13. Kidney, Water and Electrolytes

13.1 The Kidney

The kidneys are essential for life. They eliminate in the urine unwanted water and solutes and so regulate both the volume and the composition of the body fluids. The urine contains (a) waste products including inactivated hormones, (b) foreign substances and their derivatives, and (c) surplus water and normal soluble constituents of the body.

The kidneys also produce a variety of humoral agents - erythropoietin, active metabolites of vitamin D, renin and prostaglandins.

Structure

Each human kidney has about 1 million *nephrons* arranged in parallel. Nephrons are similar but not identical. The chief variants are:

(a) Superficial, *cortical nephrons.* These have short *loops of Henle* reaching only into the outer medullary zone and their *efferent glomerular arterioles* supply their *peritubular capillaries.* About 80% of the nephrons in the human kidney belong to this group.

(b) Deeper, *juxtamedullary nephrons.* These have long loops of Henle, which plunge deep into the inner medulla. Their efferent glomerular arterioles supply peritubular capillaries and also venous capillary loops (*vasa recta*) which course among the loops of Henle and the *collecting tubules* deep in the medulla.

Note that all renal tubules receive only postglomerular blood, and that the vasa recta carry the sole blood supply to the inner medulla.

Renal Blood Flow

The rate of the renal circulation through both kidneys can be measured by applying the Fick principle to any substance which is transferred from plasma to urine but is not produced, destroyed or stored within the kidney. Let concentrations in arterial plasma, renal venous plasma and urine be P_A, P_{RV} and U and let urine flow per minute be V. Then the amount entering the kidney per min is the *renal arterial plasma flow (RPF) x* P_A while the amount leaving is the sum of the amount leaving in the urine (UV) and in the renal vein (neglecting renal lymph flow which is very slow). It turns out that the volume of the urine per min (about 0.5 to 1 mL/min) is so much smaller than that of the plasma entering the kidneys each minute (about 650 mL/min), that the renal venous plasma flow can be taken as equal to the arterial plasma flow, RPF. Therefore, the amount of the substance leaving the kidney in the veins equals RPF x P_{RV}. Thus

$$\mathbf{RPF \times P_A = UV + (RPF \times P_{RV})}$$

or

$$\mathbf{RPF = UV / (P_A - P_{RV}).}$$

Catheterization studies have shown that *para-aminohippurate (PAH)* is nearly completely removed from the plasma of blood passing through the kidney when its arterial concentration is sufficiently low that the transport mechanisms in the proximal tubule is not saturated. Its extraction, E_{PAH}, which equals $(P_A - P_{RV})/P_A$, is 0.9. Thus 90% is excreted and 10% remains in renal venous plasma. We can, therefore, write

$$\mathbf{RPF = UV / (P_A - P_{RV}) = (P_A / (P_A - P_{RV})) \times (UV / P_A) = (1 / E) \times (UV / P_A).}$$

Hence three practical possibilities for the determination of RPF are:

(a) Measure U, V, P_A and P_{RV}.

(b) Measure U, V, P_A for PAH and assume E=0.9.

(c) Measure U, V, and P_A (for which mixed venous blood suffices), assume E=1.0 and use the formula ERPF = UV/P, where ERPF stands for *effective renal plasma flow* (and is, of course, the clearance of PAH). This assumes that all PAH that reaches tubular epithelium is secreted and that the other 10% of renal plasma flow bypasses the nephrons. This is unlikely. Alternatively diodone can be used instead of PAH. *Diodone* (introduced into radiology because its high content of iodine makes it radio-opaque) labelled with ^{131}I is easier to estimate than PAH and is also vigorously secreted, but its extraction (0.73) is less complete.

Typical results for ERPF in an adult man are 630 mL/min estimated from UV/P for PAH. Assuming E=0.9, then RPF=630/0.9=700 mL/min. If packed cell volume is 0.44, *total renal blood flow (RBF)* is RPF/(1-PCV)=700/(1-0.44)=700/0.56=1250 mL/min. Thus the kidneys, which form 0.5% body weight but account for nearly 10% of the total oxygen consumption of the body, receive 20% of the resting cardiac output. Renal arteriovenous difference for oxygen is low. The large blood supply reflects the need to supply fluid for filtration, not to supply oxygen for renal metabolism.

Variations in and Control of Renal Blood Flow

Experiments measuring transit times of dyes, wash-out of gases like ^{85}Kr and lodgement of microspheres in the renal vessels in experimental animals, suggest that the cortex receives 94%, the outer medulla 5% and the inner medulla 1% of the total RBF.

Between 80 and 180 mmHg arterial blood pressure as such has little effect on RBF. This is because of *autoregulation* which is believed to be mainly myogenic, the arterioles contracting when transmural pressure increases. Severe reductions of arterial blood pressure as in shock will depress RBF. Sympathetic vasoconstrictor activity is low at rest (warm, recumbent, relaxed) but increases with changes of posture, cold, pain, emotion and exercise and so reduces RBF. The reductions associated with the erect posture and exercise are often exaggerated in patients with cardiac failure. Pathological processes that destroy nephrons clinically reduce RBF.

The renal regulation of the volume and composition of the body fluids involves three processes: (i) filtration at the glomerulus, (ii) tubular reabsorption, and (iii) tubular secretion. For any substance the algebraic sum of the amounts filtered, reabsorbed and excreted by the tubules is equal to the amount excreted in the urine.

Glomerular Function

In capillary tufts blood is exposed at about 40% of mean aortic pressure to a filtering membrane of over 1 m^2 (more than half the external surface area of the body) which separates the plasma from *Bowman's space*. The capillary endothelium is *fenestrated.* Its basal lamina (0.2 to 0.3 microm thick) is composed of loose fibrillar glycoproteins with fixed negative charges. Slit pores between the foot processes (pedicels) of investing podocytes (specialized epithelial cells) provide a diffusion path between plasma and Bowman's space which does not cross cell membranes of cytoplasm. Solutes up to 10000 M_r pass through the filter freely. For these solutes, which include the ions and metabolites of the extracellular fluid, the concentrations in the filtrate in Bowman's space equal those in the plasma (slightly modified for ions by the Gibbs-Donnan distribution). With larger molecules diffusion is increasingly restricted and ceases around 70000 (for albumin, an elongated molecule with negative charge) to 100000 (for uncharged molecules). For the same head of hydrostatic pressure, an ultrafiltrate passes through this barrier about 100 times faster than through capillaries elsewhere. Thus glomerular capillaries behave as if they possessed pores of the same size as in muscle capillaries but occupying 10% instead of 0.1% of their surface. Glomerular capillaries are also unlike others in that they form a capillary bed in the course of an arteriole. Contrast this with the situation in other capillaries where arteriolar and venular ends show the Starling equilibrium between ultrafiltration and osmosis. In Munich Wistar rats, which are unusual in that their glomerular capillaries are accessible to micropuncture, hydrostatic pressures around 45 mmHg have been measured, with little drop from the afferent to the efferent end.

The glomerular filtration rate (GFR) should be equal to

$$\mathbf{J_v = L_pA((P_{gc} - P_t) - pi_{gc}),}$$

where P_{gc} and P_t are the hydrostatic pressures in glomerular capillaries (45 mmHg) and in Bowman's space (10 mmHg), and pi_{gc} is the colloid osmotic pressure in glomerular capillaries (25 mmHg at their afferent arteriolar ends).

Substituting the measured values of hydrostatic pressure,

$$\mathbf{GFR = L_pA((45\text{-}10)\text{-}pi_{gc}),}$$

which indicates that filtration should continue until pi_{gc} rises to about 35 mmHg. Such increases have been measured in some strains, so that glomerular filtration may not continue along the whole length of their glomerular capillaries. In other species, including man, so large an increase in colloid osmotic pressure cannot be expected because relatively less ultrafiltrate is formed from the plasma.

Regardless of whether filtration proceeds to equilibrium, it is generally accepted that the glomeruli continually produce and ultrafiltrate (less pedantically by common consent called 'filtrate'), and that this is the first step in the production of the urine. Note that the energy required for filtration is supplied by the heart, not by the kidney.

Measurement of Glomerular Filtration Rate

For a substance which has the same concentration in glomerular filtrate and plasma *and is neither removed from nor added to the urine by the tubular epithelium*, the amount filtered per minute, *the filtered load,* must equal the amount excreted per minute, i.e. P x GFR = UV or GFR = UV/P. where P and U are concentrations in plasma and urine *in the same units,* and V is the volume of urine produced per minute.

In dogs inulin (polyfructosan, M 5000), ferrocyanide, creatinine, mannitol and thiosulphate all have the same value of UV/P when excreted simultaneously. Excretion at the same rate for identical concentrations in plasma argues against metabolic handling by cells and favours a common physical process such as filtration through glomerular capillaries and simple conduction along tubules. Other substances which are excreted more rapidly (because tubule cells add them to the urine) or more slowly (because tubule cells remove them) have larger or smaller values of UV/P which approach that for inulin if tubular cells are poisoned or chilled, or if active transport is saturated by overloading at high plasma concentration. *Inulin* is regarded as the most reliable test substance for the measurement of GFR, but accurate measurement needs continuous infusion to establish a steady state with constant plasma concentration and rate of excretion. Simpler methods based on single injections, usually of radioactively labelled substances, may serve for routine use. Note that, since the amount of inulin excreted is directly proportional to plasma concentration, the plot of UV/P against plasma concentration gives a line parallel to the x-axis, i.e. UV/P for a substance which is filtered and neither reabsorbed nor secreted is independent of its concentration in the plasma.

Glomerular Filtration Rate in Man

In adults GFR is about 125 mL/min or 180 L/day, of the order of fifty times the volume of plasma in the body. This value represents the sum of the contributions of the individual nephrons from both kidneys. About 650 mL of plasma flows through the kidneys each min (renal plasma flow, RPF). Of this about one-fifth is filtered. The remaining four-fifths pass into the peritubular capillaries. That is to say, the ratio GFR/RPF, called the *filtration fraction,* is about 0.2. Contrast this with the ratio of ultrafiltration to flow through typical systemic capillaries, which is about 0.005. GFR is better related to body surface area than to weight. From about 2 years of age until after middle age, when the GFR declines slowly, the average value is 120 mL/min for each 1.73 m^2 (average body surface area). With 1 million nephrons in each kidney the average filtration rate per nephron (single nephron glomerular filtration rate comes to 60 nanoL/min or 90 microL/day.

Creatinine, produced in the body from the creatine of skeletal muscle, is often used in man to estimate GFR. It is produced at a relatively constant rate throughout the day so that a 24 h-urine collection and one plasma sample may be used for the calculation. In fact, renal

tubules do secrete a little creatinine but, fortuitously, plasma creatinine is overestimated by the usual chemical method because plasma contains a non-filtered chromogen which reacts as creatinine. Thus the ratio UV/P for creatinine approximates that for inulin.

Variations in Glomerular Filtration Rate

The constancy of GFR was probably over-emphasized by early measurements with inulin. Like renal blood flow GFR shows evidence of autoregulation when arterial blood pressure alters, although if arterial blood pressure falls below about 60 mmHg, as in shock, it ceases, leading to *anuria.* The erect posture, emotion, pain, cold, exercise and loss of blood are all associated with reductions in GFR - especially the erect posture and exercise in some patients with cardiac failure. Pathological processes which destroy nephrons reduce GFR, and GFR may increase when blood and extracellular fluid volumes are expanded, especially with saline, which dilutes plasma protein and lowers colloid osmotic pressure.

Note that the whole volume of plasma is filtered many times daily. This effectively removes waste products from the blood, but in so doing removes water and all solutes of low molecular weight at the same time. Hence a major task of the tubules must be recovery of water and solutes needed by the body.

Clearance

The expression UV/P is called *renal plasma clearance* or *clearance* for short. It has the dimensions of volume/unit time. The volume calculated from the clearance expression is the smallest volume of plasma that could yield the amount excreted per unit time if the kidney cleared the substance completely from that volume as it passed through them. The concept of clearance is useful for comparing the renal handling of different substances. The clearance of inulin, as we have seen, estimates GFR, and that of PAH renal plasma flow. If a substance that is filtered has a clearance less than that of inulin, then there must be a net reabsorption of that substance within the renal tubules. If its clearance is greater than that of inulin then there must be a net secretion by the tubular cells into the tubular fluid. This comparison can be made formally by calculating the *clearance ratio* i.e. clearance of X (C_x)/clearance of inulin (C_{In}), which compares the amounts of a substance in the urine and in the glomerular filtrate from which the urine was formed.

C_x / C_{In} = (Amount of X excreted / Amount of X filtered) in a period of time.

Hence if C_x/C_{In} is less than 1.0, there is less X in the urine that was filtered, i.e. X is *reabsorbed,* while if it is greater than 1.0, there is more X in the urine than was filtered, i.e. X is *secreted* by the tubules as well as being filtered.

Tubular Function

Tubules may reabsorb or secrete. Both processes may be either active or passive. Before discussing some examples of each process, the main factors affecting tubular function need to be summarized. The rate of epithelial transport is determined by surface area x flux, since flux is defined as amount moved/unit time x unit area.

The available surface area is enormous and offers a very favourable ration of surface area to volume (each tubule handles only about $1/(2x10^6)$ of the total volume filtered per min). In addition, especially in the proximal tubules, there is a very extensive brush border on the apical surface. In contrast to the situation in the gut, there is no motility to control contact time. Normally, the rate of filtration is such that the load presented to the tubules does not exceed their capacity to deal with it. However, if more solute than normal is filtered, or if tubular reabsorption is depressed, the increased flow rate downstream may overload the reabsorptive mechanisms for Na^+ in the distal nephron and result in an increased excretion of sodium chloride and water (*osmotic diuresis*).

In the case of passive absorption the flux of solutes depends upon the properties of the plasma membranes of the epithelial cells and of the tight junctions. The proximal tubule is lined by a typical *'leaky' epithelium.* Glucose, amino acids, phosphate and Cl^- are co-transported across the apical plasma membrane coupled to Na^+ transport and driven by the energy inherent in the electrochemical gradient for Na^+. They pass across the basolateral membrane by either simple or facilitated diffusion. Isoosmotic absorption occurs in this segment as expected for 'leaky' epithelia. The epithelia lining more distal portions of the nephron show various degrees of 'tightness' as will be discussed later. The gradient for passive absorption reflects, in part, the rate of removal of reabsorbed solutes by the peritubular capillaries. This is rapid and effective in the cortex but the arrangement of the vasa recta in long loops results in accumulation of reabsorbed solutes within the medulla.

Tubular Reabsorption

(a) *Urea.* Depending upon the circumstances, the clearance ratio for urea may vary from 0.3 to 0.7, i.e. 30-70% of the filtered load is excreted. Reabsorption of urea is always passive, from a higher concentration in the urine to a lower concentration in plasma, and is greatest when urine volume per minute is small, i.e. when urine is most concentrated and the diffusion gradient steepest. At urine flow rates greater than about 2 mL/min the clearance of urea is relatively independent of flow rate and is about two-thirds of the inulin clearance. Note that plasma urea concentration does not necessarily provide a good indication of renal function for it is dependent upon the rate of production of urea (reflecting protein intake and catabolism) as well as on renal excretion.

(b) *Glucose.* The clearance ratio of glucose normally is zero. Reabsorption is 'uphill' from the tubular fluid, where the glucose concentration reaches zero along the proximal tubule, to 5 mmol/L in the plasma. Reabsorption involves co-transport with Na^+ at the luminal membrane, is inhibited by phlorhizin and shows saturation effects typical of carrier-mediated transport. The characteristics of glucose handling are illustrated.

If the filtered load is increased by raising the plasma glucose concentration, the transport mechanism in some tubules becomes saturated and glucose begins to appear in the urine. When the transport mechanism in all the tubules is saturated the maximal reabsorptive capacity of the tubules (*tubular maximum, T_m)* has been reached. In man, when GFR is normal at about 125 mL/min, the 'threshold' plasma concentration required for glucose to appear in the urine (the *plasma threshold*) is some 10 to 12 mmol/L. Hence the transport mechanism begins to be saturated when the filtered load reaches 10 x 0.25 = 1.25 mmol/min. At plasma

glucose concentrations of about 15 mmol/L, the T_m is reached. It is thus about 1.9 mmol/min (15 x 0.125). Note that the plasma threshold reflects the limit of the rate of tubular reabsorption, it is not a fixed plasma concentration. Because filtered load equals $P_{glucose}$ x GFR, threshold concentration is inversely proportional to GFR. With GFR reduced from 125 to 50 mL/min it would take 30 not 15 mmol/L of plasma glucose to reach a critical load of 1.9 mmol/min.

The commonest cause of *glycosuria* is diabetes mellitus, in which plasma glucose concentration is abnormally high. *Renal glycosuria* occurs as an uncommon anomaly when tubular reabsorptive capacity is subnormal. Here plasma threshold will be low so that glucose appears in the urine though its concentration in the blood is not abnormally high.

(c) *Calcium.* About 50% (or 1.25 mmol/L) of calcium is bound to plasma proteins and cannot be filtered. The remainder, mostly ionized, is filtered. Of the filtered load of about 200 mmol per day, only 15 mmol or so is excreted. Some 60% is reabsorbed proximally, most of the remainder in the ascending limbs of the loop of Henle, distal tubules and collecting ducts. The mechanisms involved are not understood, though there may be some relationship between sodium and calcium reabsorption. *Parathyroid hormone* stimulates reabsorption in the distal tubule though it inhibits reabsorption proximally. Since it raises plasma ionized calcium, its net effect is often to promote urinary calcium excretion.

(d) *Phosphate.* Some 95% of the filtered inorganic phosphate is reabsorbed by co-transport with sodium, predominantly in the proximal convoluted tubule with the threshold a little above normal plasma concentration. Reabsorption is inhibited by parathyroid hormone.

(e) *Uric acid.* Filtered uric acid is mostly reabsorbed from the proximal tubule but there is also evidence of secretion from plasma to tubular fluid in this same segment.

(f) *Amino acids.* About 98% of the filtered load is reabsorbed from the proximal convoluted tubules by co-transport with sodium. There is evidence of competition within, but not between, five groups: (i) neutral amino acids, (ii) imino acids, (iii) basic amino acids and cystine, (iv) glutamic and aspartic acids and (v) glycine. *Aminoacidurias,* in which large amounts of particular amino acids are lost in the urine, result from deficiencies in specific enzymes, often genetically determined and frequently associated with abnormal metabolism of the same amino acids.

Tubular Secretion

(a) *Foreign substances.* Para-aminohippurate (PAH), diodone and some penicillins have clearance ratios of about 5; phenol red (partly bound to plasma protein and incompletely filtered) of 3 to 4. These substances are secreted by the proximal tubule almost as rapidly as the plasma presents them to the epithelial cells. Secretion is active, steeply 'uphill', and shows saturation, competition between substrates, and inhibition, all typical of carrier-mediated transport. The renal handling of PAH is summarized.

(b) *Physiologic substances.* Metabolic end-products, for example methylnicotinamide, uric acid, urobilin, aromatic sulphates, steroid glucuronides and choline, and autoacoids like

adrenaline, noradrenaline, acetylcholine and histamine are added to the urine by tubular secretion. Most of the urinary K^+ is secreted by distal tubular cells. H^+ and NH_3 are important urinary constituents which are not transferred from the plasma but produced by the tubular cells.

Renal Handling of Water and Sodium

Filtration

With a GFR of 120 mL/min and a plasma Na^+ concentration of 150 mmol/L, the filtered load of Na^+ is some 18 mmol/min or 26000 mmol per 24 h. Clearly changes in either GFR or plasma concentration will change the load presented to the tubules.

Proximal Tubules

About two-thirds of the filtered water and sodium are reabsorbed from the proximal tubules each minute, i.e. 120 L per 24 h and 17500 mmol per 24 h respectively. The sodium enters the cells across the apical plasma membrane down its electrochemical potential gradient in association with either co-transported solutes or counter-transported hydrogenium ions, and is extruded across the basolateral membrane against its electrochemical gradient by Na-K-ATPase. The sodium accumulated locally (with chloride to maintain electroneutrality) in the lateral intercellular spaces provides the osmotic force for absorbing salt and water isosmotically. The hydraulic conductivity of this segment of the nephron is so large that an osmotic imbalance as little as 1 to 2 mosm/kg of water can account for the observed rate of reabsorption. It seems likely that water follows both cellular and paracellular routes in moving from lumen to interstitium. Reabsorption of salt and water from later portions of the proximal tubules may be assisted by the increased chloride concentration resulting from reabsorption of other solutes (glucose, amino acids, bicarbonate) upstream. The increased chloride concentration in the lumen favours paracellular passive diffusion of chloride, along with sodium to maintain electroneutrality, and may contribute to salt absorption in this segment.

The amounts of salt and water reabsorbed from the proximal tubule are also influenced by the balance of hydrostatic and osmotic forces in the peritubular capillaries. An increased colloid osmotic or a decreased hydrostatic pressure in peritubular capillaries favours uptake of interstitial fluid into capillaries and assists reabsorption. Conversely, decreased colloid osmotic pressure or increased hydrostatic pressure hinders reabsorption. For example, if GFR increases with constant RPF, the filtration fraction will increase and the increased colloid osmotic pressure in the peritubular capillaries will assist in reabsorbing the increased volume of filtrate. Proximal tubular fluid reabsorption matches GFR closely over a wide range of GFR and such adjustments of capillary forces help to account for this *'glomerulo-tubular balance'*.

Loops of Henle

A volume of about 60 L containing 9000 mmol sodium leaves the proximal tubules and enters the *descending limbs of the loops of Henle* each day as an isosmotic solution. Those loops from the juxtamedullary nephrons which run deep into the medulla (some 20% in the human kidney) pass through a region in which the interstitial fluid osmolality increases

progressively from isosmotic (285 mosmol/kg water) at the corticomedullary junction to some 1200 to 1400 mosmol/kg water at the tip of the renal papilla in man. In antidiuresis sodium chloride and urea contribute in similar proportions to this gradient. The mechanism by which it is generated and sustained is discussed later. The epithelium lining this segment is quite permeable to water, but less so to sodium and chloride, and impermeable to urea. Therefore, water moves passively and progressively from the descending limb to the interstitium as fluid flows along the tubules, and some sodium and chloride enter the tubule. Each day there is a net removal of about 10 L of water from this tubular segment, much of it from the short descending limbs of cortical nephrons. There is probably no active transepithelial transport of solutes in the descending limbs.

In the medullary descending limbs fluid has equilibrated with the adjacent interstitial fluid and at the tips of the loops now has an osmolality of some 1200 mosmol/kg water, having lost water and also gained some sodium and chloride.

The *thin ascending limb of the loop of Henle* differs significantly from the thin descending limb in its permeability, being impermeable to water, highly permeable to sodium and chloride and moderately permeable to urea. Consequently, sodium chloride diffuses from the tubules to the interstitium as the fluid flows back towards the cortex, and some urea enters the tubules down its concentration gradient. By the time that the thick ascending segment is reached, much of the sodium and chloride gained in the descending limbs as the fluid flowed deeper into the medulla has been lost again passively to the interstitium, and the fluid has become somewhat hypo-osmotic compared with the adjacent interstitial fluid.

Unlike the thin ascending limb, the *thick ascending limb*, and its continuation in the cortex as the first part of the distal tubule, avidly reabsorb sodium chloride from the tubular fluid by an energy-dependent process. The unusual finding that, in this segment of the nephron, the interior of the lumen is positive with respect to the interstitium, led to the proposal that chloride rather than sodium was transported actively from the lumen. It is now realized that a combination of co-transport of chloride at the apical membrane with passive diffusion at the basolateral membrane can account for this electrical phenomenon. A possible model of transport in this segment is shown. The impermeability to water means that the removal of some 6500 mmol of sodium chloride per day (25% of the filtered load) dilutes the luminal fluid which becomes hypo-osmotic. Since in total 60 L of fluid with 9000 mmol of sodium entered all of the loops of Henle and 50 L with 2500 mmol returned to the distal tubules, sodium concentration is now about 50 mmol/L and osmolality somewhat over 100 mosmol/kg water.

Distal Tubules

It is now appreciated that the distal tubule is not a homogeneous segment. The first part is lined by an epithelium of the same type as the adjacent ascending limb of the loop of Henle and the two together comprise the *diluting segment* of the nephron. The last part is analogous in its properties to the collecting tubule which it joins. Sodium chloride is reabsorbed throughout the length of the distal tubule, some 1200 mmol of sodium (5% of the filtered load) being removed from the tubular fluid each day. The permeability of the tubule to water reflects its heterogeneity. While the early part is impermeable to water, the permeability of the last part is determined by the level of circulating *antidiuretic hormone*

(ADH). With normal hydration some 20 to 30 L of water per day are probably absorbed by the distal tubule, leaving 30 to 20 L containing some 800 mmol of sodium to pass through the collecting ducts. The distal tubule is impermeable to urea. Therefore urea entering the thin ascending limb of the loop of Henle is retained within the tubule in this portion of the nephron.

Collecting Ducts

Though only about 20% of the loops of Henle run deep into the medulla, all of the fluid remaining at the end of the distal tubules enters the collecting duct system which runs through the hyperosmotic medulla to drain into the renal pelvis and ureters. The cells lining the collecting ducts form a *'tight' epithelium.* Active sodium reabsorption continues here and sodium concentration in the urine may fall to as low as 10-15 mmol/L. On a normal diet about 600 mmol is reabsorbed each day and this reabsorption is largely independent of water handling in this segment. *Water diuresis* (the production of a large volume of a dilute urine) is associated with low circulating levels of ADH. In the absence of this hormone (as in the rare disease diabetes insipidus) epithelial water permeability is very low, little water absorption will occur and some patients have been known to excrete as much as 30-40 L of urine per day (a flow rate of about 25 mL/min) with a urine osmolality lower than 50 mosomol/kg water. ADH acts via adenylate cyclase and cyclic and cyclic AMP to increase the water permeability of the apical plasma membrane of the epithelial cells, opening a cellular, not a paracellular, pathway for water to move down the osmotic gradient from tubular lumen to interstitial fluid.

ADH increases water permeability of both cortical and medullary portions of the collecting ducts, as well as of the most distal portion of the distal tubules. With maximal plasma concentrations of ADH (*maximal antidiuresis*) as little as 0.4 to 0.5 L of urine is excreted per day (about 0.3 mL/min) with a urine osmolality about 1200 mosmol/l water. ADH also increases the permeability of the medullary (but not cortical) collecting ducts to urea and promotes its reabsorption. Therefore in antidiuresis the urea that passed from the medulla into the thin ascending limb of the loop of Henle then diffuses back into the medullary interstitium, and its recycling between medullary interstitium and tubular fluid helps to preserve medullary hyperosmolality. During water diuresis, the loss in the urine of much of the urea passing into the ascending limb contributes substantially to the reduction in the medullary osmotic gradient under these conditions. The figure summarizes the handling of sodium and water by the different tubular segments.

Generation and Maintenance of the Medullary Hyperosmotic Gradient

The generation of the gradient of medullary hyperosmolality depends primarily upon the energy-dependent reabsorption of 25% of the filtered load of NaCl in a water-impermeable region of the tubule - the thick ascending loop of the loop of Henle. The solute so reabsorbed would be washed away from the medulla were it not for the fact that the capillaries within the medulla are arranged in loops - the vasa recta, the descending and ascending limbs of which are close to each other and to the adjacent loops of Henle. Reabsorbed solute diffusing into an ascending capillary loop will tend to increase the concentration of NaCl in the capillary at that point. As it is carried towards the cortex, it flows past plasma in the adjacent descending capillary loop in which the NaCl concentration is lower. At every level there will

be a tendency, therefore, for NaCl to diffuse from the ascending to the descending capillary loop and thus to be retained within the renal medulla rather than lost to the cortex. Similarly, at every level, plasma in the descending limb coming from a more dilute region of the medulla will be slightly less hyperosmotic than plasma in the ascending limb which is emerging from the deeper, more hyperosmotic medulla. Thus, in contrast to solute, water will tend at every level to pass from the descending to the ascending limb down its osmotic gradient and, thereby, be shunted away from the deeper medulla. Urea, passing from the medullary collecting ducts into the interstitium during antidiuresis, will also be trapped within the medulla by diffusion from ascending to descending vasa recta. These diffusional movements between the two limbs of the vasa recta are often described as countercurrent exchange. Note that the arrangement of the vasa recta allows the medullary gradient to be maintained but were it not for the sources of water-free solute (from energy-dependent NaCl reabsorption in the thick ascending limb and from urea diffusion from the collecting ducts into the medulla during antidiuresis) the large osmotic gradient could not be created. The effect of the latter process is often referred to as countercurrent multiplication. What in effect happens is that some of the solute filtered at the glomerulus with water as an isosomotic solution has been trapped in the medulla while its associated water is either lost from the distal tubule in the cortex or excreted in the urine.

Diuresis

An increased rate of production of urine (diuresis) can be of two types - water diuresis and solute or osmotic diuresis. *Water diuresis* results when water is ingested or administered in excess of the body's requirements. ADH secretion is suppressed, the collecting ducts become relatively impermeable to water and the excess water is lost without solute. A typical water diuresis is illustrated. There is an inverse relationship between urine osmolality and urine flow rate. Flow rate x osmolality (the amount of solute excreted per min) is relatively constant and independent of flow rate. Thus the kidney can adjust its excretion of water without markedly affecting its handling of solutes.

Osmotic diuresis results when more solute is presented to the tubules than they reabsorb, for example, if a non-reabsorbable solute, i.e. mannitol, is filtered, or if the concentration of glucose in the plasma in diabetes mellitus rises so that the filtered load exceeds the tubular maximum, or if tubular function is inhibited (i.e. by drugs which block reabsorption of NaCl in one or more nephron segments). A typical osmotic diuresis is illustrated. In contrast to water diuresis, urinary flow rate depends upon primary solute content. Moreover, the greater the rate of solute excretion, the lower is the maximal attainable urinary concentration even with maximal concentrations of ADH, for the larger volume of water reabsorbed from the collecting ducts dilutes the medullary interstitial fluid. Faster flow through the ascending limbs of loops of Henle, especially combined with a decreased concentration of sodium when glucose or mannitol are present, may also decrease reabsorption so that less sodium is deposited to maintain the medullary osmotic gradient. This gradient cannot be demonstrated in kidneys removed during osmotic diuresis.

The roles of the different segments of the nephron are summarized in Table 13.1. Micturition is dealt with in Chapter 6. Adjustments of salt and water handling to meet the changing needs of the body and the role of the kidney in potassium homeostasis are included in the following sections.

13. Body Water and Electrolyts

Water

Distribution of Water in the Body

As outlined in Chapter 1, the average 70 kg man has a total body water of about 42 L of which 55% (23 L) is in the cells and 45% (19 L) is extracellular. This extracellular water is further subdivided into plasma (3 L), interstitial fluid (10 L) and transcellular fluids (CSF, ocular, pleural, peritoneal and synovial fluids) (1 L). The volumes of these compartments can be estimated from the volumes of distribution of substances thought to equilibrate in different compartments. For example, total body water has been estimated from the volume of distribution of urea and isotopes of water (deuterium oxide, tritiated water). Similarly, inulin, sucrose, mannitol and isotopes of sodium and chloride have been used to estimate extracellular water, and isotopically-labelled albumin to estimate plasma water. Interstitial water cannot be measured directly but is (ignoring the transcellular volume) the difference between extracellular volume and plasma volume.

The best guide to changes in body water is alteration in body weight, because water constitutes about 60% of body weight. In estimating body water from body weight, however, it must be remembered that fat contains very little water, so that the greater the body fat as a proportion of body weight the lower the body water content. Thus women, in general, have a lower body water content than men of the same body weight.

Water Balance

Normally total body water remains constant. Therefore, over a 24 h period, intake and loss of water must balance exactly. Both intake and loss are controlled, through the thirst-ADH mechanism.

Water is taken in as a liquid and in food (about 1 L per 24 h). It is also formed from the oxidation of metabolites (about 300 mL per 24 h).

Water is lost through the skin (0.5 L) and lungs (0.5 L) by evaporation and, depending upon the need to increase heat loss, as sweat. Faeces contain about 0.1 L of water per 24 h. Urinary water loss is variable. About 600 mOsmol of solutes, including end-products of metabolism, must be excreted each day in the urine. The maximal achievable urinary osmolality of about 1200 mOsmol/kg water therefore demands a minimal urinary volume of 0.4 to 0.5 L per day. Normally water intake is such that about 1.5 L of urine is excreted each day.

Regulation of Total Body Water

If water loss exceeds gain, with a reduction in total body water content, the osmolality of the body fluids increases. This excites *thirst*, so that water may be ingested, and releases ADH so that water is retained by the kidneys. Conversely, an excess intake of water expands and dilutes the body fluids. The decrease in osmolality eliminates thirst and inhibits the release of ADH, so that water diuresis rids the body of the excess. These mechanisms can

hold the osmolalities of plasma and other body fluids constant to within +/- 1% of 285 mOsmol/kg water.

Both thirst and the release of ADH appear to be triggered by the same mechanisms. Of prime importance, normally, is the osmolality of the plasma perfusing the hypothalamus. Neurones in the supraoptic nuclei synthesize ADH which is stored with neurophysin in the nerve endings in the neurohypophysis. The 'blood-brain barrier' is deficient in this region, and these neurones, or nearby 'osmoreceptors' which relay to them, respond to a local increase in plasma sodium concentration. If plasma osmolality is increased by solutes which penetrate plasma membranes, i.e. urea or ethyl alcohol, thirst and ADH release are not initiated. This suggests that changes in the volumes of the osmoreceptor cells link the changes in plasma osmolality to the response. Other mechanisms which may stimulate thirst and ADH release include (i) increased NaCl concentration in the CSF in the third ventricle, (ii) increased angiotensin II in the brain (produced locally or delivered via the systemic circulation), (iii) decreased arterial blood pressure, signalled via the carotid and aortic baroreceptors and (iv) decreased central venous pressure, signalled via the low pressure receptors in atria and great veins. The latter two mechanisms, especially (iv), are important in conditions where circulating blood volume is deficient. Water may even be retained in excess of solute in response to depleted circulating volume and the control of extracellular osmolality may be sacrificed. The figure illustrates the relationship between plasma osmolality, circulating ADH and changes in circulating blood volume.

Disturbances of Total Body Water Content

Hypernatraemia. The effectiveness of the thirst-ADH mechanism ensures that a primary loss of water (primary dehydration) only occurs when fluid intake is not possible (i.e. in the absence of a source of water, in the very young, the aged, the confused, the unconscious or in patients with severe vomiting who cannot drink and retain water). It is most likely to be seen when loss of water is excessive, i.e. with hyperventilation (particularly at high altitude where humidity is low), high environmental temperature, fever, or abnormal urinary loss either from a failure to produce or secrete ADH (neurogenic diabetes insipidus) or from a decreased responsiveness of the collecting ducts to ADH (nephrogenic diabetes insipidus).

The loss of water increases the osmolality in all fluid compartments and the concentration of sodium in the plasma (hypernatraemia). The loss is shared between the cells and the extracellular compartment, so that cell volume is decreased but extracellular volume is better preserved than when there is a loss of extracellular sodium. Nevertheless, the contraction of extracellular volume stimulates aldosterone secretion promoting renal retention of sodium.

Apart from thirst which may dominate consciousness, symptoms are vague, i.e. weakness and lethargy. The urine is scanty and concentrated except in diabetes insipidus. Plasma osmolality and sodium concentration are increased. It is important that the lost water be replaced by mouth or, if necessary, intravenously (as 5% glucose). In neurogenic diabetes insipidus ADH is also administered.

Hyponatraemia. This occurs with consumption or administration of inappropriately large volumes of water when the renal excretion of water is impaired, i.e. in anuria, with inappropriate secretion of ADH as in trauma, and with neoplasms that produce ADH. Hyponatraemia may also follow excessive loss of extracellular fluid associated with loss of blood or of sodium when the thirst-ADH mechanism is stimulated through the low pressure central venous stretch receptors, and the maintenance of normal osmolality is sacrificed in an effort to maintain circulating volume.

As the retention of water lower extracellular osmolality and plasma sodium concentration, water moves from the extracellular fluid into the cells, which swell until the osmolalities of the fluid compartments are equalized.

Depending on the degree of water retention, weakness and muscle cramps may develop, followed by confusion ('water intoxication'), convulsions, coma and death. The plasma hypo-osmolality and low plasma sodium concentration can be reversed by restricting water intake and treating the underlying cause of the condition.

Note that, unlike those of other mammalian organs, the brain cells seem to be able to regulate their volume in anisosomotic media by adjusting their solute content. In hypernatraemia they shrink initially but gradually increase their solute content over the next few hours and take up water to restore their volume towards normal. Conversely, in hyponatraemia they first swell but then lose solute and with it water, again partly restoring their volume. This ability to adjust volume has obvious value for an organ enclosed in a rigid container (the skull). However it means that hyper- and hyponatraemia ought never to be reversed to rapidly. For example, in hypernatraemia the cells contain more solute than normal and a sudden restoration of normal plasma osmolality will result in pronounced swelling of these cells, raised intracranial pressure and symptoms and signs like those of hyponatraemia.

Sodium

Distribution of Sodium in the Body

Sodium is the major cation of the extracellular fluid. A typical 70 kg man, with an extracellular sodium concentration of 150 mmol/L and an extracellular volume of 19 L, would have 2850 mmol sodium in the extracellular compartment. With an average concentration of 15 mmol/L, 23 L of cellular water would contain 345 mmol of sodium. The bones contain a further 2500 mmol of sodium, less than half of which is readily exchangeable with extracellular sodium or other cations. None of this contributes to body fluid osmolality. Total exchangeable sodium, measured by isotope dilution, is about 50 mmol/kg body weight.

Sodium Balance

On Western diets 50 to 300 mmol sodium are consumed daily. Almost all of this is absorbed from the gut (together with the much larger amount secreted into the gut) and the faeces normally contains only 5-10 mmol daily. The only additional route for sodium loss, other than the renal, is from the skin in sweat. This loss is extremely variable and depends solely upon the need to maintain a relatively constant central body temperature. Each litre of sweat contains 30-50 mmol of sodium so that the loss of a few litres of sweat can cause a

significant sodium loss from the extracellular compartment. Renal loss of sodium is adjusted to maintain sodium balance and can range from a few mmol up to about 500 mmol per day.

Regulation of Body Sodium

Most of the 'mobile' sodium that can vary in amount from day to day is in solution in the extracellular fluid. Hence day-to-day variations in the amount of body sodium represent variations in extracellular fluid volume. For every 150 mmol in sodium content there is a corresponding change of 1 litre in extracellular fluid volume. Indeed, *the volume of the extracellular fluid is regulated by regulating sodium content.*

Unlike the regulation of total body water through the thirst-ADH mechanism, which is comparatively well understood, the mechanisms involved in the regulation of sodium and thereby extracellular volume remain controversial. A brief simplified outline is all that can be provided here.

Regulation requires the monitoring of some function of the variable to be controlled, central coordination, and the appropriate control of an effector organ, in this case the kidney.

Sodium concentration is already fixed through the thirst-ADH mechanism, and neither total extracellular sodium nor volume is monitored directly. Rather, the volume of a sub-compartment - *central venous volume* - is monitored through the low pressure stretch receptors in the central veins and atria. But how can the volume of this sub-compartment adequately reflect total extracellular volume? Most of the ECF is interstitial fluid which forms a weak gel with mucopolysaccharides, largely hyaluronic acid, and does not move under gravitational forces as a free fluid, though it offers no hindrance to diffusion. One hypothesis proposes that this interstitial gel is unsaturated, because pressures as low as 6 mmHg below atmospheric have been measured within it. 'Imbibition' pressure reflects the tendency for the gel to take up and thereby immobilize fluid, and while the gel remains unsaturated the vascular system and extravascular interstitial spaces appear to have similar compliances. Consequently fluid gained or lost from the extracellular compartment will be shared between plasma and interstitial fluid, and monitoring the volume of one will monitor the volume of both. However, if sufficient fluid accumulates to saturate the gel (3-4 L), additional fluid in the interstitial compartment is free to move under gravity to the most dependent parts of the body (resulting in oedema), the total extracellular volume can no longer be monitored by measuring central venous pressure, and regulation of volume breaks down.

The carotid sinus and aortic arch baroreceptors which monitor systemic arterial blood pressure, probably only contribute significantly to the regulation of extracellular fluid volume when intravascular volume is severely depleted.

Information from the atrial low-pressure stretch receptors is processed in the brain but, unlike the well-established role of the hypothalamic osmoreceptors in the thirst-ADH response, there is no established localization within the CNS for the regulation of extracellular fluid volume as such.

The efferent limb of the control mechanism regulation of the renal handling of sodium. It has become customary to describe this as involving three factors: the filtered load,

aldosterone which acts in the distal tubules, and a group of so-called 'third factors' which are believed to act in the proximal tubule. Overall, Na+ excretion = filtered load - tubular reabsorption.

Filtered load. When Na+ concentration in the plasma remains constant, filtered load varies only with GFR. Therefore factors like blood loss or loss of extracellular volume, exercise and adoption of the upright posture, which alter GFR through cardiovascular reflexes mediated by sympathetic nerves to the kidneys and by circulating catecholamines, change filtered load.

Aldosterone. Secretion of this hormone from the zona glomerulosa of the adrenal cortex is stimulated principally by circulating angiotensin II (ANG II). Of lesser importance are decreased Na+ and increased K+ concentrations in the blood perfusing the adrenal gland. ANG II is formed through the following sequence: Renin, a specific proteolytic enzyme produced in the juxtaglomerular apparatus in the kidney, splits a decapeptide angiotensin I (ANG I) off from angiotensinogen, an alpha2-globulin synthesized in the liver. ANG I is converted in turn to ANG II (an octapeptide) by *converting enzyme* found chiefly, but not entirely, in the lungs. The rate-limiting step in this sequence is normally the release of renin from the juxtaglomerular apparatus. Three major stimuli induce this release: (i) decreased arterial blood pressure which is thought to act directly on the afferent arterioles; (ii) increased sympathetic activity (including circulating catecholamines) acting through alpha adrenergic receptors, and (iii) a decrease in the rate of delivery of fluid past the macula densa in the distal tubule.

As well as promoting the secretion of aldosterone, ANG II is a potent vasoconstrictor and can increase total peripheral resistance. An increase in mean arterial pressure, however, inhibits further release of renin. ANG II also promotes thirst and stimulates the release of ADH, by acting directly on circumventricular organs which are outside the blood-brain barrier. Thus it not only stimulates Na+ retention through aldosterone, but also the acquisition and retention of water, and so plays a major role in the regulation of extracellular volume. It is inactivated by angiotensinases in the plasma.

Aldosterone promotes the reabsorption of Na+ by a variety of 'tight' epithelia, principally the distal portion of the nephron, but also the epithelium of the colon and rectum, and the ducts of salivary and of sweat glands. In all of these it promotes Na+ absorption and K+ (and in some, H+) secretion. It appears to initiate the synthesis of protein which promotes Na+ entry to the cells by facilitated diffusion across the apical plasma membrane. This increases intracellular Na+ concentration and stimulates active Na+ transport from the cells across the basolateral membrane via the Na-K-ATPase. The increased rate of entry also depolarizes the apical plasma membrane and increases the electrochemical potential gradient favouring diffusion of cations (K+ and H+) from the cells to the lumen. The effects of aldosterone on Na+ reabsorption however take about 1 h to become apparent. Therefore, this hormone is important for long-term rather than minute-to-minute adjustments of Na+ excretion. It is inactivated in the liver.

'Third factors'. In some experimental situations the renal handling of Na+ cannot be accounted for by changes in filtered load or by aldosterone. There is some evidence that changes in the balance of the Starling forces in the peritubular capillaries will alter net Na+

reabsorption in the proximal tubules. In addition, there may be hormones, *natriuretic hormones,* which inhibit Na+ reabsorption by tubular cells; 'endoxin', which may be produced by brain, and appears to be an inhibitor of the plasma membrane Na-K-ATPase, and atrial natriuretic factor (ANF) which does not inhibit the Na-K-ATPase. Also, sympathetic innervation of proximal tubular cells has been demonstrated, and stimulation may increase Na+ reabsorption there directly. The figure summarizes current knowledge of the control of Na+ excretion by the kidney.

Disturbances of Body Sodium Content

These result in changes in extracellular volume.

Oedema. Oedema is a clinically detectable excess of extravascular ECF. It may be either localized or generalized.

Localized oedema need not imply an increased amount of sodium in the body if the affected region is small. It results from local disturbance of the Starling equilibrium by (a) increased capillary hydrostatic pressure (i.e. venous stasis, thrombosis or obstruction); (b) increased pericapillary colloid osmotic pressure (i.e. with lymphatic obstruction or inflammatory exudation); or (c) increased capillary permeability, leading to increased loss of protein to the interstitial spaces (i.e. inflammatory and allergic conditions).

Generalized oedema occurs when the disturbance of the Starling equilibrium is widespread; it is associated with increased body weight, the movement of free fluid under gravity, tissue swelling and pitting on pressure. There is typically at least 4 to 5 L of excess fluid (enough to saturate the interstitial gel and overflow) and hence there must be an excessive amount of sodium in the body - 150 mmol for each litre of oedematous fluid or kg of excess weight. This disturbance cannot occur by the mere shifting of fluid from the plasma, for there is less than 4 L of plasma, and so a patient whose whole plasma volume was shifted into the tissue spaces would not be oedematous - and would also not be alive!

Two important causes of widespread loss of fluid from blood vessels are:

1. Lack of plasma protein, especially albumin, so that the colloid osmotic pressure is everywhere low. This may occur in gross undernutrition, in diseases of the liver which impair production of albumin, and when albumin is lost in the urine faster than the liver can replace it (nephrotic syndrome).

2. A widespread increase in hydrostatic pressure in the capillaries, as in chronic cardiac failure with venous congestion.

These account for the loss from the blood vessels but excessive retention of sodium by the kidneys is required to explain the continued replenishment of the plasma and accumulation of oedema. There is no single and wholly satisfactory explanation for the development of oedema in all situations. When plasma albumin is low, and there is a diminished plasma volume, RBF and GFR are reduced so that less sodium is filtered; secretion of renin is stimulated and secretion of aldosterone is increased, promoting sodium reabsorption. In liver disease breakdown of aldosterone may be slower, increasing the

circulating concentration. When plasma albumin is depleted, retained NaCl and accompanying water do not remain in the vessels to restore plasma volume, signal the increase in total ECF volume, and turn off the enhanced retention of sodium. The kidney acts in a manner appropriate for the correction of a low volume of blood and ECF.

When cardiac failure results in decreased cardiac output and increased capillary hydrostatic pressure, reflex vasoconstriction reduces RBF and GFR, so that less sodium is filtered. Renin may be released and aldosterone secretion increased, while breakdown of aldosterone in the congested liver may be slowed. Reabsorption of sodium is therefore enhanced, sodium is retained and ECF volume increases.

Retention of sodium first tends to elevate plasma sodium concentration, but this stimulates the thirst-ADH mechanism so water is retained and, in uncomplicated oedema, plasma sodium concentration and total plasma osmolality tend to be within normal limits. The reduced colloid osmolality when albumin is deficient amount to no more than 1 or 2 mosmol/kg water and this decrease is not detected when total osmolality is measured.

Since it is dietary sodium that is retained to make oedema fluid, treatment by restriction of dietary salt is logical. It is however unpleasant, and often ineffective as it tends to mobilize aldosterone, so that a variety of diuretic drugs which inhibit renal tubular reabsorption are used to combat retention of sodium.

Depletion and deficiency of sodium. This is caused by absent or diminished sodium intake often combined with excessive loss. Under extreme environmental conditions up to 15 L per day of *sweat* wit 30 to 50 mmol/L of sodium may be lost. Normally about 8 L of fluid containing up to 1 mol of sodium are secreted and reabsorbed per day in the *gastrointestinal tract.* This sodium can be lost through vomiting, aspiration of the gut contents, by diarrhoea or through fistulae. In cholera and other secretory diarrhoeas the rate of intestinal secretion may be increased enormously. Normally losses through non-renal routes are countered by the virtual disappearance of sodium from the urine. But the *urine* can become a vehicle of loss in Addison's disease (absence of aldosterone), during osmotic diuresis, and in diabetes mellitus when sodium is lost not only as a consequence of the osmotic diuresis but also with the conjugate bases of keto-acids.

So long as osmoregulation continues, loss of sodium leads to loss of water at the rate of 1 litre per 150 mmol of sodium. This loss of water is shared between the plasma and the extravascular ECF. The cell water is unaffected or may even increase if sodium concentration is depressed. Hence for a given loss of water the consequences are much more serious for the circulation than are those of primary loss of water, since the ECF and plasma bear the brunt of the water deficiency, as shown by haemoconcentration, with increase in red cell count, plasma protein concentration and viscosity, and evidence of impaired renal function with retention of urea and other waste products as a consequence of the depressed GFR. Moreover, thirst is less prominent than in pure water loss with its increased osmolality, and the cardinal manifestation is peripheral circulatory failure. There is also the danger of a vicious cycle developing if tissue perfusion falls enough for cells deprived of oxygen to swell, taking up water and sodium, and further reducing ECF volume, circulating blood volume, and perfusion of the tissues. As the loss of ECF continues, the thirst-ADH mechanism is activated as a consequence of decreased venous volume and, eventually, of decreased arterial blood pressure.

This may result in retention of water without sodium and consequently in decreased body fluid osmolality and hyponatraemia. Thus it is possible to see hyponatraemia associated with a decreased volume of total body water as well as with pure water retention with overexpansion of total body water. Note that water alone is relatively ineffective in restoring circulating blood volume. In a 70 kg man with about 23 L of cell water and 19 L of extracellular water less than half of any additional water would be retained within the extracellular compartment, and only one-sixth of this (about 75 mL for each additional litre) would remain in the plasma in the steady state. In contrast to hypernatraemia, it is sodium not water which needs to be administered either orally (with glucose to promote intestinal absorption) or as an isosmotic intravenous solution.

Potassium

Distribution of Potassium in the Body

Potassium is the chief intracellular cation of man and of the animals and plants he eats. The body of a 70 kg man contains about 3600 mmol K+ of which 80 mmol is in the skeleton, 80 mmol is in the ECF and the rest, about 95%, is in the cells, some 3000 mmol of it in skeletal muscle. Total exchangeable K+ is about 50 mmol/kg of body weight in men and about 40 mmol/kg in women whose muscles, the largest component of the cell mass, form a lower proportion of body weight. The intracellular concentration (100-150 mmol/L) is not critical but a two- or three-fold increase or decrease in the extracellular concentration (4-5 mmol/L) can paralyse muscles or stop the heart. Rapid loss of 5% of cellular potassium into the extracellular fluid would be lethal. Extracellular potassium concentrations below 2.5 or above 7 mmol/L produce weakness of limb and trunk muscles. Below 2 and above 9 mmol/L flaccid paralysis occurs and the respiratory musculature is affected and the heart may cease to beat. These changes in cardiac and skeletal muscle activity are consequences of altered K+ gradients with a normal cellular K+. Low plasma potassium concentrations (*hypokalaemia*) increase the diffusion gradient, hyperpolarizing the plasma membrane so that the nerve and muscle cells become less easily depolarized and therefore less excitable. High plasma potassium concentrations (*hyperkalaemia*) decrease the diffusion gradient, depolarizing the plasma membrane. Initially, as threshold is approached, cells may become more excitable but, once the membrane potential has fallen below threshold, action potentials can no longer be generated and paralysis results. Hence acute alterations in plasma potassium, which leave cellular potassium concentration unaltered, are less well tolerated than are chronic alterations in which cellular potassium concentrations change in the same direction and the ratio of cell potassium to plasma potassium remains relatively constant.

Potassium Balance

Ordinary diets supply about 100 mmol. The faeces remove about 10 mmol and the urine 90 mmol per day.

Regulation of Body Potassium

Though the losses of potassium, principally in the urine, maintain body potassium constant despite variations in intake, the details of this regulation are poorly understood. Following the intake of potassium, about a half is lost in the urine over the next 6 hours or

so; the remainder largely disappears from the extracellular fluid into cells and is excreted subsequently. Insulin promotes cellular uptake of potassium and a raised plasma potassium concentration stimulates release of insulin. Insulin may therefore be used in an emergency to lower plasma potassium concentration. Other hormones including adrenaline, aldosterone and glucocorticoids may also stimulate cellular potassium uptake, though their physiological importance is unclear. The acid-base balance also influences exchange of potassium between cells and extracellular fluid. During acidosis, H+ ions enter cells in exchange for K+; during alkalosis, K+ enters cells in exchange for H+ ions. These effects are more marked with metabolic than with respiratory disturbances.

The major excretory route for K+ is through the kidneys. Of the 800 mol or so of K+ filtered per day, some 80% is reabsorbed in the proximal tubule by mechanisms which are not yet fully understood. Some of this reabsorption may be a passive consequence of the abstraction of NaCl and other solutes together with water, which concentrates luminal potassium and creates a favourable gradient for diffusion through the paracellular pathway. Some may involve uptake into the cells across the apical membrane. A further 10% of the filtered load is reabsorbed in the thick ascending limb, being co-transported across the apical membrane with Na+ and Cl- as discussed earlier.

About 10% of the filtered load reaches the late distal tubule and collecting ducts. Here, in addition to reabsorption, K+ ions are secreted. Of the two cell-types lining the nephrons in this region, it seems that the *principal cells* reabsorb Na+ and secrete K+, whereas the *intercalated cells* secrete H+ ions and reabsorb K+. Factors which promote K+ secretion by the principal cells include (i) increased dietary intake of K+, (ii) aldosterone, (iii) increased rate of Na+ delivery and (iv) increased flow rate. Factors promoting K+ reabsorption by the intercalated cells include (i) decreased cellular H+ activity and (ii) a low K+ diet. The relationship between K+ secretion and H+ secretion within this tubular segment is of great importance. Increased availability of luminal Na+ here promotes secretion of both ions, the relative contributions of each reflecting the acid-base and K+ status of the individual, but we lack a detailed understanding of the mechanisms involved.

The urinary excretion of K+, therefore, reflects the relative activities of the reabsorptive and secretory processes and the clearance ratio for K+ can range between 0.1 (reabsorption predominant) and 3.5 (secretion predominant). In K+ balance, with dietary intake of 100 mmol per day, the clearance ratio ordinarily approximates 0.15.

Potassium is also secreted by epithelial cells lining the colon. Again the mechanism is unclear but uptake of Na+ from the lumen appears to stimulate secretion of K+, and aldosterone promotes both processes.

Whereas the control of water balance and, to a lesser extent, of Na+ balance are fairly well understood, there is no comparable framework for discussing the regulation of K+ balance. Are there receptors sensitive to a function of body K+? Is there central integration of information about body K+? How is the function of the effector organs (kidney and colon) regulated so that K+ excretion matches K+ intake? These questions are still unanswered. Most discussions of K+ balance imply that the K+ concentration in cells of the distal nephron and colon mirrors that of cells elsewhere so that factors affecting cellular K+ generally affect these cells as well. Then if losses of K+ from principal cells to the luminal fluid and from

colonic epithelial cells to the gut lumen are adjusted to maintain the concentration of K+ in those cells constant, body K+ content will perforce be regulated. Whether this simple view is adequate remains to be established.

Disturbances of Body Potassium Content

Hyperkalaemia. Potassium is normally excreted by the kidneys so effectively that body K+ content remains constant, any increase in intake being matched by urinary excretion. Plasma K+ concentration *does not* necessarily parallel body K+ content. Clinically hyperkalaemia is usually caused by excessive loss of cellular K+ combined with diminished renal function as, for example, in (i) shock where the fall in mean arterial pressure, results in hypoperfusion of tissues and a loss of K+ from hypoxic cells, as well as decreased renal perfusion, (ii) crush injuries where damaged cells leak K+ into the plasma and myoglobinuria disrupts renal function, and (iii) terminal renal failure where increased tissue catabolism releases K+ from cells and depressed renal function prevents its excretion. In all of these conditions plasma K+ rises because K+ is lost from cells faster than the kidneys can excrete it. Total body potassium is actually falling, though plasma K+ is elevated.

The symptoms and signs of hyperkalaemia *per se* are vague; cardiac arrhythmias and cardiac arrest may develop before excitability of nerve and skeletal muscle has been clinically affected. Typical, progressive changes in the ECG give the earliest indications of the severity of hyperkalaemia. In an emergency plasma K+ concentration may be decreased abruptly by insulin which drives K+ (and glucose) into cells. Bicarbonate also lowers plasma K+ by stimulating cellular uptake of K+ in exchange for H+ ions.

Potassium depletion and hypokalaemia. These must be distinguished from each other. Chronic depletion of body potassium may be associated with hyperkalaemia, for example, in a metabolic acidosis where cell K+ is exchanged for the extracellular H+ ions. Conversely, body potassium may be greater than normal in the presence of hypokalaemia, for example, in a metabolic alkalosis. Normally, plasma K+ concentration remains relatively constant, any excess or deficit in body potassium being accommodated by the cells. Thus measurements of plasma K+ concentration *do not* allow conclusions to be drawn about body potassium content. Estimates of the latter have been obtained by using isotopes or by extracting potassium from muscle biopsy specimens. A recent finding that the K+ content of red blood cells mirrors that of other cells offers a simpler way to estimate total body potassium.

Chronic K+ depletion is associated with degenerative changes in myocardium and skeletal muscle as well as with functional changes (muscular weakness, paralysis). Vascular changes in renal epithelial cells accompany an impaired urinary concentrating ability and a polyuria which is insensitive to exogenous ADH.

Since the kidneys normally conserve K+, loss of body potassium usually reflects a combination of diminished intake with increased loss through the gut or kidneys. Vomiting, diarrhoea, aspiration of gastrointestinal contents, and fistulae may lead to hypokalaemia. Aldosterone and a variety of diuretics which facilitate K+ secretion by the epithelial cells through additional distal delivery of Na+ promote loss of K+ in the urine. As with hyperkalaemia typical ECG changes may give early warning of hypokalaemia.

Potassium and acid-base balance. Cells exchange K+ and H+ ions with plasma. In metabolic acidosis plasma K+ concentration increases even though body potassium may become depleted. In metabolic alkalosis plasma K+ concentration may decrease. But though cells gain K+ initially, chronic alkalosis may lead to loss of body potassium because of increased K+ secretion by renal principal cells (perhaps reflecting their higher K+ concentration) and of increased delivery of Na+ to this segment of the tubule. This encourages exchange of tubular Na+ for cell K+ with retention of K+ in the lumen to preserve electroneutrality.

Conversely, chronic K+ depletion can lead to an alkalosis, when decreased K+ secretion by depleted principal cells result in a greater portion of Na+ delivered to the distal tubule being reabsorbed in exchange for secreted H+ ion. The corresponding transfer of cell bicarbonate to the plasma may explain the paradoxical association of an acid urine with an alkaline plasma.

13.3 Acid-Base Metabolism and Regulation of pH

Cells, especially those of the nervous system, are sensitive to the pH of their surroundings. Too great an alkalinity leads to headache, lassitude, tetany and convulsions. Too great an acidity makes respiration deep and sighing, and causes drowsiness progressing to coma. Plasma pH below 7 or above 7.8 is usually lethal, so that hydrogen must remain between 16 and 100 nanomol/L.

The blood in man is normally kept blandly alkaline at pH 7.4 +/- 0.04, hydrogen = 36 to 44 i.e. 40 +/- 4 nanomol/L. Most cells are believed to be less alkaline than plasma, with pH nearer 7.0. Note how little free hydrogen there is in the body. Forty litres of body water at a pH of 7.0 would contain 4000 nanomol or 0.004 micromol of hydrogen. Holding this constant within 10% means a tolerance of 0.0004 micromol; and each day the body produces by metabolism 13000 micromol of potentially acid-forming carbon dioxide and 50-80 micromol of stronger, non-volatile acids: sulphuric and phosphoric. A little arithmetic shows that the precision of control is far better than that of a watch keeping time to 1 sec per day.

Mechanisms of Control

Stability is achieved by buffers, and by exchanges of ions with bones and cells to minimize changes of pH pending disposal of acids or alkalis by lungs or kidneys. The important buffer bases that take up hydrogen ions in the body are the conjugate bases of three weak acids - carbonic acid, proteins and phosphoric acid.

If pH tends to fall, buffer base takes up hydrogen while if pH tends to rise, buffer acid supplies hydrogen, and so pH is stabilized and changes are 'buffered'. Note also that just as much buffer is used up as hydrogen is neutralized. For example, when sulphuric acid is added to the plasma only the very weak base and the weak buffer acid remain; pH is controlled, but buffer base has disappeared.

Why plasma pH is 7.

The Henderson-Hasselbalch equation,

$$\mathbf{pH = pK' + \log_{10}(conjugate\ base/conjugate\ acid)}$$

may be applied to any of the plasma's buffer pairs, most conveniently to the predominant one, bicarbonate/carbonic acid. pK' for the sequence is 6.1 and the solubility of CO_2 in plasma is 0.03 micromol per mmHg.

Hence

$$\mathbf{pH_{plasma} = 6.1 + \log_{10}(bicarbonate/0.03P_{CO2}).}$$

With bicarbonate maintained at about 24 micromol/L by the kidney and P_{CO2} regulated close to 40 mmHg by the respiratory system

$$\mathbf{pH = 6.1 + \log_{10}(24/0.03x40) = 7.4.}$$

The Henderson-Hasselbalch equation indicates the important variables: pH varies with bicarbonate and inversely with P_{CO2}. Hence a change in pH that results from an alteration in *either* bicarbonate or P_{CO2} can be avoided or corrected by changing the *other* variable to preserve or restore the buffer ratio. In a solution in which bicarbonate is the only buffer base, doubling or halving bicarbonate or P_{CO2} shifts pH by 0.3 units. In blood, which has other buffers besides bicarbonate, doubling or halving P_{CO2} produces a smaller final change in pH of 0.2 unit because other buffers release or take up hydrogen and change bicarbonate in the same direction as P_{CO2}.

The Henderson-Hasselbalch equation also summarizes the basis for physiological regulation of pH. The kidneys control the numerator and the respiratory system the denominator. The respiratory chemoreceptors are affected by pH as well as by P_{CO2}. Adding acid to the blood lowers pH as well as bicarbonate. The reduction in pH stimulates respiration so that P_{CO2} is lowered to match the lowered bicarbonate, and pH is rapidly returned towards normal.

Finally, Henderson-Hasselbalch equation offers a rational classification of disturbances of pH. The numerator, bicarbonate, is affected by ingestion, production or loss of acid or alkali, giving rise to metabolic (including renal) acidosis or alkalosis. The denominator, 0.03 P_{CO2}, is affected by alterations in pulmonary ventilation or in the composition of inspired air, giving rise to respiratory acidosis or alkalosis. Fundamentally, disturbances of acid-base balance fall into one or other of the following categories. Clinically, mixed disturbances are sometimes seen.

1. Reduced bicarbonate: *metabolic acidosis.* This can arise from (i) ingestion of HCl or NH_4Cl (converted by the liver to urea); (ii) production of ketone acids in diabetes or of lactic acid in shock; (iii) loss of alkaline secretions such as bile, pancreatic or intestinal secretions; and (iv) failure to generate adequate bicarbonate in the kidney (*renal tubular*

acidosis). The fall in pH is largely corrected by increased pulmonary ventilation. Hence the chief effect is a reduction in buffer-base, especially bicarbonate.

2. Increased bicarbonate: *metabolic alkalosis.* This can arise from (i) ingestion of sodium bicarbonate or alkaline stomach powders; (ii) loss of acid secretions, notably gastric juice; and (iii) depletion of body potassium. The rise in pH is partly compensated by reduced pulmonary ventilation with retention of CO_2, and the chief effect is increased bicarbonate.

3. Increased PCO_2: *respiratory acidosis.* This can arise from (i) hypoventilation from, for example, respiratory obstruction or paralysis, drugs or toxins; and (ii) high inspired P_{CO2}. Retention of CO_2 increases P_{CO2} substantially. Since total anions must still equal total cations, bicarbonate can increase no more than other buffer bases are reduced with falling pH. Hence initially the main effect is a reduction in plasma pH, later to be corrected by slow replacement of chloride by bicarbonate from the kidneys if the retention of CO_2 persists.

4. Decreased P_{CO2}: *respiratory alkalosis.* This results from hyperventilation which may be (i) voluntary (experimental); (ii) nervous, with tension and anxiety (hyperventilation syndrome); or (iii) hypoxic, at high altitudes, from peripheral chemoreceptor drive (carotid body). P_{CO2} is substantially reduced, but initially alteration in bicarbonate is minimal, so that pH is increased with no change in buffer base. Later, if hyperventilation persists, the kidneys excrete bicarbonate in an alkaline urine and slowly restore pH towards normal by reducing bicarbonate.

Remember that initially, before renal compensation can be effective (which takes days), *metabolic disturbance affect bicarbonate far more than pH; respiratory disturbance affect pH far more than bicarbonate.*

Production, Initial Buffering and Final Deposition of Acids

1. *Carbonic acid.* CO_2 is produced at the rate of about 13000 mmol per day (10 mmol/min) and represents potential acid that would need 13000 mmol per day of NaOH to neutralize it to $NaHCO_3$. In fact it is excreted by the lungs, and very effectively buffered in transit from tissues to lungs in the blood, mainly by haemoglobin. Removal of O_2, as CO_2 is being taken up, replaces O_2Hb^- by Hb^- which is a much stronger base. About 0.7 mmol of hydrogen for each mmol of O_2 given up (about 25 mL at body temperature) is required to prevent the blood becoming even more alkaline than normal. Hence only about 0.1 mmol of the 0.8 mmol of CO_2 produced from 1 mmol of O_2 (with RQ of 0.8) yields hydrogen ions which need to be handled by buffers other than Hb, and venous blood is only about 0.03 pH more acid than arterial. In summary:

oxyHb + water + CO_2 ==> O_2 + Hb + bicarbonate

Because red cell bicarbonate exchanges readily with plasma chloride, CO_2 taken up into the blood is mostly carried as bicarbonate in the plasma. The process is reversed in the pulmonary capillaries.

2. *Non-volatile acids.* Each day metabolism yields a total of about 50 to 80 mmol of sulphuric acid (from oxidation of S in sulphur-containing amino acids) and phosphoric acid (derived from phosphoproteins and phospholipids). Organic acids produced in metabolism are normally oxidized to CO_2 and water. In shock, and in exercise severe enough to be partly anaerobic, oxidation may fail to keep up with production, and lactic and pyruvic acids may also accumulate. In diabetes as much as 750 mmol of acetoacetic acid and beta-hydroxybutyric acids may be produced daily. The H+ ions from all these non-volatile acids must be buffered in the body.

Mechanisms which minimize the fall in pH caused by H+ ions pending final disposal are:

(i) *Dilution in total body water.* Acid produced in cells diffuses into ECF and into other cells; each ten-fold dilution raises pH by 1 unit.

(ii) *Buffering in blood.* It would take 28 mmol of H+ to bring 1 litre of whole blood from pH 7.4 to pH 7.0. The H+ ions would be shared between the buffers of the blood as follows: bicarbonate, 18 mmol; inorganic phosphate, 0.3 mmol; plasma protein, 1.7 mmol; and haemoglobin, 8 mmol. Five litres of blood could take up 140 mmol H+ (twice the normal daily production) without lethal decrease in pH; but note that bicarbonate would be reduced from 24 to 6 mmol per litre.

(iii) *Buffering in extravascular ECF*. This is mainly achieved by bicarbonate, for there is no haemoglobin and much less protein than in the blood. The 14 L of ECF containing 24 mmol of bicarbonate per litre could take up a further 200 mmol of H+ before pH was reduced to 7.0.

Processes (i), (ii) and (iii) are rapid, being completed in minutes and total ECF including plasma could take about 350 mmol, or some five times the day's production without disaster. This is ample reserve for normal purposes.

(iv) *Buffering in cells.* Intracellular buffers, mainly proteinate and organic phosphate, can back up extracellular buffers by coping with H+ ions that enter cells which exchange them for K+ or Na+. Their capacity is probably similar to that of extracellular buffers but the process is much slower (hours) because of limited permeability of cell membranes to H+.

(v) *Carbonate in bone.* This can contribute some bicarbonate to the plasma and ECF to replace some of that which is used up in (ii) and (iii); again this probably takes hours.

(vi) *Exchange of ions with bone mineral.* Extracellular H+ ions can exchange with cations from bone and cells, i.e. H+ + M+ in bone ==> H+ in bone + M+ where M+ may represent Na+ or K+ or 1/2 Ca2+ or 1/2 Mg2+. These are slow processes, taking hours to days. Prolonged metabolic acidosis depletes bones of calcium and cells of potassium.

These six processes all add up to *whole-body buffering,* of which the effectiveness was well demonstrated in 1953 by R. F. Pitts who intravenously titrated a dog with molar HCl. The blood pH decreased from 7.44 to 7.14 after addition of 150 mL of acid (i.e. 150 mmol

H+). By contrast, the addition of the same amount of HCl to 11 kg of water (the estimated water content of the dog) brought the pH to 1.84. Since pH 7.44 corresponds to a H+ of 36 nanomol/L and pH 7.14 to a H+ of 72 nanomol/L, 36 x 11 or about 400 nanomol of the added H+ remained free in the dog, i.e. of 150 mmol or 150000000 nanomol of the H+ added to the dog, 400 remained free and 149999600 was buffered in the body. Of this about a half was buffered extracellularly and a half by cells and bone.

Final Disposal of Hydrogen Ions and Regeneration of Body Buffers

It falls to the kidneys to rectify the changes produced by additional acid or alkali in the body. Because of respiratory compensation pH may be not far from normal but the buffer stores, indicated by the concentration of bicarbonate in the plasma, may be either depleted (by acids) or augmented (by excess alkali) and these need to be restored to their normal levels.

In terms of the body's principal buffer base, bicarbonate, what happens when, for example, sulphuric acid is added, may be summarized thus:

$$H_2SO_4 \Longrightarrow SO_4 2- + 2\ H+$$

$$2\ H+ + 2\ HCO_3- \Longrightarrow 2\ H_2CO_3 \Longrightarrow 2\ H_2O + 2\ CO_2 \text{ (expired)}$$

After this, two H+ ions have become hydrogen atoms in body water. Two CO_2 have been breathed out. The pH has fallen a little, but in the plasma two bicarbonate ions have been replaced by one sulphate. The kidneys easily dispose of sulphate and the conjugate bases of other unwanted acids that are replacing bicarbonate in the plasma. These pass into the glomerular filtrate and need only to be left unreabsorbed by the tubules to be excreted in the urine. The H+ ions no longer have a free existence; they are present as parts of buffer bases or of body water, and are not directly available for excretion. Eventually, because all the buffers are in equilibrium, as the kidneys manufacture bicarbonate which they add to the plasma, H+ ions temporarily buffered by other buffer bases will finally be transferred to bicarbonate and converted to water. But for every water molecule so formed, a molecule of bicarbonate has been destroyed. The role of the kidney is to manufacture bicarbonate to replace the bicarbonate used up. The kidney cells make this bicarbonate which they restore to the plasma, with H+ which they secrete into the lumen. The H+ ions excreted in the urine are simply a by-product in the manufacture of bicarbonate. Though the mechanism may be more complex, a simple view is that in the renal tubular cells of the proximal and distal tubules and collecting ducts, carbonic acid formed from CO_2 and water (catalysed by carbonic anhydrase) ionizes to provide H+ and bicarbonate ions in equal numbers. The bicarbonate passes across the basolateral borders of the cells along with Na+ ions that are being reabsorbed, and the H+ is secreted into the tubular fluid. Because equal numbers of H+ and bicarbonate ions are formed in this way, by the time that the bicarbonate used up in coping with H+ ions in the body has been replaced, an equivalent amount of H+ formed in the kidney will have been excreted in the urine.

The Driving Force for Secretion of Hydrogen Ions

It is probable that the movement of H+ from cell to lumen involves a counter-transport of Na+ from lumen to cell, at least in the proximal and early distal tubules. The urine can be

acidified to pH 4.4, which, when the plasma remains at pH 7.4, implies a ratio $(H+)_U/(H+)_P$ of 1000:1. Such ratios are only achieved in the 'tight' distal portions of the nephron. Here the intercalated cells may secrete H+ actively, perhaps utilizing a H+-ATPase as found in gastric oxyntic cells, but this remains conjectural.

Fate of Hydrogen Ions Secreted into the Tubular Fluid

The figure illustrates three major consequences of the secretion of H+ ions into the lumen:

1. *'Reabsorption' of filtered bicarbonate* by replacement with newly formed bicarbonate from tubular cells; this consumes about 4500 mmol H+ per day. Within the lumen carbonic anhydrase attached to the brush borders catalyses the breakdown of carbonic acid and allows CO_2 to recirculate. Normally, this process is largely complete by the end of the proximal tubule and simply restores to the body bicarbonate that would otherwise have been lost in the glomerular filtrate.

2. *Production of titratable acid.* Buffers in the glomerular filtrate react with secreted H+ ions. Normally, the predominant filtered buffer is phosphate, but in diabetes mellitus and in shock, weak organic acids may make a major contribution, and as much as 250 mmol per day of titratable acid may be excreted compared with the normal 20-30 mmol per day. This process consumes less H+ than (1) but the H+ is secreted against an increasing gradient as urine is acidified, and an equivalent amount of bicarbonate is added to plasma over and above the amount rescued from the filtrate.

3. *Production of ammonium.* Ammonia formed from glutamine and amino acids in tubular cells diffuses into the lumen, where it reacts with H+ to form NH_4+ which does not readily cross plasma membranes and is trapped in the tubular fluid. Each NH_3 converted to NH_4+ allows a bicarbonate ion from the cell to be added to the plasma. From a normal range of 30-50 mmol per day up to 500 mmol per day of NH_3 can be secreted with equivalent bicarbonate added to the plasma.

Note that (1), (2) and (3) are sequential processes with some overlap and are not confined to anatomically separate segments of nephrons. H+ ions consumed in (1) are not excreted in the urine. H+ ions consumed in (2) and (3) are excreted as urinary titratable acid and NH_4+; process (3) allows excretion of more H+ in an acid urine without further lowering its pH. The kidney in effect manufactures its own urinary buffer, NH_3, and therefore more bicarbonate can be formed and more acid excreted before the minimum urinary pH (4.4 to 4.6) is reached.

The amounts of bicarbonate ions contributed by the three basic processes change under different conditions as shown by typical round figures in Table 13.2. Note that less H+ is actually secreted during metabolic acidosis than under normal or alkalotic conditions. But during acidosis, since tubular fluid pH falls to a lower level, more H+ has to be secreted against a substantial gradient. Also note that, in every situation, most of the H+ secreted is used up in the 'reabsorption' (by replacement) of filtered bicarbonate.

Measurement of Urinary Titratable Acid and Ammonia

Because urine contains buffers its pH cannot provide an estimate of total H+ excretion in the urine. Urinary titratable acid is measured by adding OH- ions to a sample of urine until its pH has been restored to the pH of the plasma from which it was formed. Since the pK of the NH_3/NH_4+ pair is 9.3, only about 1/100 of the NH_4+ has been titrated at the normal plasma pH of 7.4. The titratable acid does not therefore indicate NH_4+, and separate chemical methods are used to measure total urinary ammonia.

The importance of the measurement of titratable acid and total ammonia lies in the fact that their sum is equal to the total acid excreted in the urine, and this is equal to the total amount of additional bicarbonate added to the plasma after replacing all the filtered bicarbonate destroyed in the renal tubules. Under steady-state conditions this additional bicarbonate is equal to the bicarbonate consumed in the body in buffering H+ from non-volatile acids. That is, over a 24 h period, titratable acid + total ammonia = production of non-volatile acid.

Regeneration of Buffer Stores

Bicarbonate added to the plasma by the tubules first replaces plasma bicarbonate that had been lost temporarily in the glomerular filtrate. This conserves existing stores of bicarbonate but does not add to them. This 'extra' bicarbonate, corresponding to H+ ions excreted as titratable acid and ammonium:

(a) Replaces bicarbonate that had been used in buffering H+ ions in the body;

(b) Replaces bone carbonate that had supplied bicarbonate to the plasma and other ECF.

(c) Regenerates other (non-bicarbonate) buffer bases that had been used up:

$$\mathbf{HCO_3- + HBuf \Longrightarrow Buf- + H_2CO_3 \Longrightarrow H_2O + CO_2;}\ \text{and}$$

(d) Reverses ion exchanges with bone and cells:

$$\mathbf{HCO_3- + M+ + H+\ in\ bone\ or\ cell \Longrightarrow M+\ in\ bone\ or\ cell + H_2CO_3.}$$

Regulation of Hydrogen Ion Secretion and Urinary Acidity and Alkalinity

Most filtered bicarbonate is 'reabsorbed' from the proximal tubules and not more than about 5% ordinarily reaches the distal tubules, but process (1) must give place to (2) and (3) when filtered bicarbonate has all been destroyed within the lumen. Whether the final urine is to be acid or alkaline depends upon the relation between the rate of filtration of bicarbonate and the rate of secretion of H+ ions (Table 13.2).

The filtered load of bicarbonate equals (bicarbonate $_{plasma}$ x GFR and is proportional to the plasma bicarbonate so long as GFR remains constant. The rate of secretion of H+ ions is roughly proportional to PCO_2 in the blood supplying the kidneys. Increased PCO_2 probably

enhances H+ secretion by making more CO_2 available as a source of carbonic acid and also by lowering pH in the cells so that the gradient from cell to lumen becomes more favourable. Factors which increase sodium reabsorption in the distal tubules and collecting ducts (i.e. aldosterone, increased Na+ delivery) also promote secretion of H+.

Compensation for Metabolic Disturbances

Alterations in plasma bicarbonate provide automatic adjustments to minimize metabolic disturbances. In *metabolic acidosis,* when plasma bicarbonate is reduced, less bicarbonate is filtered, less of the H+ secreted is consumed in 'reabsorption' of filtered bicarbonate, more H+ is excreted as titratable acid and NH_4 in an acid urine, and corresponding amounts of additional bicarbonate are added to the plasma restoring bicarbonate towards normal.

In *metabolic alkalosis,* when bicarbonate is increased, the filtered load of bicarbonate requires more H+ for its 'reabsorption', and less H+ goes to form titratable acid and ammonium. When filtered bicarbonate exceeds the total rate of H+ secretion, process (1) cannot be completed, unreabsorbed bicarbonate is excreted in an alkaline urine, reducing the plasma bicarbonate towards normal.

Alterations in PCO_2 caused by ventilatory responses to changed plasma pH retard but do not prevent these automatic adjustments. Thus in *metabolic acidosis* stimulation of ventilation lowers PCO_2 and reduces the rate of secretion of H+. In *metabolic alkalosis* retention of CO_2 by reduced ventilation increases PCO_2 and the rate of H+ secretion (see illustrative values in Table 13.2). The body's store of bicarbonate is normalized more slowly than it would be if the ventilatory response did not occur. In the meantime, the ventilatory responses protect against considerable alterations in pH, and the nervous system is intolerant of pH change but relatively unaffected by alterations in the amount of buffer in the stores.

The excretion of NH_4+ ions takes up to a week to reach its maximum rate during sustained acidosis. The delay cannot be satisfactorily explained by slow adaptive increases in activity of the enzymes involved in NH_3 formation, but it might reflect a slow increase in the intensity of H+ secretion mediated by aldosterone; for sodium is inevitably lost to maintain electroneutrality in urines containing excessive amounts of conjugate bases of acids such as sulphuric or HCl (experimentally) or ketone acids (in diabetic keto-acidosis) and there is a threat of sodium depletion until sufficient NH_4+ is available to be used instead.

Renal Compensation for Respiratory Disturbances of pH

The dependence of hydrogenium secretion upon P_{CO2} provides slow renal compensation for respiratory disturbances of pH. In *respiratory acidosis* the increased P_{CO2} increases hydrogenium secretion. While the filtered load of bicarbonate is not increased appreciably, more hydrogenium is excreted as titratable acid and NH_4^+, and the corresponding bicarbonate added to the plasma increases plasma bicarbonate and slowly corrects the buffer ration.

In *respiratory alkalosis* the low P_{CO2} slows hydrogenium secretion so that the initially undiminished filtered load of bicarbonate is not completely 'reabsorbed'. Bicarbonate is

excreted in an alkaline urine and lowers plasma bicarbonate so that the excessive alkalinity of the plasma is slowly corrected.

Some Clinical Aspects

Significance of Plasma Bicarbonate

After renal compensation for respiratory disturbances note that: *respiratory acidosis* ends up with a *high* plasma bicarbonate whereas *respiratory alkalosis* ends up with a *low* plasma bicarbonate. Consequently, an *increased* plasma bicarbonate may indicate either *metabolic alkalosis* or *respiratory acidosis* (chronic and renally compensated). Laboratory differentiation between these alternatives requires the measurement of plasma pH or P_{CO2} as well as bicarbonate. The conditions can usually be distinguished clinically from observation of the patient and the history of the disturbance.

Effect of Depletion of Potassium

Potassium-depleted tubular cells are more acid than normal and, especially when stimulated by aldosterone, secrete more hydrogenium. Hence the urine tends to be acid and the plasma alkaline with bicarbonate high and chloride low.

Effect of Inhibitors of Carbonic Anhydrase

Inhibition of carbonic anhydrase slows the secretion of hydrogenium so that bicarbonate is excreted in an alkaline urine and plasma bicarbonate is lowered. This explains the otherwise mysterious acidosis seen in the early days of treatment with massive doses of sulphonamide drugs, which inhibit carbonic anhydrase. Later the potent inhibitor acetazolamide (Diamox) was tried as an alternative to mercurial diuretics because sodium excreted with the bicarbonate reduced the body's excessive load of sodium. The diuretic effect ceases however when bicarbonate has been reduced to the level at which the filtered load is not greater than the tubules can 'reabsorb' using hydrogenium produced through the uncatalysed reaction of CO_2 with water. Bicarbonate then ceases to be excreted and a new steady state is established.

13.4 Function in Diseased Kidney

Damaged kidneys may fail to maintain the normal composition and volume of the body fluids because they lose or excessively retain natural constituents of the body or else because they fail to remove waste products.

Disturbances of Glomerular Filtration

The glomeruli may leak protein or may fail to produce their normal daily quota of 180 L of protein-free filtrate for the following reasons:

1. *Protein loss from kidney.* In 'nephrotic syndromes' an abnormal glomerular basement membrane allows plasma proteins, especially albumin, to escape. The urine may contain from 5 to as much as 30 g of protein per day. When synthesis in the liver fails to keep up with

renal loss, there is less circulating albumin and its concentration in the plasma falls, so that the volume and the colloid osmotic pressure of the plasma tend to be reduced. There is a body-wide disturbance of the Starling equilibrium, with gross generalized oedema, caused and sustained by excessive retention of sodium by the kidneys.

2. *Failure to produce enough glomerular filtrate.* In chronic renal failure destruction of nephrons gradually reduces GFR. About three-quarters of person's nephrons can be destroyed before renal function is obviously impaired. The remaining nephrons hypertrophy and adapt so that total function is surprisingly well maintained. Note that for substances excreted primarily by the glomeruli, excretion depends upon the filtered load, not upon GFR as such. Consequently for creatine or urea, the rate of excretion will be maintained if plasma concentration increases in proportion to dwindling GFR. The normal amount of creatinine can be eliminated in one quarter as much glomerular filtrate if each volume of filtrate contains four times as much creatinine, i.e. if plasma creatinine concentration is four times normal. Similarly, the rising plasma urea concentration with advancing renal failure is not merely a sign of falling GFR. It is an effective compensating mechanism that allows the excretion of urea to be maintained.

In a steady state the rate of excretion of urea must equal the rate of production. If GFR suddenly falls to half, excretion will at first lag behind production and plasma urea concentration will increase until it reaches twice normal, when excretion will catch up with production. The body's urea pool will then have twice as much in it, but retention of urea will no longer be increasing. If GFR falls to 12 mL/min (10% of normal), plasma urea will have to rise to 40 mmol instead of 4 mmol and the body will contain 1800 mmol instead of a normal 170 mmol of urea, but the rates of production and of excretion will again be equal and normal at about 500 mmol or 30 g per day.

Disturbances of Tubular Function

In chronic destructive diseases renal tissue gradually disappears, but the amount of solute to be excreted every day does not diminish. The hypertrophies and adapted nephrons that are left have to do all the work that was formerly done by a full complement of normal nephrons. If 10% of the nephrons remains, each one of these must on average handle ten times as much solute as a nephron in a normal kidney, like a nephron in fact in a normal pair of kidneys excreting 5000 (instead of 500) mmol of urea each day. Under such conditions of permanent osmotic diuresis the urine cannot be concentrated. During water deprivation normal kidneys can make urine concentrated to four times the osmolality of the plasma - but at rates less than about 0.6 mL/min. The attainable concentration falls as flow rate increases during osmotic diuresis, and at 15 mL/min the urine can be little more than isosmolal. With only 10% of the nephrons remaining, the urine cannot be more concentrated than the plasma if the rate of production exceeds 1.5 mL/min. This is about output of urinary solutes, which explains the fixed osmolality and specific gravity of 1010 (isosthenuria) characteristic of renal failure.

Inability to concentrate and to reduce the volume of the urine also accounts for the *nocturia* which may be an early sign of renal damage. Note that a simple test of the power of the kidneys to concentrate the urine (such as the urinary osmolality or specific gravity after 12 h overnight without fluid) is both the simplest and the most sensitive test for detecting early impairment of renal function, especially in diseases like pyelonephritis which attack the

medulla. Note too that the increased urinary volume that results from the kidneys' loss of concentrating power promotes the excretion of urea; for the clearance of urea increases with rising flow rate and becomes maximal above about 2 mL/min.

Handling of Sodium

In contrast to urea, retention of sodium with increasing concentration in the plasma would disturb the balance between cells and ECF. Retention is avoided by reabsorbing a smaller fraction of filtered sodium and since normally more than 99% of filtered sodium is reabsorbed, there is a large margin of safety. If GFR fell to 12 mL/min (one-tenth of normal) sodium balance could still be maintained by reabsorbing 97.5% of the filtered load of 2500 mmol per day. Even with GFR reduced to 3 mL/min (one-fortieth of normal) the filtered load would be 600 mmol per day and balance could be maintained by reabsorption of five-sixths of the filtered sodium. Possible causes of reduced fractional reabsorption include: (i) osmotic diuresis, since back-flux increases as sodium concentration becomes lower in the tubular fluid, (ii) reduced availability of NH_4^+, which means that sodium must accompany conjugate bases of urinary acids, and (iii) possibly the action of natriuretic substance.

Sometimes, and especially when tubular functions have been damaged more than glomerular, too much sodium may be lost, with depletion of the volume of ECF which further depresses GFR and aggravates retention of nitrogenous substances.

Handling of Potassium

Since K+ is secreted by the tubules its conservation presents no problem to the failing kidney. Excretion of K+ may however be impaired if a grossly reduced filtered load of Na+, due to a very low GFR, delivers too little Na+ to the distal exchange site to be reabsorbed in exchange for secreted K+. Increased catabolism of tissues in uraemia releases K+ from broken-down cells, and failure to excrete this K+, may lead to a dangerous elevation of plasma K+ concentration requiring treatment by dialysis.

Handling of Hydrogen Ions

Renal acidosis is a metabolic acidosis arising from impaired capacity of the renal tubular cells to secrete hydrogenium ions and add bicarbonate to the plasma, and is accordingly often characterized as *renal tubular acidosis.* Possible causes include: (i) failure to secrete hydrogenium into the urine and add bicarbonate to the plasma at a sufficient rate, (ii) failure to bring urine to a sufficiently low pH, so that less hydrogenium can be excreted as titratable acid and ammonium, (iii) failure to supply sufficient ammonia to carry hydrogenium ions into the urine as NH_4^+, and (iv) shortage of filtered buffer to make into titratable acid. Only the last of these depends primarily upon GFR. The rest depend upon metabolic failure to transport hydrogenium ions against gradients or to provide NH_3. The biggest single factor is usually the inadequate supply of NH_3 from the dwindling of the mass of metabolizing tubular tissue as nephrons are destroyed.

In summary, the patient with failing kidneys has an undiminished need for the services of his kidneys, and so must overload a dwindling population of functioning nephrons. Nitrogen end-products accumulate (*azotaemia*) because there are too few glomeruli, and GFR

falls. Lack of tubular tissue leads to failure to secrete sufficient potassium, hydrogenium and NH_4; and sometimes to retain sodium. The overloaded remaining nephrons, possibly with a disorganized medulla with its countercurrent arrangements wrecked, cannot concentrate the urine; and so the patient must live with an increased turnover of water or risk dehydration. Two well-organized kidneys have been reduced to a motley collection of nephrons.