The delivery and efficacy of chemotherapy is integral to curing cancer, and new drug transport systems are being developed to decrease harmful side effects that interrupt dosages. Minimally invasive approaches are focused on the mechanisms of drug-dispensing nanoparticles, which are continuously dispersed into the patient and constantly deliver anticancer medication. Mathematical modeling can aid in predicting how cells react to chemotherapy. With multiple variables such as uptake, proliferation, death, and interactions between cell receptors and ligands, outlining the relationships between the inner and outer cell environments would prove valuable. This study provides an agent-based mathematical model describing cell behaviors undergoing chemotherapy via nanoparticle delivery and attempts to map biophysical changes throughout treatment.
Creating the model based on data and experimental designs used in cancer research included following current literature on the latest nanoparticle studies. Beginning with the premise of the Cell Adhesion-Migration method of micro-pipetting cells onto a substrate of nanoparticles in order to enhance their uptake\textsuperscript{13}, the model focused on the reactions of MCF-7 breast cancer cells to both raw (uncoated) and cytotoxic-coated nanoparticles. The particles acted as a delivery system by which drugs were able to enter the cell via active diffusion.

Each portion of the mathematical system involved equations depicting cells’ reactions to being exposed to ligands and included the probability that a cell would bind with a nanoparticle\textsuperscript{3} it encountered. Creating binding, proliferation, and death rates on the individual cellular level allowed the authors to predict cells’ behavior when bound to a nanoparticle. In addition to mathematical models, a computer model in NetLogo was created to mimic the relationships between receptors and ligands, as each step of a MCF-7 cancer cells experience under nanoparticle chemotherapy was depicted.

The first section depicts how a nanoparticle binds with a cell, including reaction kinetics and the Hill equation\textsuperscript{15}, and establishes a binding rate per cell. Section II shows how cells unaffected by chemotherapeutic ligands proliferate, as not every cell in a tumor is killed by the agent coating the nanoparticles. Section III presents the rate of necrosis once the cell has metabolized a toxic drug, relying on the growth of a necrotic core and changing nutrient uptake. Finally, Section IV elaborates upon the model constructed in NetLogo while Sections V-VI compare the model to the data provided in Pitchaimani et al. (2017) and provide an assessment of the program’s predictive accuracy.

I. REACTION KINETICS AND BINDING RATE

To better understand the kinetics of cellular uptake, forward and reverse reaction rates had to be developed, as they are composed of functions which also contribute to a cell’s binding probability. Interactions between receptors and ligands are treated as first-order reactions, similar to $A + B \xrightleftharpoons{k_r}{k_f} AB$, and are explained using diffusion equations, as convection is rarely found in cell binding behaviors. Reviewing the results presented by Shoup and Szabo (1982) and Lauffenburger et al. (1993), a model for a cell’s binding probability was created. According to the literature, to clearly express $k_f$ and $k_r$, the rate at which ligands and solution receptors bind must be calculated. Using the spherical coordinate system where a receptor is placed at the origin, the diffusion equation is:
\[ L(r) = \frac{D d}{r^2} \left( r^2 \frac{dL}{dr} \right) = 0. \]  
\( \text{(1)} \)

in which \( D \) is the sum of the ligand diffusivities. The above equation measures the concentration of ligands around a receptor. To obtain a rate at which the ligands are bound by the receptor, another condition must be added:

\[ k_{on} L(s) = 4\pi s^2 D \frac{dL}{dr} \bigg|_{r=s} \]
\( \text{(2)} \)

integrating with respect to \( dr \) yields:

\[ L(r) = \frac{-k_{on}sL_0}{4\pi Ds + k_{on}} \left( \frac{1}{r} \right) + L_0 \]
\( \text{(3)} \)

with the boundary conditions of \( r \to 0 \) and \( L \to L_0 \), or as the radius of the spherical cell grows, the concentration of ligands approaches its bulk concentration. The flux of the molecule, or how many ligands are traveling around a certain receptor, is the surface area of the receptor multiplied by the change in the diffusion rate:

\[ 4\pi s^2 D \frac{dL}{dr} \bigg|_{r=s} = k_f L_0 \]
\( \text{(4)} \)

Equating the overall flux \( (k_m) \) and the rate of ligand molecules binding to a receptor \( (k_{on} L(s)) \):

\[ k_f L_0 = k_{on} L(s) \]
\( \text{(5)} \)

\[ k_f = k_{on} L(s)L_0^{-1} = L_0^{-1}4\pi s^2 D \frac{dL}{dr} \bigg|_{r=s} \]
\( \text{(6)} \)

thus \( k_f = \frac{4\pi Dsk_{on}}{4\pi Ds + k_{on}} \)
\( \text{(7)} \)

Assuming molecules are not restricted by their position in space, to show how \( k_+ = 4\pi Ds \), Lauffenburger et al. (1993) solved equation 1 with the conditions of \( L=0 \) (instead of using Equation 2 as a boundary condition) at \( r=s \), and then substituted into Equation 6 to obtain the value for \( k_+ \). Simplifying the equation for \( k_f \):

\[ k_f = \frac{k_+ k_{on}}{k_{on} + k_+} \]
\( \text{(8)} \)

The new formulation of \( k_f \) shows that the overall intrinsic dissociation rate constant relies upon both the transport rate constant \( k_+ \) and the intrinsic association rate constant \( k_{on} \). From establishing relationships between \( k_f, k_+ \), and \( k_{on} \), the probability that a cell will successfully bind with a ligand was formulated to be:

\[ \gamma = \frac{k_{on}}{k_{on} + k_+} \]
\( \text{(9)} \)
This principally measures how severely a receptor and ligand association is rate-limited by \( A + B \xrightleftharpoons{\kappa_r}{\kappa_f} AB \). Several properties of \( \gamma \) include: as \( \gamma \to 0 \), the complex becomes more limited by the reaction and as \( \gamma \to 1 \), the complex relies more on the process of diffusion. The above equations relate to solution receptors, and additional revisions were added by Lauffenburger et al. (1993) to include a 2D cell surface. They incorporated a new interpretation of the rate at which ligands diffuse onto a cell’s surface, describing the rate for an individual cell:

\[
(k_+ \text{cell}) = 4\pi Da
\]  

(10)

Where \( a \) is the cell’s radius and \( D \) is the sum of the added diffusion coefficients of the ligands and cells. The literature also defined the forward reaction rate for the whole cell with \( R \) number of surface receptors as:

\[
(k_{\text{on}} \text{cell}) = Rk_{\text{on}}
\]  

(11)

In which \( k_{\text{on}} \) is the rate constant for individual receptor associations and would be determined experimentally if there were no influences upon ligand-receptor binding in free solution. Similar to Shoup and Szabo (1982), Lauffenburger et al. (1993) created a new forward rate constant. However, the more recent publication altered it to include the number of free binding cites on the entire cell, and Equation 8 becomes:

\[
(k_f \text{cell}) = \frac{(k_+ \text{cell})R(k_{\text{on}})}{(k_+ \text{cell} + R(k_{\text{on}}))}
\]  

(12)

Where \( k_+ \) is replaced by \( (k_f \text{cell}) \). If the entire cell is not under study, then the binding rate constant per receptor is:

\[
(k_f \text{cell}) = \frac{(k_+ \text{cell})(k_{\text{on}})}{(k_+ \text{cell} + (k_{\text{on}}))}
\]  

(13)

and dividing by \( R \) changes the perspective from that of the entire cell to that of individual receptors. To preserve the structure of the “capture probability” determined by Shoup and Szabo (1982), Lauffenburger et al. (1993) included \( R \):

\[
\gamma_{\text{cell}} = \frac{Rk_{\text{on}}}{(k_+ \text{cell} + Rk_{\text{on}})}
\]  

(14)

The significance of this equation is that it shows how both the probability that a receptor and a ligand will bond (9) and the association rate constant both rely on the intrinsic \( k_{\text{on}} \) and the transport \( k_+ \) association rate constants. The mathematical structures and assumptions behind the aforementioned equations created a theoretical model to depict real behaviors.
A. The Hill Equation

Incorporating both the changing rate of probability from Lauffenburger et al. (1993) and the Hill equation was the subsequent phase in creating the model. The Hill equation is:

\[ \theta = \frac{[L]^n}{[K_d]^n + [L]^n} \]  

(15)

Where \( L \) is treated as the number of nanoparticles instead of being the number of bonds and \( K_d \) is the dissociation rate of ligands and receptors. Computing \( \gamma \frac{[L]^n}{[K_d]^n + [L]^n} \) yields the binding rate, assigning each cell a probability of binding with a nanoparticle and multiplying those likelihoods by the rate at which it binds for the subsequent time steps. The formulae for \( K_d \) and the Hill coefficient \( n_H \) were based on Goutelle et al. (2008), where the equilibrium constant was formulated from the reaction: \( A + B \xrightarrow{k_c} AB \) where \( K_d = \frac{[L][M]}{[LM]} \). Instead of the numerator being concentrations or molarities, as they are in most circumstances when computing \( K_d \), \( [L] \) and \( [M] \) were replaced by the estimated concentrations of nanoparticles and cells, respectively. The approximations were retrieved from the uptake assay portion of Pitchaimani et al. (2017). The new Hill equation is:

\[ \frac{[L]^n}{K_d + [L]^n} = \frac{[L]}{(2.1 \times 10^{13}) + [L]} \]  

(16)

Where \([LM]\) is the number of cells multiplied by the average percentage of the uptake of raw nanoparticles, mimicking a reaction between the ligands and receptors. The data for creating the percentage includes nanoparticles from both sizes of 5nm (72% cellular uptake after 24 hours) and 40nm (42% cellular uptake after 24 hours). Continuing to calculate the needed constants, the Hill coefficient \( n_H \) was formulated using the equation:

\[ \frac{\log(81)}{\log(EC_{90}/EC_{10})} = \frac{\text{occupied receptors}}{\text{total receptors}} \]  

(17)

Where \( EC_{90} \) and \( EC_{10} \) are the concentrations required to obtain 90% and 10% between the minimal and maximal response from a drug, respectively. These values were determined by the number of nanoparticles on the substrate and the following expression was used for finding \( EC_{90} \) and \( EC_{10} \):

\[ EC_F = \left( \frac{F}{100 - F} \right)^{\sqrt{H}} \times EC_{50} \]  

(18)

Where where \( F \) is the response rate for which the equation is solved (90 or 10 in the current case), \( H \) is the Hill slope of 1, and \( EC_{50} \) is half the concentration of seeded nanoparticles from the cellular uptake portion of Pitchaimani et al. (2017), assuming that half the nanoparticles would produce
a 50% response rate. The values for $EC_{90} = 5.4 \times 10^{13}$ and $EC_{10} = 6.7 \times 10^{11}$ were computed and substituted into the Hill coefficients formula:

$$n_H = \frac{\log(81)}{\log((5.4 \times 10^{13})(6.7 \times 10^{11}))} = 1.0001 \approx 1$$  \hspace{1cm} (19)

The final form of the Hill equation after substituting the values for $n_H$ and $K_d$ is:

$$\theta = \frac{[L]}{(2.1 \times 10^{13}) + [L]}$$  \hspace{1cm} (20)

This Hill equation was then multiplied with the capture probability to create:

$$\frac{R_{k_{on}}}{(k_{+})_{cell} + R_{k_{on}}} \left( \frac{[L]^n}{K_d + [L]^n} \right)$$  \hspace{1cm} (21)

The above equation was confirmed for its validity through a probability matrix in MatLab and then incorporated into NetLogo to create a model for the behavior of each cell. By extending both the Hill equation and the capture probability to a model predicting cells’ individual binding rates, the future behavior of the cancer may be better anticipated. If the overall binding rate can be foretold, then patients individual chemotherapy treatments could be adjusted to their specific metabolisms.

II. CELL PROLIFERATION

Within a culture of cells treated with nanoparticles, a proportion of them either uptake the ligands, proliferate, or die, depending on their specific coating (or lack thereof) and examples of variations in nanoparticle toxicities are drawn from in vitro cytotoxicity assays in Pitchaimani et al. (2017). In the aforementioned study, cells’ viabilities were examined when samples were exposed to both 5nm and 40nm nanoparticles coated with either the harmful CTAB (cetyl trimethyl ammonium bromide), a biocompatible PSS (poly (sodium 4-styrenesulphonate)), or no coating at all. Approximately 72-73% of the MCF-7 cancer cells perished when exposed to CTAB using the CAM deposition method, however, there remains a portion that were not eradicated in the experimental period of twenty-four hours. To address the portion of cells that lived, a mathematical model of proliferation was created to mimic what occurs when a cell is not affected by treatment. In doing so, a holistic model of tumor growth was scaled to the size of a single cell using the Gompertz equation (Gompertz 1825, Casey 1934), demonstrating how its volume changes over time:

$$V(t) = V_0 e^{\frac{\alpha}{\beta}(1-e^{-\beta t})}$$  \hspace{1cm} (22)
However, the cell proliferation rate in Pitchaimani et al. (2017) is negative, and the cellular viability decreases over time for all three separate nanoparticle coatings (raw, CTAB, PSS). A different version of the Gompertz model had to be constructed; the following equation is its decreasing equivalent with a y-axis scaling factor:

\[
V(t) = V_0 e^{\frac{\alpha}{\beta} - V_0 e^{\frac{\alpha}{\beta}(1 - \frac{\beta t}{24})}} + V_0
\]  

(23)

Where \( V(t) \) is the volume of the tumor at time \( t \) and \( V_0 \) is the volume of the tumor at time 0, with the equality: \( V(0) = V_0 \). For the purposes of the current model, \( V_0 = 1 \) to represent a tumor of initially one cell. The cells proliferation would then be based on that of a tumors growth. \( T_0 \) is the discrete time interval (1, 2, ......24) in hours and \( k = (1, 2, .......) \). In addition, \( \alpha \) and \( \beta \) are parameters retrieved from linearizing the data’s graphs:

\[
\ln[\ln V_{max} - \ln V_t]
\]

(24)

plotted against time \( t \) where: \( V_{max} = V_0 e^{\frac{\alpha}{\beta}} \)

The above equation yields a line with a y-intercept of \( \ln(V_{max}) \) and slope of \( \beta \). To use the Gompertz equation in an agent-based model, its derivative was discretized in the following equations:

\[
\frac{dV(t)}{dt} = \alpha V_0 e^{\frac{\alpha}{\beta}(1-\beta t)}
\]  

(25)

\[
\frac{dV(t)}{dt} \approx \frac{V(kT_0+T_0) - V(kT_0)}{T_0}
\]

(25.1)

\[
= \alpha V_0 e^{\frac{\alpha}{\beta}(1-\beta(kT_0+T_0))} - \alpha V_0 e^{\frac{\alpha}{\beta}(1-\beta kT_0)}
\]

(25.2)

\[
= \alpha V_0 e^{\frac{\alpha}{\beta} - \frac{\beta kT_0}{T_0} - \frac{\beta T_0}{T_0}} - \alpha V_0 e^{\frac{\alpha}{\beta} - \frac{\beta kT_0}{T_0} - \frac{\beta T_0}{T_0}}
\]

(25.3)

\[
= \frac{\alpha V_0 e^{-\alpha T_0}}{T_0}
\]

(25.4)

and since \( t = kT_0 \) we obtain:

\[
= \frac{\alpha k V_0 e^{-\alpha T_0}}{t}
\]

(25.5)

Inputting values of the discrete time interval \( T_0 \) into the discretized Gompertz model yields a per-timestep version of cellular proliferation. In addition, the intuitive rate equation of:

\[
\text{cell count at time } t + 1 - \text{cell count at time } t \\
(t + 1) - t
\]

(26)
was included in the model as well. Substituting the discretized Gompertz equation at \( t + 1 \) time in for the number of cells at the second time and the discretized Gompertz equation at \( t \) time in for the number of cells at the first timestep yields:

\[
V((k + 1)T_0) = \frac{\alpha k V_0 e^{(-\alpha T_{t+1})}}{T_{t+1}} - \frac{\alpha k V_0 e^{-\alpha T_t}}{T_t}
\]  

(27)

Inputting distinct values of time, such as hours, for \( t \), and allowing \( \alpha \) and \( \beta \) to be parameters in the computer model allows for the prediction of the MCF-7 cells proliferation rates. Through computing the spread of cancer cells from a given data set, the amount of time in treatment or the toxicity of the chemotherapy may be better estimated.

### III. CELL DEATH

Modeling cells’ reactions to chemotherapy includes fatality, as nanoparticles coated with chemotherapeutic agents hinder tumor growth. There are several types of cell death including apoptosis, necrosis, and autophagy. Apoptosis is the more natural form of death, as it eliminates old or unhealthy cells and is rigorously regulated; an excess amount of can give rise to autoimmune disorders. Necrosis has its origins in infections or immune responses and results in the cell membrane rupturing or organelles breaking down. Autophagy occurs when the cell regulates the production of its own organelles and nutrient uptake, playing a role in regulating disease response\(^5\). Cell death and and the metabolism of toxic ligands are the foci of the current model’s structure.

The present study is based on the reasoning of Byrne and Chaplain (1998) and will focus on cell necrosis, as treating a cell with a toxic substance transported via a nanoparticle may be categorized as effectively poisoning it. For the purposes of clarity, the solid tumor will consist of cells in the avascular stage, as that is the time during which growth is limited by diffusive effects with an established boundary. The setup of the model also includes multicellular spheroids (MCS) and establishes cells as spheres in a reaction diffusion equation with necrosis occurring as an effect of influences outside the cell. Focusing on the latter half of the tumor’s development time after necrosis appears, the model will depict how the authors, using the models presented by Byrne and Chaplain (1998), showed that cell death affects the overall volume of the MCF-7 cells agglomeration. Their reasoning is founded upon the spherical equation:

\[
\frac{1}{r^2} \frac{\partial}{\partial r} \left( r^2 \frac{d\sigma}{dr} \right) - \Gamma H(r - r_{nec}) = 0
\]  

(28)

Where \( r \) is the radius of the cell, \( \sigma \) is the distribution of the nutrient, \( r_{nec} \) is the necrotic radius of the cell (occurring only when \( \sigma = \sigma_{nec} \)), and \( \Gamma \) is the constant rate at which proliferating cells
consume nutrients. Another equation depicts the change of the cell’s outer radius with respect to time:

$$\left( R^2 \frac{dR}{dt} \right) = \int_0^R [S(\sigma)H(r - r_{\text{nec}}) - N(\sigma) \ast H(r - r_{\text{nec}})]r^2 dr$$  \hspace{1cm} (29)

Where $r_{\text{nec}}(t)$ is the necrotic radius, $S(\sigma)$ is the cell proliferation, and $N(\sigma)$ is the cell loss from necrosis. Since the necrotic core has no proliferating cells, it has no nutrient consumption as reflected in the Heaviside function $H(r - r_{\text{nec}})$:

$$H(x) = \begin{cases} 
1 & x > 0 \\
0 & x \leq 0 
\end{cases}$$  \hspace{1cm} (30)

Thus the nutrient consumption of the proliferating cells is represented by $\Gamma H(r - r_{\text{nec}})$. From defining the change in radius of the tumor in relation to the nutrient uptake of the constituent cells in Equation 28, Byrne and Chaplain (1998) account for the balance between the proliferating and dying cells. Proliferation is bounded between $r < r_{\text{nec}} < R$, meaning that it is contained between the necrotic tissue and the outer border of the cell. The relationship between cell proliferation and natural cell death via apoptosis is illustrated in the following equation:

$$S(\sigma) = s(\sigma - \tilde{\sigma})$$  \hspace{1cm} (31)

Where $s^*$ and $\tilde{\sigma}$ are constants $> 0$ and $s(\sigma)$ and $s(\tilde{\sigma})$ are the proliferation rate and the rate of cell loss due to apoptosis, respectively. Transitioning the equations from natural to abnormal cell death, necrotic tissue is placed in nutrient-poor areas (or places in which the chemotherapy drug is present) and lies between $\sigma < \sigma_{\text{nec}}$, meaning that the nutrient distribution of the cell must be less than that of the necrotic region. Dying tissue evolves according to the equation:

$$N(\sigma) = 3s^* \lambda H(r - r_{\text{nec}})$$  \hspace{1cm} (32)

In which $s$ is a fitting parameter based on the data and $\lambda$ is another data-based parameter. If $\sigma(0, t) = \sigma_{\text{nec}}$, necrosis begins when the tumor’s radius surpasses a value $R^*$ defined as:

$$R^* = \sqrt{\frac{6(\sigma_\infty - \sigma_{\text{nec}})}{\Gamma}}$$  \hspace{1cm} (33)

Where: $\sigma_\infty$ is the constant nutrient concentration on exterior portion of the tumor. Since $\sigma_\infty$ is continuous on the tumor boundary, it ensures the continuity of $\sigma$ and $\frac{\partial \sigma}{\partial r}$. With both necrotic tissue behavior $N(\sigma)$ and the boundary $R^*$ defined, Byrne and Chaplain (1998) note that the overall change in the radius of the necrotic region is:

$$\frac{dR}{dt} = \frac{s^*}{15R^*}(3\sigma_\infty - 5\tilde{\sigma} + 2\sigma_{\text{nec}})$$  \hspace{1cm} (34)
when $R = R^*$. If $\frac{dR}{dt} > 0$, the tumor expands, and the necrotic area continues. As $t \to \infty$, for the purposes of the current model, the necrotic core will persist and Equation 34 would be assumed for each cell within the tumor and then multiplied by the probability that a toxic ligand will be consumed by the cell. The final equation run through the model is:

$$s^* \frac{R^* (3\sigma_{\infty} - 5\tilde{\sigma} + 2\sigma_{nec} R^\prime k_{on})}{15 R^* (3\sigma_{\infty} - 5\tilde{\sigma} + 2\sigma_{nec} (k_{+})_{cell} + R^\prime k_{on})}$$

(35)

Where $R^\prime$ is the number of surface receptors and is changed from the original “capture probability” in Equation 14. Thus, the change in the necrotic portion of the tumors radius as a function of its necrotic core and its uptake of outside nutrients multiplied by the chance that a bioincompatible ligand will bind with each cell (thus inciting the cells demise) yields the rate at which the MCF-7 breast cancer cells die. Predicting the frequency of cell death may give important information on dosing and the effectiveness of different drugs delivered by nanoparticles. By constructing a theoretical model describing both how cells die and their likelihood of undergoing necrosis, experimental trial results or accurate dosages may be made more consistent.

**IV. NETLOGO PROGRAMMING**

An agent-based model in NetLogo was created using all three equations for cellular binding, proliferation, and death. The code was created by defining the cell and the nanoparticle as separate agents and subsequently managing their interactions according to the experimental methods specific to Pitchaimani et al. (2017). The model was divided between the code and the interface, and certain cell attributes were defined in both regions. The interface displayed individual cells’ final binding rates from Equation 21 and their probability of becoming necrotic from Equation 35 while the code scripted for cells’ proliferation rates. The following portions A-D describe the details of the cells’ actions.

**A. Binding Rates**

For the primary part of the model concerning the binding rates, the cells were given individual attributes in the form of equations representing a Hill binding rate (Equation 15), the probability of successfully forming bonds with toxic ligands (Equation 21), and the number of surrounding nanoparticles. Parameters specific to the cell behaviors were either included as a cell attribute or displayed on the interface, where they were set to be randomly chosen within a range of all probable values. Interface variables included the free ligand concentration $L$, the number of free receptor
sites $R$, and the transport rate constant $k_+$. Several ranges of constants were retrieved from the literature, as the diffusion coefficient $D$, association rate constant $k_{on}$, and diameter of the cell $a$ (Equation 10) were obtained from Laffenburger et al. (1993). Uptaking a nanoparticle included first directing the cell to count the number of nanoparticles surrounding it and then assigning individuals a binding rate based on the Hill equation and the capture probability. After each iteration of time, the number of nanoparticles previously counted was included in the total accumulated by the cell, since several ligands may bond to a cells receptors simultaneously. The final binding rate was unique to each cell and affected individual agents’ future actions and responses.

**B. Proliferation Rates**

Cellular proliferation rates were meant to mimic actions of the cancer cells that were not killed by the chemotherapy, as not every cell is affected. Obtaining the ratio of cells that perished under CTAB treatment from Pitchaimani et al. (2017), around 28% of the original amount were found to survive twenty-four hours in the toxic environment. Those remaining cells in the model were commanded to act according to Equation 27, and their behavior was defined as a global variable. The initial number of cells was set to $V_0$, and a loop was applied to the number they proliferated. New cells were “hatched” for each integer-valued timestep, as they were asked to repeat the procedure of producing a new cell up to twenty-four times, representing a day. The variables $k$ and $t$ were set to be randomly chosen from $(1, 2, ..., 24)$ respectively and were controlled in the interface. In addition, parameter ranges retrieved from Olea et al. (1994) for $\alpha$ and $\beta$ were in the interface, and only the number of already proliferated cells was fixed in the code as a cell attribute. The virtual cells’ actions and commands were created to reflect those in actual breast cancer tumors.

**C. Death Rates**

Recreating cells’ reactions when treated with chemotherapeutic nanoparticles also included necrotic development. First, the proponents of Equation 33 were defined from Byrne and Chaplain (1998) and applied to the data from Pitchaimani et al. (2017), including the constants $s^*$, $\sigma_\infty$, $\bar{\sigma}$, and $\sigma_{nec}$, which were set between a range of values. $R^*$ was considered the boundary above which cells underwent necrosis. The change in cell death multiplied by the probability that a receptor and deadly ligand would bind successfully in Equation 35 was appointed to each cell. The higher
the binding rate, the more likely that a cell would die, since more toxic ligands would attach to receptors. While the necrotic boundary $R^*$ was fixed since it was based on constants, the change in necrosis from timestep to timestep due to the evolution of the binding rate formed the basis for tracking the spread of the dying region. Cell death in NetLogo was made to mimic that of the MCF-7 line of breast cancer cells through a combination of individual binding rates and necrotic evolution.

D. Assumptions

The following assumptions were made for model and the NetLogo program and were derived from both the authors and Byrne and Chaplain (1998):

- The percentage of treated nanoparticles that the cell uptakes is directly proportional to their response rates.
- Both cells and tumor radii were assumed to be spherical.
- The majority of cells was assumed to uptake toxic nanoparticles, if not, they proliferated.
- $\frac{\partial \sigma}{\partial r} = 0, r = 0$ for symmetry
- $\sigma = \sigma_\infty$ on $r = R(t)$ or the nutrient concentration of the tumor is equal to that of the cell boundary when the tumor radius equals the cell’s outer radius.
- $\sigma$ and $\frac{\partial \sigma}{\partial r}$ are continuous on $r = r_{nec}$, or the tumor’s nutrient consumption and its change with respect to the radius are continuous when the radius of the tumor is equivalent to that of the necrotic radius.
- $\sigma(r_{nec}, t) = \sigma_{nec}$ to define $r_{nec}$ implicitly, meaning that the nutrient uptake of the tumor with respect to its radius and time is equal to the nutrient uptake of the necrotic area itself.
- $R(t = 0) = R_0$ is the radius of the cell at time $t = 0$ is equal to the initial radius of the tumor. The initial radius for the purposes of the model is equivalent to 1.

V. RESULTS

Interpreting the results began by comparing the NetLogo model to the data on cells’ behavior under nanoparticle exposure from Pitchaimani et al. (2017). The cellular uptake, proliferation,
and death rates from the corresponding publication were calculated and compared to the program’s outputs. The model’s three equations were reviewed for their individual predictive accuracy and were judged on their integrity as a unit, since each individual equation’s output was graphed in plots, and the cells’ overall movement in the interface was analyzed. The NetLogo program was found to accurately predict the positive correlation between the binding and death rates of the MCF-7 cells. As more of the toxic ligands bound to receptors, the higher their death rates became. The proliferation rates increased along with the expansion in the cells’ population, as can be derived intuitively in the absence of chemotherapy. However, despite the Gompertz model being specially fitted to intake decreasing values of cells (since the majority of the agents should die during treatment), the model continued to output a growing cell population. This irregularity may be explained by individual cells’ low binding probabilities and rates of necrosis, expanded upon in the following section.

Despite the rates’ overall appearance matching that of Pitchaimani et al. (2017), there were aspects of the model that were unexpected. When evaluated on the individual cellular level, the binding rate based on the Hill equation, the probability of a successful binding, and the probability of necrosis developing ranged between values of $10^{-5}$ to $10^{-14}$. This discrepancy was due to the parameters being scaled on a microscopic level, yielding minuscule probabilities per cell and not truly predicting the scale at which cells interacted with nanoparticles. When analyzed as a whole, the cells acted more congruent to the established data on MCF-7 cells than evaluated individually. These results indicate that a more detailed program must be utilized due to the complexity of parameterizing cells’ attributes.

VI. DISCUSSION

From completing the project and defining the model parameters, it has been shown that NetLogo has the capacity to model the relationship between the nanoparticle uptake rates and death of MCF-7 breast cancer cells under chemotherapy treatment as depicted in Pitchaimani et al. (2017). This comparison differed from the aforementioned study regarding cells’ individual binding rates and probabilities of proliferation and necrosis. The results show that an agent-based model can be used to accurately replicate cellular behavior as a whole, but not on an individual level. Cells themselves displayed very small probabilities of binding, uptake, and necrosis due microscopic fitting parameters for the equations. Making adjustments to model by redefining agents may be considered in future renditions to the project. Potential improvements include increasing the
number of nanoparticles to which the cells are exposed and making the cell-to-nanoparticle ratio more true to the concentrations used in Pitchaimani et al. (2017). Using a realistic proportion of receptors to ligands would require a program with the ability to calculate large amounts of data, such as MatLab. Additionally, several more agents affecting nanoparticle uptake, cell proliferation, and death could be added. These include cell membranes and distinct types of coated nanoparticles, such as raw, toxic, or biocompatible. In the current model, the cells did not react to whether or not they ingested a specific type of ligand, as the nanoparticle itself was not commanded to act upon the cell. Instead of relying the proliferation and death rates on the toxic nanoparticle uptake, a fixed proportion of cells were set to act independent of their bonds. The relationship between the ligands’ coatings and cell response should be more thoroughly explored in forthcoming models.
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<tr>
<td>a</td>
<td>cell radius for binding rate$^6$ (10 $\mu$m)</td>
<td>$\mu$m</td>
</tr>
<tr>
<td>D</td>
<td>diffusion coefficient$^6$ (1.0 * $10^{-7}$ to 1.0 * $10^{-5}$)</td>
<td>$cm^2$/s</td>
</tr>
<tr>
<td>F</td>
<td>response rate for Hill equation$^{18}$</td>
<td>none</td>
</tr>
<tr>
<td>H</td>
<td>Hill slope$^{18}$ of 1</td>
<td>none</td>
</tr>
<tr>
<td>$K_d$</td>
<td>eq. disassociation constant$^6$ (2.1 * $10^{13}$)</td>
<td>[M]</td>
</tr>
<tr>
<td>$k_+$</td>
<td>transport rate const. per receptor$^6$ (4$\pi$aD)</td>
<td>$cm^3$/s</td>
</tr>
<tr>
<td>$(k_+)_cell$</td>
<td>trans. rate const. per receptor per cell$^6$</td>
<td>$cm^3$/s</td>
</tr>
<tr>
<td>$k_f$</td>
<td>association rate const. per receptor$^6$</td>
<td>$cm^3$/s</td>
</tr>
<tr>
<td>$(k_f)_cell$</td>
<td>assoc. rate const. per receptor per cell$^6$</td>
<td>$cm^3$/s</td>
</tr>
<tr>
<td>$k_{on}$</td>
<td>rate at which ligands &amp; receptors bind$^6$ (1.0 * $10^{-13}$ to 1.0 * $10^{-10}$)</td>
<td>$cm^3$/s</td>
</tr>
<tr>
<td>$(k_{on})_cell$</td>
<td>ligand &amp; receptor binding per cell$^6$</td>
<td>$cm^3$/s</td>
</tr>
<tr>
<td>$k_r$</td>
<td>disassociation rate const. per receptor$^6$</td>
<td>$min^{-1}$</td>
</tr>
<tr>
<td>$L$</td>
<td>free ligand concentration$^6$ (0 to 500)</td>
<td>ligands/area</td>
</tr>
<tr>
<td>$L_0$</td>
<td>maximum ligand concentration$^6$</td>
<td>[M]</td>
</tr>
<tr>
<td>n</td>
<td>Hill Coefficient (1)</td>
<td>unitless</td>
</tr>
<tr>
<td>R</td>
<td>number of free receptors$^7$ (0 to 20,000)</td>
<td>none</td>
</tr>
<tr>
<td>r</td>
<td>cell radius for proliferation rate</td>
<td>$\mu$m</td>
</tr>
<tr>
<td>$r_{nec}$</td>
<td>radius of necrotic region$^1$</td>
<td>$\mu$m</td>
</tr>
<tr>
<td>s</td>
<td>ligand/receptor encounter radius$^6$</td>
<td>nm</td>
</tr>
<tr>
<td>s*</td>
<td>data parameter for cell death$^1$ (100)</td>
<td>none</td>
</tr>
<tr>
<td>$\alpha$</td>
<td>data fitting parameter$^1$ (0 to 1, intervals of 0.0001)</td>
<td>unitless</td>
</tr>
<tr>
<td>$\beta$</td>
<td>y-intercept of Gompertz graphed data$^1$ (1)</td>
<td>unitless</td>
</tr>
<tr>
<td>$\Gamma$</td>
<td>constant rate of nutrient consumption$^1$ (0 to 30, intervals of 1)</td>
<td>unitless</td>
</tr>
<tr>
<td>$\lambda$</td>
<td>data fitting parameter$^1$ (0.1)</td>
<td>none</td>
</tr>
<tr>
<td>$\sigma$</td>
<td>concentration of cell’s nutrients$^1$</td>
<td>unitless</td>
</tr>
<tr>
<td>$\sigma_\infty$</td>
<td>concen. of nutrients on cell’s exterior$^1$ (0 to 1, intervals of .1)</td>
<td>unitless</td>
</tr>
<tr>
<td>$\sigma_{nec}$</td>
<td>concentration of tumor’s nutrients$^1$ (0 to 1, intervals of .1)</td>
<td>unitless</td>
</tr>
<tr>
<td>$\tilde{\sigma}$</td>
<td>positive constant based on data$^1$ (0 to 1, intervals of .01)</td>
<td>none</td>
</tr>
</tbody>
</table>
VII. REFERENCES


Software, G. Quickcalcs.

Appendix A  NETLOGO CODE

The following script takes the biochemical parameters of cellular uptake, proliferation, and death and generates a simulation of chemotherapeutic agent nanoparticle delivery.

```plaintext
;; this determines the two agent types in the model
breed [ cells a-cell ]
breed [ nanoparticles a-nanoparticle ]

;; defines the attributes for agent types
cells-own [hill-binding-rate number-of-surrounding-nanoparticles final-binding-rate probability-of-success-in-binding necrosis-probability ]
nanoparticles-own [number-of-nanoparticles]
patches-own []
globals [count-number-of-nanoparticles change-in-proliferated-cells number-of-proliferated-cells step number-of-dead-cells necrosis-boundary]

;; n is the number of bonds
;; L free ligand concentration
;; R number of free receptor sites
;; k is the Hill coefficient, equal to 1
;; Kd is the ratio between the forward and reverse reaction rates (forward/reverse), equilibrium constant
;; kplus is the transport rate constant per cell, equal to 4piDa
;; kon is the rate at which ligands are bound to a receptor
;; D is the range between: (1.0*10^-5 to 1.0*10^-7)
;; a is the radius of the cell (10)
;; gamma is the probability that a ligand and a receptor will successfully bind, equal to Rkon/(kplus + Rkon)
;; V0 is the initial number of cells in the Gompertz model
;; sigma_inf is the constant nutrient concentration outside of the tumor = 0.8
;; sigma_tilda is a positive constant, the net loss of the cell due to apoptosis = 0.31
;; sigma is the distribution of the nutrient
;; sigma_nec is the 0.4
;; lambda is a parameter estimation = 0.1
;; Ugamma is the = 25
;; s is a positive constant = 100
```
; Rtumor is the radius of the whole tumor
; r_{nec} is the radius of the necrotic region of the tumor
; $R^*$ is the critical value through which necrosis is initiated = $\left( \frac{6 (\sigma_{\text{inf}} - \sigma_{\text{nec}})}{U_{\gamma}} \right)^{1/2}$ = 0.30983
; the onset of necrosis is $\frac{dR}{dt} = \left(\frac{s}{R^*}\right)(3 \sigma_{\text{inf}} - 5 \sigma_{\tilde{\text{tilda}}} + 2 \sigma_{\text{nec}})$

to setup
    clear-all
    setup-patches
    setup-cells
    setup-nanoparticles
    reset-ticks
end

to setup-patches
    ask patches [ set pcolor white ]
end

to setup-cells
    create-cells initial-number-of-cells
    set-default-shape cells "cell"
    ask cells [ setxy random-xcor random-ycor ]
end

to setup-nanoparticles
    create-nanoparticles initial-number-of-nanoparticles
    set-default-shape nanoparticles "nanoparticle"
    ask nanoparticles [ setxy random-xcor random-ycor ]
end

to go
    if ticks >= 10000 [ stop ]
    move-cells
    move-nanoparticles
    binding-process-between-nanoparticles-and-cells
    link-between-cellular-death-and-cellular-binding
change-in-proliferation
cells-proliferation
necrosis-bound
probability-of-necrosis
cell-death
do-plots
set step step + 1
tick
end
to move-cells
  ask cells [  
    right random 360  
    forward 2.5  
  ]
end
to move-nanoparticles
  ask nanoparticles [  
    right random 360  
    forward 1  
  ]
end

;; Combine hill binding rates and probability of binding into one procedure and
  calculate the final binding rate.
to binding-process-between-nanoparticles-and-cells
  ask cells [  
    ask neighbors [  
      set count-number-of-nanoparticles count-number-of-nanoparticles + count
      nanoparticles-here
    ]
  ]
You should define `number-of-surrounding-nanoparticles` first, then calculate `hill-binding-rate`. Otherwise, the program doesn't know how to calculate the hill binding rate

because the `number-of-surrounding-nanoparticles` wasn't defined.

```lisp
set number-of-surrounding-nanoparticles count-number-of-nanoparticles ]
```

```lisp
ask cells

;; let can be used to define local variables that only store the values in the
current procedure, so I encourage

;; to use "let" to define random-float

[ let ligand-receptor-binding random-float kon
let eq-diffusion-coefficient random-float D
let number-of-receptors random-float R

set probability-of-success-in-binding
((number-of-receptors * ligand-receptor-binding)/(number-of-receptors *
ligand-receptor-binding + (40 * pi * eq-diffusion-coefficient)))

set hill-binding-rate (number-of-surrounding-nanoparticles) ^ k /
((number-of-surrounding-nanoparticles) ^ k + (2.1 * (10 ^ 13)))

set final-binding-rate probability-of-success-in-binding * hill-binding-rate
set count-number-of-nanoparticles 0
]

;; cleaning the variable so the number of nanoparticles resets every time step
end
```

find the evidences that could support the correlation between binding and cell death, or binding and cell proliferation

to link-between-cellular-death-and-cellular-binding

as binding increases so does cellular death
to change-in-proliferation
let parameter1 random-float A
let initially-number-of-cells random-float V0
let parameter2 random-float B
let time random-float t
set change-in-proliferated-cells ((parameter1 * k * initially-number-of-cells * e ^
((- parameter1) * (time + 1) * step)) / (time + 1)) - ((parameter1 * k *
initially-number-of-cells * e ^((- parameter1 * time * step))) / time)
repeat (change-in-proliferated-cells)
[ ask n-of 20 change-in-proliferated-cells
[ hatch 1
] ]
end

to cells-proliferation
;; calculate the number of proliferated cells using the revised Gompertz model
;; A is parameter alpha in the revised Gompertz equation for V(t)
;; B is parameter beta in the revised Gompertz equation for V(t)

;; define local variables representing A and B in equation (2). Why local variables
here?
let parameter1 random-float A
let initially-number-of-cells random-float V0
let parameter2 random-float B

;; define number-of-proliferated cells as global variable and calculate it for each
loop,
;set number-of-proliferated-cells ((parameter1 * k * V0 * exp((- parameter1) * (t + 1)
* step)) / (t + 1)) - ((parameter1 * k * V0 * exp((- parameter1 * t * step))) / t)
set number-of-proliferated-cells V0 * e ^ ((parameter1 / parameter2) - V0 * e ^
((parameter1 / parameter2) * (1 - ((parameter2 * t) / 24)))) + V0
;; the below procedure describes repeating (number-of-proliferated-cells) times the
following procedure: [ask one-of cells to produce a new cell]
repeat (number-of-proliferated-cells)
[ask one-of cells
[
  hatch 1
]
]
end

to necrosis-bound
let parameter5 random-float Ugamma
let cell-nutrients random-float sigma_inf
let necrotic-nutrients random-float sigma_nec
ask cells
[
  set necrosis-boundary ((( 6 * (cell-nutrients) - necrotic-nutrients))) / (parameter5) ^ (1 / 2))
]
end

to probability-of-necrosis
let cell-nutrients random-float sigma_inf
let necrotic-nutrients random-float sigma_nec
let parameter5 random-float Ugamma
let parameter4 random-float sigma_tilda
let parameter3 random-float s
ask cells
[
  set necrosis-probability ((parameter3 / ((( 6 * (cell-nutrients) - necrotic-nutrients))) / (parameter5) ^ (1 / 2)) * (3 * cell-nutrients + 5 * parameter4 + 2 * necrotic-nutrients)) * ((number-of-surrounding-nanoparticles) ^ k / ((number-of-surrounding-nanoparticles) ^ k + 2.1 * (10 ^ 13)))
]
end

to cell-death
let parameter3 random-float s
let cell-nutrients random-float sigma_inf
let necrotic-nutrients random-float sigma_nec
let parameter4 random-float sigma_tilda
let parameter5 random-float Ugamma
let time random-float t

ask n-of 50 cells
[ if necrosis-boundary >= ((6 * (cell-nutrients - necrotic-nutrients)) / parameter5) ^ (1 / 2) ] [ die ]
]

ask n-of 50 cells
[ if necrosis-probability >= ((parameter3 / ((( 6 * ((cell-nutrients) - necrotic-nutrients))) / (parameter5) ^ (1 / 2))) * (3 * cell-nutrients - 5 * parameter4 + 2 * necrotic-nutrients)) * ((number-of-surrounding-nanoparticles) ^ k / ((number-of-surrounding-nanoparticles) ^ k + 2.1 * (10 ^ 13))) ] [ die ]
]
end

;if necrosis-boundary = .096 ^ (1 / 2) [ die ] ;;the calculated value of R*
;if change-in-necrosis >= 532.6 [ die ] ;;the calculated value of dR/dt

to do-plots
  ;; create a plot named Binding rate of cell
  ;set-current-plot "Binding rate of cell"
  ;; create a plot pen named Binding rate 1
  ;set-current-plot-pen "Binding rate 1"
  ;; start to plot the hill binding rate of cell number 7
  ;plot [final-binding-rate] of a-cell 1
  set-current-plot "Cell Count"
  set-current-plot-pen "Cell"
  plot count cells
  ;set-current-plot "Death rate of cell"
; set-current-plot-pen "Death of Cells"
; plot [number-of-dead-cells] of a-cell 7

;; You can create another plot to draw binding rate for other cells, following the above procedures
end
VIII. ACKNOWLEDGEMENTS

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