# Systems Medicine Chapter 4

# Autoimmune diseases as a fragility of mutant surveillance

This chapter is in memory of Nir Friedman.

**Introduction:**

Hormone glands are prime targets for autoimmune attack. We saw how in type-1 diabetes (T1D) the immune system, namely T-cells, kills beta cells. In Hashimoto's thyroiditis T-cells kill the thyroid cells. In both cases, important hormones are lost, insulin and thyroid hormone, and this loss can be fatal unless treated by lifelong supplement of the missing hormones. The diseases often occur at a young age and are very common, 1% of the population has T1D and 2%, primarily female, has Hashimoto's thyroiditis. Why did evolution fail to eradicate these diseases? Why does the body attack itself?

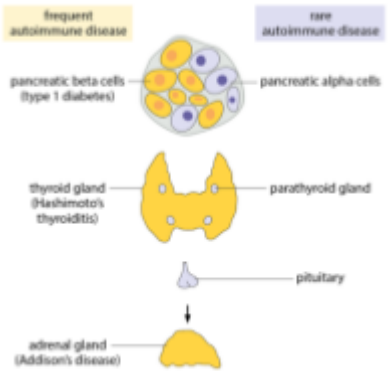


Figure 4.1: Some cell types get prevalent autoimmune diseases which specifically kill them, whereas other cell types do not.

Another question is why the immune system targets these specific cell types and not others? Fig 4.1 shows how some glands get common autoimmune diseases while other glands very rarely get these diseases. Are there rules for which organ gets attacked and which is spared?

In this chapter we will discuss from first principles why endocrine autoimmune diseases arise. Here is the main idea: as we saw, these glands have a circuit that is essential for size control and robustness; this circuit is, however, fragile to mis-sensing mutants that secrete too much hormone and proliferate. To avoid mutant take-over, we will explore the hypothesis that the body uses T-cells to remove the mutants.

These auto-immune T-cells thus serve an essential role in healthy individuals. But in some individuals they go rogue and cause auto-immune disease. Thus, there is a tradeoff between risk of autoimmune disease and risk of diseases of hyper-secreting mutant expansion. Different tissues choose among these two evils according to the evolutionary costs and benefits, and in this chapter we will deduce rules for which tissues get autoimmune diseases versus mutant-expansion diseases.

**Type-1 diabetes is a disease in which the immune system kills beta-cells**

In type-1 diabetes, beta-cells are attacked and killed by the body's own immune system. When enough beta-cells are killed, insulin levels in the blood are insufficient and glucose can't get into the cells from the blood. The cells starve, and switch to metabolizing fats, acidifying the blood below the normal pH of 7.35, which is deadly.

Until the 1920s, T1D was a death sentence for the children who got it. The discovery of insulin 100 years ago by Banting and Best allowed T1D patients to survive by injecting insulin at the proper doses and times, nowadays using an automated pump. But T1D still causes suffering and morbidity and is not easy to control. It is not known how to prevent T1D, causing special concern for people at risk, such as family members of a person with T1D. The fundamental reason that the body attacks specific

cells, beta cells in this case, is not known. As usual in medicine, when the origin is unknown, it is discussed as a combination of genetic and environmental factors.

It is remarkable that T1D is so prevalent and has such a young age of onset (peaking around age 14), because this is a huge evolutionary cost. Natural selection should have eradicated this disease, especially the self-killing immune cells. The fact that these cells are not eliminated raises the possibility that the disease represents the dark side of an important physiological process.

**Many endocrine organs have organ-specific autoimmune disease**

T1D and Hashimoto's are just two of many autoimmune diseases. Autoimmune diseases are classified into systemic diseases that attack many organs (like lupus and rheumatoid arthritis), and **cell-type-specific diseases** such as T1D. Here we focus on the latter. These diseases happen primarily in hormone-secreting organs (endocrine organs) or other secretory organs. There is a range of such diseases. Relatively common diseases with a prevalence of 0.01%-0.1% are Addison's disease of the adrenal cortex, vitiligo of the skin melanocytes, and gastritis of the stomach parietal cells (Fig 4.2). The origin of all these diseases is currently unknown: they are said to be a combination of genetic and environmental factors.

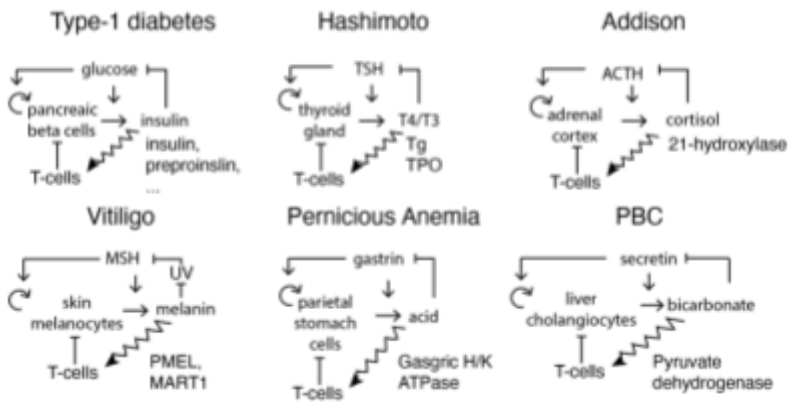


*Figure 4.2: Cell types with autoimmune diseases share the secrete-and-grow circuit motif.*

Equally puzzling is the fact that some endocrine organs virtually never get autoimmune diseases (Fig 4.1). These include the pituitary, alpha cells that secrete glucagon and parathyroid cells that secrete a hormone that controls calcium (PTH). We will try to understand why in this chapter.

All these organ-specific diseases are due to white blood cells called T-cells attacking the specific cell type that secretes the hormone. Antibodies from B-cells also participate in the carnage. Why does the immune system attack our own body cells?

The immune system is designed to protect us against pathogens like bacteria and viruses, and to eliminate cancer cells. The key players are white blood cells called T-cells, so let's learn more about them.

## Executive summary of T cell biology

The T-cells normally attack virus-infected cells, not healthy cells of the body. How do they identify which cells are infected? Cells in the body display small pieces of the proteins they make on "identity cards" on their surface, protein complexes called MHCs. Cells infected by virus therefore display pieces of the virus proteins on some of the MHCs. For example, corona virus forces the cell to make the viral spike protein, and cells display pieces of spike in MHC on their surface. The MHCs are scanned by T-cells. Each T-cell has a special receptor

called the T-cell receptor or TCR. Each TCR can sense specific pieces of protein, called **antigens**, when presented on an MHC.

T-cells that sense normal body proteins are dangerous because they might kill healthy cells. Therefore they are usually eliminated in an "education" process called tolerance. The remaining T-cells respond to foreign proteins and can kill virus infected cells (Fig. 4.3). When a T-cell recognizes a viral antigen, like the corona spike protein, it kills the cell by injecting poison and setting off suicide pathways. The T-cells also activate B-cells to make **antibodies** against the same targets, and the antibodies mark the virus for destruction. The COVID-19 vaccine works by making muscle cells produce spike protein, inducing T-cells and antibodies that can kill the virus and virus infected cells.

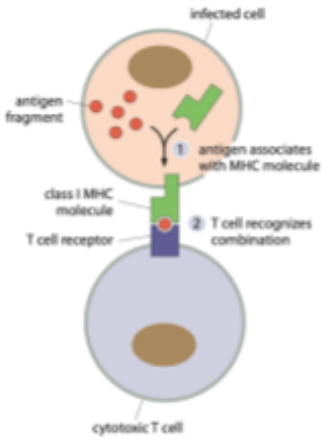


*Figure 4.3: A cytotoxic T-cell recognizes and kills a virus infected cell when the infected cell presents viral antigen fragments in MHC complexes on its surface that binds the T-cell receptor.*

Unfortunately, in T1D, T-cells recognize insulin precursors as antigens, and kill beta cells. In Hashimoto's thyroiditis, T-cells attack healthy thyroid cells. The antigens recognized by T-cells are pieces of the Tg and TPO proteins that are the final stages in production of thyroid hormone. The T-cells also activate B-cells to make antibodies against Tg and TPO. The antibodies participate in damaging the thyroid. Hashimoto's is identified in the clinic by blood tests for anti-Tg and anti-TPO antibodies.

**Autoimmune T-cells are thought to be errors**

The most common hypothesis until recently is that autoimmune diseases are mistakes, failures of **tolerance** mechanisms that eliminate T cells that attack healthy cells. The purpose of tolerance is to eliminate T-cells that detect self-proteins. This is done when T cells are made in the thymus. Their receptors are compared to a vast library of self proteins in the thymus, and self-reactive cells are eliminated or turned into regulatory T cells $T_{regs}$ which act to reduce T-cell activity.

The regulatory T cells are important elements to control against autoimmunity; rare congenital mutations in Tregs often lead to autoimmune attack of multiple endocrine organs. So do mutations that destroy selection in the thymus (AIRE mutations).

T cells that escape elimination in the thymus can still be eliminated or suppressed in the rest of the body when they are over-activated by self-proteins or activated out of context in the periphery.

Still, these processes do not eliminate all self-reactive T-cells. Research over decades has shown that there are self-reactive T-cells in all healthy people. How these self-reactive T-cells sit quiet is not fully understood (Semana et al. 1999; Madi et al. 2014; Yu et al. 2015; Culina et al. 2018; Li et al. 2022; Hs et al. 2021).

Thus, mainstream thought is that self-reactive T-cells are errors in the tolerance mechanisms. A different line of thought in immunology is that ***self-reactive T-cells play maintenance roles in the body*** (Kracht et al., 2016 ; Schwartz & Cohen, 2000; Schwartz & Raposo, 2014). As always in this book, we will go with this line of thought - that what appears to be an error or arbitrary detail

actually has a functional role. This outlook gives you the chance to make discoveries that you might otherwise miss.

**We explore the idea that T-cells can help to remove hyper-secreting mutants**

The organs that get organ-specific autoimmune disease have the same circuit motif as the beta-cells and thyroid cells. In this secrete-and-grow circuit, a signal causes the cells both to secrete a hormone and to proliferate (Fig 4.2). All of these tissues are thus sensitive to mutants that mis-sense the signal. Such mis-sensing mutants can expand and cause loss of homeostasis.

Such mutants are well known clinically. Thyroid cells with mutations in the receptor for their signal (TSH) grow into nodules that secrete too much thyroid hormone. These toxic nodules cause hyper-thyroidism which can be lethal. Incidentally, these nodules are not cancerous- unlike cancer, they don't give rise to new growths in other tissues called metastasis. They are instead adenomas which behave like normal thyroid cells, except that the mutant cells "think" there is too much signal. Similarly, certain mutations in beta cells make them think there is too much glucose as described in chapter 2.

These mutant cells are inevitable. An organ like the thyroid weighs 10g and has $10^{10}$ cells. It thus takes $10^{10}$ cell divisions to make it. Since mutation rate is about $10^{-9}$/base-pair/division, each possible point mutation will be found in about 10 thyroid cells. It is known that at least 50 such mutations cause hyper-sensing and hypersecretion leading to toxic thyroid nodules. Thus, during development every person should develop at least 10x50=500 toxic thyroid nodules secreting thyroid hormone - which would kill the person. Similarly, the $10^{9}$ beta cells are sure to get enough insulin hypersecreting mutants to kill the person from hypoglycemia. Thus, just to have functional endocrine organs requires removal of mutants.

In chapter 2 we saw a **biphasic mechanism** for removing such mutants in beta cells: glucotoxicity. Glucotoxicity causes mutants that "think" glucose is too high to kill themselves. We noted that this mechanism still leaves the range of mild mis-sensing mutants, between the two fixed points (hatched region in Fig 4.4). Other organs, like the thyroid, do not have a mechanism like glucotoxicity at all. There is no TSH toxicity, because TSH needs to vary over a 1000-fold range in normal physiology, such as when iodine levels in nutrition change. Thus, we need another mechanism to remove mutants.

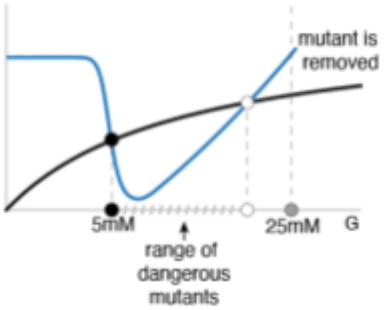


*Figure 4.4: The biphasic mechanism makes mutant cells that strongly hypersense the input signal remove themselves.*

In this chapter, we consider the idea that T-cells can help to remove mis-sensing mutants (Korem et al, 2020). To eliminate these mutants, we need a surveillance mechanism, which we will call

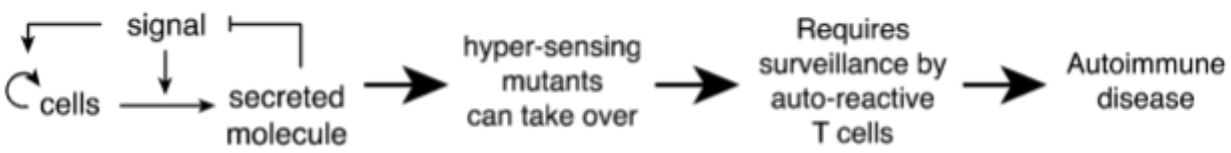


*Figure 4.5: Overview of the autoimmune surveillance theory in which auto-reactive T-cells remove hyper-sensing mutant cells.*

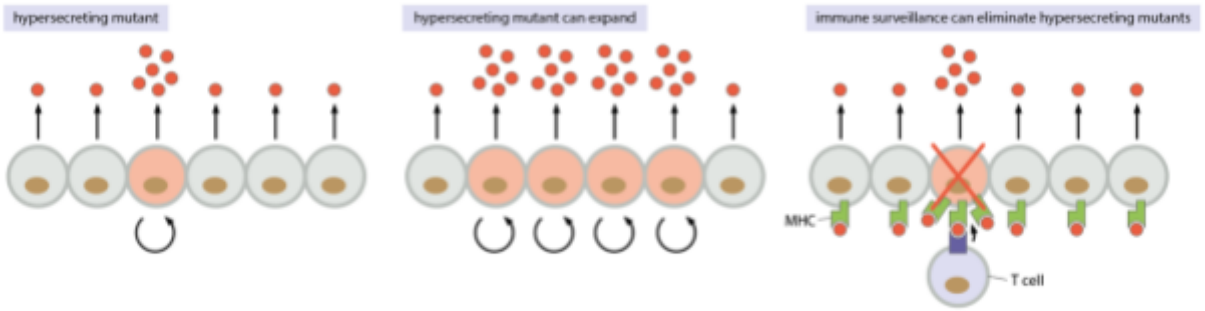


*Figure 4.6: Hyper-sensing mutant cells hyper-secrete the hormone and proliferate, expanding into a clone of mutant cells that leads to abnormal hormone levels. These cells can be removed by the autoreactive T cells in the autoimmune surveillance mechanism*

Autoimmune Surveillance of Hypersecreting Mutants (ASHM) (Figs 4.5, 4.6) or **autoimmune surveillance** for short.

This is a theory we developed with Yael Korem Kohanim during her PhD and systems immunologist Nir Friedman (Korem Kohanim et al. 2020). This chapter is in memory of Nir Friedman, who passed away in 2020. A noble, gentle, and clear thinker.

Autoimmune surveillance needs to detect the hyper-secreting cells in order to eliminate them. Thus, the antigens it detects must be in the production pathway of the hormone. In this way, cells that make more hormones than their neighbors will present more of these antigens on their surface (Fig 4.7). T cells can then detect the hypersecreting mutant cells based on their large number of presented hormone-related antigens.

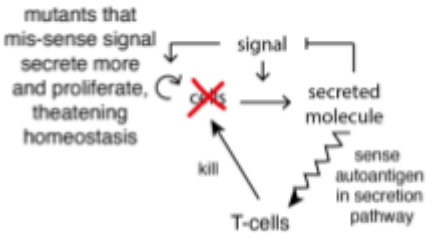


*Figure 4.7: In the autoimmune surveillance theory T cells that sense the autoantigen preferentially kill cells with more antigen then their neighbors, eliminating hyper-secreting mutant cells.*

The best idea is to detect antigens at the very end of the production pathway of the hormone. That design can capture many different mutations that lead to hyper secretion. Indeed, the antigens in T1D (called **autoantigens**) are all pieces of proteins in the insulin secretion pathway. For example, a major antigen is pre-proinsulin, the very last protein in the production pipeline, which is cleaved to make insulin. Other major T1D auto-antigens are also proteins in the secretion pathway of insulin.

Antigens from proteins at the end of the hormone production pathway are found in all the organ-specific autoimmune diseases: the autoantigen in Hashimoto's thyroiditis is the protein cleaved to make thyroid hormone (thyroglobulin Tg, the analog of pre-proinsulin), or the key enzyme that modifies this protein to make the hormone (TPO). In Addison's, the autoantigen is the key enzyme that synthesizes cortisol (21- hydroxylase). Other examples are shown in Figure 4.2 and in (Table 4.1).

| Autoimmune disease | Auto-antigens | Role of auto-antigen |
|---|---|---|
| Type 1 diabetes | Insulin, preproinsulin, PTPRN, PTPRN2, islet cell antigen-69, ZnT8, GAD65 | Insulin synthesis, storage and secretion |

| | | |
|---|---|---|
| Hashimoto's thyroiditis | Thyroid peroxidase, thyroglobulin | thyroid hormone biosynthesis |
| Addison's disease | 21-hydroxylase | Cortisol/aldosterone biosynthesis |
| Vitiligo | PMEL, MART1, tyrosinase, tyrosinase related proteins 1 and 2 | Melanin synthesis and storage |
| Autoimmune gastritis | Gastric H/K ATPase | Acid production |
| Primary Biliary Cirrhosis | PDC-E2 pyruvate/oxo-glutarate dehydrogenase | Bicarbonate production |

*Table 4.1 The autoantigens in many cell-type-specific autoimmune diseases are fragments of proteins that play a role in the production and secretion of the hormone or metabolite produced by the cell.*

The T-cells that recognize pre-proinsulin and other auto-antigens are found in the T-cell repertoire shared by all people, called the **public T-cell repertoire** (Korem et al, 2020, Madi et al. 2014).

**T-cells can tell the difference in antigen between neighboring cells**

For immune surveillance to work, the killer T-cells need to tell which cell makes more antigen than its neighbors, so that they can preferentially kill hyper-secreting cells.

Such differential sensitivity is indeed a feature of T-cells. Experiments tested the relation between the amount of an antigen that a cell presents and the probability that it is killed by a T-cell that recognizes that antigen. The probability of killing is often an S-shaped function of the number of MHCs on the cell surface that present the antigen (Fig 4.8) (Pettmann et al. 2021; Martin-Blanco et al. 2018; Halle et al. 2016 ).

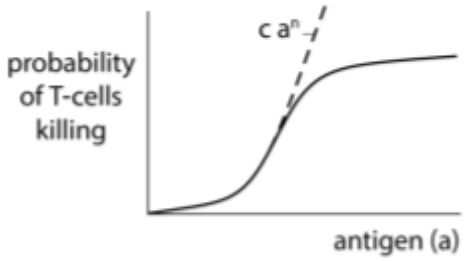


*Figure 4.8: T cells with intermediate affinity kill target cells according to a sigmoidally rising function of presented antigen. Dashed line is an approximation of the Hill function to a power law, with steepness determined by n.*

For T-cells that bind their antigen very strongly, a single antigen presented on a cell is enough. Thus, autoimmune surveillance cannot operate with very strong binding T-cells, because they would not be able to discriminate between hypersecreting cells and normal cells.

Many T cells, however, have only moderate binding to their antigens. For moderate binding, the killing rate *h(a)* goes approximately as a power law of antigen level *a*

$$(1)\ h(a) \approx c\, a^n,$$

with a large exponent n=3-5, signifying a steep relationship (Fig 4.9). We saw such steep relationships in previous chapters, for example in glucose sensing by beta cells. Steep relationships in biology are often caused by mechanisms of '**cooperativity**', for example when multiple T-cell receptors in the same T-cell cluster together and cooperate to make each other more active.

Another important property of the immune system is that it can adapt to a background level of antigen, and only respond to temporal changes in antigen. This adaptation to background is provided by **regulatory T-cells**. $T_{regs}$ provide an incoherent feedforward loop circuit that has the capacity to adapt to a constant input signal (antigen level), and to respond to exponentially increasing antigen

threat (Sontag, 2017). This circuit is explored in exercise 4.2. Other mechanisms exist to help the T cells adapt, such as molecular 'switches' on the T-cell that make them less active if they kill too often, called immune checkpoints. The result of this adaptation is that killing rate goes according to the ratio of antigen relative to the mean antigen presented by all cells, $a/<a>$, so that

$$(2)\ h(a) = c\left(\frac{a}{<a>}\right)^n$$

This killing function therefore has two parameters: the rate *c* and the cooperativity *n*.

The relative sensing explains why the T-cells don't severely attack an organ if it simply starts to produce more hormone. For example, when beta cells start making more insulin due to a change in diet or insulin resistance. More hormone is made in all the cells of the organ, more antigen is presented, $T_{regs}$ level rise and compensate by inhibiting the effector T-cells. The immune system thus adjusts to precisely cancel out the rise in antigen. It remains sensitive to individual cells that make more antigen than their neighbors.

**Autoimmune surveillance can eliminate any mutant, and can do so with a low killing rate**

To work well, autoimmune surveillance needs to eliminate any possible hyper-sensing mutant, and to do so without killing too many healthy cells. To understand how this might work, we analyze a mathematical model for autoimmune surveillance in solved exercise 1.

The main conclusion from the model is that autoimmune surveillance can work effectively and silently it can eliminate mutants while only rarely killing normal cells. The model shows that autoimmune surveillance is an '**evolutionary stable strategy**' (ESS): a mechanism in which no mis-sensing mutant cell can invade and outgrow a large population of normal cells. No matter what is the "perceived glucose" as in Fig 4.9, the mutant cell has a growth disadvantage compared to normal cells. Autoimmune surveillance thus effectively eliminates sporadic mutant cells.

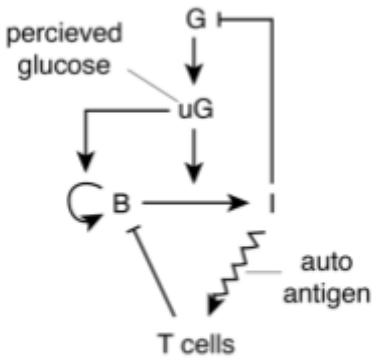


*Figure 4.9: Circuit for autoimmune surveillance in the beta cell-insulin-glucose system. A mutant cell senses u times more glucose than really exists.*

Of course, if all the cells are mutant, as in rare mutations that occur in the fertilized egg and are present in all cells of the body, autoimmune surveillance cannot detect cheater cells – it relies on differences between cells. This occurs in very rare conditions of congenital hyperinsulinemia, in which all beta cells are hyper-secreting mutants and babies are born with low glucose, necessitating removal of the pancreas.

The model also estimates the collateral killing of normal cells by autoimmune surveillance. This off-target killing is prevented by a steep T-cell recognition curve, described in Fig 4.9 by high Hill coefficients n in Eq. 2. Autoimmune surveillance kills much fewer cells than the natural turnover of the tissue when n is large, n=2 or above. Thus, only a small part of the removal of beta-cells is due to autoimmune surveillance, and the rest to natural turnover.

**Surveillance can descend to autoimmune disease in several ways**

Most people don't get autoimmune disease, suggesting that this surveillance mechanism works well. But a small fraction of people unfortunately get autoimmune disease. The risk has a sizable genetic

component. But genetics is not all, there is also a stochastic component- even identical twins have only about a 50% concordance in terms of getting autoimmune disease.

How might autoimmune surveillance fail and descend to autoimmune disease? We don't know for sure. One theory is that a viral or bacterial infection damages the tissue, causing release of self-antigen. The infection grows exponentially and provides an inflammation danger signal. The T–cells see an exponentially rising amount of self-antigen in the context of inflammation. It concludes wrongly that the self-antigen, such as pre-proinsulin, is actually of viral origin.

Genetic factors come into play, such as **MHC variants**. The MHC genes are polymorphic, meaning that people carry different variants. The MHC variants that pose a risk for autoimmune disease include HLA-DR3,4. They encode class 2 MHCs, which are used by antigen presenting cells. Such class 2 MHCs present antigens to other immune cells and play a role in activating B cells and in regulatory T cell function. The high-risk variants may help to set off the heavy guns, the **B-cells** that produce antibodies against the antigen. The antibodies coat the beta-cells, and act as a "kill me" signal. Immune cells then attack the beta-cells aggressively, thinking that they have viruses inside them (Fig 4.10).

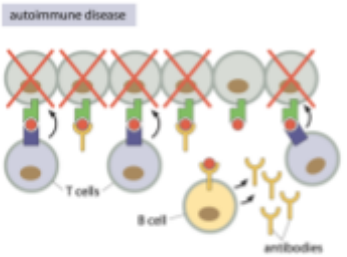

*Figure 4.10: When autoimmune surveillance falsely triggers B cells, enhanced and indiscriminate damage to the tissue can result. This is full-fledged autoimmune disease.*

Another possibility, raised by experiments in mouse, is that these high-risk MHC variants cause $T_{regs}$ to kill themselves at high antigen levels (a kind of biphasic response). When $T_{regs}$ are gone, there is less inhibition on killer T cells, unleashing a large auto-immune response.

Note that these genetic risk factors are relatively common. They were probably selected in the past because they played a beneficial role against a dangerous pathogen. By enhancing B-cell activation or reducing $T_{regs}$ inhibition, such variants set off a powerful immune response and helped to better fight the pathogen. This may explain why these genetic variants are present in a sizable fraction of the population, about 30%, quite consistently across the world population.

In a normal response to pathogens, the antibody and T-cell responses stop when the pathogen is eliminated, and the foreign antigen is gone. In the aftermath of infection, T-cells even kill each other in a process called fratricide. But in autoimmune disease, cells of the targeted tissue are attacked and killed relentlessly and the immune response is not turned off. The killing releases more self-antigen, activating more immune cells, making a **vicious cycle**. Long lasting **memory** B- cells and T-cells are formed which are easily triggered by the antigen. When about 90% of the beta-cells or thyroid cells are killed, hormone production drops so low that clinical symptoms set in.

Whatever the precise route to auto-immune disease, the presence of auto-reactive T-cells provides a potential fragility to self-attack.

**Endocrine tissues that rarely get auto-immune disease are prone to diseases of mutant expansion**

We now turn to the question of why certain organs are attacked and others aren't (Fig 4.1). The autoimmune surveillance theory predicts a tradeoff: if there is little or no surveillance in a tissue, it should get no autoimmune disease. However, it should get diseases of mutant expansion, especially at old ages when mutant cells have had enough time to grow into a large nodule called an **adenoma**.

We can test this prediction by looking at endocrine cells and organs that very rarely have autoimmune diseases - less than $10^{-5} - 10^{-6}$ lifetime prevalence. These organs include the parathyroid (PT) gland, a tiny gland that sits on top of the thyroid (Fig 4.1). Its job is to secrete the hormone PTH in order to control free blood calcium. PTH helps dissolve bone, which is made of calcium phosphate, and to regulate calcium intake from the gut, in order to increase blood calcium.

The lack of autoimmune disease in this gland suggests that it has no autoimmune surveillance or perhaps a weak version. **This predicts that the gland is prone to expansion of hypersecreting mutants.** Indeed, such mutant expansion occurs. It is a common disease with the long name *primary hyperparathyroidism*. It afflicts about 1/50 women after menopause. A hypersecreting mutant cell grows exponentially and becomes an adenoma in the gland, secreting too much hormone and pushing calcium levels up. The excessive calcium comes at the expense of bones, and the symptoms include loss of bone mass and neuronal problems. Treatment sometimes requires surgically removing the adenoma.

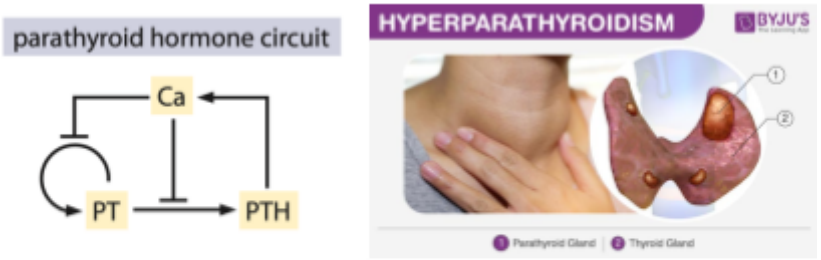


*Figure 4.11: The parathyroid hormone system shows the secrete-and-grow circuit motif and shows prevalent expansions of hypersecreting mutant cells that lead to excessive blood calcium.*

The PT gland has a circuit that is sensitive to take-over by mis-sensing mutants. The circuit is essentially the same as in the other glands, except for a sign reversal. In this circuit (Fig 4.11), the signal, calcium, inhibits both the proliferation of PT-cells and secretion of PTH. A mutant cell that mis-senses normal calcium as too little calcium (hypo-sensing mutant) is the culprit: such a mutant expands and hyper-secretes PTH, and if it grows to a sizable adenoma it leads to excessive calcium. This circuit also has biphasic mutant protection (low and high calcium kills PT cells). But intermediate mis-sensing mutants are still dangerous. It is precisely such mild mis-sensing mutations in the calcium receptor that cause the adenomas in the parathyroid gland[1].

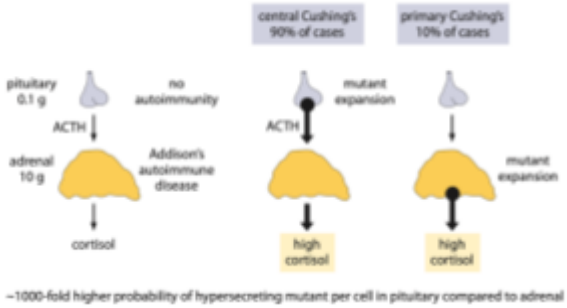


*Figure 4.12: The HPA axis shows an example of the inverse relation of autoimmune disease and hypersecreting mutant-expansion disease. The adrenal gets an autoimmune disease whereas the pituitary almost never does. Conversely, the chance per cell of a hypersecreting mutant expansion is about 1000 times larger in the pituitary.*

Thus, a tradeoff seems to exist between two evils: autoimmune disease and diseases of hypersecreting mutant expansion. An example of this mutant/autoimmunity tradeoff occurs in the HPA axis. Recall the two glands, the pituitary and the adrenal, A and P. Each has a version of the circuit motif that is fragile to mis-sensing mutants, as we saw in chapter 3. Mutants in the pituitary

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[1] It seems that the parathyroid has at least some autoimmune surveillance because autoimmune disease can be caused by certain drugs. In particular, drugs that enhance immune response against cancer, by blocking immune checkpoints, have parathyroid autoimmunity as a side effect in some patients. Thus, autoimmune surveillance in this organ may be tuned to low levels that prohibit autoimmune disease under normal conditions.

that hyper-sense hormone $x_1$, for example, grow into nodules that hyper-secrete hormone $x_2$ (ACTH), making the adrenal secrete too much cortisol (Fig 4.12). This is known as **Cushing's syndrome**, with depression, hypertension, muscle wasting and fat distribution in the face and abdomen. The same disorder can be caused by adrenal mutants that hyper-sense $x_2$. However, 90% of Cushing's syndrome is caused by mutants in P, not in A. This is surprising because the number of cells in P is smaller by a factor of 100 than in A (relevant cells in the adrenal total about 10g, in the pituitary about 0.1g).

Our theory predicts then that the adrenal A is protected from mutants by autoimmune surveillance, and hence should have autoimmune disease. Indeed, the adrenal is destroyed by T cells in an autoimmune disease called Addison's disease. The pituitary virtually never gets such an autoimmune disease (unless caused by certain checkpoint inhibitor drugs). It seems to have little autoimmune surveillance, perhaps because it is an immune-privileged site. However, as mentioned it shows relatively frequent mutant-expansion diseases- the most common form of Cushing's syndrome called central Cushing's (Fig 4.12).

Similar pituitary mutant expansion diseases plague other HP-axes. Pituitary mutant cells in the growth axis account for acromegaly and gigantism, and in other pituitary pathways to disease of hyper-gonadism and hyper-thyroidism. Again, like the adrenal, the thyroid is prone to autoimmunity, whereas its pituitary controller cells (TSH-secreting thyrotroph cells) are prone to mutant expansion.

What rules might determine if a tissue gets autoimmune disease or diseases of mutant expansion? One possibility is based on the evolutionary cost of these diseases: it pays to set things up so that the less severe disease occurs (Fig 4.13). In beta-cells, a hypersecreting mutant expansion is lethal, because it causes low glucose. Thus, it makes sense to have strong autoimmune surveillance, which can sacrifice some of the population to T1D, but can save a higher fraction of the population from lethal mutant expansion disease.

In the PT gland, in contrast, high levels of calcium caused by mutant expansion are bad but not lethal.

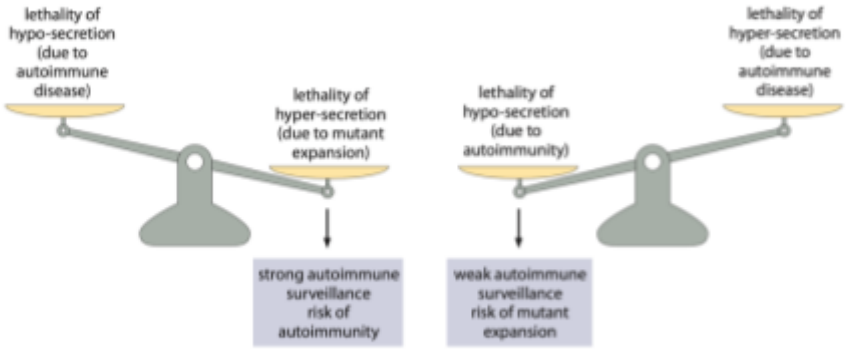
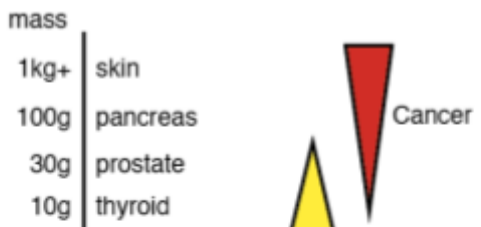



*Figure 4.13: One explanation for the prevalence of autoimmune versus mutant expansion disease is natural selection of immune surveillance levels so that the more lethal disease is rarer.*

This gland has a biphasic mechanism to protect against strong hyper-secreting mutants. The mild mutants that take over cause high calcium, but below the lethal calcium level of roughly 4 times the normal level. But a reduction in PTH, as would be caused by autoimmune destruction of the PT gland, can push calcium down to lethal levels: even a 20% reduction is lethal (below 0.9mM compared to the normal level of 1.1mM). Thus, it makes sense that autoimmune surveillance does not evolve in the PT gland, to avoid the risk of low calcium, at the price of a less severe mutant expansion disease with a late age of onset.

Perhaps the simplest explanation for which organ gets which disease stems from the number of mutations in the organ. The smaller the organ, the



fewer cell divisions are needed to make it, and the fewer cell divisions occur over life (most glands have a similar turnover time). The fewer the mutations, the less the need for autoimmune surveillance. The cutoff seems to be at a mass of about 1g, which is about 10^9 cells. Above 1g are endocrine organs with autoimmune diseases: beta cells (1g), adrenal (10g), thyroid (10g), as shown in Fig 4.14. Below 1g are glands with mutant expansion and very rare autoimmunity: pituitary cells (0.1g), parathyroid (0.3g), renin secreting cells (0.1g).

At above 10-30g, the disease spectrum shifts again, with less autoimmunity and more cancer (prostate 30g, pancreas 100g, skin several Kg), Fig 4.16. This is because there are so many cell divisions, that mutations become too frequent. The required levels of autoimmune surveillance would be too high to avoid autoimmune disease. Organs thus change strategy to stem-cell-based production, in which a single stem-cell division is amplified to make thousands of cells by transit amplifying cells. This reduces the number of mutations that remain in the stem cells of the tissue. But stem cells are more prone to cancer, being cells with high proliferative potential. In the transition zone of 10-30g one sees both cancer and autoimmunity, as in the thyroid, and prostate (Fig. 4.14)

I like the prospect of such rules for diseases, pointing towards the periodic table of diseases that will be our closing chapter. The table can predict unknown diseases based on first principles. We will expand on this theme later on. But now we turn to understand another common pathology of the immune response - inflammation and excessive scarring known as fibrosis.

**Solved exercise 1: Develop and analyze a model for autoimmune surveillance of hypersecreting mutants. Show the conditions where autoimmune surveillance can not be invaded by any mutant cell, known as an evolutionarily stable strategy.**

Since we are dealing with secrete-and-grow circuits, we use beta cells as an example. We begin with the growth equation for beta-cell mass B from chapter2, whose growth rate is controlled by glucose G:

$$(1)\ \frac{dB}{dt} = \mu(G)B$$

We consider first healthy conditions, where glucose is near the stable fixed-point $G = G0 = 5mM$. Recall that the beta cell growth rate is zero at $G0$, so that biomass growth equals removal and beta-cell mass is at steady-state (Fig 4.15).

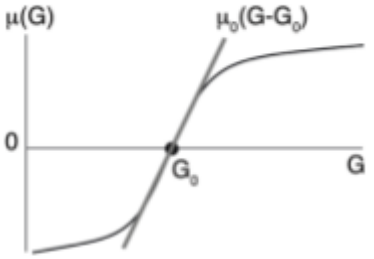


*Figure 4.15: A linear approximation for the net growth rate near the fixed point.*

Near $G0$, we can approximate the growth rate as a line with slope denoted $\mu 0$, so that $\mu = \mu 0(G - G0)$ (gray line in Fig 4.15). This linear approximation is not essential but makes the math easier and is sufficiently accurate for our purposes. Thus

$$(2)\ \frac{dB}{dt} = \mu_0\left(G - G_0\right)B$$

Now let's add autoimmune surveillance, in which beta-cells are killed by T-cells. We begin with the case in which there are only non-mutant beta-cells, called **wild-type** cells. Each cell presents $a$ copies of antigen on its surface, and the average <a>=a since we assume all cells are the same. The antigens are in the secretion pathway and hence proportional to the insulin production rate per cell. Inserting the killing term from Eq. 1 into the growth equation, we find

$$(3)\frac{dB}{dt} = \mu_0\left(G - G_0\right)B - c\left(\frac{a}{<a>}\right)^n B$$

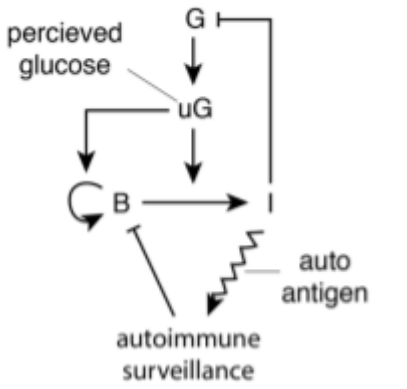


*Figure 4.16: The autoimmune surveillance circuit, where a mutant cell senses u times more glucose than reality.*

Since all cells are wild type cells, $< a >= a$, and the killing term is just equal to $c\ 1^n = c$. Thus, at steady state,

$$(4)\ \frac{dB}{dt} = 0 = \mu_0\left(G - G_0\right) - c$$

whose solution is the steady-state glucose level that is slightly shifted upwards from $G0$ due to the effect of beta-cell killing:

$$(5)\ Gst = G_0 + c/\mu 0$$

The higher the killing rate $c$, the higher the glucose because more beta-cells are killed per unit time, and hence less insulin, and thus more glucose. In extreme cases, where $c$ is very large, killing is widespread and we have very high glucose levels- this is the situation in autoimmune disease like T1D.

Now let's consider a mutant beta-cell that mis-senses glucose. It acts as though the true glucose level G is actually $uG$, where $u$ is the **mis-sensing factor** (Fig 4.16). Such mis-sensing mutants were discussed in chapter 2. The mutant cells have enhanced insulin secretion and proliferation. In the equation for such a mutant we need to replace all occurrences of $G$ by $uG$. The equation for the growth of the mutant population $B_m$ is therefore

$$(6)\ \frac{dB_m}{dt} = B_m\left(\mu_0\left(u\,G - G_0\right) - c\left(\frac{a_m}{<a>}\right)^n\right)$$

where the autoimmune surveillance killing term contains the mutant antigen level $a_m$. Initially, there is a single mutant cell surrounded by wild-type cells. Since there is only one mutant cell, the average antigen <a> is virtually unaffected by the mutant so that <a> can be approximated by the wild-type level of antigen. Similarly, glucose level G is virtually unaffected by the mutants as long as they are few.

The antigen level of the mutant is determined by its insulin production rate. Using the insulin equation from chapter 1 we have $a_m \sim q\,f(uG)$. Using $f(G)\sim G^2$, we find that mutant cell antigen level is $a_m = qu^2G^2$. The wild-type antigen level is $< a >= qG^2$, because u=1 for the wild-type cells. Thus, the mutant killing term $c\left(\frac{a_m}{<a>}\right)^n$ depends only on the mis-sensing factor u, because the factors $q$ and $G^2$ cancel out, leaving $\left(u^2\right)^n = cu^{2n}$. Thus:

$$(7)\ \frac{dB_m}{dt} = B_m\left(\mu_0\left(uG - G_0\right) - cu^{2n}\right) = \mu(u)B_m$$

We conclude that the mutant has a growth rate that is determined by its mis-sensing factor u:

$$(8)\ \mu(u) = \mu_0\,(uG - G_0) - cu^{2n}$$

In order for autoimmune surveillance to work perfectly, we need the wild type cells ($u = 1$) to have the highest growth rate, higher than all possible mutant cells. This is called an '**evolutionary stable strategy**' (ESS): a mechanism which cannot be invaded by any single mutant. For organ size control, the wild-type cells should have zero growth rate (proliferation equal removal and thus a steady population size), and all mutants should have negative growth rate and thus eventually vanish. We therefore need to find a condition such that growth rate $\mu(u)$ is maximal at $u = 1$ (Fig 4.17). This occurs when

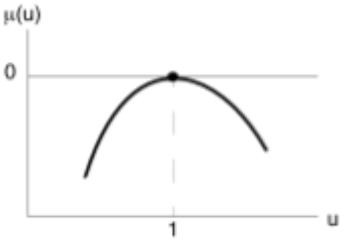


*Figure 4.17: For an evolutionarily stable strategy – a design that cannot be invaded by any mutant - net growth rate must be maximal for the non-mutant cells, which sense glucose as it is (u=1).*

$$(9)\ \frac{d\mu(u=1)}{du} = 0 \qquad condition\ for\ evolutionary\ stable\ strategy$$

and also that the second derivative is negative to ensure a maximum. Taking the derivative of Eq.8, we find

$$(10)\ \frac{d\mu}{du} = \mu_0 G\ -\ 2ncu^{2n-1}\ =\ 0$$

The glucose level G is just the glucose level for the wild-type case (Eq.5) because a single mutant cell can't affect glucose levels much. Plugging in $G_{st}$ from Eq.5, and u=1, we find the condition for ESS:

$$\frac{c}{\mu_0 G_0} = \frac{1}{2n-1}$$

This equation connects the killing rate c, normalized by the natural turnover of beta cells, to the steepness of the killing function n. Interestingly, the higher the T-cell cooperativity (steepness) parameter *n*, the lower the killing rate *c* that is required for ESS. Since the cooperativity of immune recognition is high ($n \sim 3\text{-}5$), killing rate should be small, about 10-20% of the natural turnover rate parameter $\mu_0 G_0$. Thus, these secret agent T-cells can work subtly and precisely.

**Exercises**

**4.2 An exponential threat detector in the immune system:** This exercise explores a circuit that can respond to an exponentially rising threat such as a virus or bacterium, and ignore signals that do not change with time, such as antigens from self-tissues (Sontag 2017), see Fig 4.18.

a) Consider an antigen u(t) presented by antigen-presenting cells to T cells. This activates effector T-cells T(t) that perform the response functions, and also regulatory T cells denoted R(t) that inhibit the effector T cells. Interpret the following equations and their parameters:

$$\frac{dR}{dt} = a\,u - b\,R$$
$$\frac{dT}{dt} = c\frac{u}{R} - d\,T$$

b) Solve the steady state of R and T for a given steady-state level of u. Explain why this steady-state does not depend on u.

c) Explain why a step-function change in u from level $u_1$ to level $u_2$ leads to a pulse of T activation that returns to its original level (Fig 4.21)

d) Shows that if antigen rises exponentially, $u(t) = u_0\, e^{\alpha\, t}$, the effector T cell activity does not return to the original steady state level. Show that the activation level above their original steady state is proportional to the antigen exponential growth rate parameter α.

e) What happens when the antigen rises linearly with time $u = \alpha\, t$?

f) Explain why an exponential threat detector might be useful for the immune system.

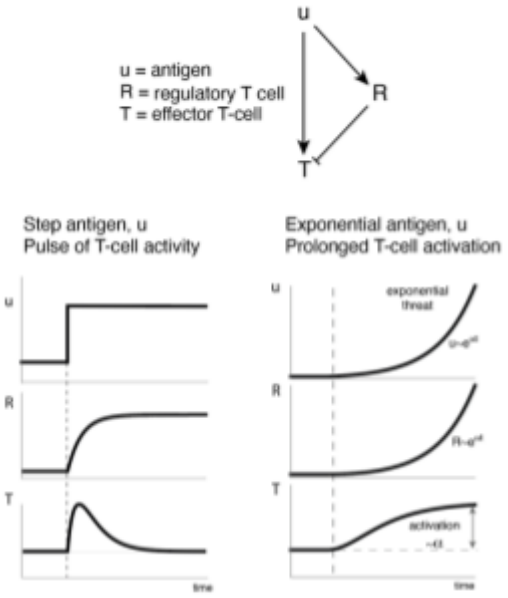



*Figure 4.18: An exponential threat detector circuit in the immune system. Antigen activates cytotoxic T cells,T, and also regulatory T cells, R, that inhibit T. A step increase in antigen causes a pulse of T that adapts back to baseline. Only an exponentially increasing antigen level, as in pathogens or early cancer, causes a prolonged rise in T without adaptation to baseline.*

**4.3 Viral dynamics:**

Consider the model for the concentrations of virus, u(t), effector T-cells, T(t), and $T_{regs}$ ,R(t):

$$\frac{du}{dt} = (\alpha_0 - c\,T)u$$
$$\frac{dR}{dt} = u - R$$
$$\frac{dT}{dt} = \frac{u}{k+R} - T$$

a. Explain the equations and the parameters $k$, $c$ and $\alpha_0$.

b. Calculate the steady-state solution.

c. Numerically solve the equations for various values of $\alpha_0$. Use $c = 1$, $k = 1$, $R(0) = T(0) = 0$, and $u(0) = 1$. Explain the meaning of these initial conditions.

d. Assume that when the virus concentration goes below a minimal dose, $u_0 = 0.01$, it is killed by the innate immune system. What is the maximal viral growth rate $\alpha_0$ for which the virus is killed by the immune system? What happens if $\alpha_0$ is larger than this value?

**4.4 Theories for autoimmunity:**

(a) Read about the hypothesis of 'molecular mimicry' for autoimmune diseases.

(b) Read about the 'hygiene hypothesis' for autoimmune diseases.
(c) Discuss their pros and cons, and compare to the 'surveillance of hypersecreting mutant' theory discussed in this chapter (200 words).

**4.5 Bistability in a simple model for autoimmunity:**
Consider this simple model: The immune system attacks healthy tissue. This releases auto-antigens, making the immune killing stronger, in a cooperative way, with Hill coefficient n=2. The variable is the amount of autoantigen a(t). The autoantigen is removed at rate γ.

(a) Explain the equation:

$$\frac{da}{dt} = c\frac{a^n}{k^n+a^n} - \gamma\, a.$$

(b) Draw a rate plot showing the fixed points. Consider (graphically) different scenarios (different parameters) with different numbers of fixed points. When is there bistability?
(c) Which scenario corresponds to an autoimmune disease? Which corresponds to no autoimmune disease?
(d) Suppose that individuals vary in their genetics in a way that affects the parameters of the equation. Does an increase in the parameter c increase the risk for autoimmune disease? Repeat for the parameters k and γ.

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