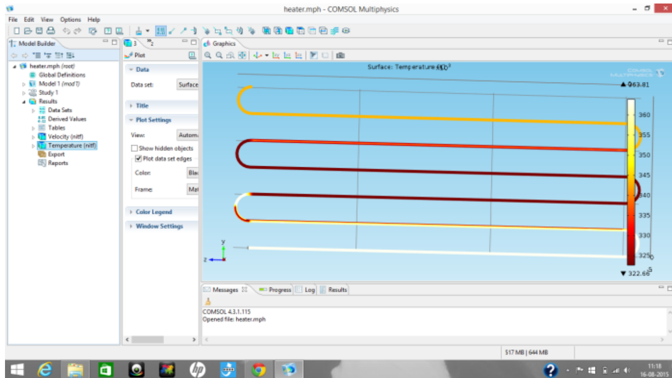

Figure 3: simulation result obtained for saliva sample

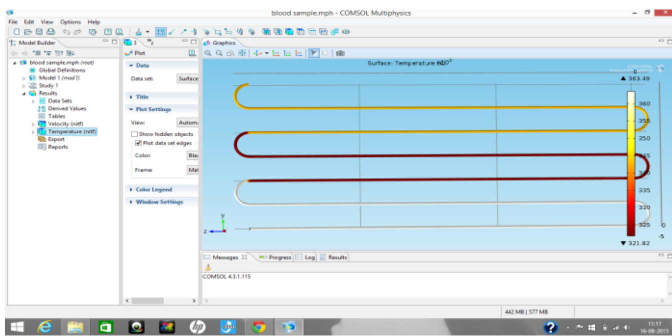

Figure 4: simulation result obtained for blood sample

The time duration for which the reaction sample is to be exposed to each temperature zone is adjusted by varying the velocity of the fluid in the micro channel. Here, the blood sample flow is through the laminar flow which is a continuous flow of fluid, since the specific temperature for each zone is applied when the fluid enters the inner wall of the micro heater, the velocity is less at the layer which adjoins the inner walls of the micro channel, as the fluid increases the velocity at the center of the micro channel is maximum as shown in the velocity scale in figure 5.

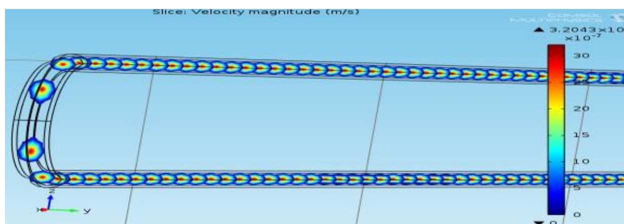


Figure 5: simulation result obtained for blood sample

## 6. Conclusion:

A novel technique to carry out DNA amplification by the process of Polymerase Chain Reaction (PCR) has been designed. Using this process a single or a few strands of sample DNA can be multiplied in orders of thousands and billions. Considerably good results are obtained with flowing blood sample in the micro channels of the designed MEMS based heaters. PDMS was used for the interconnect which is biocompatible and a simplified fabrication process. The system is reliable to with stand the pressure of 470kPa.The use of two heaters to obtain the three distinct temperatures eliminates the cross talk that would occur in previously design system. Temperature distribution in the chip is obtained by using FEM (finite element method). The so designed process can be employed for various applications from bio science to research.

## 7. Acknowledgement:

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