Body fluids are distributed between two major fluid compartments; intracellular fluid (ICF), and extracellular fluid (ECF). The ECF compartment is further subdivided into plasma and interstitial fluid

Even though particles(ions, sugars, and proteins) in the ECF and ICF differ, their concentrations are normally identical, and the **number** (not the nature) of the unequally distributed **particles per volume** determines the fluid's **osmolarity**.

And because they are similar in their osmolarities, no net movement of H2O usually occurs between compartments (although the water can move freely between them). Therefore, cell volume normally remains constant.

Molarity= #of moles/ L
(One mole of any molecule = 6.022×1023 particles**)**

Osmolarity= molarity * # of particles= #of moles/ L * # of particles
([example: 1 mole/L of NaCl equals 2 Osmol/L because 1 mole of NaCl means 1 mole of Na and 1 of Cl so its 2 Osmoles])

Note; Equivalents (Eq)=number of univalent counter ions needed to react with each molecule of substance (example: 1 mole of Ca++= 2eq because you need 2 moles of Cl- to react

Ex: a 2 g of which of the following particles would have the strongest tendency to attract water? Na+, albumin, or glucose: **(osmolarity↑ then the tendency to attract water↑).**

Osmolarity is directly proportional to the mass and inversely proportional to the MW. So, the particle with the lowest MW will have the higher tendency to attract water, and this particle is Na+ followed by glucose, and the least is being albumin.

Examples;

- **how much NaCl is needed to prepare the normal saline (isotonic solution with osmolarity of 285 mOsm/L? (notice the units) (it is the same equation above).**
  Mass = MW*osmolarity(in mOsm/L)/(#of particles *1000)=58*285/(2*1000)=9 g/L
  we find that the needed mass of the NaCl is 9 g/L = 0.9 g/100 ml
  From this the 0.9% N/S (normal saline) is named!
- **What is needed mass of glucose (MW=180) to prepare an isotonic solution with osmolarity of 285 mOsm/L?** The needed mass will be 5 (g/100 ml) dextrose/ water solution notice that the # of particles here is 1.
  (X= 180*285/1000 = 50g/L = 5g/dl = 5% O/W).

Tubular reabsorption of Na+

The importance of sodium:

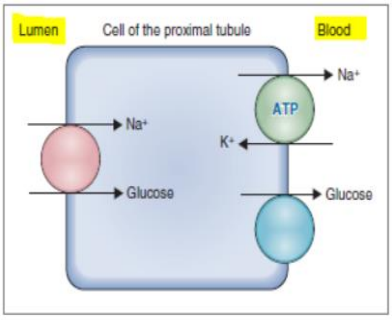
1- **It Contributes to the osmolarity of the plasma and in turn, controls fluid volumes;** We have the same number of cations and anions (the ECF is electroneutral). Sodium and its attendant anions, being by far the most osmotically active solutes in the ECF (number not nature). To calculate the osmolarity, multiply $\{Na^+\}$ by 2:. {Na+}=140 mMol/L → so the osmolarity=140 * 2=280. But because it is not the only cation, multiply its concentration by 2.1 to get 295 mOsm/L.

2- **it controls the extracellular volume;** If its concentration increases (intake), more water is retained (from the kidney) which increases the volume causing edema. But if it decreases, less water is retained which causes volume contraction(dehydration).

3- **It is important for the excitable tissue to function**; Excitability of the cell means; its ability to reverse its membrane potential by the influx of **sodium**, calcium, or both. Excitable cell is the cell that doesn't perform its function unless it is excited**.**

4- **It is essential for the kidney to reabsorb useful particles (glucose, amino acids, and other) and git rid of waste products (H+) via the secondary active transport.**

   a- The basolateral Na+–K+ pump actively transports Na+ from the tubular cell into the interstitial fluid (blood side) in order to establish the Na^+ concentration gradient between the lumen and the tubular cell.



   b- This Na+ gradient is used to

      i) co-transport the glucose, amino acids, and others against their electrochemical gradient.

      ii) counter-transport waste products such as H+ against their electrochemical gradient (from the interstitial fluid with the higher PH toward the lumen with the lower PH(Higher {H+})).

   Note; active reabsorption takes place if any step in the transepithelial transport of a substance requires energy, even if the other steps are passive.

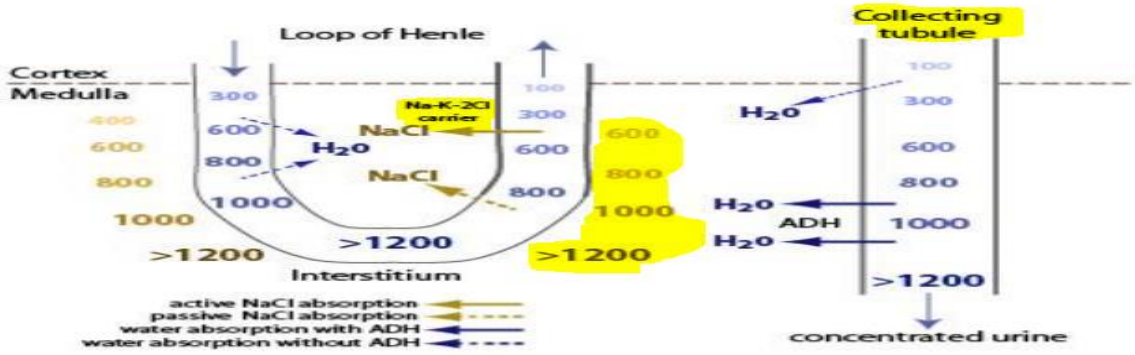
5- **It is targeted by diuretics**; They inhibit Na+ reabsorption → Na+ remains in the lumen of the tubule and keeps water with it → so you get rid of extra Na+ and water.

6- **It has a role in the formation of the corticomedullary osmotic gradient which has a main role in concentrating urine(please, go back for the online lecture before studying this point);**

The cells of the thick ascending limb are the only human cells that are impermeable to water. Na, Cl and k (by Na+:K+:2*Cl- carrier) are reabsorbed by the thick ascending limb, but water is not reabsorbed along with it, which increases the osmolarity in the surrounding peritubular interstitium and capillaries(it may reach 1200-1400 mOsmol/L)((and osmolarity is decreasing as the ascending limb ascends)) see the figure below.

The Collecting ducts passes through this osmotic gradient in the medulla from 300 mOsm/L near the cortex to 1,400 mOsm/L near the papilla. This gradient is produced by a **countercurrent multiplier(again study the online lecture to understand this better)** of the nephron loop, which concentrates ions in the lower medulla.

- In a state of hydration, ADH is not secreted and the collecting duct reabsorbs salt without reabsorbing water (it will be water impermeable like the ascending loop of Henle); the water remains to be excreted in the dilute urine(urine as hypotonic as 50 mOsm/L).
- In a state of dehydration, ADH is secreted, the collecting duct reabsorbs water (by inserting a special type of water channels), and the urine is more concentrated.



It is important for those with renal impairment to

1- **reduce the Na+ intake, as the majority of renal ATP consumption is to reabsorb Na+.**
2- **reduce their protein intake due to GFR impairment(proteins increases urea which is a waste product that the impaired kidney cannot handle).**
3- **maintain their blood pressure tightly, as hypertension causes scleroses in the basement membranes of the kidney.**

The renal function in handling sodium:

To maintain its homeostasis, body must ensure that Na+ excretion exactly equals Na+ intake, a matching process called **Na+ balance(the daily intake of sodium=155mMol;** 150 mMol will be renally excreted, and the other 5 mMol will be excreted via other mechanisms (such as GIT secretions or sweating).

1- Na+ is **freely filtered(**plasma concentration=filtrate concentration)
2- in the **proximal convoluted tubule**, two-thirds (or **65**%) of the filtered load is reabsorbed. Eater and Na are reabsorbed isosmoticly(same ratio)
3- The **descending limb doesn't reabsorb Na+ at all (0%),** but water is reabsorbed here (which increases the osmolarity of the tubular fluid gradually).
4- The **thick ascending limb of the loop of Henle reabsorbs 25%** (along with 2*Cl-, and K+ reabsorption by Na+:K+:2*Cl- carrier, it is electroneutral) of the filtered load of Na+.
   Note; The **single effect** phenomena of the countercurrent multiplier happens at this level. Na, cl, and k are reabsorbed out of the ascending limb and deposited in interstitial fluid, and water remains behind in the ascending limb. So, we end up with hyperosmolar interstitium and hypoosmolar filtrate.
5- **The early distal convoluted tubule reabsorbs approximately 7%** of the filtered load, and, like the thick ascending limb, it is impermeable to water.
6- The **late distal convoluted tubule and collecting ducts reabsorb the final 2.5%** of the filtered load and are responsible for the fine-tuning of Na+ reabsorption, which ultimately ensures Na+ balance. Not surprisingly, then, the late distal convoluted tubule and collecting duct are the sites of action of the Na+-regulating hormone **aldosterone**.
7- Approximately, **0.6 % of Na+ will be excreted**, how to calculate this percent?
   Sodium intake= output =150 mMol/day, which is less than 1% of the filtered load(FL=GFR*PNa+= 180 L/day *140 mMol/L=25,200 mMol/day), excretion fraction = excreted amount/filtered load =150/25,200 *100%= 0.6%.
   You can easily calculate the absorbed load to be 99.4% [25,050(25,200-150) divided by 25,200].

   How could the scientists determine the proportion of reabsorption in each segment?!
   - Along the nephron, the clearance of Na+ can be easily calculated as follows:

CNa+= (UNa+/PNa+)* V
=(100/140) * 1 = 0.7 ml/min

UNa+: {Na+} in urine =(150 mEq/day)/ 1.5 L
(urine output per day)= 100 mEq/L

Now how to calculate the Segmental Clearance:.

Let's take the proximal tubule as an example:
by using the micropuncture technique, by which a micropipette (25 µm) is inserted in different parts of the nephron, we take two samples the 1st one from the bowman's space and the 2nd one from the end of proximal tubules and analyze the concentration of sodium in both samples, we would find that they have same concentration, How?!
- There could be neither secretion nor reabsorption, but it is not the case here!
- What happens is that, sodium and water are reabsorbed to the same proportion through the proximal tubules then the concentration of the sodium still the same!
We can mathematically prove this by the equation below:

Clearance of Na^+ across the proximal tubule:

$$C\,(Na^+) = \frac{Na^+\ concentration\ in\ B}{Na^+\ concentration\ in\ A} X\ flow\ rate = \frac{TF(Na^+)}{P(Na^+)} X\ V$$

- Concentration of Na+ at A = concentration of Na+ in plasma =P(Na+)
- TF (Na+): concentration of Na+ in the tubular fluid
- V is the flow rate in this particular segment

So, we find it to be 140/140=1!

Now, we measure the proportion of water reabsorption to know the proportion of sodium reabsorption in the proximal tubules by Injecting the patient with inulin (neither reabsorbed nor secreted) then we take two; samples 1st in the bowman's space and the 2nd at the end of the proximal tubule. After the analysis of these two samples we calculate the clearance as below:

Clearance of inulin across the proximal tubule:

$$C\,(inulin) = \frac{inulin\ concentration\ in\ B}{inulin\ concentration\ in\ A} X\ flow\ rate = \frac{TF(inulin)}{P(inulin)} X\ V$$

Clearance of inulin= [inulin at the end of proximal tubule] / [Inulin in the bowman's space-plasma] =3/1=3 (the concentration is tripled, so we expect the water to be reduced by 2 thirds). So, 65% of water is reabsorbed, and 65% of Na is reabsorbed.

Now, to relate the Na+ to inulin across a specific segment, we use the following equation:

$$\frac{C_{Na}}{C_{in}} = \frac{\frac{T_{Na} \times V}{P_{Na}}}{\frac{T_{in} \times V}{P_{in}}}$$ Since the flow rate is the same: $$\frac{C_{Na}}{C_{in}} = \frac{\frac{T_{Na}}{P_{Na}}}{\frac{T_{in}}{P_{in}}}$$

If $C_{Na}C_{inulin}$:

a) =1 → this means that Na, is neither reabsorbed nor secreted. However, this is not necessarily true; this substance may be reabsorbed at point and secreted at another point then they cancel each other. Because of this, short segments give more accurate conclusions.

b) >1 → there is secretion of Na at this segment

c) <1 → there was reabsorption

Going back to our example:

Clearance Na / Clearance Inulin = 1/3 which is less than 1, so, Na is reabsorbed.

How can we isolate each segment in the lab?

- By injecting a dye from the cortex, we ensure that we are inside the nephron. Then, we inject 2 drops of oil separated by a moment to isolate the segment between them. After that we apply what we have learnt above.
- The micropuncture technique(micropippate): segmental function of the nephron, discovered by 1924 by Richard brothers, what makes it difficult that it should be done in vivo not vitro. also, it can isolate the cortical segments only not the medullary ones, too.
- To study the medullary segments, the scientists find a way through the pelvis of ureter to reach the kidney and take samples to study them.

Notes:

What is the most important part in handling Na+?? We can answer this question from different aspects.

- the proximal part is the most important because it reabsorbs 65% of filtered Na+
- The descending part can be considered important because it is not permeable to Na+

- **Physiologists** consider the distal part is the most important, as it is under the control(ADH & aldosterone)
- For **pharmacologists**, the most important part is the thick ascending part because it is targeted by diuretics which can be divided into:

a- Potassium wasting agents

1- **Loop diuretics like Furosemide**: works by inhibiting the (Na+: K+: 2*Cl-) carrier of the thick ascending limb of the loop of Henle. This family of diuretics is the strongest one so it is used to treat pulmonary edema (remember the 25% of Na+!).

2- **Thiazide**: works distally to furosemide by inhibiting another carrier (NaCl), it is widely used as it increases the reabsorption of Ca++, so, it reduces kidney stones

b- Potassium sparing agents (**Aldosterone antagonists**); increase potassium in the body by inhibition of aldosterone secretion. So, if a patient takes potassium wasting diuretics, he should take a banana with the drug (to compensate for potassium loss), if this does not work we give him potassium supplements, or I can give him aldosterone antagonists. We must be very careful while using diuretics, due to potassium as we will see)

Regulation of Na+ Reabsorption

Na+ homeostasis is regulated by 3 factors:

1- **GFR**: An increase in Na+ amount in the extracellular fluid will stimulate water intake which causes hypervolemia and elevates the blood pressure. In a consequence to that, GFR increases and thus more Na+ is excreted.

2- **Aldosterone**: aldosterone stimulates Na+ reabsorption in the late distal tubule and the collecting duct through Na+ channels and Na+ -K + pump.
Aldosterone acts on the distal tubules (on the principle cells there) and:
1) it inserts Na+ and K+ channels on the luminal membrane (it is steroid which work to increase transcription)
2) makes Na+/K+ pump (proteins) on the basolateral membrane.
3) makes the enzymes needed to make ATP for the pump.
4) makes sodium and potassium channels (for facilitated transport).

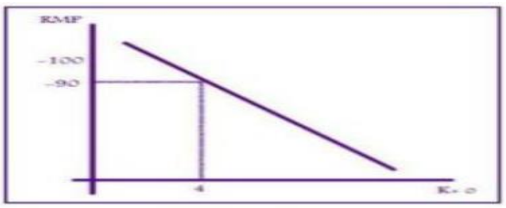
- So, when Na+ amount is high, there will not be Aldosterone secretion.

Hyperaldosteronism (Conn's Syndrome): is a disease in which the adrenal gland(s) make too much aldosterone which leads to hypertension (high blood pressure) (aldosterone cause sodium and water retention) and low blood potassium levels.

3- **ANP**: ANP is secreted by the atria in response to an increase in ECF volume and causes vasodilation of afferent arterioles which in turn increases the GFR, and decreased Na+ reabsorption in the late distal tubule and collecting ducts directly and by inhibiting aldosterone secretion. These all will increase Na+ excretion and urine output.
firstly it was called the 3rd factor, as they didn't know exactly what it does.
Then they found that it is a peptide, so called ANP.
at last, They found out that it is secreted from a place (right atrium) and transported via blood to affect another place (kidney) and due to this fact, they called it a hormone (ANH).

Why do we care about potassium?

- **It is important for the function of excitable tissue; as its concentration gradient across excitable cell membranes determines the resting membrane potential.**
- The concentration of potassium normally inside the cell = 150 mmol/L, and outside the cell = 4 mmol/L, outside the cell concentration should be constant(3.5 to 5.5 mmol/L) (potassium homeostasis), so if the intake increases, the excretion should increase too. **potassium accumulation is very dangerous(sever arrythmias)**.
- **Nernst equation:** This figure illustrates the relationship between k+ concentration outside the cell and resting membrane potential (Nernst equation).



Em=-61 log ([K+]i/ [K+]O)
EK+ = -61 log (K+ in / K+ out)
= -61 log (150/4) = -61 log 35 = -90 mV
you can notice that any change in intracellular K+ is not significant, however little change in extracellular K+ can cause a lot of change in Resting Membrane Potential (RMP), use the equation to ensure this point!

- if K+ concentration low, cells will Hyperpolarized (excitable tissue will face a difficulty to reach the threshold).

- If a patient comes with K+ levels above 7 you go for an ECG, if there are any ECG changes, go for haemodialysis immediately.

-after a meal, insulin is secreted to push glucose and potassium inside cells, and cells secrets it slowly toward the blood to be renally excreted. (extracellular potassium is more dangerous than the intracellular, as we said) so inside the cell; the concentration will be 150(normally) +3.5(form meal) =153.5 mmol/L which is not a problem