

<b>Glomerular Filtration</b>
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PA Renal, Winter 2010  
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Suggested Reading: There is no assigned reading other than this handout. However, a good reference text if you would like to buy one is Koeppen and Stanton's *Renal Physiology*, 4<sup>th</sup> edition, Mosby Elsevier, 2007, For this particular lecture (glomerular filtration), Chapter 3 is the reference chapter to read in this text.

### Learning Objectives

1. After reading Chapter 36 in Berne and Levy, my MS Word handout, and coming to class and listening to me go over my Powerpoints, you should be knowledgeable about the following:
2. Basic structure of the two common types of nephrons
3. Basic structure of the renal corpuscle
4. How is the kidney supplied with blood, particularly the nephrons?
5. What exactly is the glomerular filtration rate (GFR)? What does it encompass?
6. What is the ultrafiltrate, and how is it formed?
7. What is the basic structure of the glomerular capillaries that are packed within Bowman's capsule, and how do these glomerular capillary structures restrict filtration of proteins into Bowman's space?
8. How is the GFR measured clinically?
9. How is chronic renal insufficiency (CRI), chronic renal failure (CRF), and end stage renal disease (ESRD) defined in terms of the GFR?
10. Why is a plasma creatinine measurement by itself not useful clinically in detecting early stage renal disease?
11. How do Starling's forces apply to glomerular filtration, and how do glomerular capillaries differ from other capillaries?
12. What is tubuloglomerular feedback (TGF), and how does it help control the GFR?
13. How does vasoconstriction or vasodilation of either the afferent or efferent arteriole affect the GFR, and why?

### Overview

The term 'glomerular filtration rate' or 'GFR' is commonly used in medical practice. Many first year medical students have likely heard the term in relation to patients with known acute or chronic kidney disease, but really do not understand what the GFR encompasses, or why we bother to measure or estimate it.

As we will review in this lecture, the GFR is the sum of the rate of glomerular filtration of all the nephrons total, in both kidneys (or a single kidney, if all the patient

has is one). The GFR remains the most reliable indicator as to whether or not renal disease is present, and for this reason it is of great interest to physicians when they have patients whom they suspect have renal disease. However, it is difficult to measure as you will see, but both direct and indirect estimates of the GFR are available.

### *Types and Prevalence of Kidney Disease*

Kidney disease (or renal failure) falls into two broad categories: acute and chronic. Acute renal failure (ARF; also referred to as acute renal injury or 'ARI' in the medical literature) is defined as an abrupt or rapid decline in GFR. This condition is usually marked by a rise in serum creatinine concentration or azotemia (a rise in blood urea nitrogen [BUN] concentration), which occurs because the kidneys are no longer able to filter these compounds out of the body at a normal rate. AKI may occur in 3 clinical patterns, including the following: (1) as an adaptive response to severe volume depletion and hypotension, with structurally intact nephrons; (2) in response to cytotoxic, ischemic, or inflammatory insults to the kidney, with structural and functional damage; and (3) with obstruction to the passage of urine. Therefore, in general terms, AKI may be classified as prerenal, intrinsic, and postrenal.

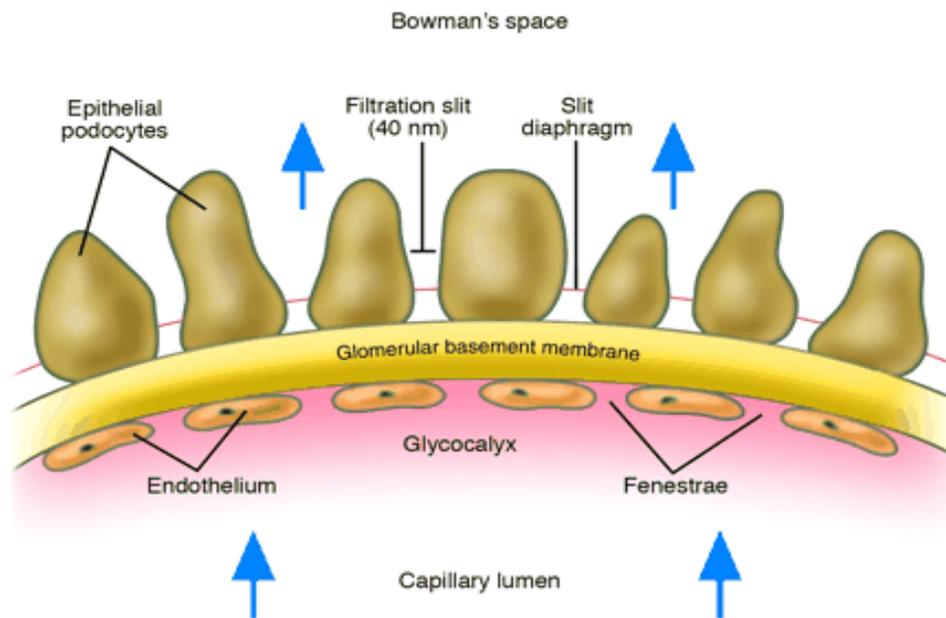
Conversely, chronic renal disease (CRD) is a progressive loss of renal function over a period of months or years. CRD is defined by the presence of a GFR that is 60 ml/min/1.73m<sup>2</sup> or lower for 3 or more months, regardless of the cause. CRD is further broken down into the following classifications:

- Chronic renal insufficiency (CRI) is the preferred term for patients with mild-to-moderate renal impairment, whose GFR falls in the range of 30-60 ml/min/1.73m<sup>2</sup>.
- Chronic renal failure (CRF) is usually reserved to describe patients whose GFR is between 15 and 30 ml/min/1.73m<sup>2</sup>.
- End-stage renal disease (ESRD), usually associated with signs and symptoms of uremia, is the term reserved for patients whose GFR has declined to levels of less than 10 to 15 ml/min/1.73m<sup>2</sup>.

Chronic kidney disease is common in the U.S., and appears to be on the rise worldwide. In the year 2000, in which we have the most reliable available estimates, early 400,000 people had to be dialyzed or transplanted due to ESRD. That number is expected to rise to 2 million by the year 2030. These data also estimated that in 2000, approximately 20 million adults in the U.S. had CRI or CRF (11.7% of the population). The most common risk factors for developing chronic renal disease are **diabetes, chronic hypertension, cardiovascular disease, family history of chronic renal disease, and age greater than 60**. Since estimating the GFR (along with other tests, such as measuring protein in the urine) can find chronic renal disease in its earlier stages when it is more amenable to therapies, teaching medical students about the GFR is important.

### Ultrafiltration

The first step in the formation of urine is the production of an *ultrafiltrate* of the plasma by the glomerular capillaries. This plasma ultrafiltrate collects in Bowman's space before entering the beginning of the proximal tubule. As with other capillaries in the body, ultrafiltration is driven by **Starling forces** across the glomerular capillaries, and changes in these forces will alter the GFR (more on this later). The glomerular capillary wall is a living ultrafiltration membrane. It permits water and small solutes to pass readily into Bowman's space, while normally rejecting albumin and other large proteins with great efficiency. As shown in the figure below (J. Clin. Invest. 114:1412-1414, 2004), the glomerular capillary wall consists of a fenestrated endothelium, the glomerular basement membrane (GBM), and the interdigitated foot processes of epithelial cells (podocytes). The filtration pathway is extracellular; that is, water and filtered solutes pass through the fenestrae, across the GBM, and through filtration slits bounded by the foot processes. The filtration slits are spanned by porous slit diaphragms:



The approximate diameter of the fenestra in the capillary endothelium is 10-30 nm, the GBM thickness is approximately 200–400 nm, and the filtration slits are about 40 nm across. The glycocalyx thickness is uncertain. The capillary endothelium, the basement membrane, and the foot processes together form what is called the **filtration barrier**. All substances filtering out of the glomerular capillaries and into Bowman's space must filter across these 3 structures.

### Capillary Exclusion of Molecules Based on both Molecular Size and Electrical Charge

The filtration of neutral molecules with a molecular radius of less than 20Å ( $1 \text{ \AA} = 10^{-10} \text{ m}$ ) are not inhibited to any extent by the filtration barrier. Water, individual electrolytes, and glucose for example are all freely filtered into Bowman's space. White cells, red cells, and

platelets, however, are not normally filtered into Bowman's space due to their size. Neutral molecules with a molecular radius of 20Å to 40Å are filtered to varying degrees, and neutral molecules with a molecular radius greater than 40Å are generally not filtered at all, due to their size.

The filtration barrier does not discriminate on the basis of size alone. The glycocalyx, is composed of glycoproteins including proteoglycans, and coats the luminal surface of the glomerular capillaries. These polyanionic glycoproteins repel large anions (i.e. proteins with a net negative charge like albumin) without significantly affecting smaller anions such as chloride or bicarbonate. In addition, negative charges are present in the proteins that compose the other components of the filtration barrier, i.e. the basement membrane, as well as the foot process of the podocytes. Most plasma proteins have negative charges associated with them. Thus, plasma proteins with a molecular radius between 20Å to 40Å are excluded from filtration based on both their size and their net negative charge (remember, similar charges repel each other). In fact, cationic and neutral molecules with a molecular radius between 20Å to 40Å will always be filtered to a greater extent than molecules of the same size with a net negative charge.

Albumin has a molecular radius of around 35Å, and as noted has a net negative charge. Normal average urinary excretion of albumin in men and women is around 7 mg/24 h. If the fixed negative charges on the filtration barrier are lost, as in some forms of glomerular disease (e.g. minimal change nephropathy), the filtration of albumin becomes inhibited only by size and not charge, and the glomerular filtration of albumin increase significantly from its normal value, causing significant **proteinuria**.

### The Emerging Role of the Slit Diaphragm

The strategic location of the slit diaphragm has long suggested that it might play a crucial role in restricting the passage of solutes on the basis of *molecular size*. Thus, disruption of slit diaphragms might also in part underlie the proteinuria that is one of the hallmarks of chronic renal disease.

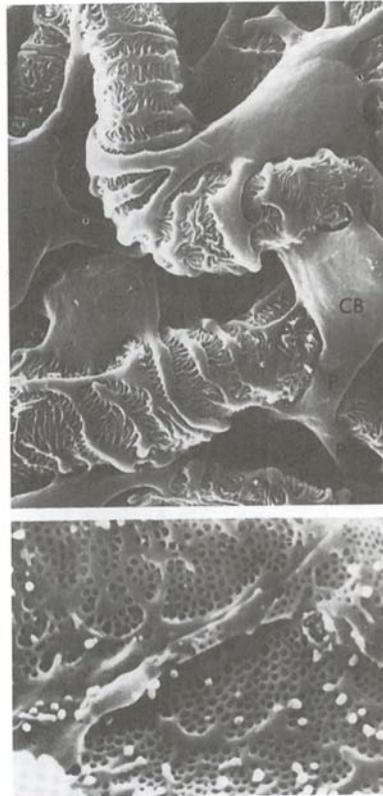
In fact, a large amount of research has been invested into the structure of the slit diaphragm. That's because the rates of chronic kidney disease are increasing in many countries, most likely due to the concomitant increases in global Type II diabetes. In almost all cases of renal failure that occurs secondary to Type II diabetes, the initial finding is an increase in urinary albumin excretion, suggesting a breakdown of the protein-excluding structures of the glomerular capillaries (including loss of negative charges in structures within the filtration barrier, as noted above). Studies of the glomerular capillaries have subsequently found that certain families of proteins that are integral parts of the slit diaphragm – specifically, the nephrins and podocins – are also in fact adversely affected by Type II diabetes (Aaltonen and Holthöfer. 2007 The nephrin-based slit diaphragm: new insight into the signaling platform identifies targets for therapy. *Nephrology Dialysis Transplantation*. 22(12):3408-3410).

### Composition of the Ultrafiltrate

Because the glomerular capillaries are less permeable to proteins than other capillaries, there is a lower oncotic pressure in Bowman's space than in the interstitial spaces in the ECF.

The end result is that the ultrafiltrate which forms in Bowman's space has *almost exactly the same composition as the plasma that is filtered*, with the exception of less protein (remember, cells are not normally filtered). The following figure is an electron micrograph of the glomerular capillaries - the top view as they would be seen from Bowman's space, and the bottom view as they would be seen from the lumen of the glomerular capillary:

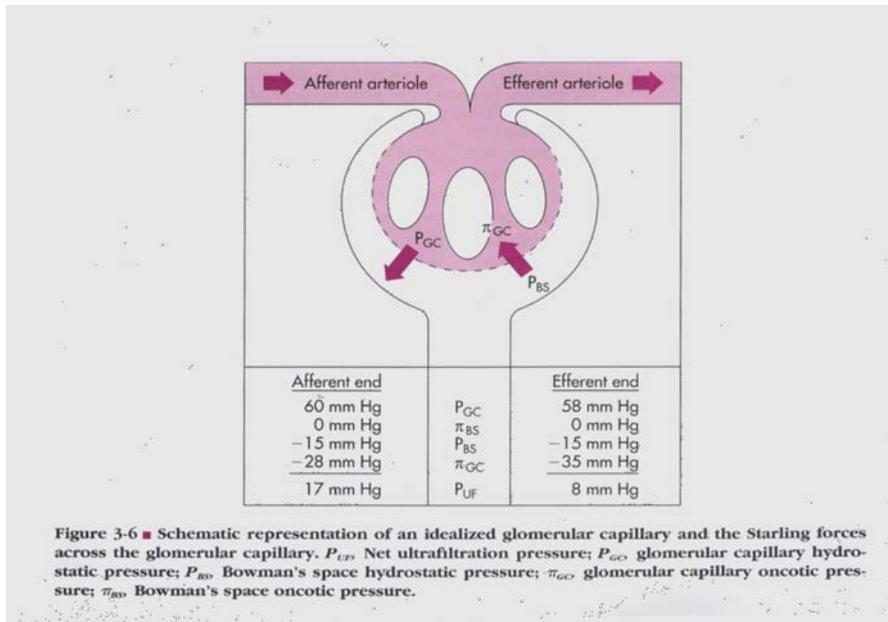
Figure 2-7 ■ A, Scanning electron micrograph showing the outer surface of glomerular capillaries. This is the view that would be seen from Bowman's space. Processes (P) of podocytes run from the cell body (CB) toward the capillaries, where they ultimately split into foot processes. Interdigitation of the foot processes creates the filtration slits. (Magnification ~ × 2500.) B, Scanning electron micrograph of the inner surface (blood side) of a glomerular capillary. This is the view that would be seen from the lumen of the capillary. The fenestrations of the endothelial cells are seen as small 700 Å holes. (Magnification ~ × 12000.) (Electron micrographs courtesy of Kriz W, Kaissling B: Structural organization of the mammalian kidney. In Seldin DW and Gebisch G, editors: *The kidney: physiology and pathophysiology*, ed 2, New York, 1992, Raven.)



### Dynamics of Ultrafiltration

As noted above, the forces involved in ultrafiltration across glomerular capillaries are similar to the forces involved with fluid exchange across non-glomerular capillaries. The figure below shows the sum of these forces across the glomerular capillaries.

Ultrafiltration occurs because Starling forces drive fluid from the lumen of glomerular capillaries across the filtration barrier, and into Bowman's space:



The hydrostatic pressure in the glomerular capillaries ( $P_{GC}$ ) is oriented to promote movement of fluid from inside the capillary into Bowman's space. Because the glomerular filtrate is mostly protein-free, the oncotic pressure in Bowman's space ( $\pi_{BS}$ ) is essentially zero. Therefore, **the  $P_{GC}$  is the only force that favors filtration into Bowman's space**, and is opposed by the hydrostatic pressure in Bowman's space ( $P_{BS}$ ), as well as the glomerular capillary oncotic pressure ( $\pi_{GC}$ ).

Please note in the above figure that numbers listed with a negative (-) sign in front of them are not negative numbers, but are pressures which oppose ultrafiltration into Bowman's space. Note also that **there is a positive ultrafiltration pressure ( $P_{UF}$ ) of 17 mm Hg** at the afferent end of the glomerular capillaries, whereas at the efferent end, the  $P_{UF}$  is a positive **8 mm Hg**. Thus, the following equation describes the net ultrafiltration pressure:

$$P_{uf} = (P_{GC} + \pi_{BS}) - (P_{BS} + \pi_{GC})$$

A major reason for the decrease in the net ultrafiltration pressure at the efferent end of the glomerular capillary is that the  $\pi_{GC}$  increases from 28 to 35 mm Hg along the length of the capillary (remember, this force opposes ultrafiltration). This is due to the fact that as ultrafiltration progresses along the capillary, lots of water and small solutes are being filtered into Bowman's space from the capillary, but little or no protein is filtered out. Therefore, the protein left in the plasma within the glomerular capillary when it reaches the efferent end of the capillary is now more concentrated (due to the loss of water), and therefore exerts a greater  $\pi_{GC}$ .

It is essentially the arterial blood pressure that forms the  $P_{GC}$  (in other words, the  $P_{GC}$  generated in the blood vessels and capillaries is mostly due to the force of the heart contracting and pushing blood through the vessels). It should be noted, however, that the arterial side of capillaries in the periphery have a "precapillary sphincter" which allow them to autoregulate the  $P_{GC}$ , in response to pressure/flow changes from the arterial side, but not the venous side. The

precapillary sphincter, however, *is not present on glomerular capillaries*. However, glomerular capillaries do have several intrinsic mechanisms for regulating  $P_{GC}$ , as you will see later.

### Surface Area and Intrinsic Permeability of Glomerular Capillaries

In addition to Starlings Forces, the GFR is affected by the **surface area and intrinsic permeability** of the glomerular capillaries. The following equation accounts for these factors in determining what the GFR will be:

$$GFR = K_f [(P_{GC} + \pi_{BS}) - (P_{BS} + \pi_{GC})]$$

Where  $K_f$  is equal to the product of the intrinsic permeability of the glomerular capillaries, as well as their surface area that is available for filtration. Glomerular capillaries filter much more plasma in a day than other capillaries (up to 180 L daily, compared to 2 or 3 L daily for non-glomerular capillaries), because they have a *very high* intrinsic permeability and a large surface area for filtration. In addition, the  $P_{GC}$  in glomerular capillaries is nearly *twice* that of non-glomerular capillaries (60 mm Hg vs around 35 mm Hg).

**IMPORTANT CONCEPT:** The GFR can be altered by changes in the Starling Forces, and / or by changes in intrinsic permeability or surface area of the glomerular capillaries. This is essential to understand, because in healthy individuals, the GFR is regulated primarily by adjusting the Starling Forces (usually the  $P_{GC}$ ). However, in many renal diseases (particularly **GLOMERULAR DISEASES**), the decrease in the GFR that is seen is due to a decrease in the intrinsic permeability and available surface area for filtration, and not due to a change in  $P_{GC}$ . Eventually, as the disease progresses and nephrons die, there are simply not enough nephrons left to generate a GFR that is adequate to maintain health.

### Factors Regulating Starling Forces and thus the GFR

As noted above, it is the  $P_{GC}$  that is primary driving force behind the glomerular filtration rate in healthy individuals – increases in the  $P_{GC}$  will generally increase GFR, and decreases in  $P_{GC}$  will usually decrease the GFR. As also noted above, it is the arterial blood pressure that is primarily responsible for dictating what the  $P_{GC}$  will be. The figure below graphically illustrates that, in the healthy kidney, *plasma flow rate into the glomerulus is an important determinant of GFR*. This is because plasma flow rate effects the Starling forces which determine the  $P_{UF}$ , which in turn affects the GFR:



The figure above illustrates some important concepts:

- **Constriction** of the **afferent arterioles** for some reason (with no change in efferent arterioles) would result in a **decrease** in plasma flow into the glomerular capillaries, and thus would decrease the  $P_{GC}$ , which in turn would decrease the GFR and overall renal blood flow. A systemic decrease in arterial blood pressure would have the same effect, because it would also decrease the amount of blood perfusing the glomerular capillaries, and thus lower the  $P_{GC}$  and therefore lower the GFR. A decrease in arterial pressure would also result in less blood overall perfusing the kidney so it would decrease overall renal blood flow as well.

- **Constriction** of the **efferent arterioles** for some reason (with no change in afferent arterioles) would result in the plasma that was flowing into the glomerular capillaries through the afferent arterioles remaining in the glomerular capillaries LONGER, and thus exerting greater hydrostatic pressure within the glomerular capillaries. This increase in the  $P_{GC}$  would subsequently increase the GFR. Overall renal blood flow would be decreased in this scenerio, however.

- **Dilation** of the **efferent arterioles** for some reason (with no change in afferent arterioles) would result in the plasma that was flowing into the glomerular capillaries through the afferent arterioles exiting the glomerular capillaries SOONER, and thus this plasma would end up exerting less overall hydrostatic pressure within the walls of the glomerular capillaries. This decrease in the  $P_{GC}$  would in turn decrease the GFR. However, this dilation of the efferent arterioles would increase the overall renal blood flow.

- **Dilation** of the **afferent arterioles** for some reason (with no change in efferent arterioles) would result in an **increase** in plasma flow into the glomerular capillaries, and thus would increase the  $P_{GC}$ , which in turn would increase the GFR and overall renal blood flow. A systemic increase in arterial blood pressure would have the same effect, because it would also increase the amount of blood perfusing the glomerular capillaries, and thus raise the  $P_{GC}$  and therefore increase the GFR. An increase in arterial pressure would also result in more blood overall perfusing the kidney so it would increase overall renal blood flow as well.

### Regulating the GFR

Physiologically, we are slightly altering our GFR all the time. Sometimes we need to increase it a little to increase the excretion rate of certain compounds. For example, in normal individuals, the body wants to excrete more Na<sup>+</sup> molecules into the urine when we become hypertensive. To do this, we first increase the **filtered load** of sodium, which simply means we filter more of it into Bowman's space, so it has the opportunity to ultimately be excreted into the urine. The filtered load for any substance in the plasma can be estimated by the following:

$$\text{Filtered load} = \text{GFR} \times [P_x]$$

Where [P<sub>x</sub>] is simply the 'free' (not protein bound; protein bound substances cannot readily filter across the glomerular capillaries) plasma concentration of the substance you want to filter out. Thus, if the plasma concentration of sodium was 140 mMol/L (and sodium is not bound to protein), and the GFR was known to be 100 ml/min, the filtered load of sodium would be:

$$100 \text{ ml/min} \times .140 \text{ mMol/ml Na}^+ = \mathbf{14 \text{ mMol / min}}$$
 (I converted mMol/L to mMol/ml here)

Therefore, in this example, the filtered load of Na<sup>+</sup> would be 14 mMol/min (ie, every minute, 14 mMol of Na<sup>+</sup> total is being filtered into Bowman's space by the glomerular capillaries). If I increased the GFR to 120 ml/min (say, by dilating the afferent arterioles), the new filtered load of Na<sup>+</sup> would be:

$$120 \text{ ml/min} \times .140 \text{ mMol/ml Na}^+ = \mathbf{17 \text{ mMol / min}}$$

This 3 mMol / min increase in the Na<sup>+</sup> filtered load may not seem like much, but there are 1440 minutes in a 24 hour period. Thus, if an individual raised their GFR from 100 ml/min to 120 ml/min for 24 hr, the difference in the filtered load of Na<sup>+</sup> in this case would be:

$$1440 \text{ min} \times 3 \text{ mMol / min} = \mathbf{4,320 \text{ mMol increase in filtered Na}^+ \text{ over 24 hr period.}}$$

There are about 14 liters of extracellular fluid (ECF) in a healthy, 70 kg individual. On average, each liter of ECF contains about 140 mMol of Na<sup>+</sup>. Therefore, the TOTAL amount of Na<sup>+</sup> in the ECF of the average 70 kg person is around 1960 mMol Na<sup>+</sup> (14 x 140 = 1960). Therefore, you can see that this **4,320 mMol increase** in the filtered load of Na<sup>+</sup> that was accomplished by increasing the GFR about 20 ml/min over a 24 hr period represents an amount of Na<sup>+</sup> that is equal to more than **twice the total amount of Na<sup>+</sup> that is present in the ECF!**

The **take home lesson** from this example is that you do not have to increase – or decrease – the GFR for very much, or for very long, to make a large difference in the filtered load of non-protein bound substances that are in the plasma.

### **Autoregulation of the GFR**

There are several hormones which can either constrict or dilate the afferent or efferent arterioles, and therefore affect the GFR by the mechanisms shown above. In addition, activation of the sympathetic nervous system can constrict the afferent arterioles (it has essentially no effect on the efferent side) and thereby decrease the  $P_{GC}$  and subsequently decrease the GFR. Decreased sympathetic input to the glomeruli allows for dilation of the afferent arterioles, and an increase in  $P_{GC}$  and subsequently an increase in the GFR (there is little if any parasympathetic effect on the glomerular capillaries).

Most of the hormones and changes in autonomic nervous system function that affect GFR are in response to changes in arterial pressure. This is important, as otherwise changes in arterial pressure could be directly translated to changes in GFR. It is important to understand that our kidneys try to tightly regulate the GFR, so that it does not change too far in either direction. Otherwise, as you saw above, the filtered loads of non- protein bound substances in the plasma can change dramatically in one direction or the other, and total renal blood flow can change significantly. In other words, without these controls, a person with chronic hypertension would walk around with a chronically increased  $P_{GC}$  and therefore a chronically increased GFR, as well as increased renal blood flow. A person who hemorrhaged significantly and had a dramatic decrease in blood pressure could have a dramatic drop in  $P_{GC}$  and therefore a drop in GFR and overall renal blood flow, if some mechanisms were not in place to try and keep the GFR in the normal range.

So what are these mechanisms that the body uses to keep the GFR from fluctuating too much in one direction or another, especially in response to changes in arterial pressure? There are two that have been studied the most: the myogenic mechanism, and tubuloglomerular feedback.

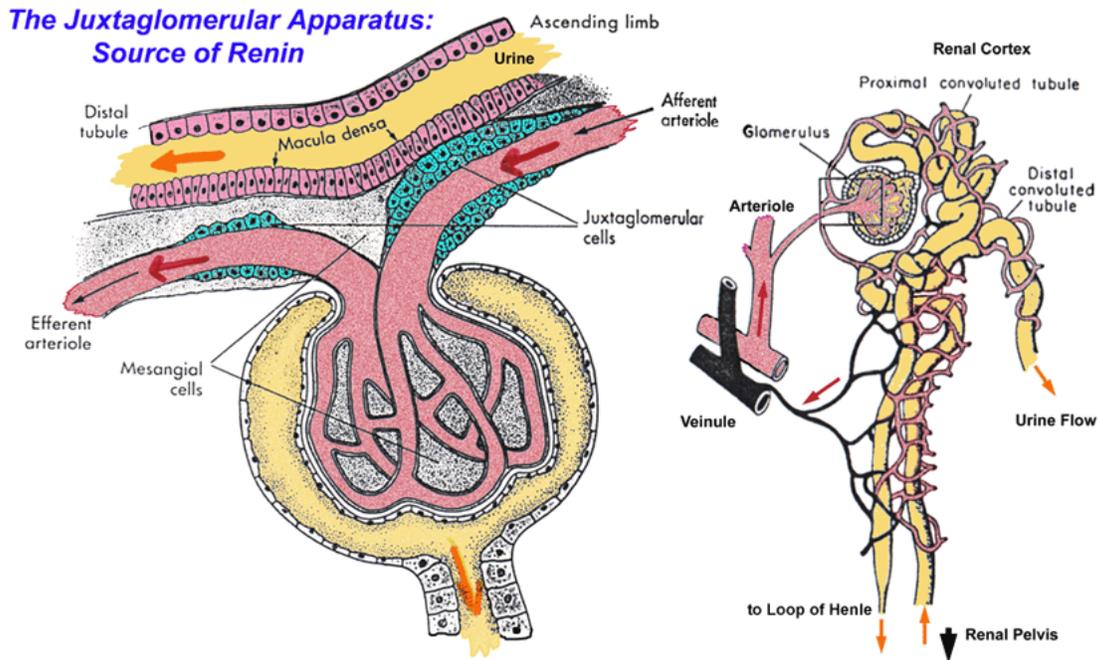
#### Myogenic Mechanism

The myogenic mechanism is not unique to glomerular capillaries. Myogenic mechanisms are intrinsic to the smooth muscle of all blood vessels, particularly in small arteries and arterioles. If the pressure within a vessel is suddenly increased, the vessel responds by constricting. Diminishing the pressure within the blood vessel causes relaxation and vasodilation.

The myogenic mechanism is most prominent in the afferent arterioles in the kidney, which are the first to sense changes in arterial pressure in the glomerular capillaries. Therefore, if an increase in systemic pressure occurs, the smooth muscle will initially be expanded in the afferent arteriole, thus increasing the  $P_{GC}$  and subsequently the GFR. However, very quickly after the smooth muscles in the afferent arterioles are forced to expand due to the increase in arterial pressure, the myogenic mechanism will be activated, causing the smooth muscle to constrict back towards their normal diameter, in response to this forced dilation. This is a homeostatic mechanism, and the diameter of the afferent arteriole will likely not be returned completely to normal, and thus the GFR will not be returned completely to normal. However, they will be returned TOWARDS NORMAL, and thus the increase in the GFR due to an increase in arterial pressure will not be as great due to this homeostatic effect of the myogenic mechanism. Thus, in this case, the GFR is prevented from increasing too far out of the normal range due to activation of the myogenic mechanism. The opposite scenario occurs if there is a drop in arterial pressure sensed by the afferent arterioles.

### Tubuloglomerular Feedback

Another autoregulatory mechanism for keeping the GFR from fluctuating too high or too low is tubuloglomerular feedback (TGF). In this mechanism, the amount of  $\text{Na}^+$  that is filtered into Bowman's space in individual nephrons is sensed by the macula densa cells that are present in the thick ascending limb of Henle's Loop. However, to understand TGF, you first have to understand something about the juxtaglomerular apparatus:



In almost all nephrons, the thick ascending limb (TAL) of Henle's loop comes back up and passes very close to the vascular pole, where the afferent arteriole enters and the efferent arteriole leaves Bowman's capsule. By doing this, the TAL puts itself into close proximity to the afferent arteriole in particular. Specialized macula densa cells line the TAL as it passes near these afferent arterioles at the vascular pole. These macula densa cells are thought to be capable of 'sensing' the delivery of certain compounds to this part of the nephron, that have been filtered into Bowman's space by the glomerular capillaries.

One theory proposes that transporters for reabsorbing  $\text{NaCl}$  and  $\text{K}^+$  in the TAL (the so-called  $\text{Na}^+/\text{K}^+/\text{2Cl}^-$  transporters) can sense abnormally increased or decreased  $\text{NaCl}$  delivery to this site, and then send a signal over to the smooth muscle cells in the afferent arterioles causing them to either constrict or to dilate. The evidence suggests that an increase in delivery of  $\text{NaCl}$  to the macula densa cells is interpreted as an increase in the GFR (which is logical; if the GFR increases, the rate of filtration of  $\text{NaCl}$  will increase, and more will reach the TAL in a given period of time). Therefore, if an increase in  $\text{NaCl}$  delivery is sensed, a signal is sent by the macula densa cells to the smooth muscle cells of the afferent arterioles that causes them to constrict, which will lower the  $P_{GC}$  and therefore lower the GFR back towards the normal range.

The figure below from Berne and Levy's 4th edition of *Physiology* illustrates this concept:

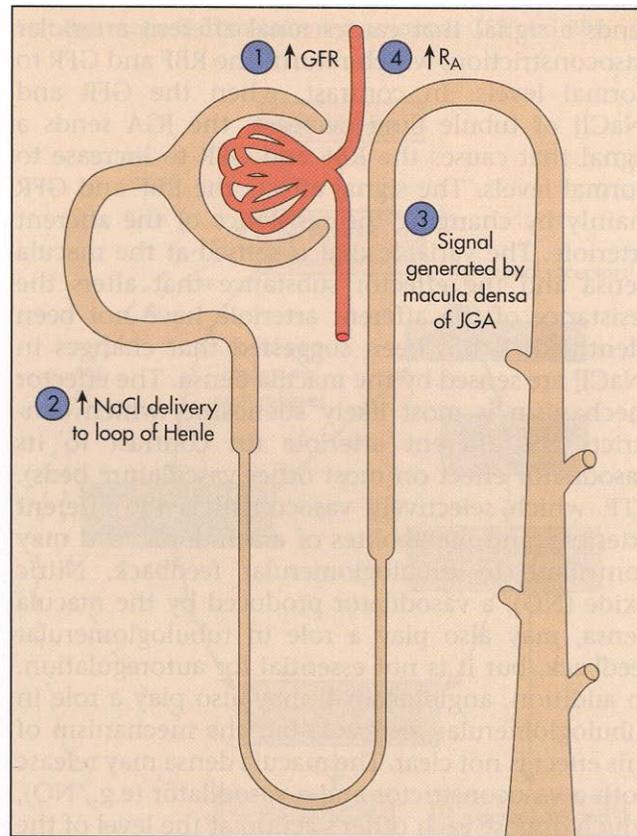


FIGURE 36–13. Tubuloglomerular feedback. An increase in the GFR (1) increases [NaCl] in tubule fluid in the loop of Henle (2), which is sensed by the macula densa of the juxtaglomerular apparatus (JGA) and converted into a signal (3) that increases the resistance of the afferent arteriole ( $R_A$ ) (4), which decreases the GFR. (Modified from Cogan MG: *Fluid and electrolytes: physiology and pathophysiology*, Norwalk, Conn, 1991, Appleton & Lange.)

### GFR in Healthy Adults

The average quantity of plasma that is filtered out of the glomerular capillaries and into Bowman's space each minute is **90 to 100 ml per  $1.73 \text{ m}^2$**  for an adult woman and **115 to 125 ml per  $1.73 \text{ m}^2$**  for an adult male. Thus, an adult male with a GFR of 125 ml/minute is filtering about **180 L** of plasma into Bowman's space daily (125 ml/min x 1440 minutes in a 24 hr day = 180 L). It is important to recognize that the GFR we speak of in medicine represents the **total** amount of plasma filtered into Bowman's space each minute, accomplished by *all the nephrons in both kidneys* (assuming you have two); the amount of

plasma filtered by each single individual nephron is called the “single nephron glomerular filtration rate”(SNGFR), which cannot be directly estimated in humans. However, the SNGFR is important in disease states, as you will see next year.

### Renal Blood Flow

The normal renal blood flow is ~1200 ml/min, equivalent to ~25% of the resting cardiac output. Given that the kidneys make up < 0.5% of the body weight, this clearly represents a massive blood flow. The magnitude of the blood supply is required not for the kidneys’ metabolic needs, but in order to maintain a high rate of glomerular filtration. The difference between flow in the renal artery and flow in the renal vein is the **urine flow**, which is small compared to total blood flow through the kidneys (urine flow averages about **1 ml/min** in most healthy adults).

**Renal plasma flow (RPF)** refers to the amount of plasma that traverses the renal vein or artery per minute. Assuming a hematocrit of around 45%, renal plasma flow averages around **600 ml/min** (total for 2 kidneys) in the normal individual. For example, in a person with a RBF of 1.2 L/min and a hematocrit of 45%, renal plasma flow would be 660 ml/min (1200 ml/min x .55 = 660 ml/min). As you know, red and white cells are not normally filtered across the glomerular capillaries into Bowman’s space...only plasma.

I stated above that about 125 ml of plasma total (high end of average for young adult males) actually gets filtered out of the glomeruli and into Bowman’s space each minute. Therefore, if only about 600 ml of plasma was actually passing through all of the glomerular capillaries each minute in an individual who had a GFR of 125 ml/min, it is obvious that only about **25%** of the total plasma flowing through all the glomerular capillaries each minute is actually getting filtered into Bowman’s space in this individual. This is termed the **filtration fraction**, and can be calculated by dividing the GFR by the RPF (GFR/RPF = filtration fraction). The filtration fraction of plasma traversing the all the glomerular capillaries each minute averages around 15 to 25% in most healthy people.

The RPF can be quantified. In the past, it was done primarily by measuring the "clearance" (more on this below) of a compound called **p-aminohippuric acid**, although the use of Doppler Ultrasound is now much more common. Whichever way it is done, once you have estimated RPF, you can quickly estimate renal blood flow (RBF) by the following formula:

$$\text{RBF} = \text{RPF}/1 - \text{hematocrit.}$$

### Estimating the GFR

The GFR is the best indicator of overall renal function. The assessment of the GFR is therefore fundamental to the diagnosis of renal glomerular pathology, to the management of drug therapy where urinary drug excretion depends on the glomerular filtration of the drug, and in chronic renal disease to both diagnose it, and to facilitate timely management decisions once it has been diagnosed (because continual decreases in GFR indicate a continual decline in renal function; stability or a slight increase in GFR in someone with renal disease suggests the current therapy is effective).

In order to understand how to measure the GFR, you must first understand the concept of clearance. Clearance incorporates the dimensions of time and volume, and represents *the volume of plasma from which all the substance of interest has been removed and excreted in the urine per unit time*. It should be stressed that the concept of clearance is by no means restricted to renal function. One can measure the rate at which the lungs, liver, or other organs clear substances from the blood as well.

Example of Renal Clearance:

If a substance is present in urine at a concentration of 1 mg/ml, and urine flow rate is 1 ml/minute, then the excretion rate of this substance is:

$$U_X \times V = 1 \text{ mg/ml} \times 1 \text{ ml/minute} = 1 \text{ mg/minute}$$

Where  $U_X$  is the concentration of substance X in the urine (usually always measured in mg/ml) and  $V$  is the urine flow rate (usually always measured in ml/min).

If the amount of this substance X is measured in plasma and found to be present at a concentration of **1 mg/dl**, then the plasma clearance can be calculated:

$$C_X = \frac{U_X \times V}{P_X} = \frac{1 \text{ mg/minute}}{1 \text{ mg/dl (.01 mg/ml)}} = 100 \text{ ml/min}$$

I converted mg/dl to mg/ml before calculating, because renal clearance is always expressed as ml/min ( $P_X$  is the concentration of substance X measured in plasma, and  $C_X$  is the plasma clearance of substance X).

Remember, it was stated above that "clearance incorporates the dimensions of time and volume, and represents the volume of plasma from which all the substance of interest has been removed and excreted in the urine per unit time". Therefore, the plasma clearance of 100 ml/min calculated above means that **every minute, 100 ml of plasma is completely "cleared" of substance X by the kidneys**.

The most accurate way to measure the GFR is to measure the renal clearance of a substance that has the following 2 characteristics:

1. It **ONLY** enters the lumen of the nephron by being **FILTERED** from the plasma of the glomerular capillaries into Bowman's space (**NO** secretion occurs).
2. Once filtered from the glomerular capillaries into Bowman's space, **NONE** of it is reabsorbed as it moves down the nephron – therefore, the amount filtered into the nephron in 24 h **EQUALS** the amount ultimately excreted into the urine 24 h.

To make things even more simple and effective, the theoretical substance above would be produced in our bodies and appear in the plasma in constant amounts – i.e., the plasma levels would not fluctuate (and it would not have to be infused in the clinic!). Also, it would not be

bound to proteins so that glomerular filtration of the substance would readily occur. Unfortunately, this is not the case. The substance that comes closest to this theoretical ideal is probably creatinine, which will be discussed below.

There are *exogenous* substances, however, that do meet the two ideal criteria listed above. *Inulin* (not to be confused with “insulin”) is a polyfructose molecule found in chicory and Jerusalem artichokes, with a M.W. of about 5000. To measure the GFR by measuring the clearance of inulin, the patient is brought into the hospital, and a continuous i.v. infusion of inulin is started until a certain plasma steady state is reached. After the steady state is reached (the amount being infused is equal to the amount being excreted at that point), urine is collected for several hours, and urine concentration of inulin as well as urine flow rate are determined. Inulin levels in a plasma sample are then quantified as well, and the values are plugged into the clearance formula that you saw previously above:

$$Cl_{in} = \frac{U_{in} \times V}{P_{in}}$$

$V$  = urine flow rate (ml/min)

$U_{in}$  = urinary concentration of inulin (mg/ml)

$P_{in}$  = plasma concentration of inulin (mg/ml)

$Cl_{in}$  = clearance of inulin

Look at the figure below:

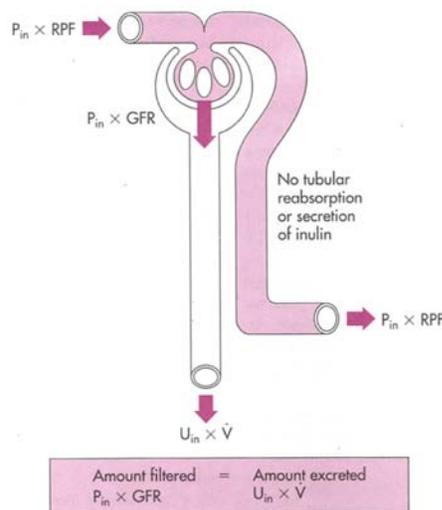


Figure 3-2 ■ Renal handling of inulin. Inulin is freely filtered across the glomerulus and is not reabsorbed, secreted, or metabolized by the nephron.  $P_{in}$ , Plasma inulin concentration;  $RPF$ , renal plasma flow;  $GFR$ , glomerular filtration rate;  $U_{in}$ , urinary concentration of inulin;  $\dot{V}$ , urine flow rate. Note that all the inulin coming to the kidney in the renal artery does not get filtered at the glomerulus (normally 15% to 20% of plasma and inulin are filtered). The portion that is not filtered is returned to the systemic circulation in the renal vein.

As you look at this figure, remember that you can measure the renal clearance of anything, but that doesn't necessarily estimate the GFR. It is ONLY because of the special renal handling characteristics of inulin described above that the measured clearance of inulin is AN ACCURATE MEASURE OF THE GFR.

Take note of the fact that in the above figure, *the amount of inulin that gets filtered into Bowman's space EQUALS the amount of inulin that is excreted in the urine* – none gets in by secretion, and none that is filtered in gets reabsorbed anywhere downstream.

Therefore, if we know how much inulin is in each ml of plasma (by measuring it in the plasma), and we know how much inulin is in each ml of urine (because we measure that as well), and we know the urine flow rate in ml/min (which we DO when we calculate clearance), then we can use the clearance formula to calculate how many ml's of plasma MUST HAVE been filtered into the nephrons each minute to put that amount of inulin into the urine each minute that we measured. This amount of plasma that had to be filtered each minute to put this amount of inulin into the urine in the same amount of time is BY DEFINITION THE GFR. Again, when doing this we make the assumptions that the substance in question (inulin, in this case) ONLY gets filtered into nephrons (NOT secreted), and is NOT reabsorbed from the nephron after it is filtered in.

#### Other Exogenous Markers to Measure GFR using Clearance

In addition to inulin, other exogenous compounds can be administered, whose renal clearance gives a very good measure of the true GFR. These include the radiolabeled  $^{51}\text{CrEDTA}$ ,  $^{125}\text{I}$ -iothalamate, and  $^{99}\text{Tc-DTPA}$ .

Measuring the clearance of inulin in a hospital setting is THE GOLD STANDARD FOR MEASURING GFR. If you wanted to know what the TRUE GFR is in your patient, this would be the test to perform.

In reality, these renal clearance tests involving administration of exogenous inulin in a clinical setting are NOT routinely performed in clinical practice due to high cost, requirement of a skilled staff, availability of assays to measure inulin, and inconvenience to the patient. Instead, if a clearance test is going to be used to measure GFR, a 24 h creatinine clearance is usually used.

#### 24 h Creatinine Clearance to Measure GFR

In practice, the renal clearance of an endogenous compound, **plasma creatinine** (a by-product of muscle creatine metabolism) is measured instead of inulin, and it gives a reasonable estimate of the GFR. The patient collects their urine for 24 h, brings the urine into the lab, and provides a blood sample at that time. Serum creatine, urinary creatinine, and urine flow rate are determined, and the same formula used for inulin clearance is applied to measure GFR.

#### *Drawbacks to a 24 h Creatinine Clearance*

Although a 24 h creatinine clearance is a reasonable measurement of GFR, it has several drawbacks. For starters, patients have to collect their urine into a container over

24 h. This is very inconvenient, and often leads to incomplete collections, which affects the accuracy of the test.

Furthermore, even in people with normal renal function, about 10% to 15% of all creatinine appearing in the urine was *secreted* into the nephron. As noted previously, the ideal clearance marker to estimate GFR is *not* secreted into the nephron at all – only filtered in through the glomeruli. If secretion occurs, then the calculated GFR will be *overestimated*.

This is an even bigger problem in patients who already have significant renal dysfunction, such as when their true GFR is below 50 or 60 ml/min. In these people therefore, **MORE** than 10% to 15% of the total creatinine excreted in the urine over 24 h is in fact **SECRETED** into the nephrons, as opposed to **FILTERED** in, thus *overestimating the calculated GFR even more than in healthy people* when GFR is measured using a 24 h creatinine clearance.

The reason for this is that, in this setting, the surviving nephrons are secreting lots of creatinine because it is the only way to keep creatinine from accumulating in the plasma in excessive amounts. Remember, creatinine is produced daily from muscle in relatively constant amounts. The person who has lost half their renal function has about half as many nephrons now to get rid of the daily load of creatinine. These nephrons cannot do the job anymore simply by filtering (there are too few nephrons now), so they must filter **AND** secrete more to try to maintain plasma creatinine levels.

One of the things that can be done to increase the accuracy of GFR measurement in a 24 h creatinine clearance in a patient with significant renal disease is to put them on a high dose of **cimetidine** prior to collecting urine. Cimetidine is a member of the H<sub>2</sub> blocker family of drugs which impairs acid secretion in the stomach. Interestingly, cimetidine also seems to block secretion of creatinine in nephrons when given at high doses, and is thus used for this purpose.

Measuring the GFR in growing children using a clearance formula has always been a problem, as creatinine production is proportional to muscle mass, and creatinine values have to be adjusted for height and weight when using them to estimate GFR in children. However, a recent paper (Bokenkamp et. al., 1998, Pediatrics, 101(5):875-881) has suggested that we should start measuring the clearance of an endogeneously-produced compound called **cystatin C** (a glycosylated protein produced at a constant rate by all nucleated body cells) in children, when we want to estimate their GFR. The authors of this paper reported that cystatin C, unlike creatinine, reflects renal function in children, independent of age, gender, height, and body composition. This is currently not standard practice, and is still being evaluated.

### **Using Equations to ESTIMATE the GFR**

Numerous formulas have been developed to estimate GFR from serum creatinine and other variables. One widely used formula to predict creatinine clearance was proposed by Cockcroft and Gault in 1976. The Cockcroft-Gault equation predicts creatinine clearance (mL/min) from serum creatinine, age, and weight. Height must also be measured to compute body surface area and express the result in conventional units (mL/min per 1.73 m<sup>2</sup>). The Cockcroft-Gault equation was derived from an investigation of 249 men with creatinine in a steady state; the subsequent companion equation for women was based on their 15% lower

muscle mass. In one study of 394 subjects (208 men and 186 women), the correlation between estimated creatinine clearance and measured GFR was excellent ( $R^2 = 84\%$ ).

### **Cockcroft and Gault Equation (1976)**

$$\text{FORMULA: } \frac{(140 - \text{age}) \times \text{LBW} \times F}{72 \times S_{\text{cr}}}$$

F = 1 for male; F = 0.85 for female

LBW = Lean body weight (kg)

$S_{\text{cr}}$  = serum creatinine

Your LBW is approximately: Height (cm) – 100

More recently, an equation was developed to predict GFR using data from 1,628 patients enrolled in the baseline period of the Modification of Diet in Renal Disease (MDRD) study.

### **II. Levey and colleagues (2000) Simplified 4-variable MDRD study formula:**

WHAT IT ESTIMATES: GFR (mL/min/1.73 m<sup>2</sup>)

$$\text{FORMULA: } \text{GFR} = 186.3 \times (S_{\text{cr}})^{-1.154} \times (\text{age, yr})^{-0.203} \times 1.212 \text{ (if patient is black)} \times 0.742 \text{ (if patient is female)}$$

Thus, both the Cockcroft-Gault and MDRD study equations provide reasonably accurate estimates of GFR. In patients with chronic renal disease (CRD), the MDRD study equation appears to provide a more accurate and precise estimate of GFR than either the Cockcroft-Gault equation or measured creatinine clearance (using a 24 h urine collection).

Recently, the National Kidney Foundation published a position statement on testing for the presence for chronic kidney disease (American Journal of Kidney Disease, vol 50(2), August 2007, pp. 169-180), in hopes of providing information to physicians on how to best identify patients who have chronic kidney disease at an earlier stage of their disease. This paper is posted on Blackboard for you to read if you wish; reading it is not required by me.

### GFR and Changes in Plasma Creatinine

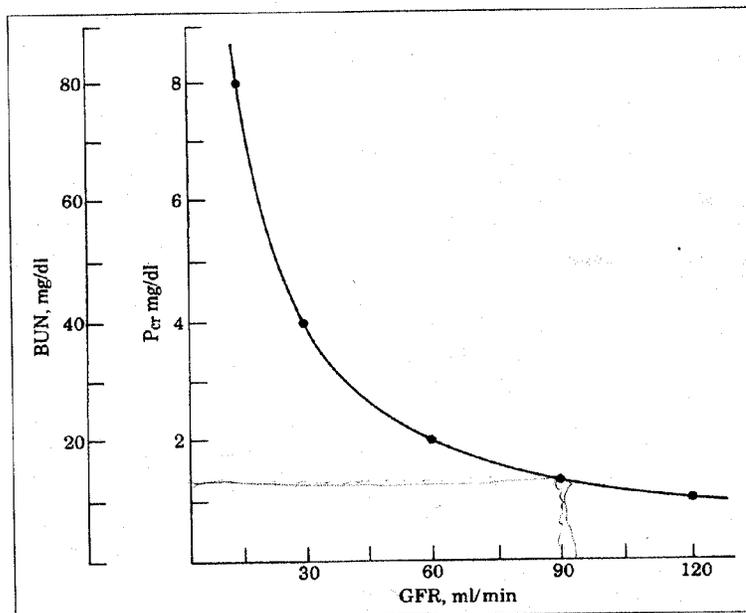
Serum creatinine values rise as GFR goes down, because the kidneys represent the major excretory route for creatinine. As we will see, however, the rise in creatinine as the GFR drops is not linear.

The range of normal plasma creatinine values in any human population is fairly variable, and this reduces the usefulness of a single plasma creatinine sample alone as an indicator of underlying renal problems. Most texts report a normal range of 0.8 to 1.3 mg/dl adult men, and 0.6 to 1.0 in adult women. However, because creatinine comes from muscle, the normal range is also affected by muscle mass (weight lifters have a high normal value) and food intake (meat, of course, contains lots of creatinine) can also affect levels.

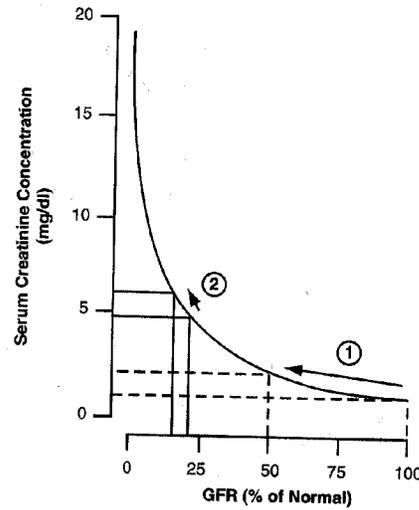
In early chronic renal disease, when the GFR has fallen around 10-20%, there will be very little increase in plasma creatinine levels to reflect this. That's because the unaffected nephrons began to *secrete* creatinine at higher rates in the proximal tubules, to allow amounts of creatinine getting excreted into the urine to remain fairly stable, and thus plasma levels remain fairly stable.

It is not until renal disease is well advanced that a single plasma creatinine measurement can raise a red flag to the clinician. For example, if a woman with normal renal function has a plasma creatinine value of 0.6 mg/dl, and then loses 50% of her functioning nephrons over ten years (due to some underlying chronic kidney disease), her plasma creatinine level would still be only approximately 1.2 mg/dl if tested. Unless she is complaining of symptoms which mimic renal disease (unlikely, since that generally does not occur until the GFR is below 15 to 20 ml/min) this is likely to be ignored. Yet, she actually has lost about half of her renal function at this point. Look at the figure below:

**Figure 13-1.** Hypothetical steady-state relationship between the plasma creatinine concentration ( $P_{cr}$ ), blood urea nitrogen (BUN), and glomerular filtration rate (GFR).



The illustration below further demonstrates this phenomena:



**FIGURE 1** A rise in serum creatinine from 1 to 2 mg/dL (①) represents a 50% reduction in GFR. Conversely, a rise in serum creatinine from 4.8 to 6 mg/dL (②) represents a decline in GFR of approximately 20–15%.

In advanced renal disease (ie, when the GFR is only 25% or so of normal), the % increase in plasma creatinine for any given % decrease in GFR is much greater than in early renal disease. At this point there are so many non-functioning or dead nephrons, that even the combination of filtering and secreting creatinine is not enough to balance the constant daily output by the muscles, and creatinine levels begin to rise rapidly as the kidneys continue to fail.

Therefore, it should be remembered that *it is generally true that plasma creatinine levels do not rise outside of the normal range until serious renal impairment exists*. Therefore, high values in the 'normal range' should arouse suspicion if a patient's history suggests renal disease.