

Diabetes is a disease that is characterized by excessive glucose in the blood stream. Currently, there is an epidemic of diabetes that has resulted from unhealthy lifestyles, which are dramatically different from how humans survived when they evolved from small nomadic hunter-gatherer societies, and food was difficult to find. There are two forms of diabetes, Type 1, often called juvenile diabetes, and Type 2, often referred to as adult onset diabetes (which now occurs in children as young as 5). For our studies here, we will concentrate on Type 1 diabetes, which is an autoimmune disease, and represents only 10% of all cases of diabetes. Type 1 diabetes is a hereditary disease, which occurs in about 4-20 per 100,000 people with peak occurrence around 14 years of age.

The classical symptoms of diabetes are increased hunger (polyphagia), increased thirst (polydipsia), and frequent urination (polyuria) [3]. These symptoms tend to develop rapidly (weeks or months) in Type 1 diabetes. The frequent urination results from the poor water reabsorption in the kidney because of the osmotic imbalance of glucose in the blood. The dehydration from frequent and dilute urine causes the symptom of being thirsty. Other symptoms include blurred vision caused by glucose absorption in the lenses of the eyes, fatigue, weight loss, and poor wound healing. Type 1 diabetes may result in diabetic ketoacidosis, where the urine smells of acetone.

Diabetes increases the risk of heart disease, especially because of atherosclerosis from low insulin. Over time, diabetes can damage blood vessels and nerves, which increases the chance of foot injury and decreases the body's ability to fight infection. These problems can become sufficiently severe as to require amputations. The osmotic imbalances over time result in kidney damage (nephropathy) and nerve damage (neuropathy). The increased pressure in the eye or on the optic nerve can lead to blindness (retinopathy). Thus, diabetes when untreated has very serious consequences.

### Glucose Metabolism

We begin with a short discussion of glucose metabolism. We ingest food to obtain energy to sustain our bodies. The carbohydrates in the food are broken down into simple sugars, which are absorbed into the blood. This raises the concentration of glucose in the blood, where cells can access it for metabolism into energy.

However, when glucose levels get too high, then pressures (osmotic?) increase that can cause problems in the tissues. For normal subjects, the rise in glucose concentration causes the  $\beta$ -cells in the pancreas to release insulin into the blood (along with a number of other hormones). Insulin affects glucose concentration in several ways, including the facilitation of glucose transport across cell membranes, especially in skeletal muscles, and conversion of glucose to glycogen in the liver, which provides a good storage of glucose for future consumption. Thus, increasing the concentration of insulin results in blood glucose concentration decreasing. This negative feedback system helps the body tightly regulate glucose levels to maintain a balance.

There are other significant hormones that are involved in the regulation of blood glucose. In response to high energy demands, epinephrine (adrenalin) is released to break down the glycogen and produce glucose. This hormone works opposite insulin to increase blood glucose. The glucocorticoids help metabolize carbohydrates, especially in the liver, and also help increase blood glucose concentrations. Growth hormone can block the effects of insulin by reducing the liver uptake of glucose and decrease muscle sensitivity to insulin. There are many other hormones that regulate glucose levels in the blood, creating a complex regulatory system that is crucial to maintenance of blood glucose for energy to all cells in the body.

Type 1 diabetes occurs when someone who is genetically predisposed to the disease incurs some unknown environmental assault that initiates the auto-immune system to attack their own  $\beta$ -cells. When the  $\beta$ -cells are destroyed, they boost the immune system to further attack more  $\beta$ -cells, leaving the body without the ability to produce insulin. This severely limits its ability to regulate glucose and results in the onset of diabetes. Because of the immune response, the body cannot regenerate new  $\beta$ -cells nor can transplants succeed.

### Glucose Tolerance Test

Our modeling efforts begin with a simple model developed by Ackerman et al [1,2] in the 1960s, which is based on the Glucose Tolerance Test (GTT). After a 12 hour fast, subject is given a large amount of glucose (1.75 mg of glucose/kg of body weight). This sugar is rapidly ingested, then blood sugar is monitored for the next 4-6 hours. The glucose concentration in the blood of the subject is measured and these data are fit to a model.

As noted above, the glucose regulatory system in the body is very complex. However, we want to develop a simple model with a few parameters that can be fit to the data generated by the GTT. For simplicity, we create a system of differential equations that follow the blood concentrations of glucose ( $G(t)$ ) and insulin ( $I(t)$ ) though the later can be thought of as the complex soup of hormones in the body regulating blood glucose. The general model is written:

$$\begin{aligned}\frac{dG}{dt} &= f_1(G, I) + J(t), \\ \frac{dI}{dt} &= f_2(G, I),\end{aligned}$$

where  $J(t)$  is the ingested source of glucose. (This is an external control function represented glucose sources coming from food ingested.) For the GTT, we can think of  $J(t)$  as a  $\delta$ -function, since we give an initial large quantity of glucose after a fast, then have no other glucose inputted to the system. More complex models that have been developed and are continually being improved often examine ideal ways to match  $J(t)$  with different foods.

The above model is very general, so gives little insight into the dynamics of glucose and insulin. We assume that the body wants to maintain a homeostasis for glucose concentrations in the blood. Also, we are assuming that  $J(t)$  is acting like a  $\delta$ -function, so only affects the initial conditions and is effectively zero away from  $t = 0$ . The homeostasis assumption means that we want to consider a local perturbation of the dynamical system away from equilibrium. Thus, create the perturbation variables,

$$g(t) = G(t) - G_0 \quad \text{and} \quad i(t) = I(t) - I_0,$$

where  $G_0$  and  $I_0$  are the equilibrium values for blood glucose and insulin concentrations, respectively. Thus,

$$f_1(G_0, I_0) = f_2(G_0, I_0) = 0.$$

We expand the general model to linear terms with these definitions, yielding the linearized perturbation model given by:

$$\begin{aligned}\frac{dg}{dt} &= \frac{\partial f_1(G_0, I_0)}{\partial g} g + \frac{\partial f_1(G_0, I_0)}{\partial i} i, \\ \frac{di}{dt} &= \frac{\partial f_2(G_0, I_0)}{\partial g} g + \frac{\partial f_2(G_0, I_0)}{\partial i} i,\end{aligned}$$

where  $g(t)$  and  $i(t)$  now represent the linearized perturbed variables.

For the next step in our analysis, we examine the partial derivatives of the functions,  $f_1$  and  $f_2$ , with our understanding of the physiology of glucose and insulin. An increase in glucose in the blood stimulates tissue uptake of glucose and glycogen storage in the liver. Also, increases in insulin facilitate the uptake of glucose in tissues and the liver. Hence, it is clear that

$$\frac{\partial f_1(G_0, I_0)}{dg} = -m_1 < 0 \quad \text{and} \quad \frac{\partial f_1(G_0, I_0)}{di} = -m_2 < 0.$$

However, increases in blood glucose result in the release of insulin, while increases in insulin only result increased metabolism of excess insulin. These physiological facts imply that

$$\frac{\partial f_2(G_0, I_0)}{dg} = m_4 > 0 \quad \text{and} \quad \frac{\partial f_2(G_0, I_0)}{di} = -m_3 < 0.$$

With these definitions, the linearized system can be written:

$$\begin{pmatrix} \dot{g} \\ \dot{i} \end{pmatrix} = \begin{pmatrix} -m_1 & -m_2 \\ m_4 & -m_3 \end{pmatrix} \begin{pmatrix} g \\ i \end{pmatrix},$$

where  $\dot{g} = dg/dt$  and similarly for  $i(t)$ .

The characteristic equation for this linear system is given by

$$\det \begin{vmatrix} -m_1 - \lambda & -m_2 \\ m_4 & -m_3 - \lambda \end{vmatrix} = \lambda^2 + (m_1 + m_2)\lambda + m_1m_3 + m_2m_4 = 0.$$

By our definitions above, this characteristic equation only has positive coefficients. From basic differential equations (think spring mass system), this implies that the solutions  $\lambda$  are either complex with negative real part or both eigenvalues are negative reals. Both situations give a stable equilibrium as we would expected from this self-regulatory system.

We are only measuring the blood glucose level in the GTT, so we only need the linearized solution for  $g(t)$ . We expect the underdamped situation with complex eigenvalues. Physiologically, you can think of the your body's response to a "sugar high" (maximum of blood glucose), which is followed after an hour or two by a "sugar low" (minimum of blood glucose below equilibrium) that encourages more eating. It follows that the general solution is given by

$$g(t) = e^{-\alpha t}(c_1 \cos(\omega t) + c_2 \sin(\omega t)),$$

where

$$\alpha = \frac{m_1 + m_3}{2} \quad \text{and} \quad \omega = \frac{1}{2} \sqrt{4(m_1m_3 + m_2m_4) - (m_1 + m_3)^2}.$$

If we take

$$c_1 = A \cos(\omega\delta) \quad \text{and} \quad c_2 = A \sin(\omega\delta),$$

then we can approximate the blood glucose level by

$$G(t) = G_0 + Ae^{-\alpha t} \cos(\omega(t - \delta)). \quad (1)$$

This solution has five unknown parameters to be fit to the data. The parameter  $G_0$  represents the equilibrium blood sugar level,  $\alpha$  measures the ability of the system to return to equilibrium state after being perturbed, and  $\omega$  gives a frequency response to perturbations. One might expect that measuring  $\alpha$  should be the primary measure of whether someone was diabetic, as people with diabetes should not be able to return rapidly to normal equilibrium levels. However, the parameter  $\alpha$  was found to have large errors from the many subjects tested by Ackerman

et al [1,2]. A more robust measure was the natural frequency of the system,  $\omega_0$ . (Recall the natural frequency from forced damped oscillators in elementary differential equations.) We define

$$\omega_0^2 = \omega^2 + \alpha^2 \quad \text{and} \quad T_0 = \frac{2\pi}{\omega_0},$$

where  $T_0$  is the natural period of the system. The natural period turns out to be a good predictor of diabetes from Ackerman et al's experiments [1,2]. In particular, they found that if  $T_0 < 4$ , then a person was generally normal, while if  $T_0 > 4$ , then the person is likely to have diabetes. Physiologically, you might relate to this by the idea that normally we get hungry every 3-4 hours.

### Examples

We examine the theory described above with a normal and a diabetic subject given the GTT. The table below gives the data collected on two subjects.

$t$ (hr)	Subject A	Subject B
0	70	100
0.5	150	185
0.75	165	210
1	145	220
1.5	90	195
2	75	175
2.5	65	105
3	75	100
4	80	85
6	75	90

Table 1: Data from the Glucose Tolerance Test. Subject A is a normal subject, while Subject B has diabetes.

The data in the table are fit to the model Eqn. (1). A least squares best fit is performed using either Excel or MatLab. Below is a table of the best fitting parameters for each of the subjects, including the least sum of square errors. The MatLab programs and Excel spreadsheet are put together in a GTT.zip file for anyone interested.

Parameter	Subject A	Subject B
$G_0$	79.1814	95.2125
$\alpha$	0.9927	0.6335
$A$	171.5467	263.1521
$\omega$	1.8127	1.0304
$\delta$	0.90056	1.51604
$LSSE$	225.6757	718.6180

Table 2: Best Fitting Parameters to GTT Model. Subject A is a normal subject, while Subject B has diabetes.

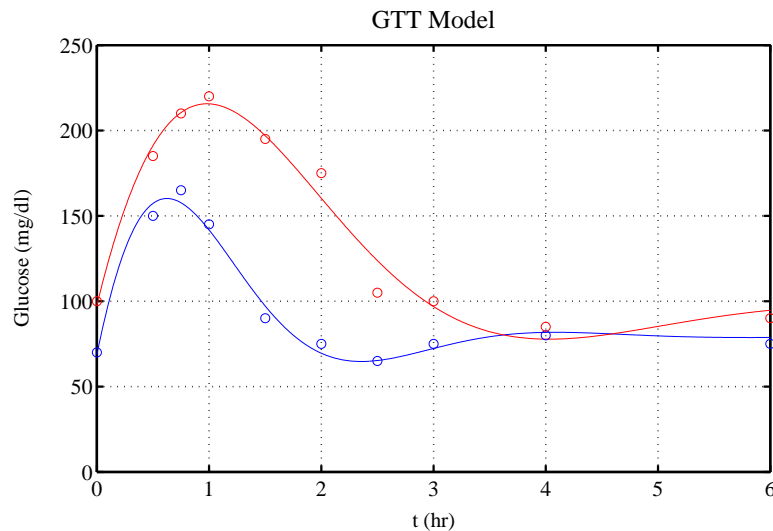
The models and the data are graphed in the figure below. One can readily see that the best parameter fit does very well matching the model to the data. From the definitions of  $\omega_0$  and  $T_0$  above, we find that for Subject A,

$$\omega_0 = 2.0668 \quad \text{and} \quad T_0 = 3.0401,$$

so according to the criterion by Ackerman et al [1,2], this subject is clearly normal. For Subject B, we find

$$\omega_0 = 1.2095 \quad \text{and} \quad T_0 = 5.1947,$$

so according to the criterion by Ackerman et al [1,2], this subject is clearly diabetic.



## Diabetes in NOD Mice

Since diabetes is such a significant human disease, an animal model has been developed to gain a better understanding in the scientific community. One important animal with a diabetic tendency is the *non-obese diabetic* (NOD) mouse. Type 1 diabetes arises in NOD mice when T cells from the immune system become primed to specifically target and kill beta-cells. These cytotoxic T cells belong to a class of lymphocytes displaying a surface marker called CD8 (denoted  $CD8^+$  T cells).

## T Cell Immune Response

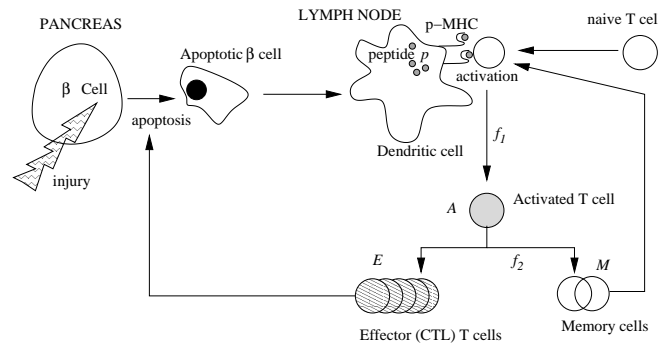
We start with a brief discussion of the T cell immune response, which is central to our model for the onset of diabetes. The T cells mature in the thymus, and most T cells that cross-react with self-proteins are destroyed to minimize autoimmune responses. Next the T cells migrate to the lymph nodes, where they interact with antigen presenting cells (APC's) that display small fragments of proteins (about 9 amino acids) primarily from foreign sources, such as viruses or bacteria. These antigens are held inside a cleft of a larger protein, the major histocompatibility complex or MHC. Depending on the strength, duration, and number of interactions that a particular T cell has determines whether it undergoes activation and issues an immune response.

When a T cell with the appropriate specificity matches a foreign protein, then the activated T cell proliferates. This activated T cell can rapidly reproduce, undergoing approximately 6

cell divisions, to create effector cells (also called cytotoxic T-lymphocytes or CTL's), which seek out and destroy target cells, which protects the host from this foreign invader. These CTL's can be dangerous in the body, so are short-lived. Alternately, the activated T cell can issue a weaker response (mostly when there is not as much of the foreign protein present) and produce long-lived memory cells, which have the same specificity, but wait until the stimulus is encountered again in larger quantities to mount an immune response that destroys the target.

### Autoimmunity for Type 1 Diabetes

An autoimmune disease, such as Type 1 diabetes, is caused by the immune cells attacking the wrong targets. In particular, the T cells attack the  $\beta$ -cells in the pancreas that produce insulin. Early in the development of NOD mice there is a wave of programmed cell death (apoptosis) of pancreatic  $\beta$ -cells. It is conjectured that the clearance of the apoptotic cells is reduced in these mice, which triggers an autoimmune response.



Recent experiments suggest that a specific peptide fragment of the protein IGRP (glucose-6-phosphatase catalytic subunit-related protein) may be the primary culprit in NOD mice. Experiments have been designed to track levels of specific T cells circulating in the blood. Specifically, some experiments of Trudeau et al.[5] track the levels of auto-reactive  $CD8^+$  T cells in NOD mice for several weeks. Surprisingly, the levels of the CTL's show dramatic fluctuations over time as seen in Fig 1.

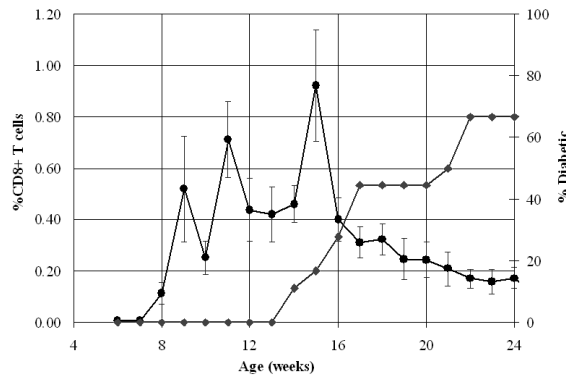
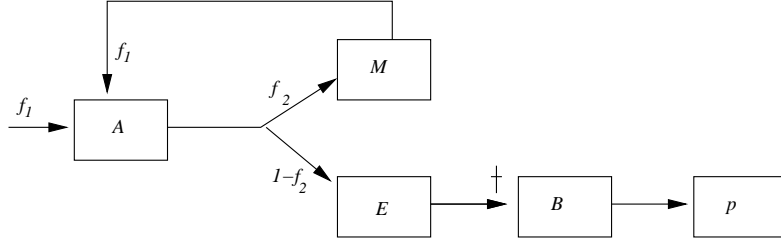


Figure 1: Periodic waves of circulating T-cells occur in mice prone to diabetes (NOD mice) in the weeks before the onset of the disease. Dark line, circles: T-cell level. Grey line, squares: percentage of the animals that became diabetic.

Not all NOD mice develop diabetes, but the presence of cyclic T cell waves for individual animals provide a predictor for the animal becoming diabetic. The data for each one of the mice were aligned to the time of onset of high-blood sugar symptoms. The the pooled data show three peaks in the level of T cells. The amplitude of the oscillations increases with time, and we observe a slight increase in the period of oscillation. Below we present and analyze a model that could shed some light on this process.

### Model for Diabetes in NOD Mice

Our full model examines five state variables. The first variable is the activated T cells, denoted  $A(t)$ . These T cells can become killer T cells (CTL's) or effector cells, denoted  $E(t)$ , or memory cells, denoted  $M(t)$ . The effector cells specifically attack the  $\beta$ -cells in the pancreas, so we track the fraction of  $\beta$ -cells that remain,  $B(t)$ . The destruction of the  $\beta$ -cells leads to the production of the specific antigen peptide,  $p(t)$ , which feedback and affects the number of activated T cells. A schematic for this model is presented below.



This mathematical model assumes that on the time-scale of interest, a system of ordinary differential equations for a well-mixed compartment model can be applied. More details on specific assumptions are given in Mahaffy and Keshet [4]. The detailed full model satisfies the first order system of differential equations given by:

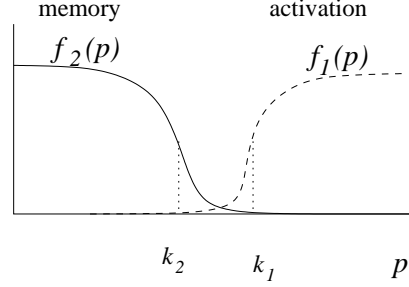
$$\begin{aligned}
 \frac{dA}{dt} &= (\sigma + \alpha M)f_1(p) - (\beta + \delta_A)A - \epsilon A^2, \\
 \frac{dM}{dt} &= \beta 2^{m_1} f_2(p)A - f_1(p)\alpha M - \delta_M M, \\
 \frac{dE}{dt} &= \beta 2^{m_2} (1 - f_2(p))A - \delta_E E, \\
 \frac{dp}{dt} &= REB - \delta_p p, \\
 \frac{dB}{dt} &= -\kappa EB,
 \end{aligned} \tag{2}$$

with nonlinear feedback functions

$$\begin{aligned}
 f_1(p) &= \frac{p^n}{k_1^n + p^n}, \\
 f_2(p) &= \frac{ak_2^m}{k_2^m + p^m}.
 \end{aligned}$$

The nonlinear feedback function,  $f_1(p)$ , represents the activation of the T cells by  $p(t)$  and is given the saturation/switching form noted above that includes the Hill coefficient  $n$  and the kinetic constant,  $k_1$ . The other nonlinear feedback function,  $f_2(p)$ , is a negative feedback function that represents how many activated T cells become memory cells. This function has

the classic Michaelis-Menten (inhibition) form with a Hill coefficient of  $m$ . Below we show the form of these functions.



The differential equation for activated T cells in (2) begins with the production of activated T cells from naive T cells (a constant  $\sigma$ ) and memory cells ( $\alpha M$ ). The activated T cells are lost by becoming effector and memory T cells (rate  $\beta$ ), decaying (rate  $\delta_A$ ), and competing with others ( $\epsilon A^2$ ). The equation for the memory cells includes the production and amplification ( $\beta 2^{m_1} f_2(p)$ ) from activated T cells and the loss due to producing more activated T cells ( $f_1(p)\alpha M$ ) and linear decay (rate  $\delta_M$ ). A similar equation describes the dynamics of the effector cells with production and amplification ( $\beta 2^{m_2}(1 - f_2(p))$ ) from activated T cells and linear decay (rate  $\delta_E$ ). The last two differential equations describe the dynamics of how the effector T cells,  $E$ , destroy  $\beta$ -cells,  $B$ , producing the protein,  $p$ , that activates T cells via the feedback function,  $f_1(p)$ . This model assumes that the  $\beta$ -cells are not replaced and that the peptide is rapidly removed (rate  $\delta_p$ ).

## Model Reduction

The full model is a highly nonlinear model consisting of five differential equations and 17 parameters. Experimental evidence limits the choice of parameters and gives insight into ways of how this system can be reduced. Biological constraints simplify the analysis to a significantly smaller region of the parameter space. However, a five dimensional system is too complex for detailed analysis.

It is important to note that the last two of the differential equations of System (2) operate on different time scales than the first three differential equations. The loss of  $\beta$ -cells is a long term process lasting weeks, so the dynamics of  $B$  can be considered more like a slow moving parameter. At the other end of the time scale is the kinetics for the peptide  $p$ , which is very rapid (on the scale of hours). This latter information allows a *quasi-steady state* (QSS) assumption. The QSS assumption on a variable is that the dynamics of this variable are sufficiently fast that it remains effectively in equilibrium relative to the other variables. This allows the differential equation to be expressed as an algebraic equation. From the equation for  $p$  in System (2), the QSS yields:

$$\frac{dp}{dt} = 0, \quad \text{so} \quad p \approx \frac{RB}{\delta_p} E.$$

If  $B$  is considered a slow moving parameter, then the dynamics reduces to the 3-dimensional system:

$$\begin{aligned} \frac{dA}{dt} &= (\sigma + \alpha M)f_1(p) - (\beta + \delta_A)A - \epsilon A^2 = F_1(A, M, E), \\ \frac{dM}{dt} &= \beta 2^{m_1} f_2(p)A - f_1(p)\alpha M - \delta_M M = F_2(A, M, E), \\ \frac{dE}{dt} &= \beta 2^{m_2}(1 - f_2(p))A - \delta_E E = F_3(A, E), \end{aligned} \quad (3)$$



where  $p = \frac{RB}{\delta_p} E$ .

## Equilibrium and Linear Analysis

The equilibria for (3) are found by solving the three highly nonlinear equations:

$$F_1(A_e, M_e, E_e) = 0, \quad F_2(A_e, M_e, E_e) = 0, \quad F_3(A_e, E_e) = 0.$$

From the form of the equations and the functions  $f_1$  and  $f_2$ , it is easy to see that one equilibrium is the disease-free or trivial equilibrium:

$$(A_e, M_e, E_e) = (0, 0, 0).$$

From the nature of the nonlinearities, there may be from zero to four other equilibria. However, there are relatively stringent biological constraints on the parameters, and in the range of biologically relevant parameters there are two additional equilibria. These can be readily found numerically, but have no analytic solution.

A linear analysis of this model is performed in the usual manner, finding the Jacobian matrix and evaluating it near the equilibria.

$$J(A, M, E) = \begin{pmatrix} \frac{\partial F_1(A, M, E)}{\partial A} & \frac{\partial F_1(A, M, E)}{\partial M} & \frac{\partial F_1(A, M, E)}{\partial E} \\ \frac{\partial F_2(A, M, E)}{\partial A} & \frac{\partial F_2(A, M, E)}{\partial M} & \frac{\partial F_2(A, M, E)}{\partial E} \\ \frac{\partial F_3(A, E)}{\partial A} & 0 & \frac{\partial F_3(A, E)}{\partial E} \end{pmatrix}$$

Provided  $n > 1$  (which is expected, since  $f_1(p)$  is a type of switch), then the linearization about the disease-free state gives the characteristic equation:

$$(\lambda + \beta + \delta_A)(\lambda + \delta_M)(\lambda + \delta_E) = 0.$$

Thus, the eigenvalues for the disease-free state are all negative, which makes this equilibrium a stable node. One would expect the disease-free state to be stable as any small perturbation of the equilibrium representing a minor assault on the body should result in the body returning to the disease-free state.

The two non-zero equilibria result in a much more complicated characteristic equation. However, solutions are readily found numerically. There exists one positive equilibrium that corresponds to a state with all immune cell levels remaining elevated. In that state, effector T cells are continuously killing  $\beta$ -cells, and this corresponds to an autoimmune attack, which eventually results in diabetes. This equilibrium has various stability properties that depend on the parameters and merit further detailed discussion below. The third equilibrium is a saddle with a two-dimensional stable manifold, which for some parameters separates the “healthy” and diseased equilibria. For these parameters, stimuli that fall on the wrong side of this separatrix will be attracted to the diseased equilibrium. For other parameter values, the unstable manifold of the diseased state connects to the stable manifold of the saddle point. In this case, almost all positive initial conditions asymptotically, approach the “healthy” state.

Before continuing the analysis of this 3D model, we examine some solution trajectories of (3) with parameters in the range predicted for NOD mice. The graph of Fig. (2) on the left shows all three equilibria with the saddle node very close to the  $M$ -axis. One trajectory from the saddle node (red) along one branch of its unstable manifold connects the saddle node equilibrium with the disease-free equilibrium at the origin. The other trajectory (green) emanating in the other direction along the saddle node’s unstable manifold wanders until it eventually approaches a limit cycle about the diseased equilibrium. The graph of Fig. (2) on the right shows a close up

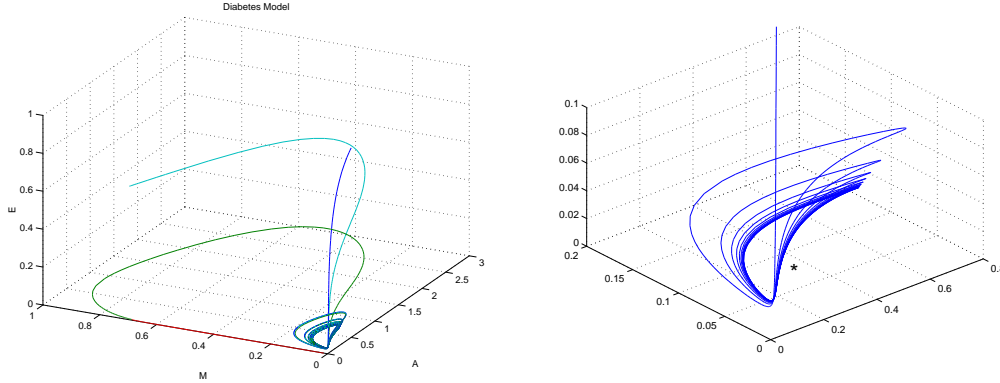


Figure 2: 3D Phase portraits for reduced model.

phase portrait near the diseased equilibrium (asterisk) with the solution trajectory approaching a stable limit cycle.

### Bifurcation Analysis

One conjecture for the onset of diabetes is that peptide fragments from the apoptosis of  $\beta$ -cells in the pancreas are not cleared sufficiently fast, which allows an autoimmune response. It follows that the bifurcation analysis should center on the parameter  $\delta_p$ . Recall that this parameter appears in the QSS approximation,  $p = \frac{RB}{\delta_p} E$ . Note also that we ignored the differential equation for the loss of  $\beta$ -cells,  $B$ , which also appears in this QSS approximation. In fact, the loss of  $\beta$ -cells is equivalent to increasing the peptide clearance parameter,  $\delta_p$ . The 3D reduced model (3) was analyzed, and bifurcation diagrams were composed with the AUTO feature of XPP, freely available software written by G Bard Ermentrout<sup>1</sup>.

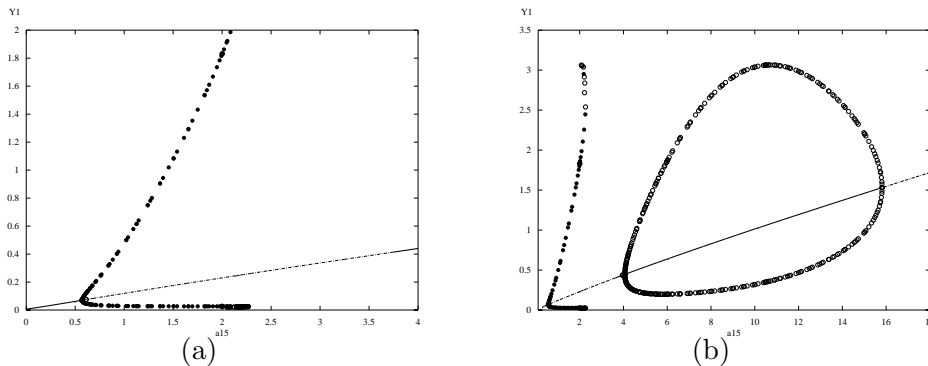


Figure 3: Bifurcation diagram for the peptide decay rate,  $a_{15} = \delta_p$ . The vertical axis is  $A$  in units of  $10^3$  cells. (a) A portion of the diagram, enlarged, shows the typical bifurcation: A Hopf bifurcation occurs at  $a_{15} = 0.5707$  spawning a stable limit cycle. A homoclinic bifurcation occurs at  $a_{15} = 2.268$ . (b) Further bifurcations on an expanded scale: another Hopf bifurcation (to an unstable limit cycle) occurs at  $a_{15} = 4.063$ . This limit cycle vanishes at  $a_{15} = 20.28$ .

The diagram given in Figure 3(a) shows the basic bifurcation behaviour of the model (and uses the default parameters values given in Tables ?? and ??). Moving across this diagram from

<sup>1</sup>XPP is freely available at [www.math.pitt.edu/~bard/xpp/xpp.html](http://www.math.pitt.edu/~bard/xpp/xpp.html)

left to right along the horizontal axis represents increasing values of the peptide decay rate  $\delta_p$ , or equivalently, a decreasing level of beta cells,  $B$ . Close to the leftmost edge, (high  $B$ , or low peptide clearance rate), we find a stable diseased state (solid line with shallow slope). The “healthy” state, also stable, and the saddle node are not indicated on the diagram. Moving towards the right, leads to a supercritical Hopf bifurcation at  $a_{15} = \delta_p = 0.571$ , spawning a stable limit cycle. Here we enter the regime of cyclic behaviour evidenced in Figure 5. The diseased equilibrium is then an unstable spiral, as predicted by the local analysis described above. The limit cycle persists, and its amplitude increases as the parameter increases (respectively, as the beta cell level decreases) up to a homoclinic bifurcation at  $\delta_p = 2.268$  (equivalently at  $B = 0.441$ , i.e., when only about 44% of beta cell mass remains). As seen in our runs, and in the upper branch of this bifurcation line on the zoomed out diagram of Fig 3(b), AUTO has difficulty resolving this global bifurcation. We discuss the nature of this dynamical shift further on.

Following the homoclinic bifurcation, the diseased state remains unstable, and the origin is the only global attractor for some range of the bifurcation parameter. Interpreting this bifurcation diagram in terms of normal and reduced levels of (peptide) clearance rates (by control vs NOD macrophages) suggests why the clearance defect itself could make the difference between healthy (control) mice versus diabetes-prone (NOD) mice: for example, as seen in Fig 3(a), a “control” peptide clearance rate of  $\delta_p = 3$  per day leads to dynamics that always resolve any initial stimulus (returning to baseline where no immune cells persist, since the limit cycle does not occur, and the disease state is unstable) whereas a factor of two decrease to  $\delta_p = 1.5$  per day (representing reduced clearance in NOD mice) puts the same system into the regime of cyclic T-cell waves and autoimmunity.

Reinterpreting this diagram in terms of the gradual decrease of beta-cell mass (from left to right starting from  $B = 1$ ) explains the following features shared by the data of Fig. 1 and the simulation of Fig. 5: (1) the increase in the amplitude of the cycles, (2) the fact that the cyclic behaviour stops abruptly (e.g., around days 80-90 in the simulation of Fig 5) when the homoclinic bifurcation occurs, and (3) the slight lengthening of the period just before this transition. It also explains why (4) the immune cells then decay to the baseline state  $A = M = E = 0$ . Thus, the bifurcation diagram can help to provide a plausible scenario for a mechanism underlying these dynamics.

## Full Model Simulation

Until further notice, this section includes only a hyperlink to a Prosper talk given on the subject and a hyperlink to the article with research associated with modeling this material.

[Prosper Talk on Diabetes in NOD Mice](#)

[Article by Mahaffy and Keshet](#)

[1] Ackerman, E., Rosevear, J. W., and McGuckin, W. F. (1964). A mathematical model of the glucose tolerance test, *Phys. Med. Biol.*, **9**, 202-213.

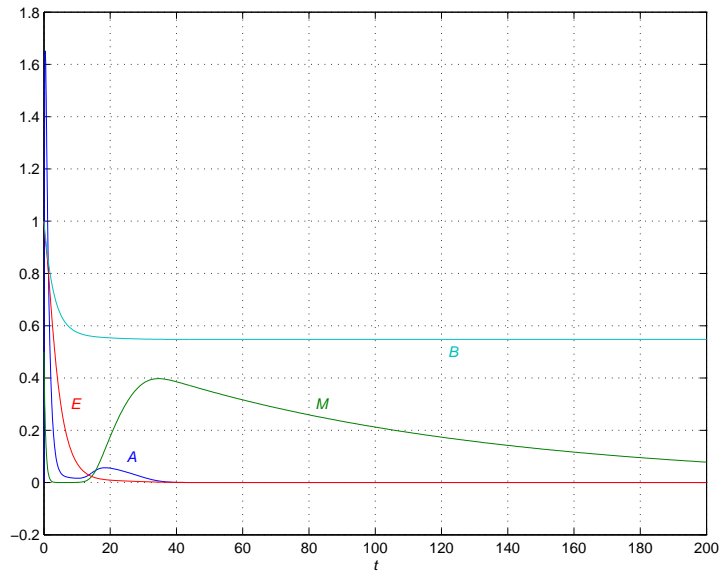


Figure 4: Simulation of the model for NOD mice that **do not** become diabetic. Number of circulating cells (scaled) vs time (days). Dark blue:  $A$  ( $\times 10^3$  cells), Green:  $M$  ( $\times 10^4$  cells), Red:  $E$  ( $\times 10^6$  cells), light blue:  $B$  (fraction of beta cell mass remaining). Simulation uses default (“NOD”) parameter values given in Tables ?? and ?. For the initial conditions  $A = 0, M = 0.5, E = 1, B = 1$ , the immune response is resolved without chronic disease or cyclic waves.

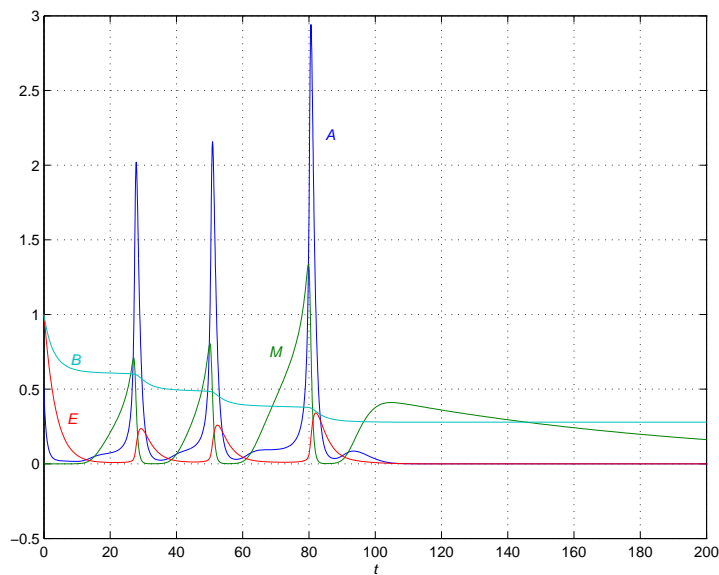


Figure 5: Simulation of the model for NOD mice that **do** become diabetic (by 80-90 days of age). Default (“NOD”) parameter values, and scaling as in Figure 4, but with initial conditions  $A = 0.5, M = 0, E = 1, B = 1$  that evoke the elevated periodic immune response. Dark blue:  $A$ , Green:  $M$ , Red:  $E$ , light blue:  $B$ . The disease progresses with cycles of T cells that cause waves of beta cell killing, as predicted by the model.

- [2] E. Ackerman, L. Gatewood, J. Rosevear, and G. Molnar, Blood glucose regulation and diabetes, Chapter 4 in *Concepts and Models of Biomathematics*, F. Heinmets, ed., Marcel Dekker, 1969, 131-156.
- [3] Cooke, D. W., Plotnick, L. Type 1 diabetes mellitus in pediatrics. *Pediatr Rev*, **29** (11): 37484 (2008).
- [4] Mahaffy, J. M. and Edelstein-Keshet, L., Modeling cyclic waves of circulating T cells in autoimmune diabetes, *SIAM J. Appl. Math.*, **67**, 915-937 (2007).
- [5] J. D. Trudeau, C. Kelly-Smith, C. B. Verchere, J. F. Elliott, J. P. Dutz, D. T. Finegood, P. Santamaria, and R. Tan, Prediction of spontaneous autoimmune diabetes in NOD mice by quantification of autoreactive T cells in peripheral blood, *J. Clin. Invest.*, **111**, 217223 (2003).