

H₂O, O₂_{aqua} circulation and **osmosis** attractors for **colligative** properties

Water circulation **colligative** properties depend only on the concentration into **water** of dissolved particles.

Used concentrations: as molar fraction N; molarity M, osmolarity and molality m.

Colligative circulation of **water** solutions don't depend on dissolved substance. Thus, any colligative circulation of **H₂O, O₂_{aqua}** has the same value, for instance, for sugar, alcohol, **NaCl** or **H₂SO₄** solutions, if the concentration of dissolved particles is the same. There is also no difference, if the dissolved particle is a molecule or an ion - it will give the same increment into the intensity of colligative circulation. **H₂O, O₂_{aqua}** circulation and **osmosis** attractors are: 1) **water**'s evaporation and condensation ,

2) **water** osmosis against concentration gradient across membranes through aquaporins.

Prior to discussion of colligative properties themselves, we have to find the interrelation between the total concentration **C_{total}** of **water dissolved solute** and the concentration **C_{particles}** of **water dissolved particles**.

I. ISOTONIC COEFFICIENT

*Isotonic coefficient **i** (or Vant Hoff's coefficient) is the proportionality coefficient between the total concentration of **water** dissolved solute and concentration of **water** dissolved particles.*

Particle concentration can be calculated from solute concentration **C_{total}** as : **C_{particles} = i • C_{total}** (3.1) ,
 where: **i** is the isotonic coefficient, **C_{particles}** is the concentration of solute particles and **C_{total}** is the total concentration of solute. In other words, **i** shows, how many times particle concentration exceeds solute concentration: **i > 1**.

In solutions of non-electrolytes, where solute doesn't dissociate into ions, the smallest particle is molecule, therefore particle concentration is equal to solute concentration and **i = 1**.

In solutions of electrolytes molecules of solute dissociate into ions and dissociation is characterized by dissociation degree **α**,

Swante Arrenius, Wilhelms Ostwalds 1886.gadā Rīgā

$$\alpha = \frac{n_{diss}}{n_{total}} = \frac{C_{diss}}{C_{total}} \quad \left| \quad \begin{array}{l} \text{where: } n_{diss}, C_{diss} \text{ and } n_{total}, C_{total} \\ n_{diss} \text{ and } C_{diss} \text{ are number and concentration of dissociated molecules respectively,} \\ n_{total} \text{ and } C_{total} \text{ are total number and total concentration of molecules respectively.} \end{array} \right.$$

Dissociation degree **α** can be expressed either as usual decimal number or in percents.

For example both expressions **α = 0.04** or **α = 4%**

mean that 4 molecules out of every 100 molecules are dissociated in ions. Nevertheless, if dissociation degree has to be used in further calculations, it has to be transformed into a decimal number.

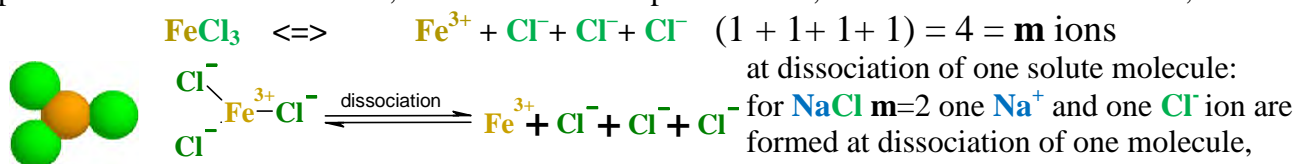
Using the above expression of **α** the concentration of dissociated molecules is found as the total concentration of solute, multiplied by dissociation degree **C_{diss} = α • C_{total}**,

Our task is to express the isotonic coefficient **i** through dissociation degree **α** and the total concentration of solute. Let us first note, that the concentration of particles includes both the concentration of non-dissociated molecules and the concentration of ions: **C_{particles} = C_{nondiss.mol.} + C_{ions}**

The concentration of nondissociated molecules can be expressed as:

$$C_{nondiss} = C_{total} - C_{diss} = C_{total} - \alpha \cdot C_{total}$$

To express the concentration of ions, let us first invent a parameter **m**, which is the number of ions, formed

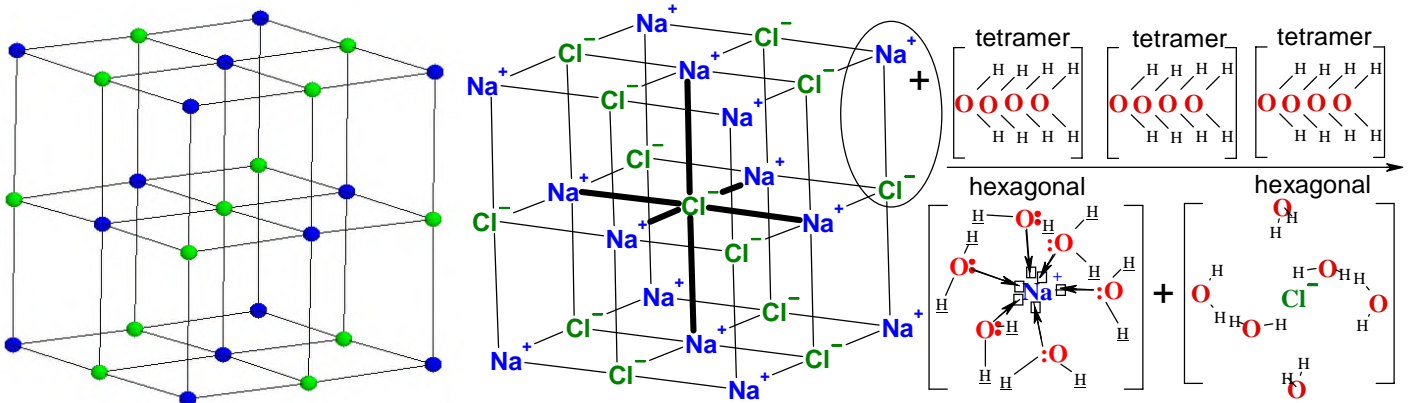


For example, for **K₃PO₄** **m = 4**, as three **K⁺** ions and one **PO₄³⁻** ion are formed. As **m** ions are created at dissociation of one molecule, concentration of ions is **m** times greater, than concentration of dissociated molecules: **C_{ions} = m • C_{diss} = α • m • C_{total}**. Inserting the meaning of **C_{nondiss}** and **C_{ions}** into (3.1), we have:

$$i = \frac{C_{total} - \alpha \cdot C_{total} + m \cdot \alpha \cdot C_{total}}{C_{total}} \quad (3.2)$$

From (3.2.) one can see, that for a non-electrolyte **i = 1**, because it doesn't dissociate into ions and therefore its **α = 0**. As soon as **α > 0** (the solute is an electrolyte), **i** becomes greater than **1** and particle concentration exceeds solute concentration.

Na⁺Cl⁻ Arrenius dissociation theory state sodium chloride in lattice node points has sodium cations **Na⁺** and chloride anions **Cl⁻** surrounded by opposite charged counter ions with coordination number 6. When tetramers dissipate the six water molecules, ligands coordinating as electron pair donors **H₂O** => □ to sodium **Na⁺** empty orbitals □ as electron acceptors.



Dissociation degree $\alpha=1$, if **Na⁺Cl⁻** dispersed in ions. Electrolyte **solution** in water can be treated as a sum of two processes :

- 1) the **separation** crystalline **Na⁺Cl⁻** into positive cations **Na⁺**, negative **Cl⁻** anions, disorder **(H₂O)₄** tetramers,
- 2) the **hydration** of ions $6\text{H}_2\text{O} \Rightarrow \square \text{Na}^+$ un $(\text{Cl}^- + 6\text{H}_2\text{O})_{\text{aqua}}$ coordinated. with six **6H₂O** molecules.

Overall dissociation process free energy change is: $\Delta G_{\text{Hess}} = \Delta H_{\text{Hess}} - T\Delta S_{\text{Hess}}$ **exoergic**,

$$\Delta G_{\text{Hess}} = 3.82 \cdot 1000 - 298.15 \cdot 43.5 = -9150 \text{ J/mol} = -9.15 \text{ kJ/mol} ;$$

$$\Delta H_{\text{Hess}} = -240.1 - 167.2 + 411.12 = +3.82 \text{ kJ/mol}$$
 endothermic heat content change cooling environment;

$$\Delta S_{\text{dispersion}} = -\Delta H_{\text{Hess}}/T = -1000 \cdot 3.82/298.15 = -12.812 \text{ J/(mol K)} ; ; \Delta S_{\text{Hess}} = 59 + 56.5 - (72) = 43.5 \text{ J/(mol K)} ;$$

$$\text{total entropy change in dissociation process } \Delta S_{\text{total}} = \Delta S_{\text{dispersion}} + \Delta S_{\text{Hess}} = -12.812 + 43.5 = 30.688 \text{ J/(mol K)}$$

Conclusions: **Na⁺Cl⁻** dissolution is **exoergic** $\Delta G_{\text{Hess}} = -9,15 \text{ kJ/mol}$ process

Strong electrolytes $\alpha \Rightarrow 1$ $\Delta G_r < 0$ negative **exoergic** are water soluble salt, bases and strong acids.

Weak electrolytes $\alpha \Rightarrow 0$ $\Delta G_r > 0$ positive **endoergic** are water insoluble salts, bases and weak acids.

Ionic strength $I=0,25 \text{ M}$, $I=0,1 \text{ M}$ and $\text{pH}=7,36$ Biochemistry standard conditions since 2003th

μ , I *ionic strength* is total electrolyte ions stoichiometric concentration sum C_{ions} half calculated of C_i times ion charge z_i exponent z_i^2 :

$$I = \mu = C_{\text{ions}} = \alpha^{1/2} \sum C_i z_i^2 .$$

Na⁺Cl⁻ measured values of α are smaller than 1 - they often are around $\alpha = 0.982$. For medical application of 0.305 M isotonic solution osmo molar concentration is necessary to keep constant $C_{\text{osm}}=0.305 \text{ M}$. Total ionic strength $I = \mu = C_{\text{ions}} = \alpha^{1/2} \sum C_i z_i^2$ of salts in to solution should be evaluated to maintain $\alpha = 0.982$ and osmo molar concentration $C_{\text{osm}}=0.305 \text{ M}$ constant.

For 0.01 M solution of **Na₂SO₄** evaluated ionic strength $I = \mu = C_{\text{ions}} = \alpha^{1/2} \sum C_i z_i^2$ calculated from electrolyte dissociation stoichiometry $\text{Na}_2\text{SO}_4 \Rightarrow 2 \text{Na}^+ + \text{SO}_4^{2-}$. Stoichiometry, molarity of total ions concentration : $[\text{Na}^+] = 2 \cdot 0.01 \text{ M} = 0.02 \text{ M}$, $[\text{SO}_4^{2-}] = 0.01 \text{ M}$. Electrolyte **Na₂SO₄** ionic strength is sum:

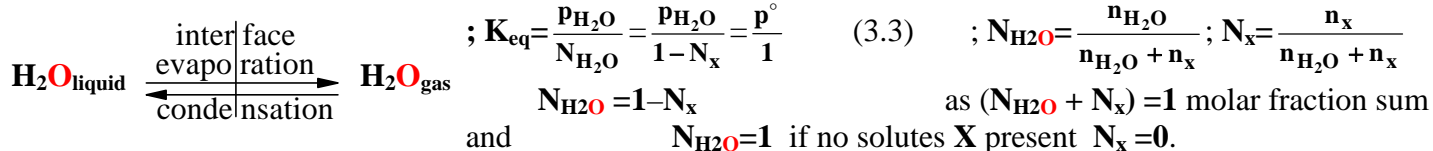
$$2 \cdot 0.01 \text{ M} + 0.01 \text{ M} = 0.03 \text{ M} = C_{\text{ions}} , \text{ if } \alpha=1 \text{ so calculated as:}$$

$$\mu = \alpha^{1/2} (1^2 \cdot 0.02 + (-2)^2 \cdot 0.01) = 1/2 (1 \cdot 0.02 + 4 \cdot 0.01) = 1/2 (0.02 + 0.04) = 1/2 (0.06) = 0.03 \text{ M} = C_{\text{ions}}$$

total ions stoichiometry molarity concentration sum $C_{\text{ions}} = 0.03 \text{ M}$ ($m=2+1=3$; $2\text{Na}^+ + 1\text{SO}_4^{2-}$).

WATERS' VAPOR PRESSURE

Equilibrium Constant K_{eq}

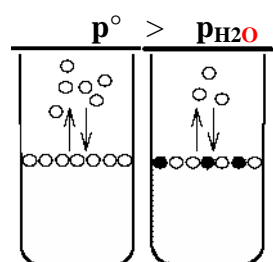


Waters' vapor pressure is the first of the colligative properties of solutions, that we have to deal with. Let us compare the vapor pressures of pure **water** and solution. When pure **water** is in contact with gas phase, two reverse processes proceed at the same time:

- 1) **water** molecules leave the surface of liquid phase (evaporate) and transfer into gas phase,
- 2) as soon as there are **water** molecules in the gas phase, they start to condense - to return back into liquid phase.

After some time an equilibrium is reached, at which the rates of evaporation and condensation are equal and a certain value of **water's** vapor pressure $p^\circ = K_{eq}$ is reached and as solute X present $p_{\text{H}_2\text{O}} / (1 - N_x) = K_{eq}$, fig.3.1.

Now let us consider a solution of a non-fugitive solute \bullet instead of pure **water**, see fig.3.1. The same two processes occur, but another equilibrium is reached. At this case the upper layer of liquid phase doesn't consist only of **water** molecules \circ (empty dots \circ in fig.3.1), but solute \bullet particles (filled dots \bullet) are present, too. For this reason the number of **water** molecules in the upper layer of liquid is smaller, than for pure **water**, the rate of evaporation is smaller, too and the equilibrium will be therefore reached at a smaller vapor pressure. Thus, the vapor pressure p above solution is smaller, than vapor pressure p° above pure **water** and difference value is depression $(p^\circ - p_{\text{H}_2\text{O}}) = \Delta p$.



Because of the reasons mentioned above the vapor pressure of **water** must be different at different concentrations of solute particles - the greater is the concentration of solute particles, the less **water** molecules remain in the surface layer of solution and, hence, the smaller becomes vapor pressure of the **water**.

Fig.3.1. Waters' vapor pressure above pure water (a) and solution (b).

The dependence of **waters'** vapor pressure on the concentration of solute is described quantitatively by Roul's I law, which states, that:

Relative depression of water's vapor pressure is equal to molar fraction of solute particles.

$$N_x = \frac{p^\circ - p_{\text{H}_2\text{O}}}{p^\circ} \quad \text{Mathematical form of this law is, where: } p^\circ = p_{\text{H}_2\text{O}} / (1 - N_x) \text{ is the equilibrium constant } K_{eq} \text{ (3.3),}$$

$$(p^\circ - p_{\text{H}_2\text{O}}) / p^\circ = \Delta p / p^\circ \text{ is called the relative depression of vapor pressure, } n_x \text{ and } n_{\text{H}_2\text{O}}$$

are numbers of moles of solute particles and **water** respectively. If the solute X is a non-electrolyte, $N_x = N_{\text{solute}}$ as the solute X is present only in molecular form, for electrolytes $N_x = i N_{\text{solute}}$, because the number of particles is i times greater than number of molecules. Looking at fig.3.1, one can also understand, that, the greater is concentration of solute, the lower will be **water's** vapor pressure.

WATER PHASE DIAGRAM Prior to discussion of next two colligative properties - boiling point raise and freezing point depression of solution, compared to pure **water**, we have to understand the **water** phase diagram, see fig.3.2. In this diagram pressure is plotted versus temperature. Three curves **AO**, **OB** and **OC** divide the diagram into three regions **a**, **b** and **c**. Considering region **a**, where pressures are high, but temperatures are low, it is likely, that **water** is solid. In region **C**, where pressures are low, **water** is gas in wide temperature interval.

Region b at medium temperatures and high pressures corresponds to liquid **water**. **Fig.3.2. Water phase**

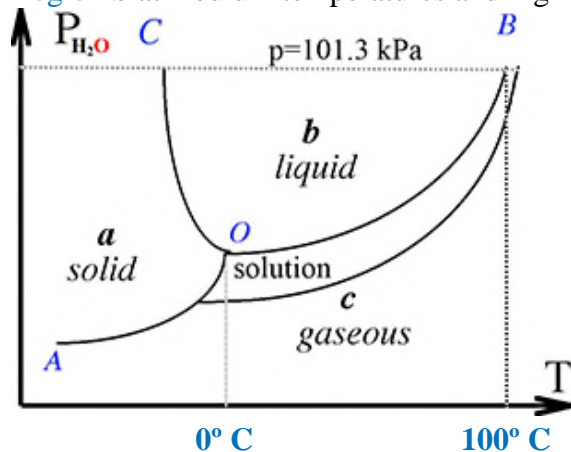


diagram. Curve **AO** separates gaseous phase **water** vapor and solid phase's ice. This curve expresses the equilibrium between solid **water** (an ice) and gaseous **water** vapor **c**. Each point of curve **A** shows the vapor pressure above ice at a given temperature. Curve **OB** is the equilibrium curve between liquid **water** and **water** vapor and each point on this curve shows the vapor pressure of liquid **water** at a given temperature. Point **O**, which is the intersection point of all three curves, is the point, at which vapor pressure above solid and liquid **water** is equal. It is the melting point of ice or freezing point of liquid **water** and the freezing temperature of liquid **water** can be found as abscissa of this point 0°C .

Evaporation point of **water** at temperature 100°C when **water** vapor pressure $p_{\text{H}_2\text{O}}$ is equal to atmosphere pressure $p = 101,3 \text{ kPa}$ starts the boiling the **water** if heat supply continues.

FREEZING POINT DEPRESSION As it was shown in the previous chapter, freezing point of liquid **water** is the intersection point of vapor pressure curves above liquid **water** and ice. In order to see difference between freezing points of pure **water** (**water**) and solution, one has to show vapor pressure curves of pure **water** (**water**) and solution in the same diagram. As it is clear from previous considerations (see fig.3.1 and comments to it), vapor pressure of solution is lower, than vapor pressure of pure **water**. As this is true at any temperature, the entire vapor pressure curve for solution lies lower than for pure **water**, see **Fig.3.3**.

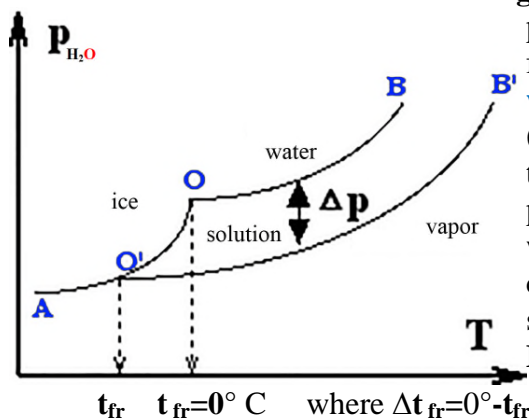


Fig.3.3. Freezing point depression. As the result of this, the intersection point between vapor pressure of solution and vapor pressure of ice (the freezing point of solution) lies at lower temperature, than for pure **water** - the freezing point is shifted towards lower temperatures (depressed). From considerations, discussed above, one can see, that the greater is the concentration of solute, the lower lies the vapor pressure curve of solution and the more freezing point of solution would be depressed when compared to pure **water**. Mathematically the connection between freezing point depression and concentration of solute is expressed by II Roul's law states: $\Delta t_{\text{freezing}} = i K_{\text{cr}} C_m$ (3.4). **Freezing point depression is proportional to molality of solute,**

where $\Delta t_{\text{fr}} = 0^\circ - t_{\text{fr}}$ is the difference between freezing temperatures of **water** and solution, C_m is molality of solute (number of solute moles in 1000 grams solution),

Freezing (cryos greekish)

$K_{\text{cr}} = 1.86$ is the *cryoscopy constant* of the **water**.

Cryoscopy constant of water 1.86 shows the freezing point depression in a 1 molal non-electrolyte solution (where $i = 1$) freezes at temperature $-1,86^\circ \text{C}$ less zero 0°C .

Cryoscopy constant is a constant value for **water** and it doesn't depend on the properties of solute, because the freezing point depression as a colligative property is affected by concentration of particles, but not by their nature. In fact, Roul's laws are strongly valid only for diluted solutions, while it is possible to ignore the interaction of solute particles. For this reason one can use molarity instead of molality for approximate calculations - $C_m \approx C_M$ in diluted solutions.

BOILING POINT RAISE Boiling point of solution, as well as the freezing point of solution, differs from the boiling point of pure **water**. To understand this, let us think a little about the boiling process.

Boiling (ebullios greekish) is the evaporation of **water** from all the liquid phase - bubbles of gaseous **water** are formed in all the volume of liquid phase and come out of it.

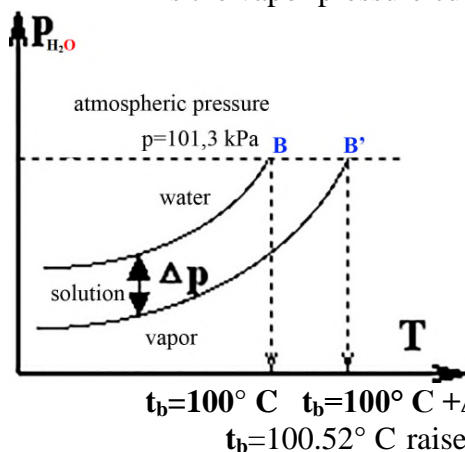
When a liquid is heated, at low temperatures the evaporation of **water** occurs only from the surface of liquid. Only at a certain temperature (100°C for **water**) bubble formation in volume of liquid phase begins.

The factor, that doesn't allow the bubble formation at lower temperatures, is the atmospheric pressure - while vapor pressure inside bubble is smaller, than atmospheric pressure, the bubble is suppressed and cannot leave volume of liquid phase.

Thus, boiling starts at a temperature, at which vapor pressure of **water** reaches atmospheric pressure, therefore the boiling point of liquid is found as an intersection point between vapor pressure curve of liquid phase and the atmospheric pressure level, see fig.3.4. The intersection point of pure **water** vapor pressure curve **OB** and normal atmospheric pressure level corresponds to temperature 100°C . **Fig.3.4. Boiling point raise.**

As the vapor pressure curve of solution **O'B'** lies lower, than the vapor pressure curve of pure **water**, its intersection with atmospheric pressure level is situated at lower temperature, than for solutio. In other words, as the vapor pressure of solution at any temperature is lower than the one of pure **water**, at 100°C vapor pressure above solution has not yet reached level of atmospheric pressure and the solution has to be heated up to a higher temperature to start boiling.

The boiling point raise (the difference between the boiling points of solution and pure **water**) is linked to solute concentration by an equation, similar to the one for freezing point: $\Delta t_{\text{boiling}} = i K_{\text{eb}} C_m$, (3.5) where $K_{\text{eb}} = 0.52$ **ebullioscopy** (**ebullios** means boiling in Greek)



constant of **water**, which shows the value of boiling point raise to value of temperature in 1 molal solution of any non-electrolyte in **water**.

CRYOSCOPIC osmo molar values determination and APLICATIONS

1. Cryoscopy and ebullioscopy is used for *quantitative analysis* of non electrolyte compounds.

In the case of non electrolytes $i=1$, thus, measuring of Δt and knowing $K_{cr}=1.86$ or $K_{eb}=0.52$ one can calculate concentration of solution. In the case of electrolytes dissociation degree and, consequently, i is different at different concentrations, therefore the equation of Roul's 2nd law contains two unknown values C and K and therefore cannot be used for quantitative analysis

2. Cryoscopy and ebullioscopy is used for determination of the **molar mass** of solute.

For ordinary compounds molar masses can be calculated, knowing the formula of compound and atomic weights of the elements, included in it. For polymer compounds, such as carbohydrates (polysaccharides), proteins and other biologically important compounds the formula of compound is not known, as the structure of a polymer is a long chain, consisting of many times repeating fragments. Molecular mass is not equal for all the polymer molecules, as the length of polymer chain can differ. Here an average value of molar mass can be determined by cryoscopy or ebullioscopy, if the molality C_m is expressed through the mass and molar mass of solute. If m^{1000} is the mass of solute, dissolved in 1000 grams of **water** and M is its molar mass, molality of solution is: $C_m = m^{1000} / M$, because at dividing of solute mass, contained in 1000 grams of **water** by the molar mass of solute, one obtains the number of moles of solute in 1000 grams of solution.

Inserting this instead of C_m into equation of Roul's 2nd law, one gets:

$$\Delta t = m \frac{1000}{M} \times K_{cr} \text{ and } M = m \frac{1000}{\Delta t} \times K_{cr}$$

At this and following applications for biological objects cryoscopy only is used, as the biological objects are denatured at boiling and application of ebullioscopy is therefore impossible.

3. Determination of **isotonic coefficient** and **dissociation degree** of electrolyte **NaCl 0.9%**.

To determine the dissociation degree of physiological solution electrolyte **NaCl** as is prepared known concentration 0.9%, its freezing point depression is measured $t_{freezing} = -0.567^\circ \text{C}$ and i is calculated from Roul's 2nd law, knowing the **water** value of cryoscopy constant $K_{cr} = 1.86$.

As $i = 1 + \alpha(m-1)$, dissociation degree can be expressed as $\Delta t_{freezing} = i \cdot C_m \cdot K_{cr}$ if is 0.9% mass fraction physiology solution of NaCl $\rho = 1.000 \text{ g/mL}$:

$$m_{NaCl} = \frac{w\% \cdot m_{solution}}{100\%} = \frac{0.9\% \cdot 1000g}{100\%} = 9 \text{ g}; C_m = \frac{m_{NaCl}}{M_{NaCl} \cdot V} = \frac{9g}{58.5g/mol \cdot 1L} = 0.1538 \text{ M};$$

$$i = \frac{\Delta t_{freezing}}{C_m \cdot K_{cr}} = \frac{0.567}{0.1538 \cdot 1.86} = 1.982 \text{ and}$$

sodium chloride NaCl dissociation degree is $\alpha = \frac{i-1}{m-1} = \frac{1.982-1}{2-1} = 0.982$ and in percentage is **98.2%**. $\alpha\%$.

4. Cryoscopy is used for determination of **osmo molarity** of **blood** $C_M=0.305 \text{ M}$ and **osmotic pressure**

This application of cryoscopy will be discussed later, see chapter IX of this part. Let us only mention here, that for biological liquids the osmotic pressure cannot be calculated, as they contain a mixture of different solutes, each of them having its own i and concentration, therefore summary concentration of particles is i times greater than number of molecules. Osmo molar concentration $C_{osm} = \sum i_n C_n$ is detected from the **blood** freezing data of microscope $t_{freezing} = -0.567^\circ \text{C}$ in cryoscopy experiment $\Delta t_{freezing} = 0^\circ - t_{freezing} = 0^\circ - (-0.567^\circ) = 0.567^\circ$:

$$C_{osm} = \frac{\Delta t_{freezing}}{K_{cr}} = \frac{0.567}{1.86} = 0.305 \text{ M}; \text{ as Roul's 2 law } \Delta t_{freezing} = K_{cr} \cdot C_{osm} = 1.86 \cdot 0.305 = 0.567^\circ .$$

For further calculation **blood** osmotic pressure at human body temperature $T=310 \text{ K}$ is:

$$\pi_{blood} = C_{osm} \cdot R \cdot T = 0.305 \cdot 8.3144 \cdot 310 = 786 \text{ kPa}$$

Osmotic pressure $J/L = kPa$ shows free osmotic pressure energy amount in one liter erythrocyte cells. If one liter erythrocyte cells are in water at $T=310 \text{ K}$ $\pi = \Delta C_M R T = 0.305 \cdot 8.3144 \cdot 310 = 786 \text{ J/L}$, than one liter volume of erythrocytes have 786 joules osmotic pressure free energy $\Delta G_\pi = \pi \cdot V = 786 \text{ J}$.

H_2O and $\text{O}_{2\text{aqua}}$ **osmosis** against concentration gradient ΔC_{osm} through aquaporins in membranes
Osmosis is flux of water through membrane aquaporins against concentration gradient.

Osmosis phenomenon are **Aquaporins** forming across **membrane** **osmotic pressure** $\pi = \Delta C_{\text{osm}}RT$, (kPa)

Water molecules transfer through aquaporin tunnel has the rate $3 \cdot 10^9 \text{ sec}^{-1}$ (AQP1).

Osmosis of H_2O and $\text{O}_{2\text{aqua}}$ flow to higher concentration ΔC_M so make pressures on membrane.

Osmotic pressure $J/L = \text{kPa}$ shows free osmotic pressure energy amount for one liter cell. If one liter erythrocyte cells are in **water** at $T = 310 \text{ K}$ $\pi = \Delta C_{\text{osm}}RT = 0.305 \cdot 8.3144 \cdot 310 = 786 \text{ J/L}$, than one liter volume $V = 1 \text{ L}$ of erythrocytes have 786 joules free energy as kJ $\Delta G_{\pi} = \pi \cdot V = 786 \text{ J/L} \cdot 1 \text{ L} = 786 \text{ J} = 0.786 \text{ kJ}$.

Aquaporins are selective channels for **water** and $\text{O}=\text{O}$, $\text{N}=\text{O}$, $\text{C}=\text{O}$ molecules across cell wall membrane, **Water** and O_2 , NO , CO molecules are smallest, neutral molecules of life and solute molecules are very large. **Aquaporin** tunnel size in constriction region is 1.5 \AA is **water** molecule size and unless greater as $\text{O}=\text{O}$, $\text{N}=\text{O}$, $\text{C}=\text{O}$ molecule sizes, therefore transfer $\text{O}=\text{O}$, $\text{N}=\text{O}$, $\text{C}=\text{O}$ molecules too.

Let us suggest, that there is pure **water** and $\text{O}=\text{O}$, $\text{N}=\text{O}$, $\text{C}=\text{O}$ in the left side of the vessel and a solution in the right one NaCl . According to the definition of membrane **water** selective aquaporins, **water** molecules can penetrate into both directions, but membrane aquaporins are closed for **water** with greater size solute molecules.

From right side large size dissociated in charged ions salt $\text{NaCl} \Rightarrow \text{Na}^+ + \text{Cl}^-$ molecules close channels for H_2O , $\text{O}_{2\text{aqua}}$ molecules. Aquaporins transfer the single-file line of 8 H_2O molecules [Illinois06.pdf](#).

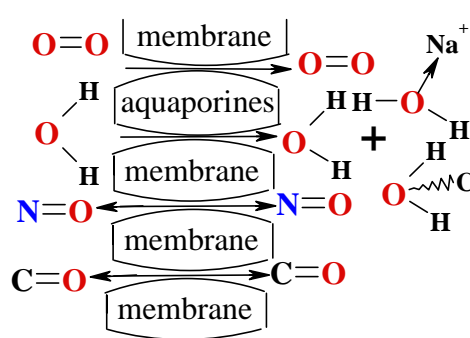


Fig.3.5. Origin H_2O and $\text{O}=\text{O}$ osmosis in to hypertonic medium Na^+ , Cl^- , H_3O^+ , OH^- right Flux **water** and **oxygen** molecules into right side of vessel is greater, than in left direction. As long as the concentration difference between both sides of vessel exists, the flow of **water** from the left to the right will continue. (Note, that the flow of **water** occurs at the direction from pure **water** towards solution, i.e. at direction, opposite to concentration of solute gradient).

The flow of **water** molecules from left to right causes

a pressure, acting at the membrane against direction as the flow of **water**. This pressure is called **osmotic pressure** and a symbol π is used for it (some books use symbol p_{osm} as osmotic pressure).

DETERMINATION OF OSMOMOLAR CONCENTRATION OF BIOLOGICAL LIQUIDS

Osmotic pressure has an important role in biological processes, as in all the cell membranes are aquaporins, therefore it is often necessary to know the osmotic pressure of a biological liquid (consider **blood**, sweat, saliva, urine, etc). Biological liquids consist of many components, having different concentrations and different isotonic coefficients. Usually the individual concentrations and isotonic coefficients of all components are not known, therefore it is impossible to calculate the osmotic pressure using Vant Hoff's law directly.



OSMOMAT 030 CRYOSCOPIC OSMOMETER 50 $\text{mL} = 0.05 \text{ mL}$ of solution.

In these cases cryoscopy is used to determine the so-called *osmotic concentration* or *osmolarity*, C_{osm} , which can be directly inserted into Vant Hoff's equation to calculate the value of osmotic pressure. This osmolarity is actually a sum of $i \cdot C$ products of all the solutes of solution: $C_{\text{osm}} = i_1 \cdot C_1 + i_2 \cdot C_2 + i_3 \cdot C_3 + \dots = \sum i_k \cdot C_k$,

where

i_k is the isotonic coefficient of selected compound,

C_k is selected compound molar concentration.

Roul's 2nd law for solution of one compound gives freezing point depression:

$$\Delta T_{\text{freezing}} = i \cdot C_m \cdot K_{\text{cr}} = K_{\text{cr}} \cdot \sum i_k C_k = C_{\text{osm}} K_{\text{cr}}; \text{ and } K_{\text{cr}} = 1.86 \text{ (degree/mol)}$$

For a solution of several solutes, this equation becomes

$$0^\circ \text{C} - T_{\text{freezing}} = \Delta T_{\text{freezing}} = C_{\text{osm}} K_{\text{cr}} = C_{\text{osm}} \cdot 1.86 \text{ (degree)}$$

CALCULATION OF OSMOTIC PRESSURE

Vant Hoff, who studied the dependence of osmotic pressure on the temperature and concentration, found out an analogy between osmotic pressure laws and gas laws. If we change the form of ideal gas equation:

$$pV = n \cdot R \cdot T$$

(**p** - pressure, **V** - volume, **n** - number of moles of gas, **R** - universal gas constant, **T** - temperature) in such a way, that volume is at its right side, we have:

$$p = \frac{n}{V} \times R \times T; \quad p = C_M \times R \times T$$

The ratio $\frac{n}{V}$ gives the number of moles in a unit of volume i.e. the molar concentration. If one wants to use volume 1 litre as it is usually done in chemistry. To introduce a coefficient 1000 for calculation from m^3 to liters:

$$p = 1000 C_M R T$$

In such a way, the last form of ideal gas equation shows, that the **pressure of gas is proportional to the molar concentration of gas and to temperature**, the proportionality coefficient being **R** if volume is measured in liters or **1000R** if volume is in m^3 .

Vant Hoff found out, that **osmotic pressure of solution is proportional to the molar concentration of solute and to temperature**, having the same proportionality coefficient (**R** or **1000R**), i.e. he showed, that the laws of osmotic pressure are similar to gas laws.

Vant Hoff's law can be formulated as follows:

The osmotic pressure of a solution is numerically equal to the gas pressure, that would be observed, if the solute was in a gaseous form at the same temperature.

Mathematical expression of Vant Hoff's law for non-electrolyte solutions is :

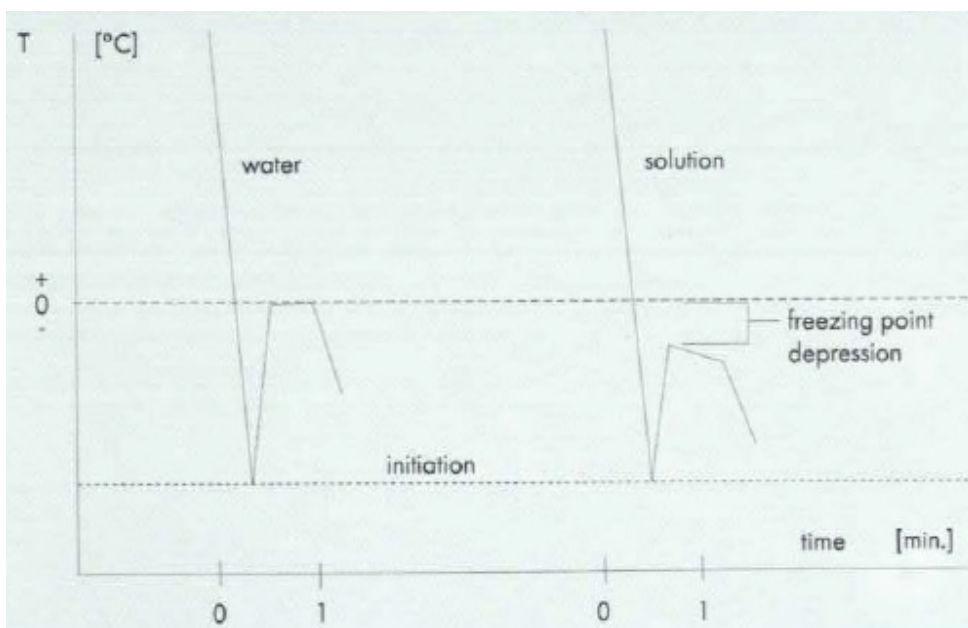
$$\pi = 1000 C_M R T, \quad (\text{Pa})$$

In electrolyte solutions the osmotic pressure is caused by all solute particles and the concentration of solute particles is found as iC_M therefore the expression of Vant Hoff's law becomes:

$$\pi = 1000 i C_M R T, \quad (\text{Pa})$$

The coefficient 1000 is inconvenient therefore it usually is not written, but one has to remember, that then osmotic pressure is calculated in kilo pascals (kPa) and not in pascals (Pa):.

$$\pi = i C_M R T, \quad (\text{kPa})$$



Now, if $\Delta T_{\text{freezing}}$ is measured, value of C_{osm} can be determined without knowing the particular isotonic coefficients and concentrations of all solutes. Inserting C_{osm} into Vant Hoff's law one calculates a correct value of osmotic pressure.

The osmotic pressure is:

$$\pi = C_{\text{osm}} R T, \quad (\text{kPa})$$

For a solution of several solutes, this equation becomes
 $0^\circ \text{C} - T_{\text{freezing}} = \Delta T_{\text{freezing}} = C_{\text{osm}} K_{\text{cr}} = C_{\text{osm}} \cdot 1.86 \text{ (degree)}$

RED BLOOD CELLS AT DIFFERENT OSMOMOLARITY IN WATER SOLUTIONS

Water and oxygen osmosis against osmo molar concentration gradient crosses cell membranes

Osmosis is organised for H_2O and O_2 movement against concentration gradients of colligative properties $\Delta C_{\text{osm}} = i\Delta C_M$ through an **Aquaporins** across cell **membranes** to form the osmotic pressure:

$$\pi = i\Delta C_M RT \text{ (kPa) ,}$$

where $R=8,3144 \text{ J/(mol}\cdot\text{K)}$ universal gas constant,

T temperature in Kelvin's degree (K) $T=t^\circ+273.15$ (if $t=37^\circ$ than $T=37^\circ+273.15=310.15 \text{ K}$).

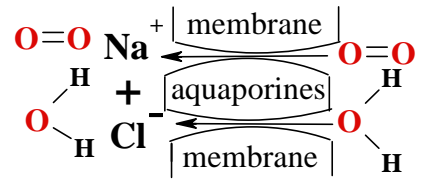
Note: Transfer **water** and oxygen molecules through membrane aquaporin tunnel in erythrocytes with rate $3\cdot 10^9 \text{ sec}^{-1}$ in both directions transfer 3000 oxygen molecules in second.

Mechanism of osmosis through membrane aquaporins

$\text{Na}^+\text{Cl}^- \Rightarrow \text{Na}^+ + \text{Cl}^-$ $m=2$ electrolyte dissociation $\alpha=1$ $i=1+\alpha(m-1)=1+1(2-1)=2$ the concentration doubled as i is 2 $iC_M = 2C_M = C_{\text{osm}}$ and pressure on membrane in plasma $C_{\text{osm}}=i_1C_1 + i_2C_2 + i_3C_3 + \dots = \sum i_k C_k = 0,305 \text{ M}$;

is $\pi = 2\Delta C_M RT = C_{\text{osm}} RT = 0,305 \cdot 8,3144 \cdot 310 = 786,5 \text{ kPa}$. Press \Rightarrow on membrane to right.

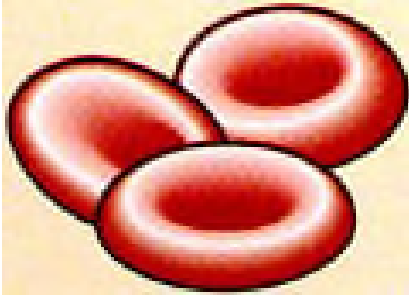
Water H_2O , O_2 oxygen flow left side against the concentration gradient from 0 to $C_{\text{osm}}=0.305 \text{ M}$ because Na^+Cl^- ions make osmo molar concentration left side $C_{\text{left}} - C_{\text{right}} = C_{\text{osm}} - 0 = C_{\text{osm}} = iC_M$ and close H_2O , O_2 flow to right side.



$C_{\text{blood}} = C_{\text{osm}} = i_1C_1 + i_2C_2 + i_3C_3 + \dots = \sum i_k C_k = 0,305 \text{ M}$

Human erythrocytes red blood cells with osmo molar concentration 0.305 M of all solutes sum $\sum i_k C_k$:

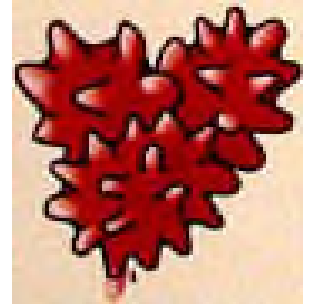
C_{blood} glucose, salts, hydroxonium H_3O^+ , hydroxyl OH^- ions, amino acids, proteins, bicarbonate etc.



Isotonic medium
 $C_{\text{blood}} = 0.305 \text{ M}$



Hypotonic medium
distilled water 0 M
or at least



Hypertonic solution
 $C_{\text{Hyperton}} \geq 0,4 \text{ M}$

osmo molar concentration $C_{\text{Hypoton}} \leq 0,2 \text{ M}$.

Hypotonic 0.2 M medium the flow of **water** is greater towards the cell against the concentration 0.305 M gradient and the cell puffs up until its membrane is broken but content leak in plasma.

$$\pi = (C_{\text{osm}} - C_{\text{Hypoton}})RT = \Delta C_{\text{osm}} RT = 0,105 \cdot 8,3144 \cdot 310 = 270,76 \text{ kPa}$$

Hypertonic salt solutions to apply for purulent wounds, because pumps **water** toxic compounds out and stimulates **blood** circulation.

Osmosis H_2O and O_2 against concentration gradient through alveolar epithelial membrane

A) **Oxygens O_2** from **AIR** 20.95% $\text{O}_2 \uparrow$ gas assimilation reaction dissolution in water to form $\text{O}_{2\text{aqua}}$ exothermic $\Delta H_r = -55,7 \text{ kJ/mol}$ and exoergic $\Delta G_r = -27,7 \text{ kJ/mol}$ as water soluble oxygen :

1) $\text{O}_{2\text{AIR}} + \text{H}_2\text{O} \rightleftharpoons \text{H}_2\text{O} + \text{O}_{2\text{aqua}} + Q + \Delta G$. Penetrate in Human body through aquaporins against concentration gradient up $[\text{O}_{2\text{aq}}] = 9,768 \cdot 10^{-5} \text{ M}$ on **arterial** $[\text{O}_{2\text{aq}}] = 6 \cdot 10^{-5} \text{ M}$ to **venous** blood $[\text{O}_{2\text{aq}}] = 0,426 \cdot 10^{-5} \text{ M}$.

2) $\Delta G_{\text{O}_2} = RT \ln([\text{O}_{2\text{Blood}}]/[\text{O}_{2\text{aqua}}]) = -4,29 \text{ kJ/mol}$ exoergic entrance human organism;

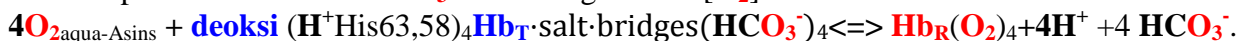
3) $\text{O}_{2\text{aqua}} + \text{H}_2\text{O} \xrightarrow{\text{Aquaporins}} \text{H}_2\text{O} + \text{O}_{2\text{aqua}} + \Delta G$ against concentration gradient 0,305 M / 0,2 M:

One liter of cells with gradient concentration $\Delta C_M = 0.105 \text{ M}$ at $T = 310 \text{ K}$ temperature

$\pi = \Delta C_M RT = 0.105 \cdot 8.3144 \cdot 310 = 286 \text{ J/L}$ have 286 joules osmotic pressure free energy $\Delta G_\pi = \pi \cdot V = 286 \text{ J} = 0.286 \text{ kJ}$.

$\Delta G_{\text{H}_2\text{O}} = RT \ln([\text{H}_2\text{O}]_{\text{right}}/[\text{H}_2\text{O}]_{\text{left}}) = -8,3144 \cdot 310,15 \cdot \ln(0,305/0,2) = -1.088 \text{ kJ/mol}$

exoergic $\Delta G_{\text{O}_2} = -5,379 \text{ kJ/mol}$. **Deoxy** hemoglobin Hb_T adsorbs 4 $\text{O}_{2\text{aqua}}$ from blood plasma of inspired fresh AIR releases four protons 4H^+ and 4 HCO_3^- stabilizing arterial $[\text{O}_2] = 6 \cdot 10^{-5} \text{ M}$ concentration



Total exothermic $\Delta H_H = -55,7 \text{ kJ/mol}$ and exoergic $\Delta G_{\text{O}_2} = -27,7 + -4,29 + -1.088 = -33.078 \text{ kJ/mol}$

Osmosis is **water** and oxygen flow left side against osmolar concentration gradient ΔC_{osm} :

$$\Delta C_{osm} = C_{osm} - C_{Hypoton} = 0.305 - 0.2 = 0.105 \text{ M with pressure energy } \pi = \Delta C_{osm} RT = 0.105 * 8,3144 * 310 = 271 \text{ J/L or kPa.}$$

Alveolar cells membrane not broken, because collagen fibers elastic frame the cells like as plant cells cellulose.

Plant roots osmolar concentration $C_{osm} = 0.1 \text{ M}$ and soil fresh water $C_{Hypoton} = 0.001 \text{ M}$ osmolar

pressure energy in one liter of cell on membrane is

$$\pi = \Delta C_{osm} RT = 0,999 * 8,3144 * 310 = 255,3 \text{ J/L or kPa.}$$

B) Breath out H_2O , CO_2 in endothermic but exoergic reactions on alveolar epithelial surface



endothermic $\Delta H_H = 9.75 \text{ kJ/mol}$; athermic $\Delta H_H = 0 \text{ kJ/mol}$; exotherm. $\Delta H_H = -9.76 \text{ kJ/mol}$; endothermic $\Delta H_H = 20.3 \text{ kJ/mol}$;

endoergic $\Delta G_H = 58.4 \text{ kJ/mol}$; exoerg. $\Delta G_H = -22.5 - 1,96 \text{ kJ/mol}$; exoerg. $\Delta G_H = -58.2 \text{ kJ/mol}$; exoerg. $\Delta G_H = -8,54 \text{ kJ/mol}$;

Physiologic pH=7,36 $Q_{aqua} + CO_{2,aqua} + 2H_2O \xleftarrow{CA} H_3O^+ + HCO_3^- + Q \xleftarrow{Membrane} H_2O + CO_2 \uparrow_{gas} + H_2O \uparrow_{gas}$ surface pH=5

endothermic $\Delta H_{Hess} = 9.75 \text{ kJ/mol}$; endothermic $\Delta H_H = 54,5 \text{ kJ/mol}$; summary endothermic $\Delta H_H = 64,25 \text{ kJ/mol}$;

endoergic $\Delta G_{Hess} = 58.4 \text{ kJ/mol}$; exoergic $\Delta G_{Hess} = -82,1 \text{ kJ/mol}$; summary exoergic $\Delta G_{Hess} = -23,7 \text{ kJ/mol}$;

Venous **deoxy Hb_T shuttle** adsorbs four **oxygen** $4O_{2Hb}$ molecules and releases $4H^+$, $4HCO_3^-$ in one cycle circulation of blood $473 * 6 \cdot 10^{-5} \text{ M} = 0,0284 \text{ M} = [HCO_3^-]$ amounts [O2Solutions.pdf](#), which shifts via membrane channels equilibrium to right $H^+ + HCO_3^- + Q \leftrightarrow H_2O + CO_2 \uparrow_{gas}$. So pH=7,36 remains constant, through respiration. promoting $CO_2 \uparrow_{gas}$ due to increased H^+ , HCO_3^- .

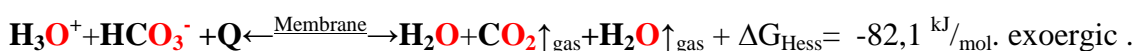
The epithelial cell surface of **lungs** has the specific building. $S = 950 \text{ nm} \times 950 \text{ nm} = 0.9 \mu\text{m}^2$ is surface area with super thin 0.6 nm **water** layer volume: $0.5415 \cdot 10^{-3} \mu\text{m}^3 = 0.5415 \cdot 10^{-18} \text{ L}$. Created acidity in thin **water** layer volume increases up to pH=5.5 if one proton H^+ crosses the membrane channels reaching the surface. Hydrogen ion concentration is: $[H_3O^+] = 10^{-pH} = 10^{-5.5} \text{ M}$. Respiration in **lungs** Hemoglobin released protons H^+ during oxygen adsorbtion for one total blood circulation cycle amount concentration [O2Solutions.pdf](#):is:

$$[O_{2Hb}] = [H_3O^+] = 473 * 6 \cdot 10^{-5} \text{ M} = 0,0284 \text{ M forms } H^+, HCO_3^- \text{ concentration gradient:}$$

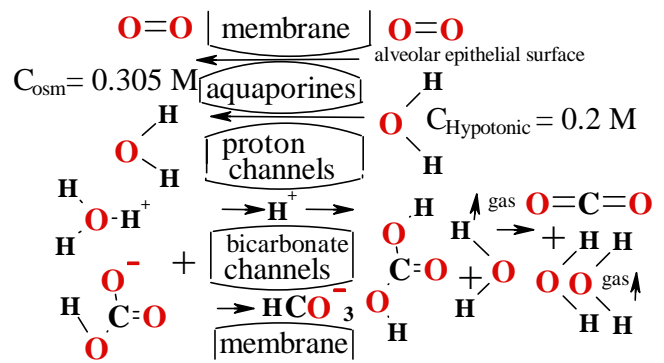
$[H_3O^+]_{right} / [H_3O^+]_{left} = 10^{-5.5} / 0,0284$, which drives exoergic $\Delta G = -22,5 \text{ kJ/mol}$ proton movement through epithelial cell membrane proton channels: $H_3O^+_{left} \xleftarrow{\text{proton channel}} H_3O^+_{right} + \Delta G$. General process

$H_2O + CO_2 \uparrow_{gas} + H_2O \uparrow_{gas}$ require heat supply endothermic $\Delta H = 54,5 \text{ kJ/mol}$ to drive spontaneous

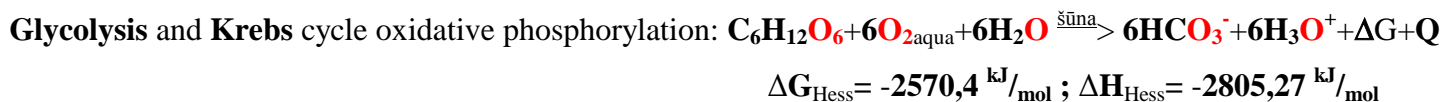
$\Delta G = -82,0679 \text{ kJ/mol}$ products evaporation $CO_2 \uparrow_{gas}$ and $H_2O \uparrow_{gas}$ keeping moisture H_2O on surface of membrane. Hydrogen ions water acidity shift endothermic $\Delta H_{Hess} = +54,5 \text{ kJ/mol}$ and exoergic $\Delta G_{Hess} = -82,1 \text{ kJ/mol}$ decomposition $H_3O^+ + HCO_3^-$ breath out to AIR $CO_2 \uparrow_{gas}$ with $H_2O \uparrow_{gas}$:



Aquaporins are wide class of **membrane crossing channel** proteins, which are **integrated** in all living organisms: **animals, plants, bacteria**. On Cell membranes effecting Physiology, Biochemistry and Health. **Aquaporins** are large families (over 450 members) that are present in all kingdoms of life.

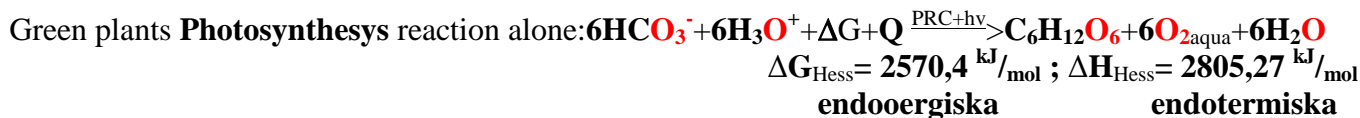
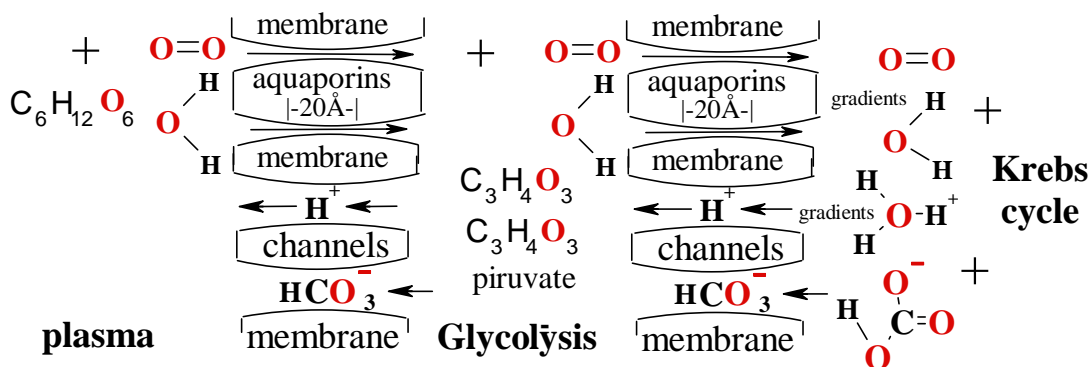


$H_2O, O_{2,aqua}$ Osmosis driving force of life across cell membrane aquaporin channels



exoergic exothermic

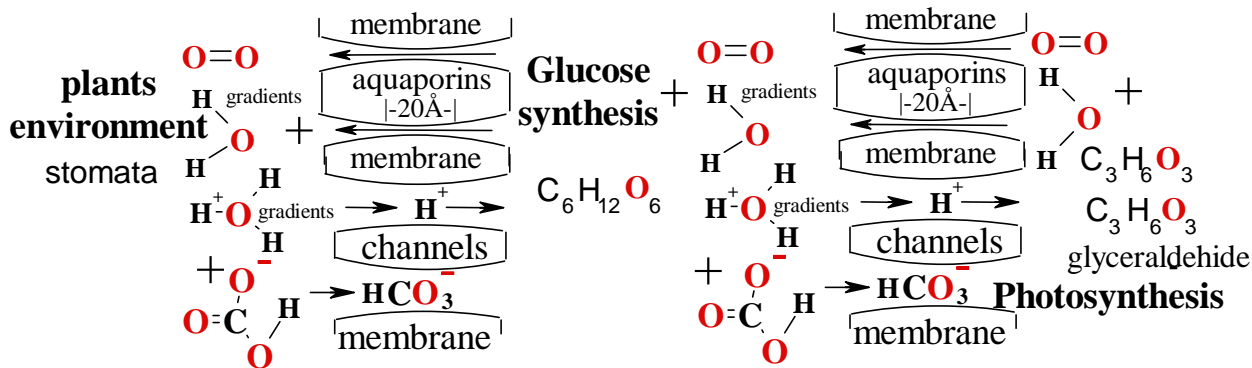
increases cell and mitochondria osmolar concentration from 1 glucose molecule to 12, forming difference 11 molecules = $\Delta Cosm$ across membrane, driving osmosis of $6O_{2,aqua} + 6H_2O$, shift oxidative phosphorylation to produce $6HCO_3^- + 6H_3O^+$, increase $\Delta Cosm$. By $H_2O, O_{2,aqua}$ osmosis into cell driven exoergic and exothermic process organism maintain the body temperature with supplied heat amount Q .



thermodynamic forbidden but joint in tandem with Photosynthesis enzyme complexes synthesises

$C_6H_{12}O_6 + 6O_{2,aqua} + 6H_2O$ products, decreases concentration from 12 molecules to 1 in chlorophyl tylakoids, forms difference 11 = $\Delta Cosm$, driving osmosis out $6O_{2,aqua} + 6H_2O$, for synthesised products.

Carbonic Anhydrase on surface of stomata complex outer sell membrane drive the reaction of dissolute $CO_{2,aqua}$ with two water molecules: $Q_{aqua} + CO_{2,aqua} + 2H_2O \xleftarrow{CA} H_3O^+ + HCO_3^-$ and creates concentration gradient for ions $6HCO_3^- + 6H_3O^+$ increasing influx in tylakoid from environment. According Le Chatelier principle shift photosynthesis equilibrium to products $C_6H_{12}O_6 + 6O_{2,aqua} + 6H_2O$. Photosynthesis drive $H_2O, O_{2,aqua}$ osmosis process out of cell endoergic and endothermic cooling surrounding as sun radiation supplied heat consuming (in summer time).



Irreversible enzyme CA reactivity for products reaching in living organism are Ilya Prigogine declared attractor for organism complex reaction five types, which inactive compounds convert to following favored irreversible process, that works as Brownian molecular engine so drive organism to evolution, homeostasis, survival.

These **channels**, allow the passive but selective movement of **Water** and **O₂**, **NO**, **CO** across **cell wall** and **subcellular membranes** like as mitochondria, endoplasmic reticulum, peroxisomes, Golgi, lysosomes.

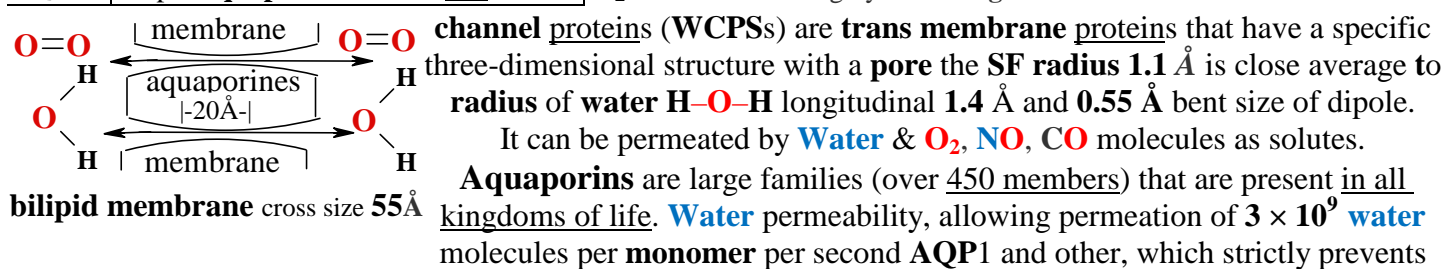
Aquaporins have been classified into **two** sub-families:

I) strict **Aquaporins** that only allow the passage of **Water**, **O₂**, **NO**, **CO** and II) the less selective **aquaglyceroporins** that **transport Water** and other neutral solutes, such as **Glycerol**, **CO₂**_{aqua} or **urea**.

Recently, the identification and characterization of a number of archaeal and bacterial **Aquaporins** suggested the existence of a **third** sub-family; one that is neither a strict **Aquaporin** nor an **aquaglyceroporin**. The function and phylogeny of this **third** family is still a matter of debate.

Water channels H₂O common O₂, NO, CO: an overview

| | |
|-------|--|
| AQP0 | + Cl⁻ , NO₃⁻ eye-lens cells; thin junctions between fiber cells AQP0 with a measured Water permeability 15-fold lower than that of AQP1 at pH 6.5 ; AQP0 is reduced a further three fold at pH 7.5 AQP0 induce a gating effect <u>close</u> conformations of <u>extracellular loop A Met176.His40</u> AQP0 becomes more constrained near the conserved Ar/R constriction site |
| AQP1- | + Cl⁻ , NO₃⁻ , Aquaglyceroporins: red blood cell (RBC), apical & basolateral membranes of epithelial brain cell, rodent brain cell AQP1-null humans kidney proximal-tubule water reabsorption gastrointestinal tract Water absorption in the teleost intestine the ovary and in the oocyte; salivary gland; |
| AQP2 | urinary bladder granular kidney cells & subcellular vasopressin regulated urine concentration (25% of the blood filtrate) trans located from the cytoplasmic pool to the apical plasma membrane of the granular cells of the pelvic patch and urinary bladder |
| AQP3 | + Aqua glyceroporins, urea: gastrointestinal tract Water absorption; rodent brain cell astrocyte end-feet Water enters in the principal cell through AQP2 and exits through located in the basolateral membranes trachea basal AQP3 & ciliated columnar AQP4 cells |
| AQP4 | Rodent-brain; basolateral membrane of ciliated columnar cells alveolar epithelium; salivary gland |
| AQP5 | stomach, duodenum, pancreas, airways, lungs, salivary gland, sweat glands, eyes, lacrimal glands, and the inner ear tears & pulmonary sub mucosal glands secretions apical membrane & rodent brain cells |
| AQP6 | + Cl⁻ , NO₃⁻ multi permeable channel; lens cells; may play a role in the body acid-base homeostasis in the intracellular vesicles of acid-secreting intercalated cells of the RCD colocalized with the H⁺-ATPase be Hg²⁺ -inhibit able Water channel function is activated by Hg²⁺ and low pH |
| AQP7 | + Aquaglyceroporins, urea; kidney proximal tubule epithelium cell glycerol reabsorption; together with AQP1 in the brush border in the concentration of urine taking place in the proximal nephron 75% of the blood filtrate which is 150–180 L per day |
| AQP8 | NH₄⁺ ; lens & kidney intracellular proximal tubule & small intestine absorptive: epithelium cell in the concentration of urine taking place in the proximal nephron also in mitochondria 75% of the blood filtrate which is 150–180 L per day & rodent brain cell |
| AQP9 | + Aquaglyceroporins, urea purines, pyrimidines & monocarboxylates, arsenite; apical membrane of brain & small intestine absorptive epithelial & rodent brain & glial cells |
| AQP10 | + Aquaglyceroporins, urea; small intestine absorptive epithelial cells |
| AQP11 | “super aquaporins” or sub cellular; kidney cytoplasm of the proximal tubule & rodent brain cells |
| AQP12 | “super aquaporins” or sub cellular |



Serine, Tyrosine, Threonine

Phosphorylation to trigger the **membrane trafficking** of AQP1, AQP2, AQP5, and AQP8, and the gating of AQP4.

Cation conductance has been induced in AQP1 by activation of cyclic GMP-dependent pathways and

<http://aris.gusc.lv/ChemFiles/Aquaporins/WCPsAQPsIUBMBLife09/AQP0-11.pdf> was blocked by **Hg²⁺**