

Gradual reaction first pathway sequences start with water 12H₂O flux from root to stomata cell membrane surface through aquaporin channels. Reaction in stomata forms pure CO₂ production thirty times less AIR CO₂↑gas in endoergic ΔG_r= 8,474 kJ/mol and ΔH_r= -20,1986 kJ/mol exothermic equilibrium moisture carbon dioxide: I) H₂O+CO₂↑gas+ΔG_{aqua}⇌Q+H₂O+CO₂↑gas; K_{eq}=[CO₂↑gas]/[CO₂↑gas]=10^{-1.485}=0,0327; 1/0,0327=30,55:
 ΔG_{aqua}=ΔG°_{aqua}+R•T•ln(K_{eq})=8,47+8,3144*298,15*ln(10^{-1.485})=0 at equilibrium zero value.

Carbonic Anhydrase CA equilibrium H₂O^{CA}/CO₂/H₃O⁺+HCO₃⁻ assimilation reaction with water produce proton and bicarbonate concentration gradient driving exoergic through membrane crossing channels HCO₃⁻ and H⁺:
 2H₂O + CO₂↑gas ⇌ CO₂↑gas + 2H₂O + Q ←^{CA}→ H₃O⁺ + HCO₃⁻ ←^{MembraneChannels}→ H₃O⁺ + HCO₃⁻ ;
 exothermic ΔH= -20,1986 kJ/mol; endothermic ΔH= 9,7576 kJ/mol; athermic; ΔH=0 kJ/mol; total exothermic ΔH= -10,441 kJ/mol;
 endoergic ΔG=+8,474 kJ/mol; ; endoergic ΔG=+58,36 kJ/mol; exoergic ΔG_r= -10,618-4,055=14,7 kJ/mol;
 ΔG_{HCO3}=RTln([HCO₃⁻]_{right}/[HCO₃⁻]_{left}) = 8,3144*298,15*ln(0,003/0,0154)=-4,0549 kJ/mol.....
 ΔG_H=RTln([H₃O⁺]_{right}/[H₃O⁺]_{left})= 8,3144*298,15*ln(10^{-7,36}/10^{-5,5})=-10,61682 kJ/mol.....

First pathway finish with ENZYME complex Photo Synthetic Reaction Center (PRC) of green plants.

Photosynthesis in thylakoid cell reach exothermic ΔH= -10,441 kJ/mol six plus six 6H₃O⁺+6HCO₃⁻ components.

Second pathway II) start with 1) (PRC) Photo Synthetic Reaction Center ENZYME complex of green plants products. Photosynthetic Reaction Center (PRC) using the red and blue light photons absorbed energy accumulate in glucose C₆H₁₂O₆ and oxygen 6O₂ energy endothermic ΔH= +2805,27 kJ/mol, ΔG= 3049.55 kJ/mol endoergic. Sun radiation, is consumed also the heat energy to shift evaporation endothermic processes of oxygen 6O₂ and water 6H₂O out of green plant stomata open hole. So substrates oxygen 6O₂ and water 6H₂O

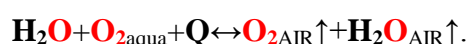
1) C_{osm} = 1 C₆H₁₂O₆ increase osmotic pressure outside cell 12 times as osmolar:
 C_{osm} = 6+6=12 concentration in cell from init C_{osm}=6+6=12 decreases to one glucose
 (PRC) $\xrightarrow[\text{BioEnergetic}]{\text{light red blue photo synthesis } E=h\nu \text{ PRC}}$ C₆H₁₂O₆ molecule C_{osm}=1 and flow intensity through aquaporins
 increases out of green plant cells. Osmotic pressure is proportional
 to osmolar concentration C_{osm} = 12 M twelve
 6HCO₃⁻+6H₃O⁺+Q C₆H₁₂O₆+6O₂↑gas+6H₂O times higher pressure π= C_{osm}•R•T (kPa).

6O₂ and water 6H₂O pushed out of cells with twelve times higher pressure as concentration gradient through:

2) aquaporins H₂O+O₂↑gas ←^{Aquaporins}→ O₂↑gas + H₂O ΔG= -6,41 kJ/mol exoergic and athermic ΔH=0 kJ/mol:

ΔG_{channel}=RTln([O₂↑gas]_{right}/[O₂↑gas]_{left})=8,3144*298,15*ln(1/12)= -6.41 kJ/mol.

3) Finally exoergic ΔG= -7,76 kJ/mol and endothermic ΔH=55,3 kJ/mol to form in AIR O₂AIR as evaporation

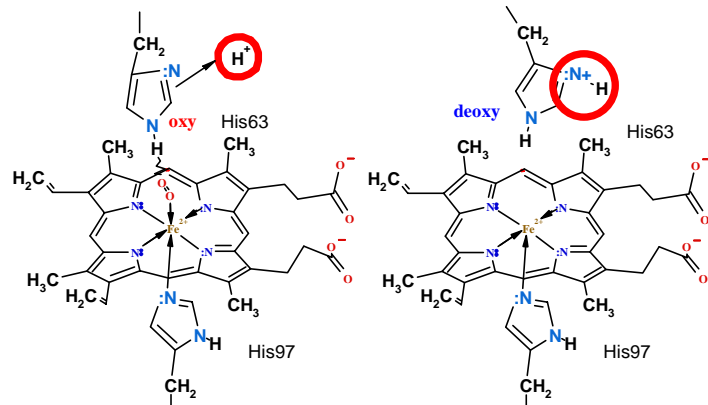


Plants need supply energy as heat to support evolved and evaporated substrates products oxygen O₂AIR↑ as well water H₂O_{AIR}↑ vapor through stomata open hole. According Le Chatelier principle heat as energy supply shift endothermic equilibrium evaporation of O₂AIR↑ and H₂O↑ outside → through green plant stomata open hole. Opening the stomata holes depends on environment temperature and humidity. Evaporation velocity increases at higher temperature as heat supply if at enough water supply usually from roots. Closing of stomata holes decreases the evaporation velocity in plants, so to prevent drying the organism as loose the water in evaporation.

Gradual reaction sequences start with osmosis through membrane aquaporin channels inside stomata holes.

I) Oxygens O₂ from AIR 20.95% **O₂**↑gas assimilation reaction through stomata open hole membrane aquaporins: $\Delta H = -55,7 \text{ kJ/mol}$ exothermic $\text{O}_{2\text{AIR}} + \text{H}_2\text{O} \xleftarrow{\text{Aquaporin}} \text{H}_2\text{O} + \text{O}_{2\text{aqua}} + \text{Q}$ $\Delta G = -31,8 \text{ kJ/mol}$ exoergic. $C_{\text{osm}} = 0,305$ and alveolar $C = 0,2 \text{ M}$ $\Delta G_{\text{aquaporin}} = RT \ln([\text{H}_2\text{O}]_{\text{right}} / [\text{H}_2\text{O}]_{\text{left}}) = -8,3144 * 298,15 * \ln(0,305/0,2) = -1,05 \text{ kJ/mol}$ exoergic.

Plant **deoxy** Leghemoglobin **LHb_T** from water medium adsorb inspired through stomata open holes fresh AIR **O₂** releases proton **H⁺**: $\text{oxyLHb}_R(\text{His63O}_2) + \text{H}^+ \leftarrow [\text{O}_2] = 6 \cdot 10^{-5} \text{ M} \rightarrow \text{deoxy}(\text{H}^+\text{His63})\text{LHb}_T + \text{O}_{2\text{aqua}}$;



Equilibrium $\text{oxyLHb}_R(\text{His63O}_2) \leftrightarrow \text{deoxy}(\text{H}^+\text{His63})\text{LHb}_T$ stabilize water concentration $[\text{O}_{2\text{aqua}}] = 6 \cdot 10^{-5} \text{ M}$ and store multiple reserves on $\text{oxyLHb}_R(\text{His63O}_2)$. Each proton sticks to water $\text{H}^+ + \text{H}_2\text{O} \rightarrow \text{H}_3\text{O}^+$ forms hydroxonium ion and from fresh AIR in stomata open hole acidify water medium with H_3O^+ , promoting metabolic **CO₂** breathe out in AIR.

Second **II)** pathway begins with carbonic anhydrase **AC** equilibrium shifted to right after Krebs cycle oxidative phosphorylation in Mitochondria increase product **CO₂** and capturing protons **H⁺** $\text{deoxy}(\text{H}^+\text{His63})\text{LHb}_T$ stabilizes $\text{pH} = 7,36$ concentration H_3O^+ as instead **CA** made new.

II) Q_{aqua} + CO₂↑gas + 2H₂O $\xleftarrow{\text{CA}} \text{H}_3\text{O}^+ + \text{HCO}_3^- \xleftarrow{\text{Membrane}} \text{H}_3\text{O}^+ + \text{HCO}_3^- \leftrightarrow \text{H}_2\text{O} + \text{H}_2\text{CO}_3 + \text{Q}_{\text{gas}} \leftrightarrow \text{H}_2\text{O} + \text{CO}_2 \uparrow_{\text{gas}} + \text{H}_2\text{O}$ endothermic $\Delta H_r = +9,59 \text{ kJ/mol}$; athermic $\Delta H_r = 0 \text{ J/mol}$ exothermic $\Delta H_r = -9,76 \text{ kJ/mol}$; endothermic $\Delta H_r = +20,29 \text{ kJ/mol}$;

gradient $1/1,1$ $\Delta G_{\text{channel}} = -RT \ln([\text{HCO}_3^-]_{\text{right}} / [\text{HCO}_3^-]_{\text{left}})^6 = -RT \ln((1/1,1)^6) = -0,245 \text{ kJ/mol}$; $R = 8,3144$, $T = 310 \text{ K}$. endoergic $\Delta G_r = +58,19 \text{ kJ/mol}$; exoergic $\Delta G_r = -0,245 \text{ J/mol}$; exoergic $\Delta G_r = -58,205 \text{ kJ/mol}$; exoergic $\Delta G_r = -8,538912 \text{ kJ/mol}$.

Total sum in sequence **endothermic** $\Delta H_r = +9,5876 + 0 - 9,76 + 20,291 = +20,1 \text{ kJ/mol}$ and free energy change

exoergic $\Delta G_r = +58,19 - 0,245 - 58,205 - 8,538912 = -8,8 \text{ kJ/mol}$ spontaneous total sum in sequence.

The ENZYME carbonic anhydrase (**CA**) from **CO₂** make acid/base equilibrium:

$\text{CO}_{2\text{aqua}} + 2\text{H}_2\text{O} \xleftarrow{\text{CA}} \text{H}_3\text{O}^+ + \text{HCO}_3^-$ value $K = [\text{H}_3\text{O}^+] \cdot [\text{HCO}_3^-] / [\text{CO}_{2\text{aqua}}] / [\text{H}_2\text{O}]^2 = 2,901 \cdot 10^{-11}$ is constant but water constant concentration $[\text{H}_2\text{O}] = 55,3 \text{ M}$ is included in $K_{\text{eq}} = K \cdot [\text{H}_2\text{O}]^2 = [\text{H}_3\text{O}^+] \cdot [\text{HCO}_3^-] / [\text{CO}_{2\text{aqua}}] = 10^{-7,0512}$ equilibrium constant value and its exponent is $\text{p}K_{\text{eq}} = -\log(K_{\text{eq}}) = 7,0512$. Weak acid **CO₂** and its salt is ubiquitous buffer system in living organism. It's reveals the key role of Mitochondrial oxidative phosphorylation final product **CO₂** for **HOMEOSTASIS 7,36 = pH = pK + log([HCO₃⁻]/[CO₂]) = 7,0512 + log(2,0263/1)** stable $\text{pH} = 7,36 \pm 0,01$ value according Henderson Haselbalh equation.

CA equilibrium is endothermic $\Delta H_r = +9,59 \text{ kJ/mol}$ and endoergic $\Delta G_r = +58,19 \text{ kJ/mol}$ so Mitochondrial oxidative phosphorylation exothermic and exoergic reactions in Krebs cycle produced energy is used for shifting **II)** pathway equilibriums to right for reaching **stomata** spongy mesophyll cell surface to evaporate **CO₂**↑gas.

Through bicarbonate **HCO₃⁻** and proton **H⁺** channels crossing membranes transport ENZYMES in athermic $\Delta H = 0 \text{ kJ/mol}$ and exoergic $\Delta G = -0,245 \text{ kJ/mol}$ processes form carbonic acid **H₂CO₃** molecules as ENZYME Carbonic Anhydrase **CA** is absent: $\text{H}_3\text{O}^+ + \text{HCO}_3^- \xleftarrow{\text{Membrane}} \text{H}_2\text{O} + \text{H}_2\text{CO}_3 + \text{Q}$.

In **stomata** spongy mesophyll cell surface (out side organism) with absence **CA** decomposes carbonic acid **H₂CO₃** to gas **CO₂**↑gas in endothermic $\Delta H_r = +20,29 \text{ kJ/mol}$ but in exoergic $\Delta G_r = -8,54 \text{ kJ/mol}$ reaction:

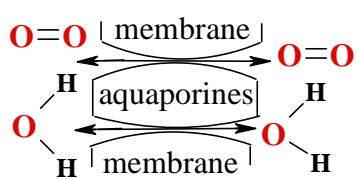
$\text{H}_2\text{CO}_3 + \text{Q} \rightarrow \text{H}_2\text{O} + \text{CO}_2 \uparrow_{\text{gas}}$. Heat supply shifts equilibrium right side → according Le Chatelier's principle. Plants have the **stomata** spongy mesophyll cell surface located inside stomata and equipped with heat accumulating cells in leaves faced to sun shine as heating promote breath out **CO₂**↑gas, so keeping constant $\text{pH} = 7,36 \pm 0,01$ in organism. If climate temperature warm so more intensive exchange of gases in nature.

Each adsorbed oxygen **O₂** on Leghemoglobin releases proton **H⁺** which increases acidity on **stomata** spongy mesophyll cell surface. The **stomata** spongy mesophyll cell surface has the specific building: super thin **0,6 nm water** layer on surface $S = 950 \text{ nm} \times 950 \text{ nm} = 0,9 \mu\text{m}^2$ as square within $0,5415 \cdot 10^{-3} \mu\text{m}^3 = 0,5415 \cdot 10^{-18} \text{ L}$ small volume. Created acidity in thin **water** layer volume increases up to $\text{pH} = 5,5$ if one proton **H⁺** crosses the membrane channel reaching the surface and that cause fast decomposition of carbonic acid **H₂CO₃** to evolving **CO₂**↑gas maintaining high concentration which support the photosynthesis gradual sequence shifting to production of glucose **C₆H₁₂O₆** and oxygen **O₂**. Additionally epithelial cell surface $\text{pH} = 5,5$ has anti-bacterial as well as anti-septic properties to prevent infection in organism. Carbonized mineral water has anti-bacterial as well as anti-septic properties.

O₂ exchange to **CO₂** Physiology, Carbonic Anhydrase pH=7.36 stability and **deoxy** Leg**HEMOGLOBIN**, **oxy** Leg**HEMOGLOBIN** controlled oxygen concentration [**O₂_{aqua}**]=6·10⁻⁵ M stability

Before any discussion of the mechanism, we have to know the sequence of strengths of the three acids, involved in the three buffer systems. The strongest one of these three acids is **oxy**LegHemoglobin, next one is **carbonic anhydrase CA** made acid with value pK=7.0512 and the weakest one is LegHemoglobin:

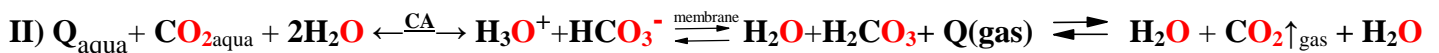
K_{LHbR(His63O₂)}+H⁺ > **K_{H₂O}^{/CA/}CO₂_{aqua}** > **K_{LHbT(H+His63)}**. The sequence of acid strength will be necessary for whole process exchange of AIR **O₂** to BREATHED OUT AIR **CO₂** is performed in two **I** and **II** pathways of gradual reactions: First **I) pathway** first gradual process *in stomata* spongy mesophyll cell surface through cell wall membrane aquaporins water **H₂O**, oxygen **O₂** with rate 10⁹ sec⁻¹ reach organism water significant changes



accumulated oxygen reserve in hemoglobin. Oxygen influx first gradual process concentration gradient via concentration stabilization in water [**O₂_{aqua}**]=6·10⁻⁵ M begins **I) pathway** 1) **O₂_{AIR} + H₂O** $\xrightleftharpoons{\text{aquaporin}}$ **H₂O + O₂_{aqua}** with parallel concentration

[**O₂**]=6·10⁻⁵ M sensitive equilibrium drive LegHemoglobin **O₂** adsorbtion ↔ desorbtion to left side:

O₂_{aqua} + LHb_T(H⁺His63) ↔ LHb_R(His63O₂)+H⁺ and releases proton **H⁺**. LegHemoglobin acid **HLHb** adsorbs oxygen **O₂_{aqua} + LHb_T(H⁺His63) ↔ LHb_R(His63O₂)+H⁺** promoting stronger acid formation with release proton from (**H⁺His63**)**LHb_T** per each adsorbed oxygen molecule **O₂**. As **O₂** adsorbtion on **deoxy Hb_T** release **H⁺** as **H₃O⁺** per one **HCO₃⁻** *in plant cells* performing four gradual sequences product **CO₂_{aqua}** on **II) pathway** reactions:

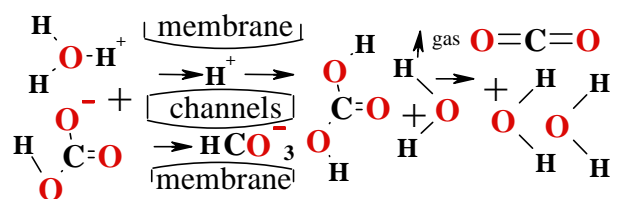
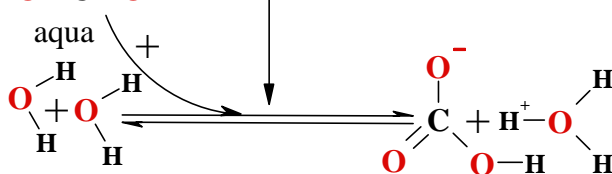


Metabolic **CO₂_{aqua}** exchanged per **O₂** consuming are by ENZYME Carbonic Anhydrase (**CA**) converted to **HCO₃⁻** and **H₃O⁺** at stabilized **blood** pH=7.36±0,01 has *alkaline reserve* 2.036/1=[**HCO₃⁻**]/[**CO₂**].

1) first gradual reaction ENZYME Carbonic Anhydrase (**CA**) converts metabolic **CO₂_{aqua}** to bicarbonate anion driven by Krebs cycle oxidation product **CO₂_{aqua}** concentration in reaction of endothermic as well as endoergic:



O=C=O carbonic anhydrase



2) Second gradual athermic ΔH= 0 kJ/mol **H₃O⁺ + HCO₃⁻** $\xleftarrow{\text{Membrane}}$ **H₃O⁺ + HCO₃⁻** + ΔG; ΔG= -24,44 kJ/mol exoergic concentration gradients ΔG_{HCO₃⁻}=RTln([**HCO₃⁻**]_{right}/[**HCO₃⁻**]_{left})=RTln((0,0154/0,0338919))= -1,9554 kJ/mol; and ΔG_H=RTln([**H₃O⁺**]_{right}/[**H₃O⁺**]_{left})=RTln((10^{-5,5}/0,02754))= -22,5 kJ/mol where R=8,3144, T=310 K. Special transport ENZYMES catalyzes bicarbonate **HCO₃⁻** and proton **H⁺** move through channels crossing membranes.

3) Third gradual exothermic and exoergic reaction **H⁺ + HCO₃⁻ → H₂CO₃ + Q**; exothermic ΔH_r=-9,76 kJ/mol; exoergic ΔG_r=-58,2 kJ/mol forms Carbonic acid **H₂CO₃**.

4) Fourth gradual reaction on *stomata* spongy mesophyll cell surface (out side organism) decomposes carbonic acid **H₂CO₃** to gas **CO₂↑_{gas}** in endothermic: **H₂CO₃ + Q(gas) → H₂O + CO₂↑_{gas}** reaction ΔH_r= +20,291 kJ/mol; as well as exoergic reaction ΔG_r= -8,539 kJ/mol. Total endothermic ΔH= 20,2886 kJ/mol; exoergic ΔG= -32,819 kJ/mol;

Each adsorbed oxygen **O₂_{aqua}** on LegHemoglobin releases proton **H⁺** which increases acidity on *stomata* spongy mesophyll cell surface. Surface has the specific building: super thin 0.6 nm **water** layer S=0.9 μm² on surface as square within small volume 0.5415·10⁻³ μm³ =0.5415·10⁻¹⁸ L created acidity increases up to pH=5.5 if one proton crosses the membrane channel reaching the surface and that cause fast decomposition of carbonic acid **H₂CO₃** to evolving **CO₂↑** gas evaporate out.

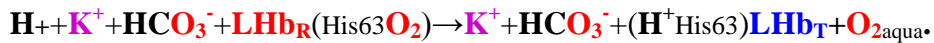
1) $H^{++} Lhb_R(His63O_2) \rightarrow (H^{+His63})Lhb_T + O_{2\text{aqua}}$ loses oxygen through aquaporins crosses membrane into mitochondria oxygen concentration little decreases below $[O_{2\text{aqua}}]=6 \cdot 10^{-5}$ M. Each desorbed oxygen adsorbs one proton to distal histidine H^{+His63} in LegHemoglobin $(H^{+His63})Lhb_T$ decreases produced metabolic product $CO_{2\text{aqua}}$ acidity effect keeping pH=7.36 constant.

2) $CO_{2\text{aqua}}$, which is a product of metabolism comes from **mitochondria** reacts with water, forming **carbonic anhydrase** made $Q + CO_{2\text{aqua}} + 2H_2O \leftarrow CA \rightarrow H_3O^{++} + HCO_3^-$ **endothermic** equilibrium.

ENZYME Carbonic Anhydrase (CA) equilibrium shifts reaction towards bicarbonate anion to prevent of carbonic dioxide accumulation, according Le Chatelier's due to high water $[H_2O]$ concentration 55.3 M and low hydroxonium ion concentration $[H_3O^{++}]=10^{-7.36}$ M as well as constant value is $pK=7.0512$. Carbonic acid constant $pK=6.11$ shifts to one fold more acidic hazard for bubbling $Q + H_2CO_3 \rightarrow H_2O + CO_2 \uparrow_{\text{gas}}$.

ENZYME Carbonic Anhydrase (CA) absent as out side plant organism enclosed with cell membranes.

3) As enzyme Carbonic Anhydrase CA made equilibrium acid is a stronger, than $(H^{+His63})Lhb_T$, bicarbonate and hydrogen ion H^+ reacts with salt $Lhb_R(His63O_2)$:



In this way lower $[O_{2\text{aqua}}]=6 \cdot 10^{-5}$ M concentration repeat cycle of the **first I) pathway first gradual** process in **stomata** through aquaporins from AIR 20.95% $O_2 \uparrow_{\text{gas}}$.

Let us consider now, why this sequence of acid strengths (given in the beginning) is necessary.

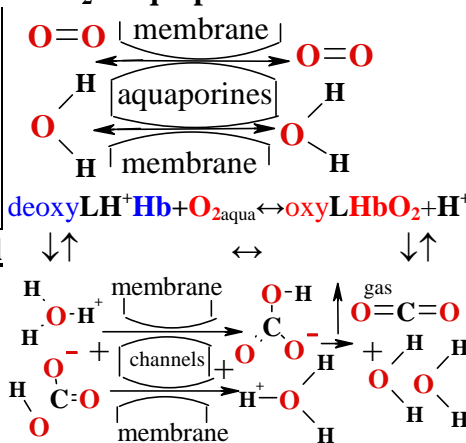
First, if it happened, that $H_2O/CA/CO_{2\text{aqua}}$ was a **stronger** acid, than $(H^{+His63})Lhb_T$. $CO_2 \uparrow_{\text{gas}}$ would stay in its water soluble transport forms $CO_{2\text{aqua}}$, $K^+ + HCO_3^-$ and could not be liberated up to **stomata** bicarbonate HCO_3^- and proton H^+ channels out of cells to **stomata** spongy mesophyll cell surface (out side organism).

Second, if $(H^{+His63})Lhb_T$ was a **weaker** acid than $H_2O/CA/CO_{2\text{aqua}}$, $(H^{+His63})Lhb_T$ not react as H^+ with $K^+ + HCO_3^-$ and is transported to stomata in the form of bicarbonate. Opposite is dangerous, because accumulation and formation of $CO_2 \uparrow_{\text{gas}}$ bubbles could occur damage of plant cells.

For moisture membrane proton channels are protons H^+ permeable, unless H^+ impermeable for dry **proton channels** $\rightleftharpoons \times \leftarrow$ H^+ . Therefore membrane is equipped by aquaporins, which are **water** and solute oxygen

$O=O$ permeable in both directions ($O=O + H_2O$ aquaporin channel $\rightleftharpoons H_2O + O=O$):

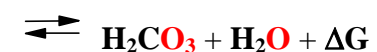
For protons crossing the membrane through proton channels, necessary **water** molecules locate both side of the membrane and aquaporins are supplier of **water** H_2O molecules to moisture **spongy** cell surface stomata.



Free energy change $\Delta G = -60$ kJ/mol for Reaction of H_2CO_3 formation is **exoergic** $\Delta G < 0$ negative therefore promotes neutralization reaction $H_3O^{++} + HCO_3^- \rightleftharpoons H_2CO_3 + H_2O + \Delta G$

Inside the cell-cytosol
 CA with **water** producing heat +Q
 $CO_2 + 2H_2O + Q \leftarrow CA \rightarrow H_3O^{++} + HCO_3^-$
 aqua **endothermic**

spongy surface stomata consuming +Q heat and evolving **water + H₂O** supporting surface moisture
 $H_2CO_3 + Q \rightleftharpoons CO_2 \uparrow_{\text{gas}} + H_2O$
endothermic



Plant cytosol Henderson Haselbalh pH equation Homeostasis

As it was mentioned before, three main buffer systems act in plants: **deoxy LegHemoglobin (H⁺His63)LHb_T**
oxy LegHemoglobin LHb_R(His63O₂)

Carbonic Anhydrase CO₂ aqua – bicarbonate H₂O^{/CA/}CO₂ aqua/ K⁺+HCO₃⁻ buffer systems

When these buffer systems struggle with acidic products CO₂ aqua of metabolism, more and more of the acid forms of the buffer systems are produced. For this reason, the acid forms have to be transported out of organism.

It is easy to imagine, that LegHemoglobin cannot be evolved out of organism, therefore there is only one buffer system, suitable for regulation of acid form's presence by breathing out CO₂ ↑ gas, that stabilize pH of plants pH=7.36 by metabolic waste CO₂ ↑ gas decreasing problems for organism through pH of plant should be maintained constant 7.36. Reaction is negligible slow CO₂ aqua + 2H₂O slow → H₃O⁺ + HCO₃⁻ and shifts to fastest reversible carbonic acid formation. CO₂ ↑ gas + 2H₂O ⇌ H₂CO₃ + H₂O + Q ⇌ H₃O⁺ + HCO₃⁻ + Q **exothermic** reaction which provides accumulation of CO₂ ↑ gas because carbonic acid pK=6.11 value is one fold more acidic as Carbonic Anhydrase (CA) pK=7.0512 value.

Carbonic anhydrase make conversion of CO₂ aqua to bicarbonate anion HCO₃⁻ in to **water** medium fast and establish acid-base Q + CO₂ aqua + 2H₂O ← CA → H₃O⁺ + HCO₃⁻ **endothermic** equilibrium as producing right side → reaction products H₃O⁺ + HCO₃⁻ - Q **endothermic** demanding to heat. So sun radiation Heating +Q shifts equilibrium right side → and as soon as H⁺ concentration decrease pH > 7.36 as each CO₂ aqua form one H₃O⁺, Carbonic anhydrase equilibrium is shifted to left ← too and CO₂ transported out together with H⁺ as HCO₃⁻ by evaporation in *stomata* spongy mesophyll cell and acid concentration [H⁺] decreases to **homeostasis** pH=7.36 level. If concentration H⁺ decreases pH > 7.36, Carbonic anhydrase equilibrium is shifted to the right and the extra amount of HCO₃⁻ through *stomata* spongy mesophyll cell surface (out side organism) passes out to AIR and pH returns to **homeostasis** pH=7.36 level according Le Chatelier's theorem.

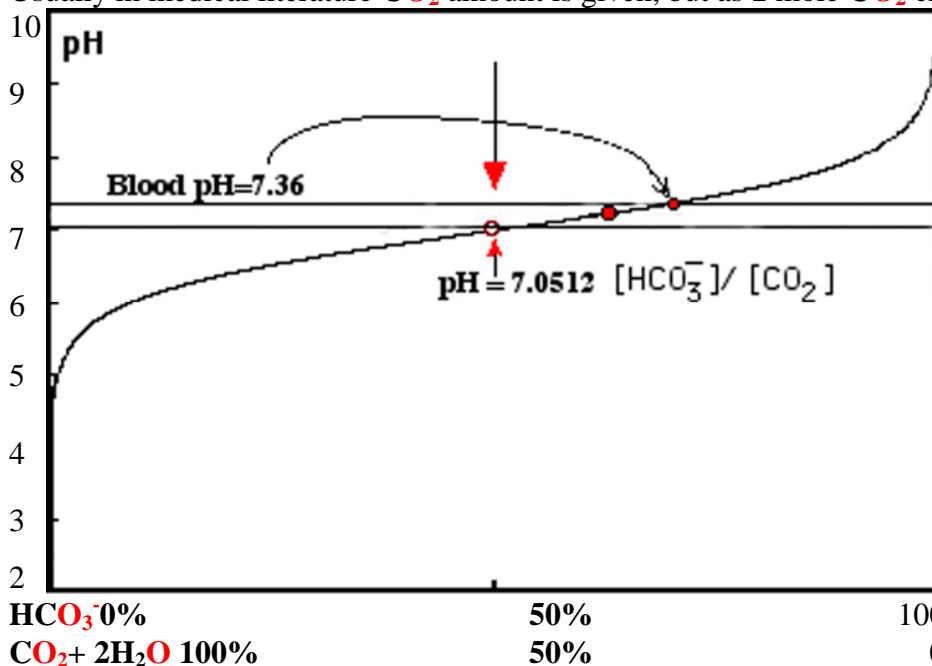
The Brensted acid is **water** solution of gaseous CO₂. The dissolved into **water** H₂O carbonic dioxide CO₂ occurring in cell converted with carbonic anhydrase CA to H⁺ + HCO₃⁻. The **water** H₂O and carbonic dioxide CO₂, finally, is in direct acid-base equilibrium with its ions H⁺ and HCO₃⁻.

Carbonic anhydrase equilibrium constant pK=7.0512 decreases concentration acid form CO₂ into **water** H₂O (avoid carbonic acid H₂CO₃ formation which is one fold more acidic pK=6.11) therefore hydrogen carbonate HCO₃⁻ and hydrogen ions H⁺ are involved into pH formation according buffer solution

Henderson-Haselbalh homeostasis: $7.36 = \text{pH} = \text{pK} + \log \left(\frac{[\text{KHCO}_3]}{[\text{CO}_2]} \right) = 7.0512 + \log \left(\frac{[\text{KHCO}_3]}{[\text{CO}_2]} \right);$

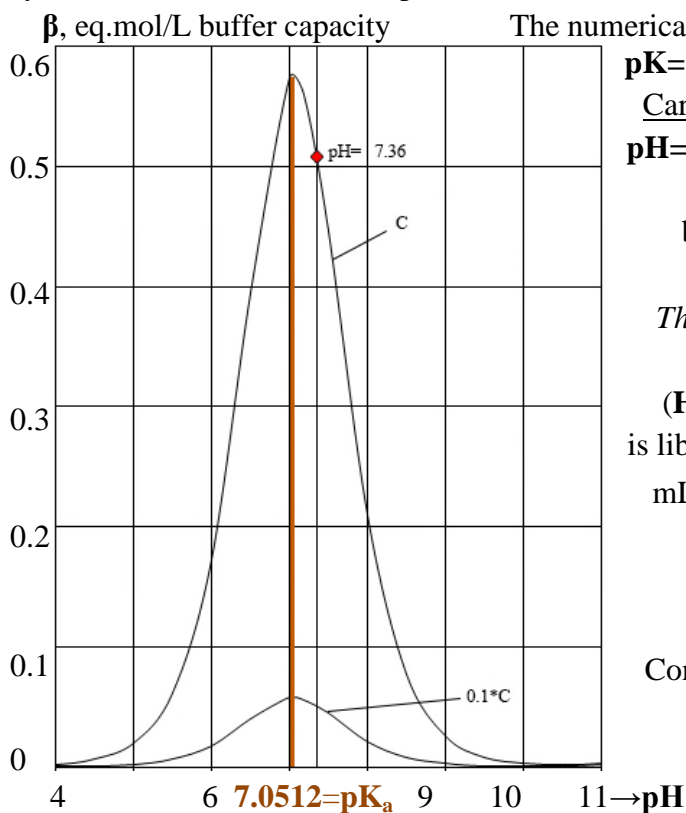
$$\frac{[\text{KHCO}_3]}{[\text{CO}_2]} = 10^{(\text{pH}-\text{pK})} = 10^{(7.36-7.0512)} = 10^{0.3088} = \frac{2.0263}{1} \text{ the ratio } [\text{KHCO}_3]/[\text{CO}_2] \text{ being approximately } 2/1.$$

Usually in medical literature CO₂ amount is given, but as 1 mole CO₂ creates 1 mole H₂O^{/CA/}CO₂, it is the same.



Buffer region middle point is the over inflection point in graph ○:
pH = pK_a = 7.0512 as
 $[\text{KHCO}_3]/[\text{CO}_2] = 1$
 is one as well buffer component concentrations are equal
 $[\text{KHCO}_3] = [\text{CO}_2]$ as well as bicarbonate salt of potassium [KHCO₃] concentration is equal to Brensted weak acid dissolved CO₂ concentration [CO₂].
 Alkaline reserve at 7.36 = pH is **normal** if $\frac{[\text{KHCO}_3]}{[\text{CO}_2]} = \frac{2.0263}{1}$.

As soon as H^+ concentration grows for some reason, Carbonic anhydrase equilibrium is shifted to left and CO_2 transported out as H^+ with HCO_3^- by respiration through *stomata* spongy mesophyll cell surface and acid concentration $[H^+]$ decreases. If concentration H^+ decreases, Carbonic anhydrase equilibrium is shifted to the right and the extra amount of HCO_3^- through *stomata* cell surface passes transported out. Bicarbonate channels in *stomata* spongy mesophyll cell surface channel transport are open at high values of $pH > 7.36$ from inside of cytosole circulation, but are sopen to outside for evolving CO_2 at low values $pH < 7.36$.



The numerical value **7.0512** is more basic about value of carbonic acid $pK = 6.11$ which is almost one unit less. This value $pK = 7.0512$ is Carbonic anhydrase made equilibrium constant very friendly to $pH = 7.36$. As metabolism products CO_2 are acidic, the organism has to have a reserve of alkalinity. For this reason the ratio between $KHCO_3$ and CO_2 concentrations is 2/1 and the pH value of physiological conditions is 7.36. *The alkaline reserve* $2.036/1 = [HCO_3^-]/[CO_2]$ at $pH = 7.36$ can be controlled by adding H_2SO_4 to a sample of plant juice (H_2SO_4 reacts with HCO_3^- and the CO_2 , included in salt, and is liberated). If 56.23 mL of gaseous CO_2 are liberated from 100 mL of juice, the *alkaline reserve* in **homeostasis** is normal and total *alkaline reserve* amount concentration $0.023M = [HCO_3^-] + [CO_2]$ is in **homeostasis normal** as sum of $[HCO_3^-] = 0.0154 M$, $[CO_2] = 0.0076M$. Controlled instructions *the alkaline reserve* of the organism by adding H_2SO_4 to 100 mL sample. As H_2SO_4 reacts with $KHCO_3$ as CO_2 , included in salt is liberated.

If 50-60 mL of gaseous CO_2 is liberated

from 100 mL of sample in **homeostasis**, the *alkaline reserve* is normal.

Two types of diseases occur, if the acid-base balance is distorted in the organism alkalosis and acidosis.

1) *Respiratory alkalosis* occurs, if *stomata* open holes AIR has low less 0.0015% CO_2 . If CO_2 concentration decreases $pH > 7.36$ alkalose. 0.0015% (150 ppm) concentration is deadly low for plant survival.

For good planting is necessary to use mixtures of AIR with higher 0.3% (30000 ppm) CO_2 in greenhouse planting.

2) *Respiratory acidosis* occurs in the cases, when the concentration of CO_2 in the air is increased. If increased CO_2 content in the air lasts long, a metabolic acidosis can occur $pH < 7.36$ acidose. In the case of metabolic acidosis the ability of LegHemoglobin to bound oxygen is lowered as shifting equilibrium to left:



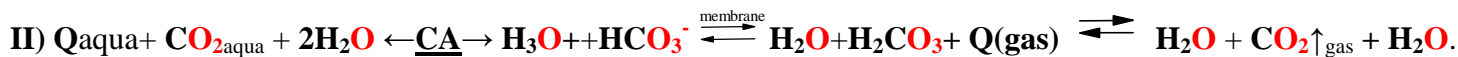
For this reason only the concentrations of carbonic dioxide CO_2 into **water** H_2O (to avoid carbonic acid H_2CO_3 formation) and hydrogen carbonate HCO_3^- + hydrogen ions H^+ are included into equation for pH :

There are two Pathway sequences of equilibria, which have composed of ENZYME governed gradual reactions:



Concentration gradient in water solution $[O_{2\text{aqua}}] = 9,768 \cdot 10^{-5} M$ to **venous** blood $[O_2] = 1,85 \cdot 10^{-5} M$ O2Solutions

exoergic $\Delta G_{os} = RT \ln([O_2]_{\text{right}}/[O_2]_{\text{left}}) = 8,3144 * 298,15 * \ln(1,85/9,768) = -4,13 \text{ kJ/mol}$. athermic $\Delta H_{os} = 0 \text{ kJ/mol}$.



Concentration gradient in water solution

If 56.23 mL of gaseous CO_2 are liberated from 100 mL of plant sample, the alkaline reserve is normal $2,0263/1 = [HCO_3^-]/[CO_2] = 2.0263$; $[HCO_3^-] = 0.0154 M$, $[HCO_3^-] + [CO_2] = 0.023 M$ and $[CO_2] = 0.00757 M$.

Pathway II) First gradual reaction governed by ENZYME Carbonic anhydrase CA made equilibrium and pH=7,36: Free energy change ΔG for reaction: $\text{CO}_{2\text{aq}} + 2\text{H}_2\text{O} + \Delta\text{G} + \text{Q} \xleftarrow{\text{CA}} \text{H}_3\text{O}^+ + \text{HCO}_3^-$

Enthalpy change ΔH for reaction endothermic as heat consumed: $\Delta\text{H} = \Delta\text{H}^\circ_{\text{H}_3\text{O}^+} - \Delta\text{H}^\circ_{\text{HCO}_3^-} - 2\Delta\text{H}^\circ_{\text{H}_2\text{O}} - \Delta\text{H}^\circ_{\text{CO}_2} = -285,81 - 689,93 - (2 \cdot -285,85 - 413,7976) = -975,74 + 985,4976 = +9,7576 \text{ kJ/mol}$.

Krebs cycle releases $[\text{CO}_{2\text{aq}}] = 0,02527 \text{ M}$ and total amount $[\text{HCO}_3^-] + [\text{CO}_2] = 0,023\text{M} + 0,02527 \text{ M} = 0,05054 \text{ M}$.

Entropy change ΔS_r < 0 negative as ENZYME Carbonic Anhydrase CA governed reaction:

$\Delta\text{S} = \Delta\text{S}^\circ_{\text{H}_3\text{O}^+} + \Delta\text{S}^\circ_{\text{HCO}_3^-} - 2\Delta\text{S}^\circ_{\text{H}_2\text{O}} - \Delta\text{S}^\circ_{\text{CO}_2} = -3,85 + 98,32 - (2 \cdot 69,956 + 117,57) = 94,47 - 257,483 = -163,013 \text{ J/mol/K}$.

$\Delta\text{G} = \Delta\text{H} - \text{T} \cdot \Delta\text{S} = 9,7576 + 298,15 \cdot 0,163013 = +58,36 \text{ kJ/mol}$ endoergic free energy accumulated in products driven by high concentration $[\text{H}_2\text{O}]^2 = (997,07/18,0153)^2 = 3063 \text{ M}^2$ and by low $[\text{H}_3\text{O}^+] = 10^{-7,36} \text{ M}$ concentration.

$\Delta\text{S}_{\text{dispersed}} = -\Delta\text{H}_r/\text{T} = -9,7576/298,15 = -32,727151 \text{ J/K/mol}$;

$\Delta\text{S}_{\text{total}} = \Delta\text{S} + \Delta\text{S}_{\text{dispersed}} = -163,0134 - 32,7271 = -195,7405 \text{ J/K/mol}$;

$\text{T} \cdot \Delta\text{S}_{\text{total}} = -0,1957405 \cdot 298,15 = -58,36 \text{ kJ/mol}$ lost energy for initial compounds $\text{CO}_{2\text{aq}} + 2\text{H}_2\text{O}$.

CA acid/base Brensted equilibrium constant pKa=7,0512 value thermodynamic calculation $\Delta\text{G}_r = \Delta\text{G}^\circ_{\text{H}_3\text{O}^+} + \Delta\text{G}^\circ_{\text{HCO}_3^-} - 2\Delta\text{G}^\circ_{\text{H}_2\text{O}} - \Delta\text{G}^\circ_{\text{CO}_2} = -213,275 - 586,94 - (2 \cdot -237,191 - 385,98) = +60,147 \text{ kJ/mol}$ endoergic.

Carbonic anhydrase make the equilibrium constant to $K_{\text{eq}} = 10^{-7,0512}$ or exponent $\text{p}K_{\text{eq}} = 7,0512$ constant very close to stabilized pH value 7.36. Water concentration $[\text{H}_2\text{O}] = 55,3 \text{ M}$ is constant included in K_{eq} . Start with equilibrium $\text{CO}_{2\text{aq}} + 2\text{H}_2\text{O} \xleftarrow{\text{CA}} \text{H}_3\text{O}^+ + \text{HCO}_3^-$ constant $K = [\text{H}_3\text{O}^+][\text{HCO}_3^-]/([\text{CO}_{2\text{aq}}] \cdot [\text{H}_2\text{O}]^2) = 10^{-10,224}$. included water concentration $[\text{H}_2\text{O}] = 997,07/18,0153 \text{ M}$, $K_{\text{eq}} = [\text{H}_2\text{O}]^2 \cdot K = [\text{H}_3\text{O}^+][\text{HCO}_3^-]/[\text{CO}_{2\text{aq}}] = 10^{-7,0512}$, $K = \text{EXP}(-\Delta\text{G}_r/\text{R}/\text{T}) \cdot [\text{H}_2\text{O}]^2 = (10^{-10,5372}) \cdot [\text{H}_2\text{O}]^2 = (10^{-10,5372}) \cdot (997,07/18,0153)^2 = 10^{-7,0512} = 10^{-\text{p}K_{\text{eq}}}$; give the exponent value of constant $\text{p}K_{\text{eq}} = 7,0512$.

Special transport ENZYMES catalyzes parallel electrochemical reactions for bicarbonate ion HCO_3^- and for proton H^+ channels penetrating membrane are athermic $\Delta\text{H} = 0 \text{ kJ/mol}$ and $\Delta\text{G} = -64,699 \text{ kJ/mol}$ exoergic.

$\text{H}_3\text{O}^+ + \text{HCO}_3^- \xleftarrow{\text{Membrane}} \text{H}_3\text{O}^+ + \text{HCO}_3^- + \Delta\text{G}$ concentration gradients as well proton gradient across membranes

$\Delta\text{G}_{\text{HCO}_3^-} = \text{RTln}([\text{HCO}_3^-]_{\text{right}}/[\text{HCO}_3^-]_{\text{left}}) = \text{RTln}(0,0154/0,0338919) = -1,9554 \text{ kJ/mol}$;

$E_{\text{HCO}_3^- \text{ Mitochon}} = -\text{P} \cdot \text{lg}([\text{HCO}_3^-]_{\text{blood}}/[\text{HCO}_3^-]_{\text{Mitochon}}) = -0,0591591 \cdot \text{lg}(0,0154/0,0338919) = 0,0202664 \text{ V}$;

$\Delta\text{G}_{\text{H}} = \text{RTln}([\text{H}_3\text{O}^+]_{\text{right}}/[\text{H}_3\text{O}^+]_{\text{left}}) = \text{RTln}((10^{-7,36})/0,01695) = -31,903 \text{ kJ/mol}$ where $\text{R} = 8,3144$, $\text{T} = 310 \text{ K}$. ;

$E_{\text{H}} = \text{Pln}([\text{H}_3\text{O}^+]_{\text{right}}/[\text{H}_3\text{O}^+]_{\text{left}}) = 0,0591 \cdot \text{lg}((10^{-5})/10^{-7,36}) = 0,0591 \cdot \text{lg}(10^{2,36}) = +0,13961549 \text{ V}$;

$E_{\text{membr}} = E_{\text{H}} + E_{\text{CO}_2} = 0,13962 + 0,0202664 = 0,1193 \text{ V}$; $\Delta\text{G}_{\text{F}} = \text{nFE} = -1 \cdot 96485 \cdot 0,1193 = -11,5154 \text{ kJ/mol}$;

Total $\Delta\text{G} = \Delta\text{G}_{\text{HCO}_3^-} + \Delta\text{G}_{\text{H}} + 2 \cdot \Delta\text{G}_{\text{F}} = -1,9554 + (-31,903) + 2 \cdot (-11,5154) = -64,699 \text{ kJ/mol}$.

Carbonic acid formation reaction : $\text{H}_3\text{O}^+ + \text{HCO}_3^- \rightarrow \text{H}_2\text{O} + \text{H}_2\text{CO}_3 + \text{Q}$ exothermic $-9,76 \text{ kJ/mol}$.

$\Delta\text{H}_r = \Delta\text{H}^\circ_{\text{H}_2\text{O}} + \Delta\text{H}^\circ_{\text{H}_2\text{CO}_3} - \Delta\text{H}^\circ_{\text{H}_3\text{O}^+} - \Delta\text{H}^\circ_{\text{HCO}_3^-} = -285,85 - 699,65 - (-285,81 - 689,93) = -985,5 + 975,74 = -9,76 \text{ kJ/mol}$.

$\Delta\text{S}_r = \Delta\text{S}^\circ_{\text{H}_2\text{O}} + \Delta\text{S}^\circ_{\text{H}_2\text{CO}_3} - \Delta\text{S}^\circ_{\text{H}_3\text{O}^+} - \Delta\text{S}^\circ_{\text{HCO}_3^-} = 69,956 + 187 - (-3,854 + 98,324) = 256,956 - 94,47 = +162,486 \text{ J/mol/K} \dots\dots$

$\Delta\text{S}_{\text{dispersed}} = -\Delta\text{H}_r/\text{T} = +9,76/298,15 = +32,735 \text{ J/K/mol} \dots\dots$

$\Delta\text{G}_r = \Delta\text{H}_r - \text{T} \cdot \Delta\text{S}_r = -9,76 - 298,15 \cdot 0,162486 = -58,2052 \text{ kJ/mol}$ is exoergic, spontaneous out as CA absent.

$\Delta\text{G} = \Delta\text{G}^\circ_{\text{H}_2\text{O}} + \Delta\text{G}^\circ_{\text{H}_2\text{CO}_3} - \Delta\text{G}^\circ_{\text{H}_3\text{O}^+} - \Delta\text{G}^\circ_{\text{HCO}_3^-} = -237,19 - 623,17 - (-213,275 - 586,94) = -860,36 + 800,215 = -60,145 \text{ kJ/mol}$.

$\Delta\text{S}_{\text{total}} = \Delta\text{S}_r + \Delta\text{S}_{\text{dispersed}} = +32,735 + 162,486 = 195,221 \text{ J/K/mol}$; $\text{T} \cdot \Delta\text{S}_{\text{total}} = 195,221 \cdot 298,15 = +58,205 \text{ kJ/mol}$

bound $\text{T} \Delta\text{S}_n \leftarrow$ lost free energy

II) third gradual reaction is NON-ENZYMATIC decomposition two branches

$\text{H}_2\text{CO}_3 \leftrightarrow \text{CO}_{2\text{aq}} + \text{H}_2\text{O}$ anenergetic weak exoergic or close neutral; $\text{H}_2\text{CO}_3 \leftrightarrow \text{CO}_2 \uparrow_{\text{gas}} + \text{H}_2\text{O}$;

$\Delta\text{G} = \Delta\text{G}^\circ_{\text{H}_2\text{O}} + \Delta\text{G}^\circ_{\text{CO}_{2\text{aq}}} - \Delta\text{G}^\circ_{\text{H}_2\text{CO}_3} = -237,19 - 385,98 - 623,17 = -623,17 + 623,17 = -1,246 \text{ kJ/mol}$ is weak exoergic;

$\Delta\text{G} = \Delta\text{G}^\circ_{\text{H}_2\text{O}} + \Delta\text{G}^\circ_{\text{CO}_2\text{gas}} - \Delta\text{G}^\circ_{\text{H}_2\text{CO}_3} = -237,19 - 394,359 - 623,17 = -623,17 + 623,17 = -1,255 \text{ kJ/mol}$ is weak exoergic.

Enthalpy change decomposition reaction of carbonic acid $\text{Q} + \text{H}_2\text{CO}_3 \leftrightarrow \text{CO}_2 \uparrow_{\text{gas}} + \text{H}_2\text{O} + \Delta\text{G}$ endothermic

Substance	$\Delta\text{H}^\circ_r, \text{kJ/mol}$	$\Delta\text{S}^\circ_r, \text{J/mol/K}$	$\Delta\text{G}^\circ_r, \text{kJ/mol}$	
H_3O^+	-285.81	-3.854	-213,275	$\Delta\text{H} = \Delta\text{H}^\circ_{\text{H}_2\text{O}} + \Delta\text{H}^\circ_{\text{CO}_2} - \Delta\text{H}^\circ_{\text{H}_2\text{CO}_3} = +20,291 \text{ kJ/mol}$
HCO_3^-	-689.93	98.324	-586,94	$= -286 - 393,509 - (-699,65) = -679,509 + 699,65 = +20,291 \text{ kJ/mol}$
H_2O	-285.85	69.9565	-237,191	is endothermic exactly with the cooling effects.....
$\text{CO}_2 \uparrow_{\text{gas}}$	-393.509	213.74	-394,359	$\Delta\text{S}_{\text{dispersed}} = -\Delta\text{H}/\text{T} = -20,291/298,15 = -68,056 \text{ J/K/mol} \dots\dots$
$\text{CO}_{2\text{aq}}$	-413.7976	117.5704	-385,98	$\Delta\text{S} = \Delta\text{S}^\circ_{\text{H}_2\text{O}} + \Delta\text{S}^\circ_{\text{CO}_2} - \Delta\text{S}^\circ_{\text{H}_2\text{CO}_3} = +96,696 \text{ J/mol/K}$
H_2CO_3	-699,65	187,00	-623,17	$= 69,956 + 213,74 - (187,00) = 257,482 - 94,47 = +96,696 \text{ J/mol/K} \dots\dots$

$\Delta\text{G}_r = \Delta\text{H} - \text{T} \cdot \Delta\text{S} = -8,53891 \text{ kJ/mol}$ exoergic, spontaneous

$= 20,291 - 298,15 \cdot 0,096696 = -8,53891 \text{ kJ/mol}$; $\Delta\text{S}_{\text{total}} = \Delta\text{S} + \Delta\text{S}_{\text{dispersed}} = -68,056 + 96,696 = +28,64 \text{ J/K/mol} \dots\dots$

$\text{T} \cdot \Delta\text{S}_{\text{total}} = 28,64 \text{ J/K/mol} \cdot 298,15 \text{ K} = +8,539 \text{ kJ/mol}$ bound $\text{T} \Delta\text{S}_n \leftarrow$ lost free energy $\Delta\text{G}_{\text{reverse reaction}} \leftarrow \dots$

endoergic $\Delta\text{H}^\circ_{\text{reaction}} = +20,291 \text{ kJ/mol}$; $\text{Q} = -20,291 \text{ kJ/mol}$; exoergic, spontaneous $\Delta\text{G}^\circ_{\text{reaction}} = -8,539 \text{ kJ/mol}$