THE KIDNEY AND THE CONCEPT OF CLEARANCE*

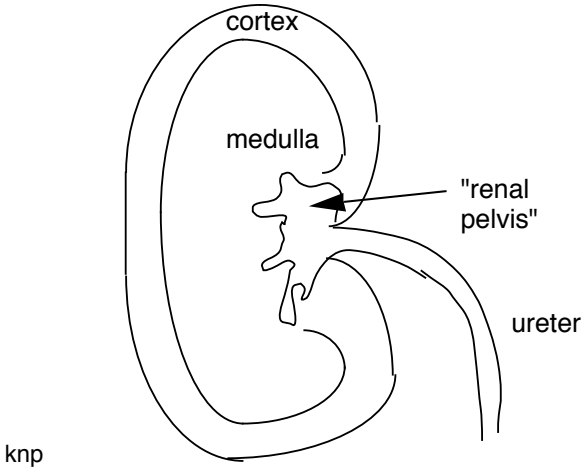
I. The Anatomy of the Mammalian Kidney

A. Gross anatomy:

1. The outer region of the kidney = the **CORTEX**
2. Inner region = the **MEDULLA**
3. The area where all of the urine collects is called the **PELVIS OF THE KIDNEY**

a. The urine leaves the kidney's pelvis via the URETERS

b. The pelvis is also where the blood supply enters and leaves the kidney via the **RENAL ARTERY AND RENAL VEIN**. Incidentally, about 25% of the cardiac output flows through these vessels.



B. Histology

1. The functional units of the kidney are called **NEPHRONS**. Of these, there are two types:

a. **CORTICAL (SUPERFICIAL) NEPHRONS**: The entire nephron is located in the outer region, the cortex. These function primarily in secretion and reabsorption of different substances.

b. **JUXTAMEDULLARY NEPHRONS**: these nephrons start in the Cortex and extend down into the medulla, in some cases to near the pelvis of the kidney. Besides being involved in secretion and reabsorption of materials, their unique role is in concentrating the urine.

c. Morphologically, the difference between cortical and juxtamedullary nephrons besides their size and location is the size of their **LOOPS OF HENLE** (see below) -- Cortical nephrons essentially lack a long loop of Henle.

d. The **Ratio of Cortical to Juxtamedullary nephrons** is an index of the ability of an animal to concentrate its urine. The more juxtamedullary nephrons, the more the urine can be concentrated. We will take this up in more detail in the next class.

2. Histology of a Juxtamedullary Nephron

a. **GLOMERULUS**: The **filter** of the nephron. Essentially, it consists of two parts:

1. A **capillary tuft** that is highly convoluted; it is surrounded by:

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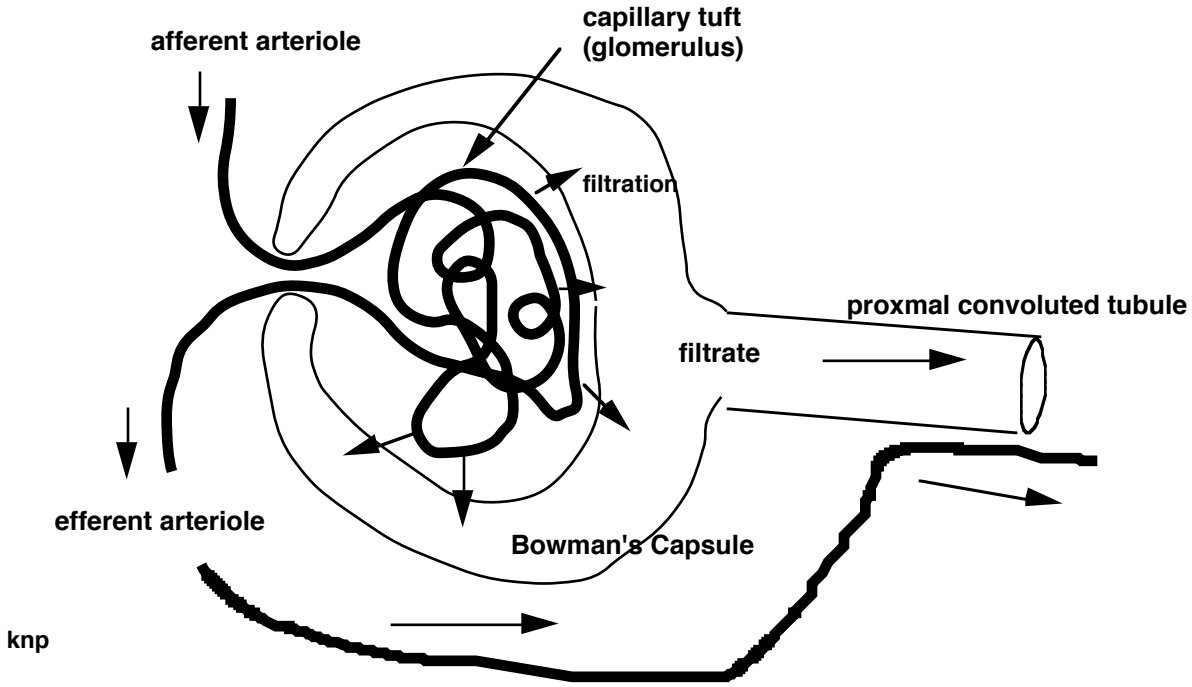
2. A cup-like layer of epithelial cells called **Bowman's Capsule**. Notice in the diagram below that the capsule can be seen as a hollow ball that has been pushed in with a fist where the fist is mostly surrounded by the ball. The fist represents the capillary tuft:



3. The action of the capsule will be reviewed later in detail, but suffice it to say that water and solutes are filtered through the capillary walls and the walls of the capsule and end up in the **CAVITY OF THE BOWMAN'S CAPSULE** as **Glomerular FILTRATE** or simply **FILTRATE**.

b. **PROXIMAL TUBULE**: This is an area where materials are passively and/or actively secreted and re-absorbed. Most of the water and solutes (67%) that were filtered are passively an actively reabsorbed here.

c. **LOOP OF HENLE:** This is very long in the juxtamedullary nephron. It has a very complex function that will be reviewed later. It consists of:

1. Two thin tubules that are linked together. The cells lining these tubules are very thin and lack large numbers of mitochondria; they are not involved in active transport.

a. The **DESCENDING THIN LIMB** which carries filtrate towards the pelvis of the kidney where it makes a **HAIRPIN TURN** and becomes the ascending thin limb (see below). No significant solute reabsorption occurs here; however this section of the loop of Henle is **permeable to water.** For reasons that will be explained later, about 17% of the filtered water is reabsorbed here.

b. the **ASCENDING THIN LIMB** which carries filtrate back towards the cortex and the thick ascending limb (see below). No significant transport occurs here. It is **water impermeable**.

3. **THICK ASCENDING LIMB:** an area where much active transport of solutes occurs. For instance, about 25% of the filtered Na^+ is reabsorbed here. This section is also **water impermeable** and as a result of the fact that water cannot move while solutes are pumped out, the osmolarity of the fluid leaving this section is extremely low (about 150 mOSM).

d. **DISTAL CONVOLUTED TUBULE (DCT)**: this section is the last region of the nephron and is primarily concerned with the regulation of Na^+, K^+ and H^+ via secretion and reabsorption. Its actions are under the control of the steroid hormone **ALDOSTERONE**. More about aldosterone in the next class.

Together with the collecting duct, about 12% of the filtered Na^+, Cl^- and 15% of the filtered water are reabsorbed in the DCT.

e. Not really part of the Nephron, but connected to it is the **COLLECTING DUCT**, a tube that takes filtrate from the distal convoluted tubule and moves it to the pelvis of the kidney. In the process, depending on the levels of the hormone **VASOPRESSIN** (also called **VASOTOCIN** and **ANTI-DIURETIC HORMONE** or **ADH**), water may be removed from the filtrate and the urine made more concentrated.
The endocrine system has an extensive role in regulating the secretion and reabsorption processes of the kidney. Below is a table that summarizes the hormones and their targets and effects. We will look at some of these in detail later:

Segment	Hormone	Effects
Prox. Conv. Tubule	Parathyroid Hormone Angiotensin	↓NaCl and water reabsorption ↑NaCl and water
Thick Ascending Loop	Aldosterone, Calcitonin, Glucagon, Vasopressin, Parathyroid Hormone	↑NaCl reabsorption
Distal Tubule and Collecting duct	Calcitonin Vasopressin Aldosterone Prostaglandins Bradykinin	↑NaCl reabsorption ↑Permeability to water and NaCl reabsorption ↑NaCl reabsorption ↓NaCl reabsorption ↓NaCl reabsorption

3. CIRCULATION:

a. Blood enters the kidney via the RENAL ARTERY

b. This eventually splits into a large number of small ARTERIOLES, many of which serve as the blood supplies of the glomeruli. Thus, their blood will be filtered.

c. The actual arteriole that enters the glomerulus is called the **AFFERENT ARTERIOLE**.

d. The blood then moves through the glomerular capillaries and is filtered.

e. The blood leaves the region of the Bowman's capsule via the **EFFERENT ARTERIOLES**

f. Most of this blood now enters the **PERITUBULAR CIRCULATION**, a group of vessels that surround the convoluted tubules and the loop of Henle and collect the materials

that come out and also act as the source for materials that will be secreted. They also supply oxygen and other substances and removes wastes from these tissues.

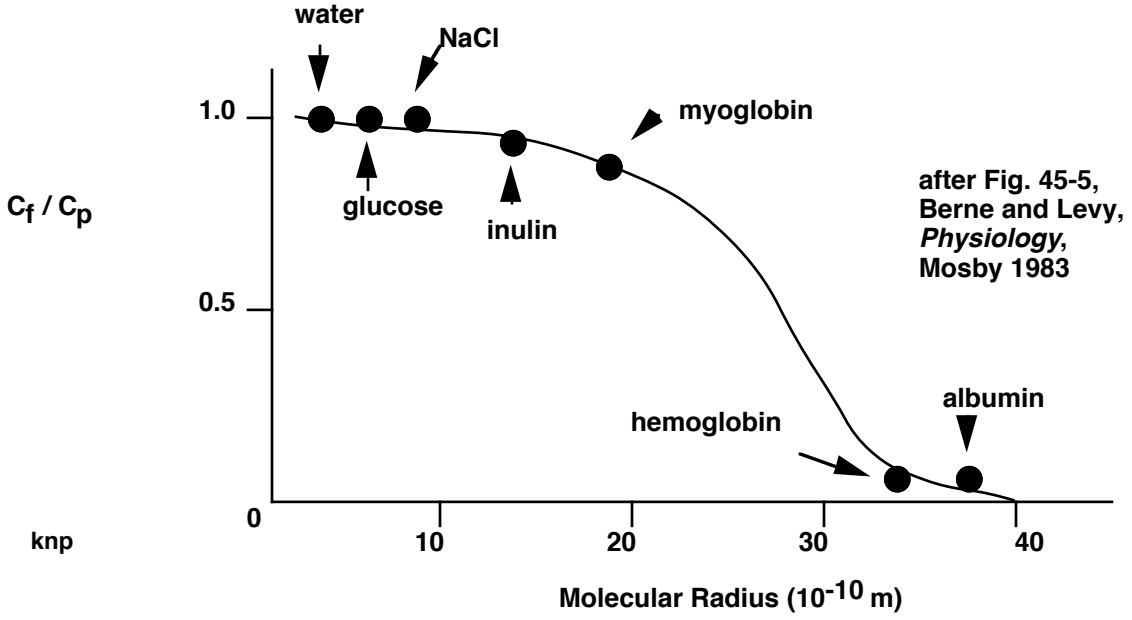
g. In juxtamedullary nephrons, about 0.7% of the blood circulates through a series of ascending and descending vessels called the **VASA RECTA.** These are associated with the loops of Henle, these vessels are arranged as a counter-current exchanger (see diagram on the next page). They perform an important task in helping to concentrate the urine. (This will be mentioned in more detail in the next lecture).

II. FORMATION OF THE GLOMERULAR FILTRATE:

A. Formation of the filtrate is a simple **physical process identical to the formation of interstitial fluid and lymph** that we discussed earlier with the circulation. The main difference is that the **pores in the filtering capillary (glomerulus) are a bit larger** than in regular capillaries and the "other side of the filter", t**he lumen of the Bowman's capsule, can be through of as having a relatively low hydrostatic pressure** as compared to the interstitial fluid. In any case, **as with the other capillaries, filtration is governed by: hydrostatic and colloidal osmotic pressure**s and by the **size of the filtered molecules** relative to the pore size in the filter and by the **charge** of the filtered particles.

Here **we will focus primarily on the filtration of water**; realize that other materials that are small enough to pass the membrane will come along for the ride. Most of this will be review, with the exceptions being a more explicit discussion of charged particle size and more on regulation of blood flow.

B. **The influence of particle size and charge on filterability** can be illustrated with an experiment where dextrans (harmless, non-metabolizable carbohydrates of different molecular weights) with various amounts of ionic groups attached are added to blood and their concentration in the Bowman's space is measured. Here is a table that summarizes the ability of materials to pass through the glomerular filter. The figure below shows the ratio of the concentrations of a given material that is in the filtrate (C_f) to that in the plasma of the tubule (C_p). Thus a ratio of C_f / C_p that is near unity implies that the substance is freely permeable, if it is less than one the material is only partially permeable or is impermeable ($C_f/C_p = 0$).



C. Assuming that a material is filterable, the rate that material passes through the filter, as we learned previously, is described by the **STARLING HYPOTHESIS**:

1. Filtration Rate = $k * (P_{GC} + \Pi_{BS}) - (P_{BS} + \Pi_{GC})$

where ***k*** is a fudge factor coefficient that describes how fast a fluid moves across the glomerulus at a given filtration pressure, $\boldsymbol{P_{GC}}$ is the hydrostatic pressure of the glomerular capillary, $\boldsymbol{P_{BS}}$ is the hydrostatic pressure in the Bowman's space (Bowman's capsule), $\boldsymbol{\Pi_{GC}}$ is the glomerular capillary colloidal osmotic pressure and $\boldsymbol{\Pi_{BS}}$ is the same measure for Bowman's space.

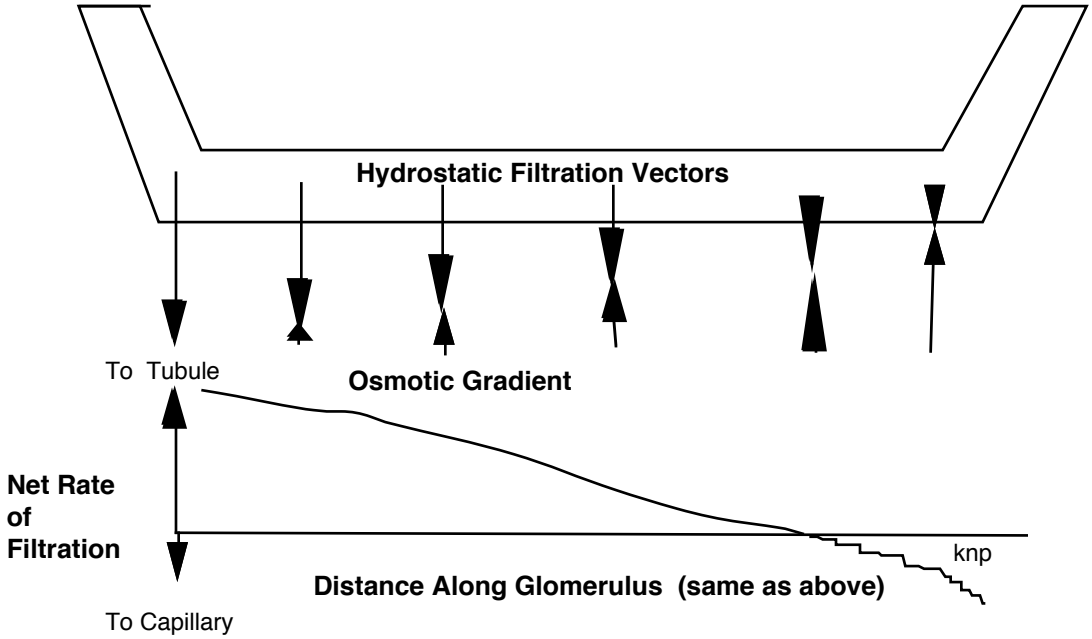
1. Let's look at this equation in detail.

IF EVERYTHING ELSE REMAINS CONSTANT:

2. if the capillary hydrostatic pressure (P_{GC}) increases or the tubular hydrostatic pressure decreases, there will be a greater pressure tending to drive materials across the membrane and the filtration rate will increase.

3. By contrast, a relatively higher Bowman's space osmotic pressure (Π_{BS}) will tend to encourage fluid to leave the capillary as will a relative lower tubule osmotic pressure.

4. We can envision the net forces of each of the hydrostatic and osmotic pressures as:



NOTE: By an exactly analogous mechanism, the blood forms lymph in the tissues of the body.

IN SUMMARY, REALIZE THAT THE GLOMERULUS-BOWMAN'S CAPSULE ACTS AS A SIMPLE PHYSICAL FILTER AND THAT FOR FREELY FILTERABLE MATERIALS, THE CONCENTRATION OF THE MATERIAL IN THE FILTRATE IS EQUAL TO THAT IN THE PLASMA.

D. AUTOREGULATION:

1. Most organs are capable of regulating their blood flow, independent of any external neural or hormonal controls. This process is referred to as **AUTOREGULATION**.

2. In the kidney, there are two mechanisms of autoregulation. They generally work together:

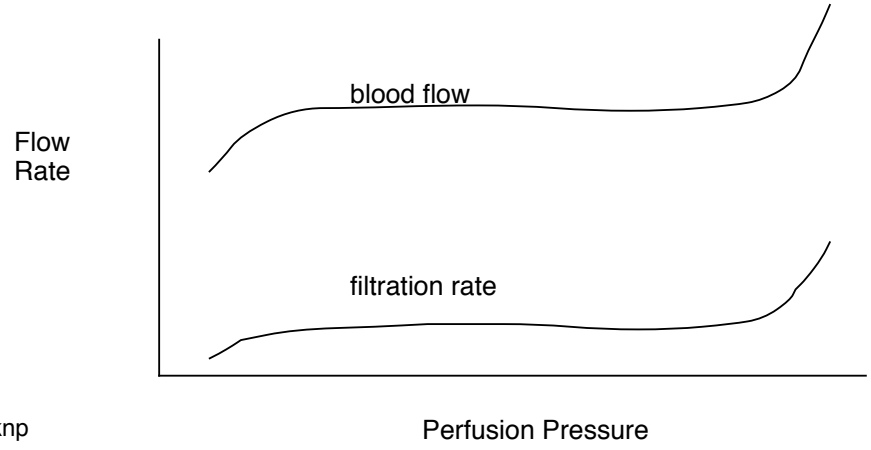
a. **Myogenic mechanism**: When arterial pressure rises (such as in the afferent arteriole), vascular smooth muscle contracts as it stretches. This increases resistance of the arteriole thereby lowering the pressure and flow through the glomerulus. As the renal blood flow (RBF) is lowered, the glomerular filtration rate (GFR) is also reduced. Thus high pressure will not result in proportionately high rates of urine production. Likewise, as pressure drops, arterioles will relax, *R* will decrease, *P* is reduced less and *Q* remains high. The rate of urine production remains high even while the blood pressure is somewhat reduced.

b. **TUBULOGLOMMERULAR FEEDBACK**: Flow of fluid (or some solute in the fluid) through the glomerulus is sensed by cells that make up the **JUXTAGLOMMERULAR APPARATUS**. Their exact identity(ies) are not known. Nor is the exact means by which they communicate with the smooth muscle cells of the afferent and efferent glomerular arterioles. It could be the renin-angiotensin system (these cells produce renin), bradykinin, catecholamines, or prostaglandins. Nevertheless the results are well known:

1. If the GFR drops, the efferent arteriole diameter decreases, this increases resistance and causes an increase in the glomerular capillary, thereby increasing the GFR.

2. By contrast, if the GFR gets too high, then efferent arteriole pressure decreases and filtration drops.

Here is a graph of the overall pattern of autoregulation:



? What things are controlled to achieve a relatively constant GFR? Why is a relatively constant GFR desirable?

III. TUBULAR SECRETION AND ABSORPTION MECHANISMS

A. A large number of substances are absorbed from the filtrate or secreted into it by the cells lining the **Proximal and Distal Convoluted Tubules**.

1. We will not consider (at this point) which substances are moved at what rates and where. We instead will be concerned with general mechanisms.

2. In general, all mechanisms of re-absorption and secretion will involve some sort of **protein mediator** and therefore all of these processes are potentially **SATURABLE**.

a. Therefore, we can characterize each transport protein in terms of its K_m and V_{max}.

Recall that:

b. K_m is largely a measure of affinity between the carrier protein and its substrate and is set by its particular structure and the physical-chemical environment.

c. The V_{max} is largely determined by the total number of transport proteins that are available (and also their individual kinetics). Obviously, the more transport molecules that are available, the higher the potential rate of transport.

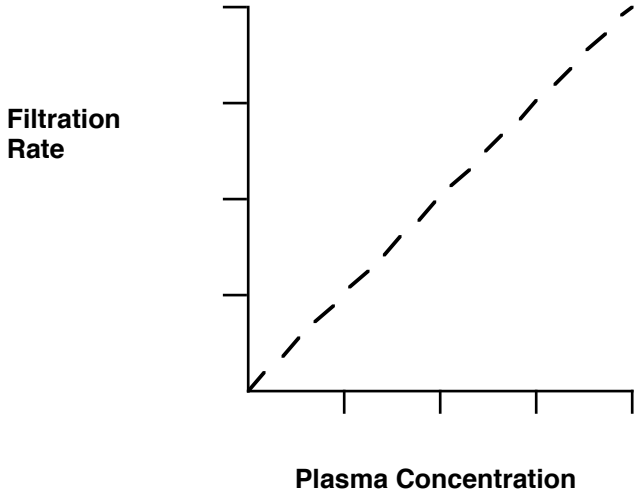
3. The actual carriers themselves can be divided into different classes, largely according to the type of materials they transport and the mechanism they use.

B. PATTERNS OF SECRETION AND RE-ABSORPTION

1. The most basic graphical treatment of secretion and reabsorption is done by plotting the **EXCRETORY RATE** (usually in amount of material/time) vs. the **PLASMA CONCENTRATION** (in amount/vol.). There are several other types of useful graphs; we will discuss each at appropriate times.

2. <u>GRAPHS OF DIFFERENT PROCESSES</u>:

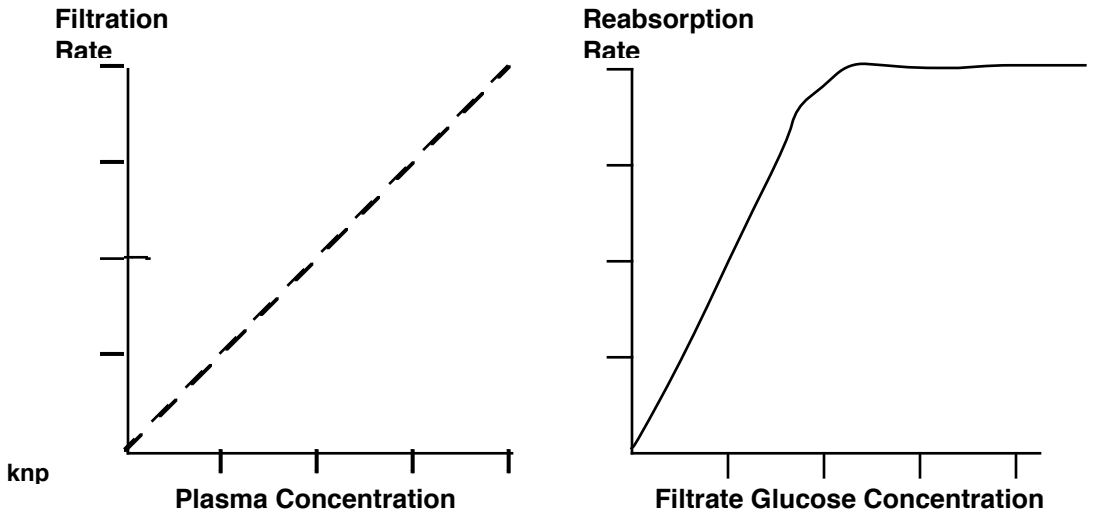
a. **FILTRATION ONLY**: If a material is only filtered, there will be a direct relationship between the plasma concentration of the substance and the amount removed (also referred to as the amount **CLEARED**):



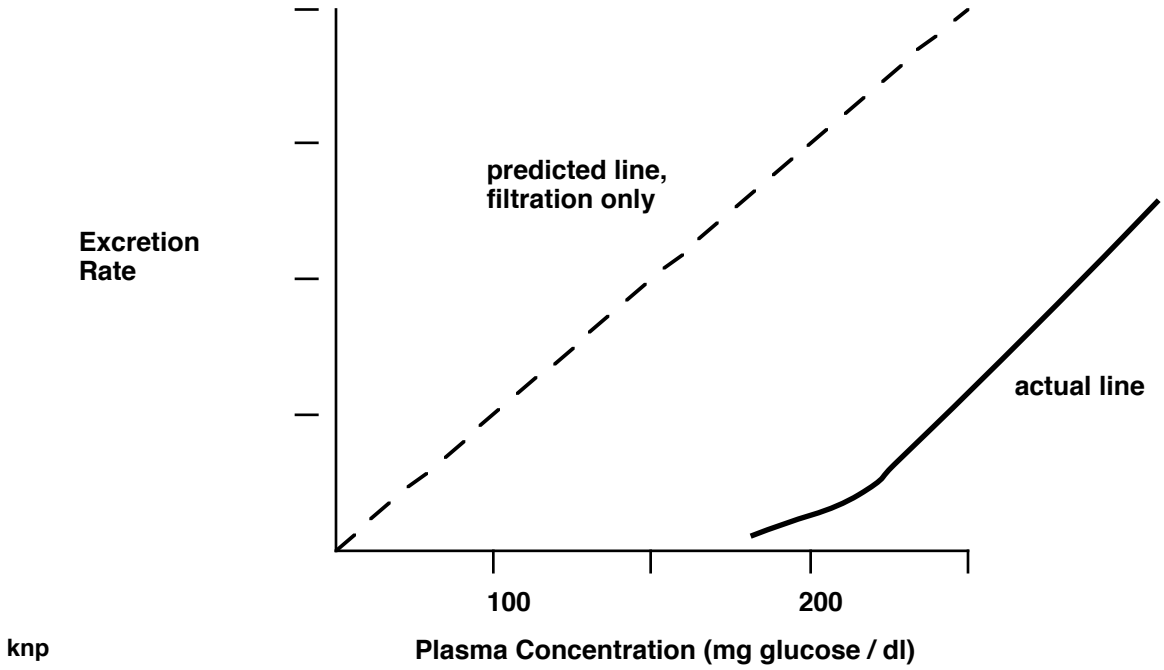
b. What will happen if a substance is being reabsorbed by the tubules? An example would be glucose.

1. The substance (we'll talk about glucose) is filtered in a way that would give a curve identical to the one shown above.

2. When it enters the proximal convoluted tubule, there are many glucose transport proteins located on the inner wall of the tubules that transport glucose out of the tubule. Thus, we have two processes that are taking place in the body: one in glomerulus that results in glucose getting in the filtrate in proportion to its plasma concentration and the other that transports glucose out of the tubule back into the blood according to the K_m and V_{max} of the transport mechanism:



3. The summed process is shown below. The removal of glucose results in a decreased [glucose] in the filtrate.



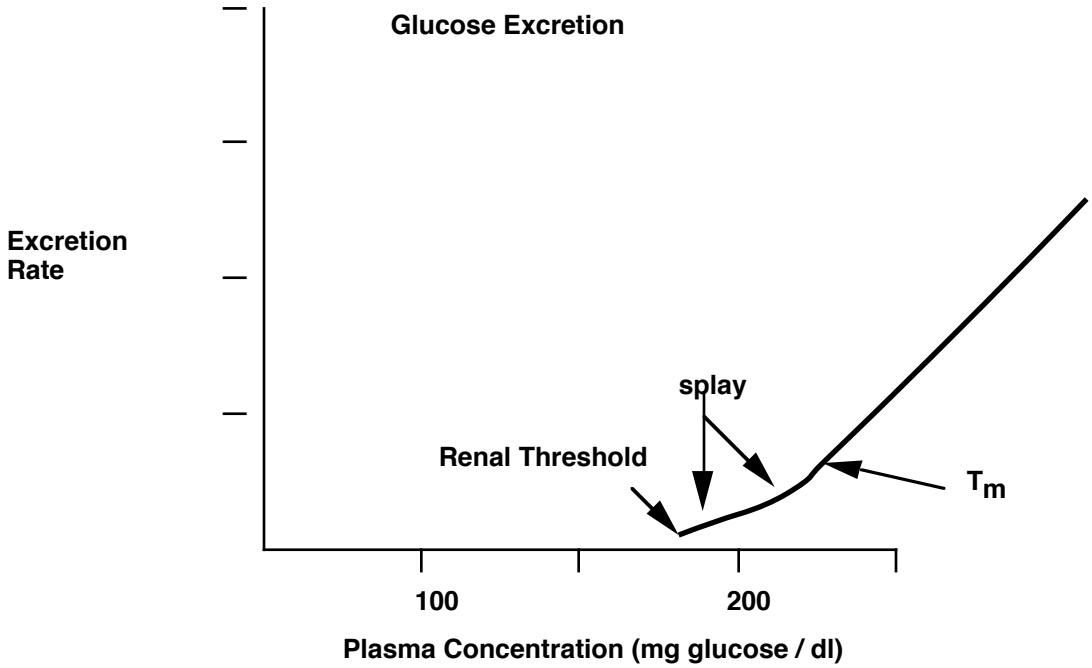
a. The removal mechanism is so effective that at low plasma [glucose] (< ~180 mg/dl), no glucose is actually excreted even though there is abundant glucose in the filtrate.

b. Eventually a concentration is reached where the amount of glucose in the tubules is so high that it overwhelms the transport mechanisms.

1. The point where this first starts to happen is called the **RENAL THRESHOLD** and the concentration where V_{max} is reached is called the **TUBULAR MAXIMUM** or $\boldsymbol{T_m}$.

2. Above the T_m, the rate of excretion changes the same as the rate of filtration: however there is a difference between the filtration and actual filtrate concentration that equals the amount cleared by filtration - the amount reabsorbed.

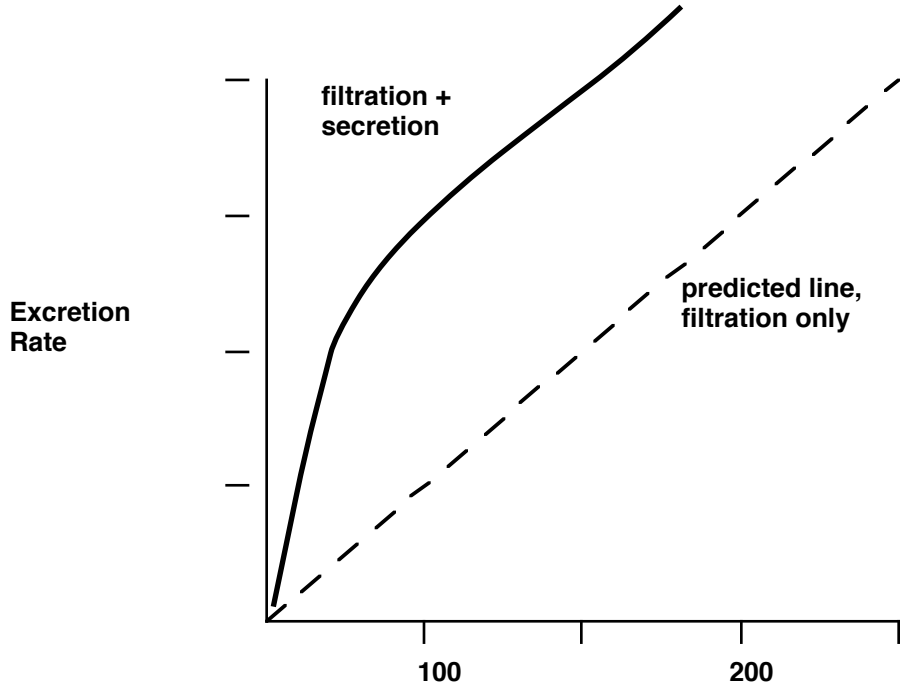
3. In the area between the T_m and the renal threshold, the concentration of the material in the filtrate changes at a different rate than after the T_m. This is called **SPLAY**. The reason for the splay is obvious from an inspection of the re-absorption graph shown previously.



? If the K_m of the transporter is greatly increased what would it mean in terms of glucose remaining in the filtrate at various concentrations? What if the total number of transporter proteins increased? What would happen to V_{max} and the glucose that was being excreted? Draw graphs to explain your answer.

c. What happens when a substance is actively being **SECRETED** from the blood into the filtrate?

If the substance is also being filtered, the process is identical to that of reabsorption shown above EXCEPT that now the TRANSPORT CURVE IS ADDED TO THE FILTRATION CURVE:



? If the substance is not filtered and is only secreted, what would the excretion curve (amount cleared/min vs. plasma conc.) look like? What is the basis for your answer?

IV. THE CONCEPT OF CLEARANCE:

A. Clearance can be thought of as the removal of a substance from the blood (and the body). Let's first consider the possible things that can happen to some substance (glucose for example) when it arrives at the kidney.

1. There is **one avenue of entry** -- the plasma of the renal artery and therefore the amount that enters will be the product of the **renal arterial plasma flow, *RPF*$^{\mathbf{a}}$** and the **renal arterial plasma concentration of the substance (let's call it substance x),** $P_{\mathbf{x}}^{\mathbf{a}}$. Thus:

1. Rate Substance arrives at kidney = $P_x^a * RPF^a$

2. There are two possible avenues of exit from the kidney, the r**enal vein and/or the ureter.**

a. **Exit through the renal vein** can be calculated in a manner similar to that which enters via the renal artery, it will be the product of the concentration of x and the renal vein plasma flow:

2. Rate substance leaves via the renal veins = $P_x^v * RPF^v$

b. **Exit via the urine into the ureter** will be calculated in a similar manner, in this case the product of the rate of urine flow, $\boldsymbol{V}$[1], and the concentration of the substance in the urine $\boldsymbol{U_X}$.

3. RATE EXCRETED = $U_X * V$

Thus:

4. $$P_x^a * RPF^a = P_x^v * RPF^v + (U_x * V)$$

This represents a complete description of the handling of any substance by the kidney.

B. Renal physiologists use the notion of clearance to mean totally removing something from the blood. If something is completely cleared, then all of it leaves the kidney via the ureter and none via the renal veins; $\boldsymbol{RPF^v}$ = 0. Rearranging eq. 4 and making this substitution, we find

$$\begin{aligned} U_x * V &= P_x^a * RPF^a - P_x^v * RPF^v \\ &= P_x^a * RPF^a - 0 * RPF^v \end{aligned}$$

5. $$U_x * V = P_x^a * RPF^a$$

this equation expresses the equality between the amount of substance entering the kidneys in the renal artery plasma and that leaving in the urine.

If we now define the **clearance rate** as the v**olume of renal artery plasma per time** needed to account for all of the substance found in the urine, then, rearranging equation #5 we find:

5. $$Clearance\ x = RPF^a = \frac{U_x * V}{P_x^a}$$

where clearance has the same units as the renal plasma flow (usually ml*min^{-1} in human physiology).

? As you know, in normal individuals, no glucose is lost in the urine. Although quite a bit goes through the glomerular filter, 100% is reclaimed by transport in the proximal convoluted tubule and therefore $\boldsymbol{RPF^a}$ = $\boldsymbol{RPF^v}$ and $\boldsymbol{U_x}$= 0. What is the clearance of glucose in ml/min? ANS: in a normal individual it is zero – none of the glucose is removed from the body by the kidney. We will further explore this concept in the next set of notes.

[1] This should really be symbolized as $\dot{V}$ since it is a rate, but for reasons unknown to be renal physiologists do not use this notion. So in renal phys., $\boldsymbol{V}$ is a volume per time!