**Script - The Cardiovascular System Part 1: Blood**

## Introduction

In previous chats, I introduced the basics of chemistry, cells, networks, drugs, how they work and how to think about them. We also discussed platforms for drug discovery and how they Differ. Finally, we focused on what we do at n-Lorem and ASO Technology.

Today, we begin a series of chats about organs. Organs reflect how complex multicellular organisms organize their components to meet the needs of the entire organism. Each organ typically has several key functions and some secondary responsibilities. To an extent, the architecture of each organ reflects the main task that the organ must perform.

It is essential to understand the main responsibilities of each organ and how each organ meets those responsibilities in order to understand diseases and therapeutic strategies because they are based on what the organs do normally as well as what changes when they fail. Once you think about organs this way, you will find that it is very easy to understand how organs can become dysfunctional and cause disease. Further, you will find that it is easy to understand how a disease in any organ is measured, and how drugs to treat that disease might be created.

The first organ system which we will focus on is the cardiovascular system. I really like teaching about this organ system because it is well understood, and simple. Plus, diseases of the cardiovascular system are the most common causes of death in developed economies, and there are many genetically caused diseases of this system. Some of these are rare enough to be mutations that we try to address at n-Lorem. Today, I

will introduce the components of the cardiovascular system, focus on the roles of blood vessels, and a very important component of the system, blood.

## Components of the Cardiovascular System

The cardiovascular system is made up of a pump, two types of pipes, and a spigot. Obviously, the pump is the heart. The two types of pipes are the arterial pipes and the venous pipes. These two sets of pipes differ significantly because they have very different jobs. The spigot is the kidney. A fourth component, the liquid that flows through the pipes, that I would argue should be considered an organ is blood. Blood is a remarkable organ because it is never stationary and every component in blood is made by a different organ.

I am sure you already know the key functions of your cardiovascular system. They are, of course, to supply tissues with the oxygen and nutrients they need to function and to remove waste material from each tissue or organ and deliver it to an organ that can dispose of waste.

KEY POINT 1:
The components of the cardiovascular system are the heart, arteries, veins, kidneys, and blood.

KEY POINT 2:
The main job of the cardiovascular system is to provide oxygen and nutrients to tissues and rid the body of waste.

## Blood Functions

Let's start with blood. Blood has many different functions. For the moment, we will focus on four main functions related to the jobs of the cardiovascular system.

In the lungs, blood must exchange oxygen for carbon dioxide. It must then deliver oxygen and nutrients to all the tissues or organs. Then, it must collect waste, which includes carbon dioxide, from each of the tissues and deliver the waste to the appropriate organ for removal from our bodies. Finally, at the end of each cardiovascular cycle, blood must exchange the carbon dioxide, which it just collected from all the tissues it passed through, for oxygen in the lungs.

After collecting oxygen from air in the lungs, blood then delivers oxygen and nutrients to all the tissues of the body. As blood passes through the tissues, it exchanges oxygen for $CO_2$, which the tissues have generated as a result of the chemical reactions of life. Finally, blood must re-exchange the $CO_2$ collected from tissues for oxygen in the lungs. If that sounds like a cycle to you, you are correct. It is a cycle – the oxygen cycle.

So how does the oxygen cycle work? Of course, you know that you have hemoglobin, and that it resides in red blood cells. Red blood cells are tiny, simple cells. When they are mature, red blood cells don't even have a nucleus. That means that mature red blood cell cannot divide and are, in fact, in the process of dying. That means that you must make many billions of red blood cells constantly in your bone marrow.

Just as proteins do the work of cells, proteins do the work in blood. Hemoglobin is a beautiful protein that carries iron-hemo, the technical syllable for iron, globin, a globulary protein. Why does hemoglobin have iron and why must you have hemoglobin? If you ever took basic chemistry, you know that iron has two ionic states or valence states. Iron can be a metal which has no charge or Iron can carry two positive

charges, Iron plus two, or it can carry three positive charges, Iron plus three. In the periodic table, Fe is the symbol for iron. Importantly iron can be oxidized (increase the charge it carries from two to three) or reduced (decrease the charge it carries from 3 to 2) at normal physiological conditions and this process releases energy.

Hemoglobin uses iron and some specific amino acids to form its active center. Remember, that oxygen and carbon dioxide diffuse easily and very rapidly across cellular membranes. Of course, you know that oxygen levels are quite high and carbon dioxide levels are low in the air that you breathe. So, in the lung, oxygen levels (or tension) are high. The oxygen you breathe in then simply displaces carbon dioxide in hemoglobin, because there is very little carbon dioxide in air, but lots of oxygen. Then blood flows to tissues. Because the tissues are using a lot of oxygen for the chemical reactions of life, and those chemical reactions produce carbon dioxide, carbon dioxide levels accumulate in tissues, and therefore, in tissues, carbon dioxide replaces oxygen in hemoglobin. Blood then flows to the lungs and the cycle is repeated. It is really that simple. In the lungs, you have a lot of oxygen, it replaces carbon dioxide. In tissues the process is reversed.

<u>KEY POINT 3:</u>
A specialized protein in the blood, hemoglobin, supports the oxygen cycle. This cycle is essential to life.

Given how vital this process is, not surprisingly, it is very tightly controlled. For example, you must constantly make red blood cells in the bone marrow. Your body constantly assesses how many red blood cells are circulating in blood. If the level of red blood cells is too low, a protein, erythropoietin, which you may know by its trade name, Epogen, is made in the liver. It is dispatched to the bone marrow and tells the marrow to make more red blood cells. One of Amgen's largest

selling drugs is Epogen. It is given to cancer patients being treated with anticancer drugs that are toxic to bone marrow.

If you don't have enough iron, no matter how many red blood cells there are, they can't function in their oxygen cycle role. If your body senses that the tissues need more oxygen, more erythropoietin is made in the liver, stimulating more red blood cell production. This doesn't help much because you already have plenty of red blood cells. Your problem is a lack of iron. Not only does making more red blood cells not help, but eventually the bone marrow wears out and you have iron deficiency anemia. Another good way to be anemic is to bleed excessively. Of course, in females, the menstrual cycle results in the loss of blood every month. If blood loss is excessive, that also will produce an anemia that can be improved by taking iron. That's why most women take iron.

Anemia means inadequate numbers of red blood cells. It is cool to realize that if hemoglobin's shape changes in the absence of iron, that deforms red blood cells. So, when a trained person looks at your blood under a microscope, the red blood cells look different, and iron deficiency anemia can immediately be called the diagnosis.

Hemoglobin is essential for life and there are hundreds of mutations in hemoglobin that cause a very wide range of diseases. Humans are thought to have begun in southern Africa, then they migrated up to the Mediterranean. Malaria is endemic in Africa and over the centuries has killed billions of humans. A mutation in hemoglobin that prevents the malaria parasite from undergoing its normal reproductive cycle protects the humans with that mutation from malaria, but at a high price. That mutation causes sickle cell anemia. Why is it called sickle cell anemia? Because the red blood cells are deformed. Instead of being round, they look sort of like a banana, at least to me.

You can also become unhealthy if you have too much iron or make too many red blood cells. If you have too much iron in cells other than red blood cells, it is very toxic because it is so chemically reactive. Metallic iron is also toxic. Chronic iron overload is called hemochromatosis. Too many red blood cells can cause problems on their own as well. Having too many red blood cells chronically is called polycythemias - meaning too many red blood cells. One manifestation of too many red blood cells is that the blood becomes viscous and no longer flows easily and therefore, in the small arterioles, the red blood cells clog these tiny blood vessels. That blocks oxygen and nutrients from getting into tissues. The lack of oxygen and nutrients kills cells. Cell death in an organ caused by lack of blood flow is called an infarction. When an infarction happens in the heart, that's a heart attack. Since blood flow is necessary for every organ, if blood flow is blocked, that organ will have an infarction.

KEY POINT 4:
The oxygen cycle is very tightly controlled.

KEY POINT 5:
Any reduction in the $O_2$ carrying capacity of blood is sensed and to compensate, erythropoietin is dispatched from the liver to direct the bone marrow to make more red blood cells.

KEY POINT 6:
Many diseases are caused by mutations in hemoglobin, including anemias and thalassemia, such as sickle cell disease.

KEY POINT 7:
Both low iron and high iron levels cause diseases. Low iron causes iron deficiency anemia and high iron causes hemochromatosis.

## Clotting

The fourth main job of blood is to respond to leaks or obstructions in pipes.

How do leaks in pipes happen in your world? Sometimes leaks are caused by events outside the pipes. The dreaded break in a big water carrying pipe that happens when some construction worker digs in the wrong place and the inconvenience caused are things most of us have experienced. Similarly, there can be externally caused leaks in the pipes we use to get rid of waste, the sewer system. Blockages of pipes can be caused by external events too. For example, sometimes tree roots can obstruct water pipes or sewers. As you would expect, externally caused obstructions are more common in sewers than water pipes because sewers are low pressure pipes while water pipes distribute water at a higher pressure.

Externally caused leaks of blood vessels are typically caused by trauma. You cut your finger, or get stabbed, or you have an accident, and you bleed. Any time you bleed, you react to it as an emergency because you know that if you allow bleeding to go unchecked, you “bleed to death” (We will return to this topic, and I will show you what “bleed to death” really means). You also react very differently to trauma that causes a leak from an artery vs a vein. Why? Because arteries are high pressure pipes and blood will literally spurt out of an artery, so you know that you have less time to stop the bleeding before you “bleed to death”.

Externally caused obstructions of blood vessels can happen when another organ or mass impinges on blood vessels. The most common cause of this is cancer, but sometimes normal organs can grow too large and obstruct a blood vessel. In our bodies, just like everyday life, the low-pressure pipes, the veins, are more likely to be externally obstructed than the high pressure, rapid flow pipes, the arteries.

Leaks and obstructions of blood can also happen without an external cause. Just as water mains break because the water pressure is too high for the pipes to manage, some leaks happen when a person's blood pressure is too high, causing a pipe to rip open. When that happens in a human, the rip in the artery is called an aneurysm. Aneurysms can occur in the large arteries like the aorta and are a surgical emergency because the person could bleed to death. Rips can also occur in the tiny arteries that are called arterioles, and, in this case, the manifestations depend on where the arteriolar leak occurs. Any disruption in the supply of oxygen to any organ leads to rapid death of cells in the site that are no longer getting oxygen and that is called an infarction.

As a general rule, the most common and severe issue caused by rips in arterioles due to high blood pressure are in the brain and those infarctions are strokes, but infarctions can occur in any tissue if oxygen supply is disrupted.

A more common cause of arterial leaks, particularly of the larger arteries, is due to build up of gunk on the arterial wall. Over time, that weakens the wall of the artery. The gunk is fat, or lipids, and you know all about those lipids. They are "bad cholesterol" or LDL and other types of lipids. When lipids deposit on the wall of the artery, they cause inflammation which then weakens the wall. The process is called atherosclerosis, which means stiffening, sclerosis, of the artery, athero. Atherosclerosis - stiff artery. No matter the cause, rips of big arteries or aneurysms are surgical emergencies. Rips in little arterioles cause disruption in oxygen supply to a tissue and that leads to an infarction. Of course, an infarction in the heart is a heart attack, and an infarction in the brain is a stroke, but infarctions can occur in any organ and produce disease.

Now, think about the challenge that blood has. It must flow easily through all blood vessels and yet it must be ready to stop a leak of blood instantly. That is a tough job. The main players in this extraordinarily important process are a set of proteins made in the liver, and some cells called platelets. I am sure that if you think about it, you could design the system we use every second of every day.

The first problem to solve is to be alerted that there is a leak in a vessel and that you cannot wait until you have lost so much blood that it is obvious. When a blood vessel springs a leak, it releases chemical signals that tell the platelets that you have a problem and they stop in their tracks and sit down on the site that is leaking producing a plug, exactly like what you would do if your water pipe leaked. Of course, you would turn the water off, then plug the leak. We don't have that option. The leak must be plugged while blood is still flowing. Step one is for the platelets to sit down on the leak site. Step two is that the platelets signal a set of proteins that are called clotting factors, which are made in the liver and circulate constantly in blood, to get busy building a permanent patch made up of a strong mesh, fibrin. That then traps more platelets and clotting factors till a solid patch is made. When you cut yourself, you see the patch. It's a scab. With modern methods, we can watch all this happen in blood vessels in real time.

What are clotting factors and how does the system work to assure that normally blood flows smoothly, yet blood can respond to a leak instantly by stopping blood flow at the leak? The way we would design the system is that we would have some proteins that normally are not active but can become active immediately circulating constantly in blood. Indeed, that is how the system works.

Clotting factors are all enzymes. Remember an enzyme is a protein that helps chemical reactions go faster. All clotting factors are proteases - proteins that degrade other proteins and sometimes themselves. So

clotting factors are inactive proteases. They are inactive because they have an extra bit of protein that blocks the enzymatic center. Such proteins are called pro-proteins, meaning inactive, but easily converted to an active form, in this case, by other clotting factors. There are twelve clotting factors that are made in the liver. These clotting factors comprise a multistep cascade.

The first clotting factor is present both in tissue and in blood and you can think of it as the first responder. It degrades the bit of protein that blocks the active center of the second clotting factor and then that process is repeated step-by-step. Again, a multistep cascade is a very common solution in biological systems because it provides multiple opportunities for control. You want to be in control of when you clot and when you don't. This is the system you use.

Given how important platelets are, you can guess that the level and activity of platelets are tightly controlled. If platelet levels are too low, a factor made in the liver, thrombopoietin, is dispatched to the bone marrow to stimulate production of more platelets. Once again, one of Amgen's major products is thrombopoietin and it is used to help cancer patients have less bleeding during chemotherapy that is toxic to the bone marrow.

Not only is platelet number monitored and carefully regulated, but platelet activity is also carefully regulated. Platelet activity is simple, they must aggregate. They must stick to a blood vessel wall and create the initial plug. So, platelet activity tests are called platelet aggregation tests. Platelet aggregation is caused by two or three chemicals that are constantly made in the body, ATP and adenosine. Platelet activation can also be stimulated by solid substrates that you might find on the vessel wall. In platelet activity tests, platelets are asked to aggregate by adding one of those stimulates of platelet aggregation.

Similarly, the production of clotting factors in the liver is very tightly controlled. The entire pathway that leads to the production of the clotting factors in the liver can be thought of as one of the many critical pathways that are regulated as an entire pathway. The production of proteins that are necessary for the performance of blood and other organs is one of the two main jobs that the liver does. Of course, the other job that the liver does is the exact opposite. It filters venous blood and extracts waste and potentially toxic materials from the blood, metabolizes and excretes them in bile.

So now you know that platelet number, activity, and the levels of clotting factors are carefully regulated. As we have seen time and again, these complicated, highly regulated systems can break down. Fortunately, when there are problems with the clotting system, we have a good many drugs that we use to treat patients with these problems.

One interesting way this system can break down is that, on occasion, you can produce antibodies that recognize your own platelets, rather than foreign invaders. That can lead to consumption of platelets so severe that the bone marrow can't make up for it and that is called thrombocytopenia - thrombo, platelets, cytopenia - cell deficiency. Such antibody-caused reductions in platelet levels can be quite severe and can even be fatal. Antibody-caused platelet reductions can also be very difficult to treat and sometimes requires the use of immunosuppressant agents, like steroids.

Similarly, production of clotting factors in the liver can fail. Untreated, loss of clotting factors as you would expect can lead to bleeding. People with diseases that make the liver dysfunctional often bleed to death. Alcohol and other toxic chemicals destroy liver cells and over time the liver in these patients fail. If you starve or have a terrible diet, clotting factors can't be made and, once again, the manifestation is bleeding. Clotting factors can have mutations that make them not work or work in

an uncontrolled fashion. Hemophilias are caused by mutations that prevent production of clotting factors or cause them not to work. Other mutations can cause too much clotting. Often these issues manifest themselves where blood flow is slow - the deep veins, and patients have deep vein thrombosis – thrombosis - clotting.

Blood clots in the deep veins (this usually happens in the veins in the legs), are dangerous because they damage local tissue, but more, because they can break apart and small clots then can travel in blood to the lungs and the heart. The effects in each organ have different names, but they really represent the same process and problem. A clot blocks an artery leading to an area of an organ supplied with $O_2$ by that artery. A lack of $O_2$ causes cell death and you now know that is called infarction. In the lung, that is called a pulmonary embolus, and in the heart, it is called a heart attack.

Another source of a fragmented clot-like material happens in patients with atherosclerosis. As I said, if you have elevated bad cholesterol (or other types of lipids), the lipids accumulate on the surface of blood vessel walls. An individual site of accumulation of lipids, platelets and other material is called a plaque because that is what they look like. The medical term for plaque is atheroma (a thrombus-like spot on a vessel wall.) The whole process is called thrombo-embolic disease. Thrombo - a blood clot; embolic - a piece of a clot breaks off and travels with blood till it gets trapped in a blood vessel.

<u>KEY POINT 8:</u>
Blood must flow easily but must be able to respond to leaks immediately.

<u>KEY POINT 9:</u>

Clotting is managed by clotting factors made in the liver. Clotting factors are proteases that are purposely made to be inactive. When needed, the protease activity of clotting factors activates the clotting system.

KEY POINT 10:
The second major component of the clotting system is a set of cells, called platelets.

KEY POINT 11:
When clotting is not properly regulated, serious, often fatal diseases are caused.

Before we leave blood, let me describe in general physiological terms what bleeding to death actually means. The sudden loss of blood generates an emergency response in the body just as it would generate a 9-1-1 call. The sudden loss of blood causes a rapid reduction in blood pressure that means that blood returning to the heart from the veins is reduced. This means that the right atrium doesn't fill and because of that the right ventricle is unable to pump appropriate amounts of blood to the lung. So, less blood gets oxygenated. The same problem exists only worse for the left atrium and left ventricle. The output of blood from the left ventricle is reduced and that is called reduced cardiac output. The autonomic nervous system responds to that by increased adrenergic activity which means more norepinephrine (noradrenaline) is released causing an increase in heart rate, constriction of the major arteries, and dilation of the tiny arterioles. All of that is designed to get more oxygen to more tissues, but of course it fails. Eventually, a fatal arrhythmia of the heart occurs and the patient dies.

## Drugs Used to Affect the Activities of Blood that are Related to Blood Flow

Before we close the book on these functions of blood, let's review some of the classes of drug used.

Blood itself can be a drug and is given any time blood levels are seriously low.

Blood cells can be drugs. RBCs, white cells, and platelets are often administered when any of those cells are low.

Anemia treatments include blood, Iron, and erythropoietin. And for thalassemias, which cause blood to flow poorly, things like urea are also used.

Low platelet levels are often treated with thrombopoietin or the administration of platelets.

Hemochromatosis is often treated with CHELATORS-agents. These are chemicals that bind iron and then are excreted in urine.

Thromboembolic diseases are often treated with "Blood thinners". Blood thinners do not actually make the blood thinner, rather they reduce the ability of blood to form clots.

The oldest drug used to reduce clotting is coumarin-the rat poison. It works by preventing the liver from making clotting factors. It has a TI (therapeutic index) of 1, and you know from earlier chats that a TI of 1 means coumarin is as likely to cause side effects as it is to cause benefit, so it is not often used these days. There are far better drugs that reduce only key clotting factors that can be lowered safely. In development, there are many other selective inhibitors such as the Ionis factor XI drug. The goal of these drugs is to prevent unwanted clotting yet allow clotting to happen when you cut your finger.

There are also a good many agents that alter the ability of platelets to form clots. Of course, the granddaddy of them all is aspirin.

Finally, there are lipid lowering drugs, like statins, used to prevent the atherosclerotic process, but we will discuss those in later chats.

## Conclusion

To wrap this up, blood serves many other functions, but we will cover those in conversations about other issues such as fighting infections. Now that we are well grounded in how blood is made and how it functions regarding blood flow, next time, we will deal with the pump, the pipes, and the spigot.

## Glossary

**Hemo:** Iron
**Hemoglobin:** An iron containing blood protein whose main function is to collect $O_2$ from the air we breathe, deliver $O_2$ and other nutrients to tissues, exchange $O_2$ for $CO_2$, return to the lungs, and re-exchange $CO_2$ for $O_2$.
**Anemia:** Reduced ability to carry out the $O_2$ cycle.
**Thalassemia:** A disorder caused by mutations in hemoglobin.
**Clotting factors:** Proteases made in the liver that can be activated to form clots.
**Atherosclerosis:** The accumulation of clot-like plaques in arteries.
**Thromboembolic disease:** Pieces of a clot or plaque that break off and are carried by blood until they lodge in a vessel and cause infarcts.
**Infarct:** Tissue death caused by a lack of $O_2$.