Biogeochemical cycles of elements (nitrogen, sulfur, metals)

L.P. Wackett, A. G. Dodge, L. B. Ellis, 2004
Microbial Genomics and the Periodic Table, Appl. Environ. Microbiol., 70, 647 - 655
The elements/metals of life

www.webelements.com

<table>
<thead>
<tr>
<th>Essential Transition Metals</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fe: 4.5 g</td>
</tr>
<tr>
<td>Zn: 2.3 g</td>
</tr>
<tr>
<td>Cu: 0.072 g</td>
</tr>
<tr>
<td>Mo: 0.009 g</td>
</tr>
</tbody>
</table>

| C: 17250 g                  |
| N: 1950 g                   |
| O: 45750 g                  |
| P: 825 g                    |
| S: 150 g                    |
| Se: 0.00375 g               |

Ca: 1.2 kg
K: 150 g
Na: 70 g
Mg: 20-30 g
References - Biogeochemical cycles of elements

A. Butler, 1998
Acquisition and Utilization of Transition Metal Ions by Marine Organisms, Science, 281, 207-210

F. M. M. Morel and N. M. Price, 2003
Science, 300, 944-947, The Biogeochemical Cycles of Trace Metals in the Oceans

L.P. Wackett, A. G. Dodge, L. B. Ellis, 2004
Microbial Genomics and the Periodic Table, Appl. Environ. Microbiol., 70, 647 - 655

M. Rudolf, P.M.H. Kroneck, 2005
The nitrogen cycle: Its biology. Metal Ions in Biological Systems, 43, 75-103

Oceanography 20, 101–109, What's New in the Nitrogen Cycle?

Oceanography 20, 117–123, The Sulfur Cycle

SCOR Working Group, 2007
GEOTRACES – An international study of the global marine biogeochemical cycles of trace elements and their isotopes, Chemie der Erde, 67, 85–131

G. M. Gadd, 2010
Metals, minerals and microbes: geomicrobiology, and bioremediation, Microbiology, 156, 609–643
http://en.wikipedia.org/wiki/Sulfur_cycle

http://www.britannica.com/EBchecked/topic/65875/biogeochemical-cycle

Oceanography 20, 117–123, The Sulfur Cycle

E.M. Cameron, 1982
Nature 296, 145-148, Sulfate and sulfate reduction in the early Precambrian oceans

G. Wächtershäuser, 1988
Syst Appl Microbiol, 10, 207-210, Pyrite formation, the first energy source for life: a hypothesis

R.K. Thauer, A. K. Kaster, et al., 2010
Annu Rev Biochem, 79, 507-36, Hydrogenases from methanogenic archaea, nickel, a novel cofactor, and H₂ storage

T. Urich et al., 2006
Science 311, 996 – 999, X-ray Structure of a Self-Compartmentalizing Sulfur Cycle
As the Earth is essentially a closed system with respect to matter, we can say that all matter on Earth cycles.

Generalized biogeochemical cycle. The major parts of the biosphere are connected by the flow of chemical elements and compounds. In many of these cycles, the biota plays an important role. Matter from the Earth’s interior is released by volcanoes. The atmosphere exchanges some compounds and elements rapidly with the biota and oceans. Exchanges of materials between rocks, soils, and the oceans are generally slower by comparison.
Forms of Life – From *Anaerobic* to *Aerobic* anoxic conditions (-O\(_2\)) vs oxic conditions (+O\(_2\))*


![Graph showing estimated evolution of atmospheric O\(_2\)](image)

**Estimated evolution of atmospheric O\(_2\)**

The red and green lines represent the range of the estimates: stage1: 3.85–2.45 Gyr (Ga), stage2: 2.45–1.85Ga, stage3: 1.85–0.85Ga, stage4: 0.85–0.54Ga, stage5: 0.54Ga–present

[www.globalchange.umich.edu/globalchange1/current/lectures/Perry_Samson_lectures/evolution_atm/index.html](http://www.globalchange.umich.edu/globalchange1/current/lectures/Perry_Samson_lectures/evolution_atm/index.html)
Today’s Energy Conservation: Reduction of O₂ to H₂O
Humans are Aerobes

![Diagram of energy conservation process involving NADH, O₂, H₂O, and E (Volt)]
“The deep past of the Earth is unobservable. Therefore, the problem of the origin of life can only be approached by a theory.”

Quote from *Origin of Life: RNA World versus Autocatalytic Anabolist*  
See lecture by T. Cech, Nobel Prize in Chemistry, 1989
Emergence of life on the ocean floor
hydrothermal vent, Russel and Hall, 1997

Atmosphere ≤10 bars of CO₂ with CO, SO₂ and N₂ but very low O₂

ANCIENT OCEAN
(acidic due to high content of carbon dioxide)

Volcanic oceanic crust
(mafic / ultramafic)

Descending seawater

Oxidation

Hydration

Carbonation

Volcanic

OCEAN FLOOR

Hydrothermal solution
150°C
pH ~ 9

H₂
HS⁻
NH₃
CH₃COO⁻
C₂H₅S⁻
[W]

Reverse Krebs cycle

Acetate
+ HS⁻
+ RS⁻
+ H₂

Phosphate

Membranous froth of FeS and proferredoxins (10-100μm)

Emergence of life on the ocean floor
4.2 thousand million years ago

90°C
pH ~ 5.5
H₂CO₃

4Km

9

Emergence of life on the ocean floor
hydrothermal vent, Russel and Hall, 1997

Atmosphere ≤10 bars of CO₂ with CO, SO₂ and N₂ but very low O₂

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+ H₂

Phosphate

Membranous froth of FeS and proferredoxins (10-100μm)
Basic Redox Couples
Biogeochemical Cycles

- **Metabolism**: All organisms transform (different) compounds

- **Metabolic pathways** constitute closed global cycles, otherwise intermediates will accumulate. The larger the amount of the accumulated intermediate, the better substrate it will be for organisms. On a geological time scale, a new metabolic pathway will develop which will convert the intermediate.

- **Metabolic pathways** can be organized and classified according to different aspects, such as with regard to the **ELEMENTS**.
Elements in the Ocean

A. Butler, 1998
Acquisition and Utilization of Transition Metal Ions by Marine Organisms, Science, 281, 207-210
Essential Metal Sites (V, Fe, Cu, Mo)

A. Butler, 1998
Acquisition and Utilization of Transition Metal Ions by Marine Organisms, Science, 281, 207-210
Vertical profiles of dissolved zinc and iron concentrations in the north Pacific Ocean.
(A) Release of complexing agents and metal ligand complexes from marine plankton: CdX, phytochelatin-Cd complex released by diatoms; CuY, peptide complexes of Cu released by coccolithophorids; CuZ, unidentified Cu ligand complex released by Synechococcus; sid, siderophore released by heterotrophic bacteria and cyanobacteria; L, unidentified Co complexing agent released by Prochlorococcus; C, Cys; E, Glu; G, Gly; Q, Gln; and R, Arg.

F M M Morel, N M Price Science 2003, 300, 44-947
The N cycle, illustrating the metal cofactors in each enzymatically catalyzed step. (B) Primary metal requirements for C, N, and P acquisition and assimilation by phytoplankton.

F M M Morel, N M Price Science 2003, 300, 944-947
A Cadmium enzyme from a marine organism
Lane et al., NATURE, 435, 42 (2005)

Active Cd-Carbonic Anhydrase
(purified by chromatography; metal analysis by X-ray spectroscopy)

Note: Until most recently, this enzyme has been regarded as a classical Zn-dependent enzyme. And indeed, usually Cd is highly toxic, replacing Zn in many important proteins.
The Nitrogen Cycle: Note Activities of Microorganisms

http://en.wikipedia.org/wiki/Nitrogen_cycle
Important Reactions and Metal Enzymes of the Nitrogen Cycle
# Oxidation states of N

<table>
<thead>
<tr>
<th>Oxidation State</th>
<th>Compound</th>
</tr>
</thead>
<tbody>
<tr>
<td>+5</td>
<td>NO₃⁻</td>
</tr>
<tr>
<td>+3</td>
<td>NO₂⁻</td>
</tr>
<tr>
<td>+2</td>
<td>NO</td>
</tr>
<tr>
<td>+1</td>
<td>N₂O</td>
</tr>
<tr>
<td>±0</td>
<td>N₂</td>
</tr>
<tr>
<td>-1</td>
<td>H₂NOH</td>
</tr>
<tr>
<td>-3</td>
<td>NH₃</td>
</tr>
</tbody>
</table>

**New intermediate: hydrazine, H₂NNH₂**

Kuypers et al. (2003) „Anaerobic ammonium oxidation by anammox bacteria in the Black Sea”, Nature 422, 608-611
# Essential Metals of the N Cycle

<table>
<thead>
<tr>
<th></th>
<th>Fe</th>
<th>Cu</th>
<th>Mo/V</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>- Nitrate Reductase</td>
<td>- Nitrite Reductase</td>
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<td></td>
<td>- Nitrite Reductase</td>
<td>- Nitrite Reductase</td>
<td></td>
</tr>
<tr>
<td></td>
<td>- NO Reductase</td>
<td>- N₂O Reductase</td>
<td>- Nitrogenase</td>
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<tr>
<td></td>
<td>- Nitrogenase</td>
<td></td>
<td>- Nitrogenase</td>
</tr>
<tr>
<td></td>
<td>- Ammonia Mono-oxygenase</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>- Hydroxylamine Oxidoreductase</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Nitrate Reductase

\[ \text{NO}_3^- + 2\text{H}^+ + 2\text{e}^- \rightarrow \text{NO}_2^- + \text{H}_2\text{O} \]
Mo bound to Molybdopterin
(a dithiolene – coordinates both Mo and W)

Numerous different forms of nitrate reductase depending on the organism
http://www.ebi.ac.uk/thornton-srv/databases/cgi-bin/enzymes/GetPage.pl?ec_number=1.7.99.4

PDB code: 2NAP
Dissimilatory Nitrite Reductase

\[
\text{NO}_2^- + 8\text{H}^+ + 6\text{e}^- \rightarrow \text{NH}_4^+ + 2\text{H}_2\text{O}
\]
Cytochrome c Nitrite Reductase

Nitrogen Uptake

- Nitrogen is an important constituent of biopolymers and numerous essential biomolecules. The only N compound which can be taken up by higher organisms is Ammonium, NH$_4^+$. 

**Amino acid synthesis: Amination of α-Ketoglutarate**

(assimilatory process vs dissimilatory process)
Biological N\textsubscript{2} Fixation

Microorganisms can do the job under „normal conditions“ (T, P)

- free living soil bacteria, e.g. \textit{Azotobacter vinelandii}
- Cyanobacteria with specialized cells, e.g. \textit{Anabaena sp.}, \textit{Nostoc sp.})
- \textit{Rhizobia} which live in special plant organelles (root nodules)

The process, however, is costly. Plants have to deliver up to 25\% of their photosynthetically produced ATP to N\textsubscript{2} fixing bacteria in the root nodules.

Note: N\textsubscript{2} Fixation is an anaerobic process
**N₂ Fixation**

http://en.wikipedia.org/wiki/Nitrogenase

\[
\text{N}_2 + 8\text{H}^+ + 16\text{Mg}^{2+}\text{ATP} + 8\text{e}^- \rightarrow 2\text{NH}_3 + \text{H}_2 + 16\text{Mg}^{2+}\text{ADP} + 16\text{P}_i
\]
The chemical speciation of arsenic in the stratified water column of Mono Lake, California (left) as explained by the metabolism of arsenic by microbial populations present in the water column (right).

R S Oremland, J F Stolz Science 2003, 300:939-944
Phylogenetic diversity of representative arsenic-metabolizing prokaryotics.

R S Oremland, J F Stolz Science 2003, 300, 939-944
The Biogeochemical Cycle of Sulfur
Life without Dioxygen – Respiration with S-Oxides Reduction of $\text{SO}_4^{2-}$ to $\text{H}_2\text{S}$

http://www.scienceclarified.com/Oi-Ph/Oxygen-Family.html
Reactions of the Biogeochemical Sulfur Cycle

- Sulfate Reduction (Assimilatory)
- Organism Sulfur
- Mineralization
- Elemental Sulfur
- Sulfur Oxidation
- Sulfate Reduction (Dissimilatory)
- Sulfur Reduction
- $H_2S$
- $SO_4^{2-}$
Oxidation/Reduction of Sulfur Compounds Coupled to Energy Conservation Evolved Early in Prokaryotic Evolution
Tracking Footprints of Early Life
Nature’s Toolbox to handle Dihydrogen

\[ \text{H}_2 \rightleftharpoons 2\text{H}^+ + 2\text{e}^- \]
Overall structure of a Fe-only H₂ase from (Handbook of Metalloproteins)

\[ H_2 \Leftrightarrow 2H^+ + 2e^- \]

uni- vs bidirectional/

Ni,Fe vs Fe-only
Presence of *Toxic* Ligands at Active Site
Analysis of CO and CN$^-$ by FT IR

**FTIR bands of *D. vulgaris* Fe-hydrogenase**

- $H_2$
- $H_2/Ar$
- $H_2/CO$
- Air exposed

Wavenumber (cm$^{-1}$):
- 2100
- 2050
- 2000
- 1950
- 1900
- 1850
- 1800
Fe-only vs Ni,Fe Hydrogenase

H-Cluster der Fe-only Hydrogenase

[NiFe] Cluster der Ni,Fe Hydrogenase
Life without O$_2$ - Sulfur Respiration

Reduction of SO$_4^{2-}$ to H$_2$S, a multi-electron, multi-proton transfer process coupled to energy conservation.

\[ 2 \text{H}_2 + 2 \text{e}^- + 2 \text{H}^+ \rightarrow \text{H}_2\text{S} \]
SO$_4^{2-}$ must be first activated, then microorganisms can eat it

$$APS \rightleftharpoons AMP + SO_3^{2-}$$

Activation of sulfate ($E^{\circ'} = -516$ mV) to

adenosine-5’-phosphosulfate = APS ($E^{\circ'} = -60$ mV)
APS Reductase from thermophilic *Archaeoglobus fulgidus*

APSR has two [4Fe-4S] – Clusters which interact

$E'_o = -60 \text{ mV}$

$E'_o = -520 \text{ mV}$

Magnetic field [mT]
Chemistry of Sulfite Reduction

Sulfite reduced to sulfide (directly or via trithionate/thiosulfate) by sulfite reductase in a six-electron transfer process.

Dissimilatory (dSIR) Sulfite Reduction

Assimilatory (aSIR) Sulfite Reduction
SIR contains Siroheme

- Siroheme = heme-type prosthetic group
- Catalyzes the six-electron reduction of sulfite to sulfide, and of nitrite to ammonia
- Formed by methylation, oxidation, and Fe insertion into uroporphyrinogen III
Sulfite Reductase (36 Fe) from *Archaeoglobus fulgidus*

\[
\text{SO}_3^{2-} + 6e^- + 8H^+ \rightarrow H_2S + 3 \text{H}_2\text{O}
\]


EPR (X-Band), resonances between \(g\) 18 and \(g\) 1.9

Unique cofactor: Siroheme coupled to \([4\text{Fe}-4\text{S}]\)
Overall structure of *A. fulgidus* dSIR $\alpha_2\beta_2$ complex

- Final model contains four siroheme and eight [4Fe-4S] centers = 36 Fe
- N- and C-terminal ends involved in intradimer and interdimer formation
- Tetrameric state may help to survive in hot environments
- One $\alpha\beta$-unit contains the catalytic siroheme center

$\gamma$-subunit DrsC, interacts with weakly with $\alpha_2\beta_2$ complex in *A. Fulgidus*, however strongly in *D. vulgaris*
Cofactors of *A. fulgidus* dSIR

Functional and structural siroheme-[4Fe-4S] centers in one αβ dimer; both sirohemes are arranged towards each other.

Minimal distance between the acetate carboxylates 9.6 Å; distance 3.9-4.3 Å from the siroheme to next [4Fe-4S] cluster.
Cofactors of Sulfite Reductase

Schiffer et al., J. Mol. Biol. (2008), 379, 1063-74

- Catalytic (left) and structural siroheme-[4Fe-4S] center (right) in one αβ dimer: note the *blocking* tryptophan in the structural center
SO$_3^{2-}$ Adduct and the Substrate Channel

Positively charged residues guide the substrate. Parey et al. (2010) Biochemistry 49, 8912-21
Reactivity in solution & *in crystallo* of *A. fulgidus* dSIR

Structure of a Self-Compartmentalizing Sulfur Cycle
T. Urich et al. (2006) Science 311, 996 – 999

C
(1) Oxygenase \( S + O_2 + H_2O \rightarrow HSO_3^- + H^+ \)
(2) Disproportionation \( 3 S + 3 H_2O \rightarrow HSO_3^- + 2 HS^- + 3 H^+ \)
(3) Sum \( 4 S + O_2 + 4 H_2O \rightarrow 2 HSO_3^- + 2 HS^- + 4 H^+ \)
(4) Non-enzymic \( S + HSO_3^- \rightarrow S_2O_3^{2-} + H^+ \)