Capstone 2011: Microfluidic Lab-on-a-Chip for Delivering Targeted Alpha Therapy

Presented to the Materials Science and Engineering Department, University of Maryland by:

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**Table of Contents**

Title  
Table of Contents  
Abstract  
Motivation  
Materials Science and Engineering Aspects and Review of Prior Work  
Design Goals  
Technical Approach  
Ethics and Environmental Concerns  
Intellectual Merit  
Broader Impact  
Simulation Results and Discussion  
Conclusions  
Acknowledgements and References
Abstract:

The goal of this project is to design a microfluidic device that will treat tumor cells. Targeted Alpha Therapy (TAT) is used to deliver the radiation source as individual radioisotopes to the desired tumor cells in the body. The chelating agent, DTPA, will be mixed with a monoclonal antibody (mAb) in a solution of water. Actinium$^{225}$, which has a half-life of 10 days, is then transferred to the device using alpha recoil, resulting in implanted Bismuth$^{213}$. Basic modeling was done for the dissolution of the sucrose film that captures the Bismuth, and Fluent was used to model the mixing behavior of the fluid.

Motivation:

Our motivation for this project is to design a microfluidic device that will administer Targeted Alpha Therapy (TAT) quickly and efficiently to a patient. TAT itself is currently in clinical trials for a broad range of cancers, but is extremely expensive, with the cost of one dose ranging from $10,000-$40,000. Another important problem with TAT is the need to have a generator on-site because of the short half life of the radio-isotope, Bismuth$^{213}$ (45 min). A lab-on-a-chip device would significantly reduce the cost (to $7.77 per generator) and make the treatment available to a much wider range of hospitals and as a result, patients.

Materials Science Engineering Aspects and Important Prior Work:

As materials science and engineering students our main objective was to design a practical system with an emphasis on the interconnection between the structures of the materials we intended to use, the way we would process them based on our design, the properties they would have as a result of the processing and the way the materials performed based on their properties.

Our design required a sucrose film that had a thickness of 1μm and based on our discussion with Dr. Phaneuf and what we learned from kinetics ENMA 471, a polycrystalline film with a small crystallite size would be desirable because these properties will aid the dissolution rate of sucrose microprocessing. Based on our skills acquired from taking ENMA 465, a class on nanoscale and microscale processing of materials with an emphasis on thin film processing for advanced technologies, we now spin coated sucrose on a silicon wafer to make a film of 1μm thickness at the FabLab on campus.

Materials characterization: We now had to verify if the fabrication process yielded the properties of the sucrose film we desired, so based on what we learned from ENMA 310, a materials laboratory, we used x-ray diffraction to determine the crystal structure and
crystallite size of the sucrose film. We had also learned about the theoretical principles of x-ray diffraction from ENMA 460, *solid state physics*.

Our knowledge of *kinetics* and our discussion with Dr. Phaneuf enabled us to understand that the precipitation and then heterogeneous nucleation at the surface of the film would be governed by kinetics and *thermodynamics*, ENMA 461. The team’s knowledge of *solid state physics* also enabled us to understand that phonons are generated through collisions of Bi atoms into sucrose film by simulating ion implantation. The kinetic Monte Carlo simulation of nuclear decay and time step, the diffusivity of Bi into collection area and the diffusion and convection of Bismuth across channels are all materials science and engineering aspects based on *kinetics*.

Targeted Alpha Therapy involving the use of radioisotopes like Ac$^{225}$ and Bi$^{213}$ for biomedical applications and particularly cancer has been investigated extensively by the scientific community [20]. It is currently in clinical trials and has been proven to work. The application of Bi$^{213}$ as the radionuclide to be delivered as the dose to the patient and its generation from Ac$^{225}$ is also well documented in literature [22]. This therapy targets the tumor cell within the body. The antibodies are administered to the body which target and bind to the tumor cells and the radioisotope decays and becomes benign after releasing an alpha particle which kills parts of the tumor cell per dose. The separation of the alpha-emitting isotope is done through recoil-ion separation. As the decay proceeds, Th$^{229}$ produces an alpha particle and recoil Ra$^{225}$ ion. This occurs through conservation of momentum and energy. This energy is enough to dislodge the Ra$^{225}$ ion into a neighboring substrate. Then the decay continues on until becoming stable at Bi$^{213}$, which is benign to the body. [18] This treatment has a range of 70 microns. TAT has been tested against melanoma, leukemia, colorectal, breast, ovarian, prostate, and pancreatic cancers. The reaction kinetics of Bi$^{213}$ with the chelation agent, DPTA has also been well documented [23]. The kinetics of adsorption and desorption of Bismuth from aqueous solutions was also studied [25]. The kinetics, the rate of diffusion and the role of the Nernst layer in understanding the dissolution process of sucrose has also been of interest to researchers [2]. The mathematical calculation of the Diffusion coefficient in liquids such as water has also been done in the past [24]. The role of the crystallite size in the rate of sucrose dissolution and understanding the role of the phase change and microstructure of the sucrose film was also found in literature [26]. The difference between the current technology and our project is the size and containment of the dose. Using lab-on-a-chip to administer this therapy makes it more widely available, affordable and able to be mass produced.

**IV Design Goals:**

Our goals for this device are primarily limited by time, because we only have 3 half lives (2.25 hours) to make and deliver the radio-labeled antibody successfully to the patient. However, we still have several substantial goals for the device as a whole. Because it is a microfluidic device we must therefore be able to fit several on a standard wafer, no more than 3.5 square centimeters for the dissolution chamber and mixing channels. Additionally, in order
to maximize the overall device efficiency, and given we have a good amount of size to work with; we make 100% mixing our goal within the device. Also, since we plan on using an IV pump, the device must be able to handle a volume flow rate of 5-20 mL/hr. These goals are all comparable with previous devices [3, 4, 5, 8, 16]. However, the goals for the newest part of our design, the sucrose dissolution chamber, have been set based on our modeling. We want the sucrose layer to dissolve quickly enough so that the Bismuth can easily diffuse across the system and be mixed with plenty of time to react with the chelate-antibody conjugate (less than 2 or 3 minutes).

V Technical Approach:

Oak Ridge National Lab sells Actinium$^{225}$ for $1,450 per milliCurie, which greatly simplifies the process of purification before insertion into the device. We will only need to do one step of recoil separation: from the actinium thin-film generator to the sucrose collection layer. In order to prove that our system is feasible in terms of this material cost, we did a simulation to calculate the available quantity of Bismuth for dosages.

The first step in knowing how many Bismuth atoms have been implanted into the sugar is to know how many atoms have recoiled in a particular time interval with a particular starting amount of Ac. The simple solution to this problem is to start from a film of 100% pure Ac and project the quantities of each respective daughter as they continue down the chain. The resulting chart is a Poisson distribution, and since the Ac has the smallest decay rate, this decay becomes the limiting factor.

The kinetics of the recoil separation process are nontrivial and result in a certain loss between the generator and the sugar. However this simulation is concerned only with the 'ceiling' of available Bi as decided by the exponential nuclear decay. Thus far in the project, the numbers calculated here are most useful for feasibility estimates, such as the potential size of the sucrose film on the chip, or the estimated cost of the Ac per chip.

Since the isotopes are constantly in a state of flux from higher to lower order isotope (at least until Bi$^{209}$), we created a basic Kinetic Monte Carlo simulation to model individual atoms. The time that a certain atom exists as an isotope before decaying is defined using the logarithm of a random number divided by the constant decay rate, $\lambda$.

$$t = -\frac{\ln < r >}{\lambda_{\text{element}}}$$
By simulating 10 million atoms, we can see that the amount of Bi$^{213}$ existing in the mixture of elements approaches a steady-state at about 3 orders of magnitude less than the quantity of Actinium. This number allows us to calculate the maximum amount of Bi in the sugar film as a function of the starting amount of Ac in the generator.

Generator-Collector Interaction

The previous section details the theoretical maximum amount of active isotope that is available to our system, which we can hold constant due to the limits of the decay rates. While we can change the starting amount of Ac$^{225}$ in our generator, the available Bi$^{213}$ will always be a fraction of that quantity.

In order to now understand the loss of the alpha-recoil method, we simulate the interaction with the energetic Bi$^{213}$ ions as they travel through the three regimes of the recoil step on the chip: the generator which holds the Ac$^{225}$, the air that separates the two films, and the sucrose layer that collects the ions. The energy of the recoil particles is a constant due to the nature of the alpha decay- the decay releases energy from 5.6 to 5.8 MeV, and due to conservation of momentum the recoil particle must have an energy from 101 to 105 keV.
Using the package Stopping and Range of Ions in Matter (SRIM), we can find the penetration depth value of Bi ions at 100 keV travelling through each of our three regimes. We can use the depth values to calculate an energy loss per distance travelled, dE/dx, or stopping power. In our PMMA generator film, the stopping power was found to be 1380 keV/µm; in air, 1.3 keV/µm, and in sucrose, 1250 keV/µm.

Using Matlab, we simulated the dimensions of the two films as they would be while the sucrose layer was ‘charging’ on the generator, as seen in figure 2a.

![Image of Collector-Generator Arrangement and Monte Carlo Simulation of Ion Densities](image)

**Figure 2:** (a): Schematic of Collector-Generator Arrangement showing recoil and the face that is displayed in the Monte Carlo Simulation (b) Monte Carlo Simulation showing relative densities for two distinct generator thicknesses. Simulation was based on stopping powers found in literature.

Assuming that the particles decay in random directions, we know that more than half of the particles will fire in the wrong direction. Either the particles go in the direction opposite to the collection plane or they are close to the edge of the generator and fire outwards. Since the aspect ratio of the plane area versus the distance is very large, close to 200, we can classify
these cases as somewhat trivial. This immediately lowers our collection efficiency to roughly half of the particles that decay on the generator. Furthermore, we know that a certain percent of ions will decay with a direction very close to parallel to the plates, and some will never have enough energy to traverse the distance from the bottom of the generator to the top. These cases also constitute losses. The way to minimize these losses was simulated by reducing the film thickness of the PMMA generator. By changing this depth, particles have a much higher probability of leaving the film. With a thickness of 50 nm, approximately 20.3% of the ions remain stuck in the generator. With a 10 nm film, this number is reduced to 4%. Similarly we can reduce the stopping power $dE/dx$ or density of the generator film to maximize our ion flux between films. The relative density of ions in the three ranges are given in figure 2.

For the 10 nm generator film, still 41.4% of the ions lose their energy while in the space (air) between the films. However since they are charged particles and the sucrose film is biased to -200V, we can assume that the majority of the ions will move across the gap with the electric field.

Behavior of Bi Isotope within Channels

After the sucrose film has been dissolved, the fluid begins down the channel path in the sucrose. The goal of the channels is to mix the bismuth with the chelated antibody as thoroughly as possible. Originally we wanted convective mixing, but this is impossible on the length and flow rate scales desired on such a small chip. The flow conditions inside the channel are entirely laminar, where viscous forces dominate over inertial. Since the fluid is moving slowly, we cannot assume convective mixing of the bismuth. Instead, we used the Hayduk-Laudie relation to approximate the diffusivity of the Bismuth ions within the water. With this we can get an approximate mass transfer.

\[
D_{12} = \frac{13.26 \times 10^{-5}}{\mu_2^{1.14} \nu_1^{0.589}}
\]

Hayduk-Laudie equation

The diffusivity allows us to solve Fick’s 2nd law and create a concentration curve as a function of time and distance from the bottom of the channel. Since the source of bismuth is finite, the solution to Fick’s law is a gaussian function and approaches a uniform concentration with time. Our diffusivity was high, so after only three minutes we can assume a nearly uniform gradient.

Fick’s and Gaussian EQs

\[
\frac{\delta \phi}{\delta t} = D \frac{\delta^2 \phi}{\delta x^2} \quad C(x, t) = \frac{c_o}{\sqrt{4\pi D t}} \exp \left( -\frac{x^2}{4Dt} \right)
\]
VI Ethics and Environmental Concerns:

The societal impact represented by successful implementation of this system for cancer treatment is obvious. Targeted alpha therapy has been shown to be effective in treating cancer but because of the need for a radio isotope source in house it has proven very expensive and the facilities which can provide such treatment are greatly limited. Our system would centralize the production of radioactive species and the necessary materials to administer targeted alpha therapy into one compact micro-fluidic system. Despite these major societal benefits the fact that our system is disposable presents ethical concerns in the form of waste generation. Materials that were implemented into the design were evaluated to ensure that they are not harmful to the environment and will not cause any long term disposal issues. Polydimethylsiloxane (PDMS) is used in the channel design. PDMS does not pose any environmental threats when disposed. PDMS has shown to degrade to lower molecular weight compounds when in contact with soil. It will degrade to Me2Si(OH)2 after only a few weeks [21]. We used Silicon based wafers for the fabrication of the chip. Silicon is not known to cause
any adverse effects on the environment or to the body because it is an inert material. Water and sucrose are not harmful. Small amounts or UV epoxy containing urethane acrylate were used in the design of the chip. Urethane acrylate if released into any aquatic environment can cause long term effects in aquatic organisms. The Materials Safety Data Sheet for urethane acrylate states that it is important to avoid disposal into drains, soil or surface water. Disposal of this agent is subject of regulations and requires chemical disposal that ensures regulatory compliance. SEC gel is agar based, and so biodegrades readily. The Bi²¹³ isotope degrades readily to a non-radioactive species Bi²⁰⁹ which is non-toxic, non bio-accumulative, and is the least toxic of all heavy metals. The concentration of it is so small that it will not pose any significant dangers. The isotope generator is provided by Oakridge National Lab (ORL) and is embedded in PMMA. Appropriate precautions should be taken when the generator has to be disposed of because of remaining radioactive species and their heavy metal decay products that could remain as remnants in the generator. The biological effects were considered thought the design project to ensure that all materials that were going to be injected into the body are compatible. This is very important because patients could have side effects or possible death if the materials were not deemed compatible. Overall the chip design and process proves not to induce any major environmental effects or safety problems.

**VII Intellectual Merit:**

Prior to this work, the use of alpha radiation for cancer treatments has been limited to only a handful of hospitals which can produce these short lived sources on-site. The main intellectual merit of this project has been the consideration of materials science, nuclear physics, and fluid dynamics to streamline the process into a microfluidic system.

The recoil separation process, with which we designed our isotope separation group, has very little previous research. We investigated the possibility of using it on a lab-on-chip and decided it definitely has the possibility of competing with resin- or strong acid-based systems. It still has major detractions: half of the ions recoil away from the collector film, and the lack of convective fluid movement inside the channels makes mixing very difficult. Recoil separation is an unresolved topic; mostly because the alpha emitters that are required to test such a system are rare and expensive. Our treatment of the subject makes a strong argument to delegate a certain amount of Ac²²⁵ to cancer research to put our system to trial. Since our cost analysis puts dosages on the order of $10, there is a great monetary benefit to exploring our design.

One basic insight that was gained during the process was the ability to coat a thin film of sucrose on a silicon wafer. From what we saw, spin coating sucrose had not been done before.
While it was a relatively simple process, we found that it was quite easy to make thin and uniform coatings of sucrose down to one micron. This ability can be used in recoil separation applications and possibly other applications where a biocompatible sacrificial layer is needed.

Our system benefits medicine and science because it expands the base of knowledge for lab-on-chip devices in general. We have furthered the understanding of how microfluidic properties can permit the delivery of drugs. Our concept allows for scientists and doctors to rethink the types of treatments that are possible. It reduces the need to worry about time sensitivity and provides an effective model for how such treatments can be made on the industrial scale.

**VIII Broader Impact:**

Should this design prove successful, it will have significant broader impact. It goes without saying that a great amount of research towards cancer treatment is required. The potential for enormous reductions in cost and increases in availability suggested by this design could provide a new standard for delivery of complex medical treatments, especially for cancer therapy. Currently the most effective cancer treatments are very expensive, and those that are highly effective and low risk even more so. This design could herald in a new trend of making those top end treatments cheaper through miniaturization.

**IX Simulation Results and Discussion:**

The solid consists of a square film of surface of area $x^2$ and thickness $f$, so $S=x^2$

If the solid mass is written in terms of the density, $\rho$, then $m=\rho x^2 f$

From this, $S = \frac{m}{\rho f}$, and the rate can be modeled as

$$-\frac{dm}{dt} = \frac{DC_s}{\rho h} \left(\frac{1}{f}\right) m$$

From Antonel

$\rho=1.4 \text{ g-cm}^{-3}$

$C_s=0.67 \text{ g-cm}^{-3}$
From Fluent Simulations, one can determine the Nernst Layer based on the maximum velocity. It was assumed that \( \frac{v_{\text{max}}}{10} \) would provide the minimum velocity required for transport limiting kinetics. Prior to the Nernst Layer due to the no flow boundary condition, the surface reaction is rate limiting and dissolution takes the form of an error function. By estimating this as a linear concentration gradient, one can easily obtain the distance of this boundary. After the Nernst Layer, the concentration of the solid in the bulk solution remains constant in a well stirred solution. The following figure displays a schematic of the Nernst layer.

By comparing one-tenth of the max velocity (from velocity profiles obtained with Fluent simulations) with the diffusivity of sucrose in water, one can obtain a characteristic length, \( h \), that we have defined as the Nernst Boundary Layer as described above.

\[
\frac{D}{h} = \frac{v_{\text{max}}}{10} = \frac{4 \times 10^{-4} \text{m/s}}{10} = 4 \times 10^{-3} \text{cm/s} \Rightarrow h = \frac{6.1 \times 10^{-6} \text{cm}^2/\text{s}}{4 \times 10^{-3} \text{cm/s}} = 1.5 \times 10^{-3} \text{cm}
\]

This results in a sucrose dissolution rate of 0.27 \( \mu \text{g}\cdot\text{s}^{-1} \) assuming entire surface coverage in a well stirred solution. With 14 ng of sucrose (1cmx1cmx1\( \mu \text{m} \) film), the dissolution time should take 52 ms.

Crystallite size also has an effect on dissolution rate. Using the Avrami equation, one can simulate the effect of grain size on the dissolution time. By defining dissolution as the opposite mechanism of crystallization, the fraction of solid present at a specific time and temperature during crystallization is given by:

\[
f_s(t, T) = 1 - \exp\left(-\frac{T}{\frac{5}{3}Nv^3t^4}\right)
\]
where N is the nucleation rate and v is the crystal growth rate. This equation was derived based on the assumption that nuclei are nucleated throughout the transformation at a constant rate, N and are expanding spheres, also at a constant rate, v. At small time, t, this equation tends toward 0 and at large time, f_s tends toward 1 as expected. In order to model dissolution with this equation, we must take 1-f_s. The equation can be written in the form
\[(1 - f_s) = 1 - (1 - \exp(-kt^4))\]

where k is \(\frac{\pi}{3}Nv^3\). To simplify the model, v is assumed constant during an isothermal transformation. In this case, k is directly proportional to the nucleation rate, N, which is inversely proportional to grain size (more sites for nucleation results in smaller and more numerous grains). By plotting the dissolution equation from the Avrami equation for different values of k, we can make a qualitative comparison of how grain size effects dissolution time. The following plot shows curves for varying orders of magnitude of k. It is apparent that as k increases (N increases) the dissolution time decreases. Since N is increasing, grain size must be

![Avrami Equation: 1-1+exp(-k t^4)](image)

Figure 5: Qualitative results of the dissolution model as a function of nucleation rate and growth rate applied to an Avrami equation. Increasing values of k simulate increasing nucleation rate for a constant growth rate. Increasing nucleation rate leads to smaller grain size. From the figure it is apparent that as grain size decreases, so does dissolution time.
smaller. Therefore, smaller grain sizes will result in faster dissolution times.

**Fluid Dynamics Simulations and Calculations**

The simulations team has contributed several essential portions of the project. These include understanding fluid dynamics, verifying the Fluent and Gambit simulation software, modeling the radioactive isotope decay, modeling the isotope concentration profile in the sucrose film, and designing and modeling the fluid flow in the chip. Each of these contributions will be summarized.

We have decided to use Fluent© to model fluid flow and mixing in our microfluidics chip, and Gambit© for geometry and mesh generation. The decision to use these particular pieces of software was based entirely on availability. We contacted several experts (see professors visited), and the takeaway points were that COMSOL© would be overall more useful, and more flexible. COMSOL© Multiphysics Classkit is currently available on the computers in the materials engineering computer lab. However, after contacting the COMSOL© licensing department, it was determined that the license had already expired and the software was being used abusively. The cost for a student to obtain a single license for COMSOL Multiphysics is $1,595. Additionally, the add-on fluid dynamics package is $1,595 per license. The price of this software greatly exceeded our budget, so we decided that Fluent© and Gambit© would be acceptable alternatives because they are available.

The first step for the simulations committee was to familiarize ourselves with the general principles of fluid dynamics. To do this, we studied the Navier-Stokes equations, and derived an exact solution for 2D Poiseuille flow. 2D Poiseuille flow assumes that we have hard wall boundaries in the up and downy directions, and infinitely spaced walls in the z direction. The flow is in the positive x direction by the coordinate system in Figure 11. The solution to the Navier –Stokes equation presented below also assumes a no-slip boundary condition, which means that there is a no-flow region directly next to the walls of the channel. Also assumed for the analytic solution is that we have completely laminar flow. Due to the dimensions of the channel (1mm height and 1m length) and the average velocity of the fluid, we can be sure that we are in the laminar regime. For a non-circular duct, the Reynolds number can be approximated by (source “fluid flow resistance” blackboard document) Re=(ρ*v*D)/µ where ρ is the density of the fluid, v is the average velocity, D is the characteristic Diameter, and µ is the fluid absolute viscosity. For our duct, the characteristic diameter can be approximated by D=2*h=2mm. For water, we have µ=.001003 (kg*m^−1*s^−1), ρ 998.2 (kg*m^−3) and set the average velocity to .001 m/s. Therefore we expect a Reynolds number of approximately 2, which puts us well within the laminar flow regime.
We have decided to use 2d flow to model our channels due to time and computer power limitations. Based on the Reynolds number, we expect that diffusional mixing will be the major contributing factor to the mixing of our reagents, and we will therefore need to have long channels so that our fluids will be in contact for a long time. Long channels necessitate lots of nodes, and it is easy to see how a 3d mesh would quickly strain our computers. The 2d models will therefore provide us with a fundamental understanding of the mixing and flow parameters, but specifics will still need to be measured and tested in the lab.

The flow between the upper and lower plates can be modeled by the following apparatus in figure 11. The channels are assumed to be horizontal with the flow in the x-direction and the width between the channels in the channel in the y-direction.

![Figure 6: Poiseuille flow geometry](image)

\[ v_x = v_{x \text{ max}} \left[ 1 - \left( \frac{y}{y_0} \right)^2 \right] \]  

(5)

**Verification**

An exact solution for 2D Poiseuille flow was obtained from the Navier-Stokes (NS) Equation. The derivation from the NS solution assumed a no slip boundary condition which was likewise modeled in the Fluent simulation. The velocity profiles are plotted both analytically and numerically. For low Reynolds numbers, there is strong agreement between the results, however, as the Reynolds number increases, slight discrepancies occur which is thought to be caused by turbulent effects. An extension of this derivation to a 3D case along with corresponding Fluent simulations will be used to verify the final chip geometry with appropriate channel cross section. The channel parameters used in the verification include a channel width of 0.001 m, a channel length of 0.1 m, fluid properties corresponding to water ($\mu=0.001003$ kgm\(^{-1}\)s\(^{-1}\) and $\rho=998.2$ kgm\(^{-3}\)). From the verification, the 2D mixing simulation implementing a switchback channel geometry can be confidently modeled to maintain a well mixed solution for
8-10 minutes for appropriate chelation. The pressure drop across the channel is expected to be linear according to Poiseulle’s equation which is what resulted from the Fluent simulations. Figures 12 and 13 display the plotted analytical calculations and numerical simulations.

Figure 7: Velocity profiles for the indicated channel for inlet pressure differences of 8.016 Pa, 80.16 Pa, and 160.32 Pa.
Figure 8: Pressure profile across the length of the channel. This profile shows the pressure change for the 8.016 Pa inlet pressure difference.

Upon having simulated and verified Poiseulle flow of a single fluid in a rectangular channel, analytically with Matlab and numerically with Fluent, it is necessary to model the mixing of two separate fluids. Multiphase considerations within the fluids (suspensions/solutions) may also be required for accurate modeling.

Under the assumption that we are able to achieve turbulent mixing, the channels may be fabricated on a larger scale and a diffusional mixing model will not be necessary. In the case where turbulent mixing cannot be achieved, channels can be fabricated sufficiently narrow, and of sufficient length to allow for optimal diffusional mixing. Previous work has shown that a zig-zag patterned channel causes the greatest degree of mixing most efficiently [Jeon, W. 2009]. Figure 14 from Jeon [Jeon] show various geometrical configurations that cause mixing. Also shown are contour plots displaying volume fraction of phase in fluid (gold nanoparticles in this case). From the contours, one can quickly observe the degree of mixing resulting from the various channel geometries. This can be further described quantitatively for a more concrete mixing capability analysis.
Figure 9: Mixing contours (displaying volume fraction of gold nanoparticles) showing effects of differing geometries (as shown in Figure 10) at specified time intervals. By inspection it is clear that the zig-zag type geometry is most efficient [Jeon].

To establish a quantitative means to describe the degree of mixing, Fluent is capable of producing contour plots of volume fraction and molar concentration of components, which, along with a discrete scale, can be used to measure the degree of mixing based on a certain channel geometry. Once a value can be assigned to the degree of mixing, this can be compared to the size of the channel to determine a mixing efficiency. Channel geometry can be altered in order to optimize mixing efficiency. We define mixing efficiency as having a homogeneous concentration across the channel, which is discussed below in the section “Chip Design.”

Figures to follow display the results from the Fluent mixing simulation. Velocity vector and contour plots clearly show laminar flow with the no-slip boundary condition. The channel width was assumed to be one millimeter and the fluid, liquid water. One sample of water was designated at inlet 1 and another sample of water was introduced at inlet 2. The mass fraction of water corresponds to the first sample (i.e. 100% water (inlet 1) at inlet 1 corresponds to 0% water (inlet 2) at this inlet.

Chip Design

The results from the isotope decay simulations indicate that we will only have a usable dose in the chip for about 45 minutes once it is removed from the actinium source. The actinium source must be brought to every hospital that plans on treating patients using the chip, and this means that the physical chip assembly must be done in hospital to ensure that the dose is delivered in a timely manner. The antibodies and chelating agents will be mixed and reacted in a very small container (on the order of 1mL in volume) off the chip to save time, and
will be subsequently pumped into the chip via IV pumps. The mixed solution will then dissolve the sucrose containing the isotopes, and will then be mixed and reacted in switchback mixing channels. The design of the channels was influenced by the fact that according to a source (McDevitt et al) the chelating agent must be in a well-mixed environment with the isotopes for 8-10 minutes to achieve 80% reaction efficiency. Also, the flow constraints of certain IV pumps were taken into consideration; for example, the Abbot Labs Plum XLD Pump IV Infusion has a minimum volumetric flow output of 1ml/hour. In total we expect the chip to be 3.5 cm x 3.5 cm, with most of the chip area corresponding to the mixing domain, but with one square centimeter in the “upper left” corresponding to the sucrose film dissolution chamber. A schematic of an approximate flow velocity and mixing are shown below in figures 15, 16 and 17. The channel width was assumed to be one millimeter wide. For flow rate purposes, the channels are assumed to be 1mm. One sample of water was designated at inlet 1 and another sample of water was introduced at inlet 2. Both of these are connected together in the beginning of the mixer. The mass fraction of water corresponds to the first sample (i.e. 100% water (inlet 1) at inlet 1 corresponds to 0% water (inlet 2) at this inlet. This allows for the mixing to be displayed as a function of mass fraction.
Figure 10: 2D simulation of pure water through the mixing chamber at volumetric flow rate of 10ml/hour. This figure shows the example flow velocity profile.
Figure 1: 2D simulations of pure water through the mixing chamber at volumetric flow rate of 2ml/hour. This figure shows the blown up velocity profile.
Figure 12: 2D simulations of pure water through the mixing chamber at volumetric flow rate of 10ml/hour. This figure shows the Example mixing

The simulations shown in Figure 15, 16 and 17 were done with pure water, which is not the fluid we will actually use. The actual fluid will close to the viscosity of water because the reactant concentrations are on the order of nanograms per milliliter. Future simulations will include the solution with the appropriate viscosity. For all of the simulations, a mass flow inlet was used because that is the capability of Fluent. The volumetric flow was converted to the mass flow rate by using the density of water, which is 998.2071 kg/m^3.

Fixing the inlet flow rate to 10ml/hour yields a maximum center-line velocity of 5.25 mm/s, and with a travel length of 534.5 mm, we expect a minimum reaction time of about 2min. This falls short of the 8-10 minutes required for mixing. However, we have decreased the flow rate to 2ml/hour (which is within the capability of standard IV pumps). Figure 16 shows that (using a finer mesh) that the maximum velocity changes to 1.13 mm/s, which puts the minimum reaction time near ideal to at 7.88 min, and the average directly in the ideal range. More expensive IV pumps can get finer flow control, and could be implemented to better “center” the flow velocity into the ideal range.
Figure 18 shown below is a schematic of the sucrose dissolution chamber. This chamber was created in order to get sufficient surface coverage and uptake of sucrose and bismuth. The large open area in the middle is for the inlet hole that will be punched in the glass.

Figure 18: Schematic of the sucrose dissolution chamber.

The main “square” of the chamber is 1cm x 1cm in area, and 1mm in height. The channels are all .5mm x 1mm in cross section. The channels were designed so that one quarter of the flow would be directed into each channel. Due to time constraints, we were not able to complete 3D fluent simulations for this design. However, we assumed that the flow would be one quarter of the total flow (2ml/hour), and we ran a Matlab simulation assuming laminar flow to display the velocity cross-section. Figure 19 is the velocity contour of this simulation.
The white border around the colored profile represents the Nernst layer which will be discussed in the next section on sucrose dissolution. The Nernst layer represents section of the velocity profile that fall below 10% of the maximum velocity. The Matlab code was written based on an exercise by Martin Pederson (Martin Pedersen, Technical University of Denmark). Flow analysis indicates that for the channel region, we expect a Reynolds number of about ½. This puts us well within the laminar flow regime, which is exactly what we see in the simulation. It is a bit distorted from a traditional laminar profile because the cross-section is a rectangle, not a square or circle. This simulation indicates that the maximum velocity is about .4 mm/sec in these channels.

Although the Fluent simulations did not directly represent materials design aspects, they were integral to the overall design of the chip. The flow profiles allowed for yield calculations, and mixing estimations could be used to determine reaction efficiency. Gambit served both as mesh generation software for Fluent, and for CAD applications showing the schematic of the sucrose mixing chamber.

**Facilities, Materials & Prototyping**

The design and fabrication of our targeted alpha therapy lab-on-chip device will utilize numerous software and laboratory resources to ensure desired operation. In designing our device the simulations committee has performed work using the ANSYS software package Fluent. The goal of these simulations has been twofold, first to confirm the applicability of the
software for our microfluidic design and second to model our system in such a way as to ensure that proper design parameters are met. The parameters of interest include flow rate, mixing and general dimensional requirements. To optimize these parameters factors such as system topology, channel diameter and length, etc are considered in our computer simulations. From these simulations the final device design will be determined and fabrication will follow.

Our device will be fabricated using lithographic techniques and it therefore becomes necessary that a lithographic mask be designed and procured so that production can begin. We have designed and sent out the mask to FineLine Prototyping for fabrication and are currently awaiting its arrival for device production to begin. Especially important in this early work will perfecting the patterning of zigzagged channels into the Si substrate as the mixing which occurs in our device will rely almost entirely on these patterns. We must be sure that we are able to create the desired 1mm channel size on Si and that they will be free of obstructions for proper flow. Our mask was designed using the CorelDRAW X4 software package.

Our microfluidic system will be fabricated on a silicon chip. There are four major fabrication aspects which require consideration: chip surface features, the sugar/Bi film, Size Exclusion Chromatography (SEC) chamber and glass top. A mask of the features is shown in Figure 20. The various surface features required for the proper functioning of the system will be molded using the polymer PDMS. This polymer will be patterned by using a Si wafer with the chip design etched into the wafer’s native silicon dioxide layer. This process was explained to us by graduate student Mariana Meyer and from her guidance we have learned that all of the patterning required can be performed relatively simply. In order to complete patterning we first surround the patterned wafer with an aluminum foil ‘boat’ and then pour the PDMS over the wafer surface. Once cured the patterned PDMS can be peeled from the Si mold and inspected for imperfections. This process represents the lion’s share of fabrication time and effort due to its inherently complicated design and need for precision when compared with the other construction aspects. The chip(s) used for fabrication will be supplied through the FABLAB which maintains a large stock of blank Si wafers.
The second major consideration lies in the sugar/Bi film which will contain the active bismuth for alpha therapy. Once the dissolution behavior of this film is determined it should be relatively simple to spin coat a sugar film which can be implanted with Bi atoms for introduction to our chip. We have determined that we can easily pattern such a film with the desired thickness (~1 micron range) by spin coating. Thus far we have completed trials using 33 and 50 wt % sucrose in water solutions and both have viscosity properties which have resulted in films in the micron range according to measurements made using the n&k analyzer device in the FABLAB. The sucrose films were spin coated at 4000rpm for 40sec., based on a known process for a similarly-viscous 1812 photo resist. In these trials we were also able to determine that by using plastic backed vacuum tape we could simply cover the parts of the chip that we do not wish to have sucrose on, spin on the film and remove the tape. Using this technique we are able to quickly and easily make any sized sucrose film on the Si wafer. You can see a finished sucrose film in Figure 21.
We also found that after a few days the films began showing regions of crystallite nucleation. To confirm or deny this observation we characterized the sucrose films with XRD. We took measurements of the regions of the film that were still as-spun and of the regions that appeared to be crystallizing. The as-spun region showed no significant peaks (excluding the large peaks from the Si substrate), suggesting that the as-spun sucrose film is amorphous. The crystallized regions showed no conclusive results from a regular XRD, so we took a wider angle range scan of that region in a powder diffractometer/XRD. The diffractometer would be able to detect crystalline peaks for a poly-crystalline sample, where the normal XRD would average over all of the crystallites and show inconclusive results as we saw. The powder diffractometry results are shown in Figure 22. The crystallized region had many peaks in common with the sucrose reference in the XRD databanks, suggesting that the crystallized region was in fact poly-crystalline. The strong presence of peaks related to the (1 0 0) family of sucrose, which aligns at (4 0 0) with Si (0 0 2) suggests that there may be some epitaxial effects on the nucleation from the Si substrate. Further testing along this idea would have to be done to conclusively conclude one way or another.
**Figure 17:** XRD Results. The bottom scan is the as-spun region, and the top scan is the crystallized region.

The SEC chamber is another area requiring major consideration in the fabrication of our device. First of all the dimensions of this chamber must be dialed in so that the desired separation can occur by the SEC gel used. This chamber will be placed off of the chip in order to ensure that the proper dimensions can be achieved. Otherwise it would be difficult if not impossible to have both dimensions and volume for the separation column. Secondly the proper gel must be selected for both exclusion size and coarseness again for resolution and also for flow rate through the device. We have chosen to use Bio-Gel A-0.5m SEC gel from Bio-Rad which is a agarose based material commonly used for the separation of antibodies and other proteins. For desired resolution we need 5-10:1 length-to-diameter and 4-10:1 bed-to-sample volume ratio column. Based on the information from our simulations we have determined that we need a column which is approximately 5x1x1 mm which is correct for both necessary ratios.
Crystallite size was estimated using optical microscopy. Figure 23 shows two pictures taken with the optical microscope in the teaching lab in the FabLab. The smallest nucleating crystallites were needle-like structures approx. 5um in width. Larger crystallite structures were up to 800um, with groups of radially-oriented needle-like structures forming circles multiple millimeters in diameter. The smallest crystallite size is the important size with respect to dissolution, since it would be on that scale that the sucrose would be dissolving into the water solution.

Figure 18: Optical Microscope pictures. 5x mag (left) shows some nucleating needle-like structures. 20x mag (right) shows fully-grown, larger, more grain-like structures.

The final major consideration in fabricating our device is in fusing the top PDMS to the bottom Si wafer. This process should be relatively simple, but it is necessary to be very careful and precise to ensure that the final device does not leak internally or externally. To bond the PDMS and glass we have chosen to use a UV curing epoxy UV 30-27 series from Loxeal. This choice was made for 3 main reasons: fast curing time (~5 min or less), high bond strength (20-30 N/mm^2) and medical use certification (ISO10993). Speed is important because once the sucrose film is activated with bismuth the clock begins counting down until the required dosage of radiation is no longer present. High bond strength is of obvious importance as the device will be subject to pressures as fluids flow through and mix within the system. This strength guarantees that the device will neither leak nor, even worse, fail totally. Finally the medical use certification ensures that we are not introducing potentially harmful components to the system by overlooking the composition of the sealant used for the device. The prototype may be sealed with partially-cured PDMS instead of the epoxy for ease and availability (not having to purchase and wait for specialty epoxy).
For testing we have discussed using streptavidin and biotin as model antibody and chelate with Dr Phaneuf. These specific molecules will be useful for testing because of their fluorescence properties once chelation has occurred. A successful trial will result in high fluorescence in the outflow stream from our chip indicating that the biotin/streptavidin chelation has occurred.

To test the sucrose dissolution rate against our models, we would have to run an experiment to measure the mass dissolved per unit time. One idea was to put the sucrose film in contact with mixed water for a fixed amount of time. After each time interval, the film would be taken out, dried, and measured for thickness. Knowing the surface area (1x1cm square) and density, we would plot dm(dt) and compare with the results of the theory. One problem with this method is introducing water to the film in a controlled manner. A microfluidic channel system would accomplish this, but requires extra work ahead of time creating one. A second problem is quickly and evenly drying the film without affecting the film itself. After being submerged in water, the film will be partially hydrated. Drying the film fast enough to effectively cease dissolution would be difficult, especially so since the film cannot be taken past 90C without degrading the sucrose.

Over the course of the semester the prototype goals have changed drastically. At the midterm point we decided to focus our energies on the modeling and prototyping of the sucrose film while putting the production of an actual microfluidic channel system as a secondary objective. We made sure at every step of the way that the prototyping aspect of our project did not sacrifice time or manpower from the primary focus of the capstone project: the design. The aspects of the prototype that we finally decided upon were those that we believed would help verify and support the design portions of the project.

**X Conclusions:**

Our design met with great success when our results are compared with our design goals. We met them all, and are able to provide a dose of radio-isotope at minimal cost and potentially, wide availability. Our simulations resulted in rapid mixing (100% mixing within 2 mm of channel length), the sucrose dissolution confirmed a rapid dissolution rate, and the Bismuth diffusion confirmed a significant spread across the channel given the assumptions made for our model.
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References:


