MATHEMATICAL MODELING OF THE TUBULOGLOMERULAR FEEDBACK MECHANISM IN THE KIDNEY

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Contents

Summary

This chapter provides an overview of mathematical models of the hemo- and hydrodynamics of the kidneys. These models have been developed over the last two decades, mainly motivated by experimental evidence of spontaneous tubular pressure oscillations in the nephron tubule, the functional unit of the kidney, thus extending the existing models of a steady state nature. More recent observations reveal that the oscillations in single nephrons are synchronized with neighboring nephrons, and to get a complete picture of the physiological mechanisms, models incorporating the interactions between ensembles of nephrons should be considered. Here merely single nephron models are considered, which are the building blocks of more complex models, only giving models of coupled nephrons a short treatment in the end.

1. Introduction

The kidney has long been known as an organ of homeostasis. Homeostasis implies constancy, and a steady state would seem to be the most appropriate dynamical state in which to view the kidney. But like the heart, another organ of homeostasis, the kidney

has an intrinsic beat, and there are disorders of renal rhythms that occur in at least one disease, chronic hypertension (high blood pressure). The renal beat arises in the operation of intrarenal mechanisms that provide autoregulation of blood flow, i.e. the intrarenal mechanisms that minimizes changes in renal blood flow in response to changes in the arterial blood pressure. Studies of the temporal dynamics and interactions of different parts of these systems have provided insight into the mechanisms. The blood pressure, the principal stimulus to autoregulation, is highly variable, and if not compensated for these fluctuations would lead to corresponding fluctuations in renal blood flow. Since renal blood flow is an important variable when it comes to determining renal function, knowledge of the dynamics of autoregulation is important for understanding how the kidney responds to this variation in its environment.

Physiological control systems often involve negative feedback regulation, which in principle should be stable. However, due to delays in the system the regulation can become unstable in many cases, producing self-sustained oscillations and other nonlinear phenomena. The possible biological advantages of such non-stable behavior are controversial. One clear advantage is that the instabilities arise in the operation of nonlinear systems which can generate a number of interesting properties of potential physiological significance, including amplitude and frequency modulation, synchronization, and bifurcations to chaos. Each of these properties has been described in the renal circulation. It is also possible that dynamic behaviors, avoiding steady states, may protect a system against long-term drift or protect it from being trapped in suboptimal conditions.

The kidneys also have a negative feedback system with delay, namely the tubuloglomerular feedback mechanism (TGF). Early micropuncture experiments in halothane anesthetized rats by Leyssac, Baumbach and Holstein-Rathlou demonstrated that this feedback regulation could become unstable and generate self-sustained oscillations in the proximal intratubular pressure with characteristic periods of 30–40 s. The appearance of oscillations depends on the experimental conditions, since rats anesthetized with the commonly used anesthetic inactin do not show similar oscillations in the proximal tubular pressure. This is due to the specific effects of inactin, since oscillations in tubular pressure has now been observed in rats anesthetized with such diverse anesthetic agents as isoflurane, sevoflurane and amytal, and in mice anesthetized with chloralose/ketamine. It is still an open question what type of tubular pressure dynamics is present in conscious animals and humans.

2. Anatomy and Physiology of the Kidney

The kidneys are the main organs that regulate the composition and the volume of the body fluids by excretion of salts, water and metabolic end products. The main tasks are regulation of osmolarity of all body fluids and the volume of the extracellular fluid; regulation of the electrolyte balance by controlling excretion of ions, e.g. Na^+ and Cl^-; balancing of acids and bases; excretion of waste products such as urea, uric acids and creatinine; secretion of externally supplied substances such as drugs or environmental

toxics; and finally synthesis and secretion of hormones, mainly renin.

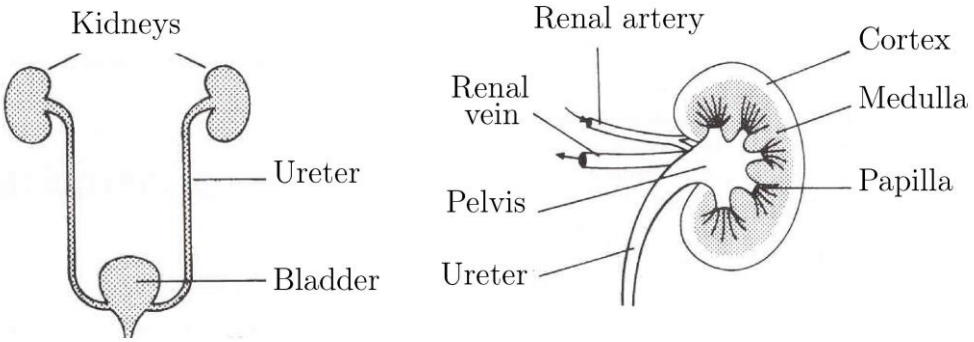

Figure 1. The anatomy of the urinary system. To the right, a cross section of a kidney.

The kidney consists of three major parts: the cortex, the medulla and the pelvis (see Figure 1). Blood enters through the renal artery, has its composition altered by filtration at the glomerulus and transport of water and solutes by the tubules, and leaves through the renal vein. It is the organ with the highest blood perfusion in the organism: in 24 hours 1,000 liters of plasma and 800 liters of red blood cells pass through the kidneys, which corresponds to 1/5 of the total cardiac output at rest. Thus, the total blood volume passes through the kidneys around 300 times a day. The urine leaves the kidney through the renal pelvis and ends up in the urinary bladder via the ureter.

The kidneys need a reasonably stable blood flow to function properly, and therefore make use of effective autoregulatory mechanisms to compensate for fluctuations in arterial blood pressure. For instance, variations in arterial blood pressure between 80 and 180 mmHg do not affect renal blood flow in humans.

Two control mechanisms have been proposed to explain autoregulation: TGF and the myogenic mechanism. Both act to increase (decrease) the hemodynamic resistance of the preglomerular vessels in response to an increase (decrease) in the arterial blood pressure. By changing the vascular resistance these mechanisms will minimize the changes in blood flow in response to the variations in arterial pressure. The myogenic mechanism is an intrinsic response of the preglomerular vessels, where an increase in arterial blood pressure increases wall tension, which induces a reflex vasoconstriction, and an increase in vascular resistance. This will decrease the blood flow back towards the value prior to the pressure increase. Conversely, a decrease in arterial blood pressure causes vasodilatation, decreasing vascular resistance, and an increase in blood flow back towards the control value.

The TGF mechanism reacts to the NaCl concentration in tubular fluid at a location in the tubule called the *macula densa* that is sensitive to the concentration of chloride, a low concentration (and thus a low flow rate of tubular fluid) signals the afferent arteriole to dilate, which increases blood flow, and vice versa. Both mechanisms work at the level of the individual *nephron*, (see Figure 2). A human kidney contains around one million nephrons, a rat kidney around 30,000.

2.1. Anatomy of the Nephron

Each nephron consists of a *glomerulus* and a *tubule*. The glomerulus is a capillary network inserted between the afferent and the efferent arteriole. The glomerulus is surrounded by *Bowman's capsule*, from where the tubule starts. The tubule is 20 - 30 mm long and has a diameter of 15-60 μm (see Figure 2). The tubule is divided into the *proximal tubule* (closest to the glomerulus); the *loop of Henle*, which is a hairpin like structure that can be divided into a descending and an ascending limb; and the *distal tubule*. Tubules from many nephrons merge to form *collecting ducts*. As these descend through the kidney medulla they continue to merge, ultimately forming a few thousand *papillary collecting ducts* which empties into the renal pelvis at the tip of the *papillas*. All tubules together are called the *tubular system*.

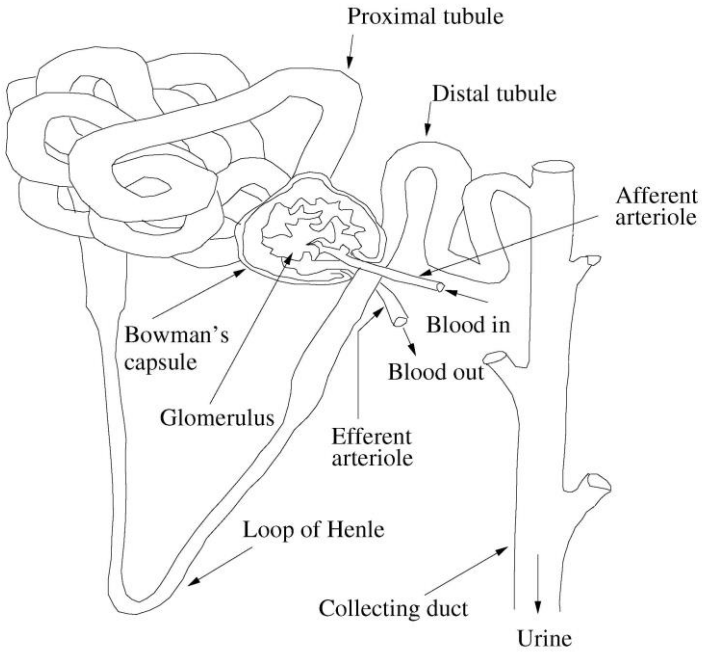

Figure 2. The nephron.

The loop of Henle returns to its own glomerulus. Part of the loop of Henle lies close to the corresponding afferent arteriole, and together they constitute the *juxtaglomerular apparatus*. It consists of several structures, including the *macula densa*, which are tubular epithelial cells in the wall of the ascending limb of the loop of Henle; and juxtaglomerular cells in the afferent arteriole. The close contact between the afferent arteriole and the loop of Henle constitutes the structural base of the TGF mechanism.

2.2. Physiology of the Nephron

The nephron is the functional unit of the kidney, and where the filtering of blood occurs. Three basic transport processes take place in the nephron: glomerular filtration, tubular reabsorption and tubular secretion, resulting in the formation of the final urine.

In the glomerulus, blood passes through a capillary network where plasma water together with substances with a molecular weight under 5,000 g/mol are filtered through the glomerular membrane and into Bowman's capsule. Plasma substances with a molecular weight above 70,000 g/mol, mainly proteins, are almost entirely retained in the vessels. Substances in between are partly passed. This process is called *glomerular ultrafiltration*. The ultrafiltrate contains almost no protein. It is isosmolar with plasma, containing the same concentration of low molecular weight substances.

Ultrafiltration in the glomerulus increases with increasing hydrostatic pressure difference between the capillaries and the capsular space, whereas the colloid osmotic pressure in the glomerular capillaries impedes the filtration. Since the ultrafiltrate contains no protein, the protein concentration increases gradually as plasma water is filtered, and thus, the colloid osmotic pressure increases as the blood flows through the glomerulus from the afferent to the efferent arteriole. If the colloid osmotic pressure becomes equal to the hydrostatic pressure difference filtration stops. This is termed filtration equilibrium, and has been observed to occur in certain rat strains. However, it is generally assumed that under most physiological conditions filtration equilibrium is not attained.

The ultrafiltrate, the nascent urine, flows from the capsular space to the tubular system and continues to the renal pelvis. On its way, volume and composition are changed as some substances (e.g. water, NaCl, and glucose) are transported from the tubular fluid back to the blood (in the peritubular capillaries and in vasa recta), a process called *tubular reabsorption*, while other substances are transported in the opposite direction from the blood to the tubular fluid, denoted *tubular secretion*.

In humans, around a fifth of the plasma passing through the glomeruli is filtered into the tubular system, which means that 180 liters of ultrafiltrate is produced daily. However, only 1-2 liters are excreted as urine, so that around 99% of the ultrafiltrate is reabsorbed from the tubular system and returned to the circulating blood. Tubular reabsorption of electrolytes and water from the more distal segments of the nephron is regulated by hormones from the adrenal gland (aldosterone) and the pituitary gland in the brain (antidiuretic hormone). Through these and other hormonal control systems, the composition and the volume of the final urine is adapted to the physiological needs of the body, maintaining the composition and volume of the extracellular fluid, the electrolyte balance and the balance of acids and bases within strict limits.

The proximal tubule reabsorbs approximately 70% of the filtrate into the surrounding capillaries. The composition of tubular fluid remains nearly unchanged in this part of the nephron. In the descending limb of the loop of Henle another 15% of the water is reabsorbed, while urea is secreted into the tubule. Hence the composition becomes more hyper-osmotic the deeper into the renal medulla the nephron reaches. The ascending limb of the loop of Henle is nearly impermeable to water, whereas salts, primarily NaCl, are transported out of the tubule into the interstitial space. Therefore the tubular fluid turns from hyper- to hypo-osmotic as the tubule returns to the renal cortex. Another 10%

of the water is reabsorbed in the distal tubule, so that around 5% of the ultrafiltrate reaches the collecting duct, and the fluid is iso-osmotic with plasma at this point. In the collecting duct, which receives tubular fluid from many nephrons, the tubular fluid is transformed into the final urine by epithelial transport of water, salts and urea, so that only around 1% of the original ultrafiltrate is excreted. In the presence of antidiuretic hormone (vasopressin), the osmolarity of the fluid in the collecting duct increases on its way down from the renal cortex, through the renal medulla, and to the end in the renal pelvis at the papillas, with an osmolarity of 1.400 osmol/l compared to a plasma osmolarity of 300 osmol/l. Aldosterone, from the cortex of the adrenal gland, promotes Na^+ reabsorption and K^+ secretion in the distal tubule and collecting ducts.

2.3. Autoregulation of Renal Blood Flow

Together with the brain, the kidney has the most efficient autoregulation of blood flow. Due to autoregulation even rather large fluctuations in the arterial blood pressure will only lead to small changes in renal blood flow. A perturbation of the arterial blood pressure will induce rapid changes in the vascular resistance that tends to stabilize the renal blood flow. The kidneys dispose of two autoregulatory mechanisms to stabilize the renal blood flow, namely the myogenic mechanism and TGF. Each of the two mechanisms is frequency dependent and the contribution each makes to autoregulation therefore depends on the frequency of the perturbation of the blood pressure. Both contribute to autoregulation and there is a complex interaction between them.

The myogenic mechanism. An increase in the arterial blood pressure leads to a rapid contraction in the smooth muscles of the vessels. As pressure rises, the smooth muscle cells within the arteriolar wall will sense an increase in wall tension, to which the myogenic mechanism responds by contracting the arteriole, thus reducing the blood flow back towards the control value. The myogenic mechanism reacts within one or two seconds, and is hence related to fast autoregulatory changes.

The TGF mechanism. TGF a negative feedback loop, is unique to the kidney. By changing the renal vascular resistance, the TGF regulates the renal blood flow, the glomerular filtration rate (GFR), the pressure in the glomerulus and in the proximal tubule, and the flow into the loop of Henle. Its most important task is to maintain the stability of salt and water delivery to the distal part of the nephron so that the distal tubule and the collecting duct can regulate hormone dependent salt and water excretion and reabsorption optimally for the homeostatic needs of the body.

An increased blood pressure will transiently increase both the renal blood pressure and the glomerular capillary pressure. Both changes will increase the filtration rate, and thereby increase the flow into the loop of Henle. TGF is activated by increased flow into the loop of Henle, because of the resulting increase in the NaCl concentration at the *macula densa*. The *macula densa* cells monitor the NaCl concentration and generate a signal that is transmitted to smooth muscle cells in the wall of the afferent arteriole, causing the afferent arteriole to contract. The details of the signaling process is still not

fully understood, but it appears that an increased concentration of NaCl in the tubular fluid at the site of the *macula densa* causes an increased uptake of NaCl into the *macula densa* cells through the action of the $Na^+/K^+/2Cl^-$-cotransporter present in the luminal membrane of the cells. Through unknown mechanisms the increased uptake of NaCl causes secretion of ATP by the *macula densa* cells into the mesangial region, where it is rapidly converted to adenosine by the extracellular enzyme ectonucleotidase. Adenosine then reaches the vascular smooth muscle cells of the afferent arteriole causing constriction of the vessel. Because of the vasconstriction renal blood flow and glomerular filtration pressure decreases, this then reduces GFR and inflow to the loop of Henle. A causal loop diagram of the TGF mechanism is illustrated in Figure 3. Due to the transition time through the tubular system, the salt concentration at the *macula densa* does not change instantaneously after a change in glomerular filtration rate, but some 8-10 seconds later. An additional delay of 4-5 seconds is associated with the transmission of the signal from the *macula densa* cells to the arterial wall. The response time for the TGF mechanism is thus related to slower autoregulatory changes of around 12-15 seconds.

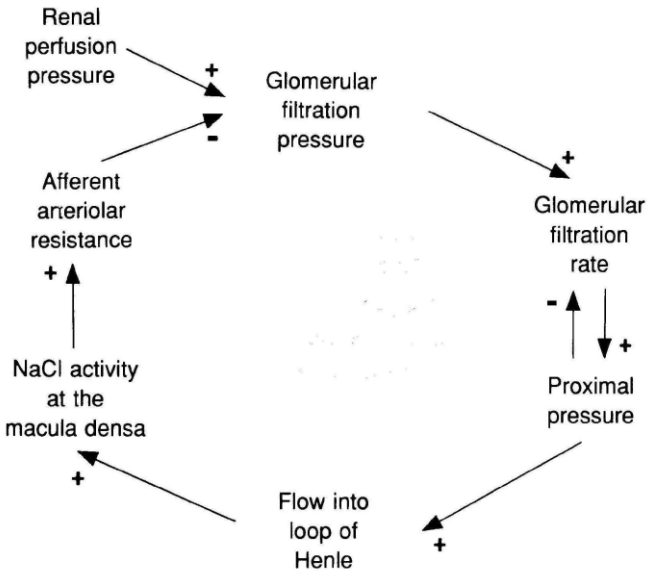

Figure 3. Causal loop diagram of the TGF mechanism. From Holstein-Rathlou and Marsh, Physiological Reviews, 1994. Am Physiol Soc, with permission.

Experiments have shown that spontaneously hypertensive rats have a stronger TGF response, i.e. the increase in resistance is greater and occur at lower concentrations of NaCl, when compared to normotensive rats. The exact mechanism underlying this change is unknown.

Dynamics of tubular pressure and flow. Tubular and vascular pressures can be measured experimentally on the surface of the kidneys of anesthetized rats. The data we use for illustration are obtained by Yip, Marsh and Holstein-Rathlou.

The experimental data show two fundamental oscillations in tubular pressure and flow,

one with a frequency of 25 to 45 mHz (see Figure 4), and another with a frequency of 100 to 140 mHz. The slower of the two is due to the operation of the TGF mechanism, and the faster is due to the myogenic mechanism.

Data show a clear difference in dynamic behavior of the tubular pressure oscillations between normo- and hypertensive rats. In normotensive rats the TGF mediated oscillation is regular, and is seen in 80-90% of the nephrons studied, Figure 4. In contrast in rats with a genetic form of hypertension (spontaneously hypertensive rats) the fluctuations are highly irregular, Figure 4. Why the rhythms are different is at present unknown, but as will be seen below, comparing the results of computer simulations of TGF models to experimental data can help suggest causal explanations.

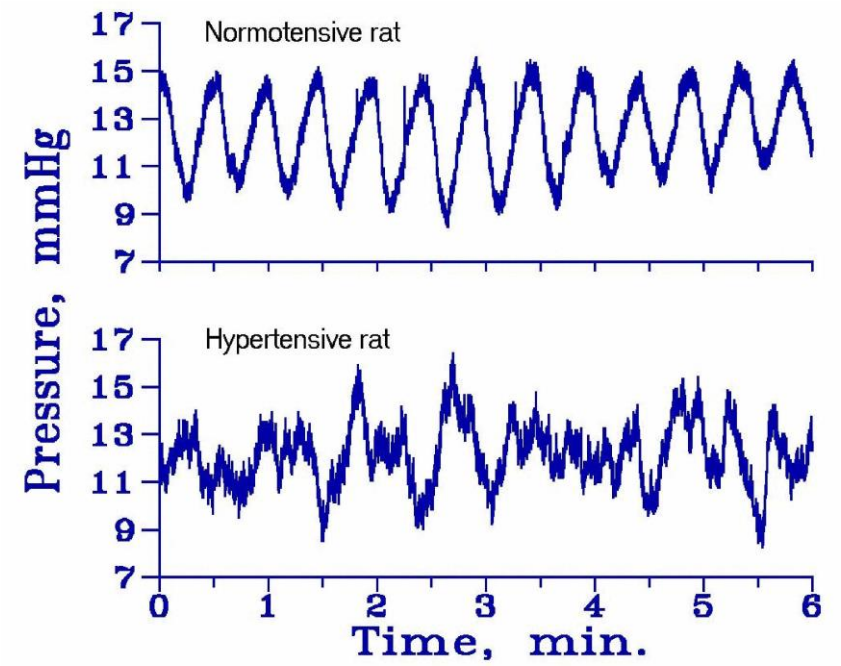

Figure 4. Typical fluctuations of proximal tubular pressure. Upper panel: normotensive rat. Lower panel: spontaneous hypertensive rat. From Holstein-Rathlou and Marsh, Physiological Reviews, 1994. Am Physiol Soc, with permission.

The kidneys play a central role in the regulation of the arterial blood pressure, and it is well known that renal dysfunction can lead to hypertension. Experiments have shown that transplanting a kidney from a hypertensive rat into a normotensive rat induces hypertension in the otherwise healthy rat, and that hypertension can be alleviated by transplanting a kidney from a normotensive rat to a rat with hypertension.

Despite the clear evidence for a central role of the kidneys in the regulation of arterial pressure, it has not been possible to identify changes in kidney function in most subjects (humans and animals) with hypertension. It remains an open question whether the observed change in dynamics of the TGF system is associated with the changes in renal function that ultimately lead to the development of high blood pressure. The change in

the dynamics of TGF will lead to changes in the response to the fluctuations in arterial pressure, and this could lead to changes in the excretory function of the kidneys.

An understanding of the consequences of the intrinsic dynamics in the nephron for kidney function may therefore be central to an understanding of the role of the kidney in blood pressure regulation. This endeavor will require the development of detailed dynamic models of the nephron and its associated regulatory mechanisms.

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Biographical Sketches

Susanne Ditlevsen graduated in 1999 with a MSc in mathematics from Universidad Nacional de Educacion a Distancia, Spain and a MSc in statistics in 2000 from University of Copenhagen, Denmark. She got a PhD in 2005 from Department of Biostatistics, University of Copenhagen, where she became Associate Professor in 2007. She is now Professor at Department of Mathematical Sciences, University of Copenhagen, and head of the research group Statistics and Probability Theory. Her major fields of research are statistical inference for stochastic processes, computational neuroscience and mathematical modeling of physiological dynamics.

Donald Marsh received the MD degree in 1958 from the University of California, San Francisco. He was Assistant Professor of Physiology and Biophysics at New York University from 1963-67, and Associate Professor from 1967 to 1970. He was Professor of Biomedical Engineering at the University of Southern California from 1970-1992. In 1978 he also became Professor of Physiology and Biophysics at the University of Southern California. In 1992 he became Professor of Molecular Pharmacology, Physiology, and Biotechnology at Brown University, and from 1992-2002 Dean of Medicine and Biological Sciences.

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