OXYGENOLOGY
IN OUTLINE

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COVER: Earth planet. Earth is an enormous beautiful blue globe because oxygen in atmosphere causes a selective absorption of waves in light spectrum.

CONTENTS

INTRODUCTION

Oxygenology has been defined by Stępniewski and Stępniewski (1998) as a scientific discipline related to the presence and role of oxygen in nature on Earth. It constitutes a branch of environmental sciences and comprises issues of storage, transport, generation, absorption, turnover, functions and measurement of oxygen content in the environment. The name of this new discipline is analogous to that of hydrology.

The proposal of identification of oxygenology as a separate branch of science is justified not only by the unique role of oxygen as the most abundant component of the lithosphere and as an exceptional component, on the cosmic scale, of the atmosphere of our planet, but also by the need for a holistic approach to oxygen related problems faced in aquatic, wetland and dry-land ecosystems because of their common nature and structure.

Oxygen, being a dominant final acceptor of electrons, plays an essential role in the life of all macro- and microorganisms, as well as in the biochemical and chemical processes occurring in the environment.

Having limited our interest only to Earth's oxygenology (*i.e.* without oxygenology related to other planets), and more precisely only to the contemporary oxygenology, such subbranches as atmospheric, aquatic, lithospheric, bio- and human oxygenology have been distinguished. The aim of this book is to present only an outline of the basic problems and examples of the issues involved in oxygenology, as presentation of the entire scope of this discipline would require several books and specialistic meetings. An illustration of this fact is the book devoted solely to soil oxygenology (Gliński and Stępniewski, 1985) that made reference to more than 820 publications and the book with 9 presentations on the soil – plant – atmosphere aeration and environmental problems (Gliński *et al.*, 2004).

The last book and two meetings were realized within the EU Centre of Excellence AGROPHYSICS (Workpackage 3 "Soil-plant-atmosphere aeration problems"):
- Summer School on "Soil-Plant-Aeration and Environmental Problems" (26-29 July, 2004) organized in Stuttgart, Germany by the Institute of Agrophysics in Lublin and the Hohenheim University in Stuttgart.
- Symposium on "Gas Exchange in Soil" organized in Freiburg, Germany (4-12 September, 2004) during the International Congress EUROSOIL.

Moreover, the other extension of oxygenology was creation of Website http://oxy.ipan.lublin.pl which includes main information about this new discipline.

1. HISTORY OF RECOGNITION OF OXYGEN PRESENCE

The discovery of oxygen is connected with controversial opinions. This element, most popular in the lithospheric composition (46.6%) or in the biosphere (53%), as well as absolutely needed for men so that we would be dead in minutes without it, was a secret of a long time. From an old book of chemistry of Klaproth it is known that in the 13th century the Chinese already knew the composition of water and the nature of the atmosphere. They knew that the air had an element which had a tendency to bond with many metals and sulfur and carbon, but not with gold. According to them, oxygen could be prepared by heating up *saltpeter* (potassium nitrate) and certain minerals like pyrolusite.

The role of the air in combustion was observed by Leonardo da Vinci (1452-1519), and in 1669 by Mayow who stated that the *spiritus nitro-aereus* (oxygen) caused a mass increase in the metals when heated up.

Michał Sędziwój (1556-1636)

The discovery of oxygen and recognition of its role in the environment is connected with the Polish alchemist Michał Sędziwój (known in Europe as Michael Sendivogius Polonus) who, in 1604, wrote that "Man was created of the Earth, and lives by virtue of the air; there is in the air a secret food of life... whose invisible congealed spirit is better than the whole Earth" (After Lane, 2003). Sędziwój proposed that this "aerial food of life" circulated between the air and earth by way of an unusual salt – nitrate or saltpetre. When heated above 336°C, during decomposition it released oxygen which was known as *aerial nitrate.* This discovery had the name of the Elixir of Life and according to the author "without which no mortal can live, and without which nothing grows or is generated in the world". Sędziwój explained his method of producing oxygen to his Dutch fellow, Cornelius Drebbel, who demonstrated the role of oxygen in 1621 by constructing the world's first wooden submarine that stayed under the water for three hours with twelve oarsmen (Lane, 2003).

In 1678, oxygen was extracted from <u>saltpeter</u> by Borch; in 1731, from the same substance, by Hales; and in 1774 from the mercury oxide by Bayen.

Determining the oxygen content in the air is mostly connected with three very famous names in the eighteen century: the English chemist Joseph Pristley (1774), who obtained oxygen by focusing sunlight onto an oxide of mercury, the Swedish apothecary Carl Scheele, who prepared oxygen from nitrates, and the French very modern chemist Antoine Lavoisier, who proved that the gas consumed during combustion and during the breathing of animals was the reactive constituent of air, and that the two processes had the same purpose. However, neither <u>Priestley</u> nor <u>Scheele</u> was able to know the true nature of new element, from the reason of their "flogisto" theories. This work was done by <u>Lavoisier</u>, who derived the name of "oxygen" from the Greek for "acid-former", in the mistaken belief that in all acids a content of oxygen was necessary.

<u>Priestley</u>, with discovery of oxygen, foresaw not only its medical application but also its potential danger, which was published in his: "Experiments and Observations on Different Kind of Air" in 1775. His words contain the suggestion, not documented for the next hundred years, that oxygen can accelerate ageing (Lane, 2003).
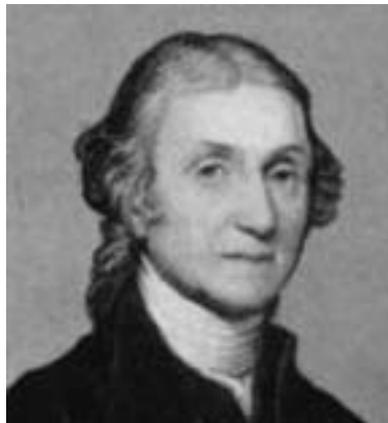
Joseph Pristley (1733-1804)   Carl Wilchelm Scheele (1742-1786)

The use of oxygen therapy on a large scale was performed by Thomas Beddoes who founded, in 1798, the Pneumatic Institute for inhalation gas therapy in Bristol.

Interest in oxygen therapy was suggested in many reports that higher pressure of oxygen did affect health. For citizens of Mexico City, where oxygen pressure is low, the conditions with higher pressure of oxygen give a better chance of recovery. To have higher barometric pressure, the American physician Orval Cunningham

constructed, in 1928, the hyperbaric chamber in Cleveland with twice the atmospheric pressure at sea level. He used not oxygen but compressed air, which was not beneficial for patients with diabetes, cancer or pernicious anemia. Another expert in oxygen inhalation was the Scottish physiologist John Scott Haldane who published a book "Respiration", where he explained that oxygen therapy gives the body an opportunity to recover the healthy equilibrium. Haldane also observed the risk of oxygen therapy which, especially under pressure, can cause a shocking reaction. It was found that breathing oxygen at high concentration or pure oxygen causes convulsions and sometimes death. At atmospheric pressure breathing pure oxygen for a few days can evoke serious lung damage and slowing down the heart beat and producing fewer red blood cells as a result of adaptation. In effect, the most important is unchanging of oxygen levels in the body except when concentration of oxygen is pathologically deprived.

Antoine Laurent Lavoisier (1743-1794)

2. APPEARANCE OF OXYGEN IN THE ENVIRONMENT (PALEOOXYGENOLOGY)

The appearance of oxygen in the atmosphere and in the oceans is connected with oxygenic photosynthesis by cyanobacteria and by plants. Its presence in the atmosphere promotes oxidative weathering of rocks, and the accumulation of sulphates in the oceans as well as the formation of iron oxides during weathering on land are two substantial geochemical expressions of the presence of oxygen in the atmosphere. Another indicator is sedimentary sulfur isotope record due to its isotopic fractionation (Farquahar and Wing, 2003).

Variation in the amount of atmospheric oxygen has been a matter of intense study and controversy among geologists and evolutionary biologists since the suggestion by Cloud (1972) that Archean atmosphere contained much less oxygen then now. In one scenario, presented by Ohomoto (1997), oxygen started to accumulate by the earliest Archean. He summarized the evidence that atmospheric oxygen pressure was at levels of at least 50% of the present atmospheric level (PAL) during the last 4 Ga (billions years).

In another scenario, oxygen started to accumulate much later – in the early Proterozoic – and the present-day levels have been reached in the Neoproterozoic, 0.54-1 Ga ago (Canfield et al., 2000). The papers of Holland (1994, 1999) have summarized the arguments supporting the view that oxygen was absent, or present at very low concentrations, until ~2.3 Ga before present. According to these data, oxygen concentration after that increased rapidly causing so called Great Oxidation Event, and by 2 Ga ago the atmospheric oxygen pressure reached 10-15% PAL.

Recent papers seem to provide evidence for the second scenario. The paper of Yang et al. (2002) indicates that the 2.76 Ga Mt. Roe (Western Australia) paleosol was developed under atmosphere which contained very little or no oxygen. Other papers by the same authors (Holland and Yang, 2000; Yang and Holland, 2000) contain evidence that significant amounts of oxygen were present in the atmosphere ~ 2.25 Ga. According to Canfield et al. (2000) and Kasting (2001) there is evidence for the presence of oxygenic photosynthesis due to the appearance of cyanobacteria by 2.7 Ga, and for the existence of a protracted period from >2.7 Ga to 2.2 Ga with little net oxidation of the Earth's surface. A recent paper by Bakker et al. (2004) presented evidence that atmospheric oxygen concentration was extremely low before 2.45 Ga and that the rise of atmospheric oxygen to a considerable level – higher than 10^{-5} PAL – had occurred by 2.32 Ga ago. Also

Huston and Logan (2004) support the view that increasing atmospheric oxygenation took place around 2.4 Ga ago.

Recently, Farquhar and Wing (2003), on the basis of the analysis of sulfur isotopes, distinguished three stages in the history of Earth. Stage I extended before 2.45 Ga and was characterized by oxygen levels $< 10^{-5}$ PAL. Stage II extended from 2.45 to 2 Ga and was characterized by intermediate oxygen levels of 10^{-2}-10^{-5} PAL. Stage III extends from 2 Ga until the present, with relatively high oxygen concentrations. It should be added that a possibility of oscillation of atmospheric oxygen levels in the time interval corresponding to stage II has been also suggested by Wing *et al*. (2002).

Different models of atmospheric composition in the past are presented in Figures 2.1-2.3. According of one of theme (Fig. 2.1) oxygen content reached the highest 35% level on the brake of Carboniferous and Permian periods, and then fell to the level of 15% at the end of Permian period. Oxygen content rose again in Cretaceous period. Carbon dioxide changes showed a reverse tendency but the amplitude of these changes was much lower.
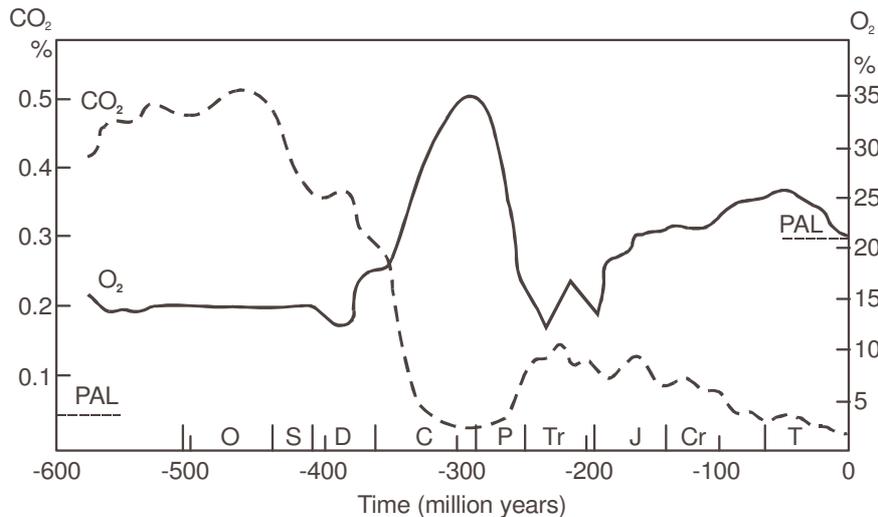
Fig. 2.1. Changes in the concentration of oxygen and carbon dioxide in the atmosphere during the last 600 millions of years on the basis of models by Berner and Canfield (1989). Geologocal periods: O – Ordovician, S –Sidurian, D – Devonian, C – Carboniferous, P – Permian, Tr – Trassic, J – Jurassic, Cr – Cretaceous, T – Tertiary (After Lane, 2003, modified)
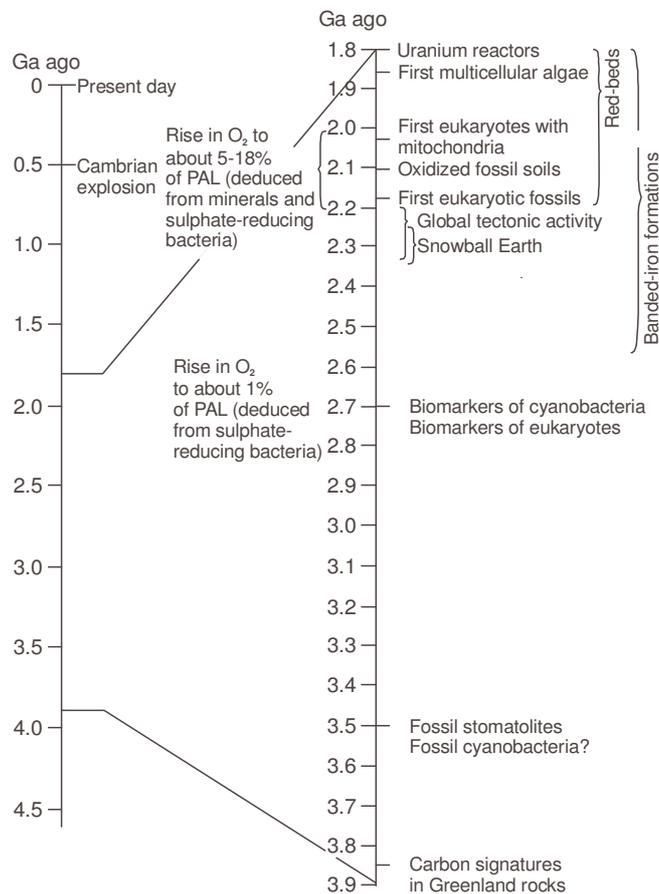
Fig. 2.2. Development of atmospheric oxygen concentration on Earth in the geological time scale with particular emphasis on the mid-Precambrian period (Archaean and early Proterozoic). The burst of biological activity in the period of 2.3 to 2.0 Ga ago when oxygen level rose to 5-18% of PAL (After Lane, 2003, modified)

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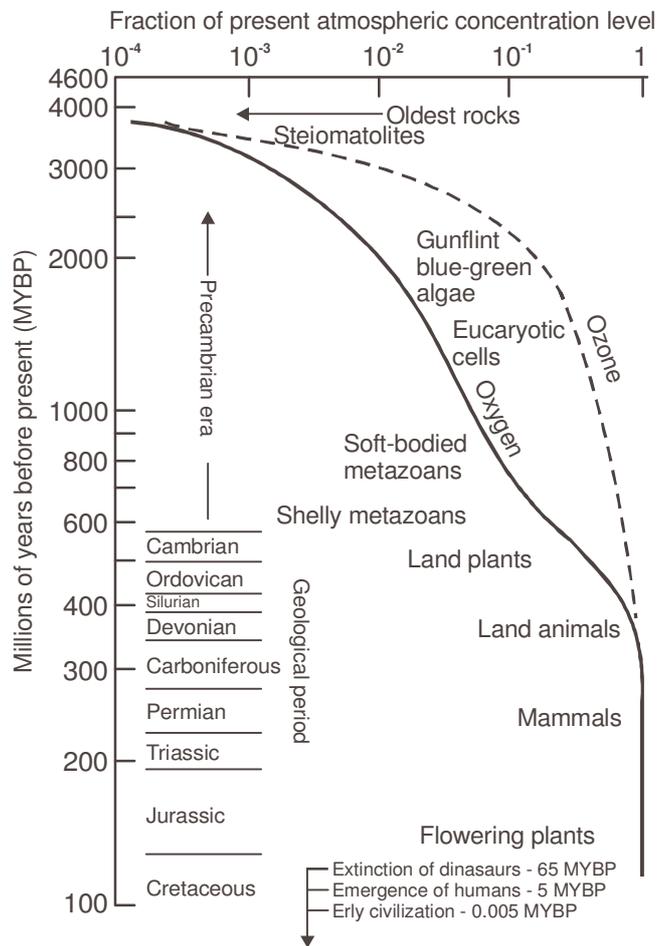
Fig. 2.3. The changes of atmospheric oxygen and ozone content on the background of development of different forms of living organisms during particular geological periods (After Iowa State University, Dept. of Geological and Atmospheric Sciences, Geocourse, 2005, modified)

3. OXYGEN FORMS AND PROPERTIES

The particular properties of oxygen are connected with the specificity of O_2 chemistry (Halliwell and Gutteridge, 1999), which is denoted in literature as a paradox. In its ground state, molecular O_2 is relatively unreactive due to the parallel spin of the electrons in the two outer uncompleted $\Pi*2p$ orbitals:

$$:\overset{\uparrow}{\underset{..}{O}}:\overset{\uparrow}{\underset{..}{O}}: \tag{3.1}$$

All aerobes consume O_2 by using it as a terminal electron acceptor. Reduction of O_2 to H_2O requires high activation energy and at ambient temperature could proceed very slowly.

$$O_2 + 4e^- + 4H^+ \rightarrow 2H_2O \tag{3.2}$$

The utilization of molecular oxygen may proceed by an alternative route — consecutive monoelectron transfers generating partially reduced oxygen to high reactivity intermediates, which may have damaging or even lethal effects (Halliwell and Gutteridge, 1999). The oxygen paradox is thus defined as an obligate dependency for aerobes on oxygen reduction and an inevitable hazard, which is connected with reduction of oxygen at the same moment.

This situation restricts the acceptance of two electrons at a time from the molecules to be oxidized which contain electron pairs with opposite spins. This is in accordance with Pauli principle postulating that only electrons with antiparallel spins (*i.e.* different quantum numbers) can form an electron pair.

$$:\overset{\uparrow}{\underset{..}{O}}:\overset{\uparrow}{\underset{..}{O}}: \;+\; \overset{\uparrow\downarrow}{X} \;\longrightarrow\; :\overset{\uparrow\uparrow}{\cancel{O}}:\overset{\uparrow\downarrow}{\underset{..}{O}}: \;\longrightarrow\; :\overset{\uparrow}{\underset{..}{O}}:\overset{\uparrow\downarrow}{\underset{..}{O}}: \tag{3.3}$$

| Ground O_2 state | Molecule to be oxidized | Forbiden transfer | Allowed transfer | Superoxide free radical |
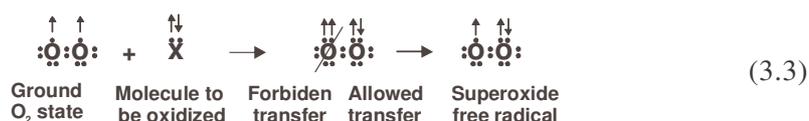
From the reason of electronic chemistry restrictions, reduction of O_2 at ambient temperature is going on consecutive with single electron transfer (Fig. 3.1).

The transformation of oxygen into H_2O molecule is going on in endothermic and exothermic 4 steps:

Step 1 is superoxide generation by acceptance of one electron. This step is endothermic and hence rate-limiting.

Step 2 is exothermic and hence spontaneous. The protonation of superoxide gives rise to hydroperoxyl radical, HO_2*. The superoxide is reduced by accep-

tance of one electron and protonated by two H^+, this resulting in H_2O_2 formation.

Step 3 – H_2O_2 undergoes heterolytic fission in which one atom oxygen receives both electrons from the broken covalent bond. This moiety is protonated yielding one molecule H_2O. The other moiety receives one electron and is transformed in hydroxyl free radical (OH*).

Step 4 – OH* (hydroxyl radical) receives one electron and, after protonation, yields one molecule H_2O.
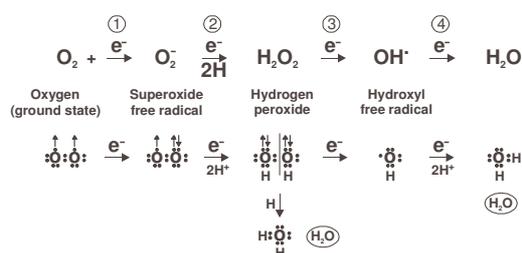
Fig. 3.1. Consecutive four-step monoelectron reduction of dioxygen yielding reactive oxygen intermediates and $2H_2O$. (After Edreva, 2005)

$$(3.4)$$
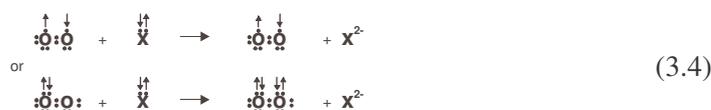
These O_2 products are highly reactive and in literature are nominated as "reactive forms of oxygen" or "reactive oxygen species" (ROS) (Halliwell and Gutteridge, 1999, and Mittler, 2002). They include in the category of ROS different types of free radicals as well as non-radical molecules of high reactivity such as H_2O_2, singlet oxygen ($^1O_2$), O_3, etc. The high reactivity of ROS is connected with the specificity of their electronic configuration because free radicals having unpaired electrons in the outer orbitals (Fig. 3.1) easily form pairs with other electrons characterized with antiparallel spin. In this way free radicals can react with non-radical compounds, transforming them into new radicals and initiating a chain of free radical reactions. By input of energy, spin reversal is produced in the ground state oxygen (O_2), where Π^* electrons have antiparallel spins:

$$(3.5)$$
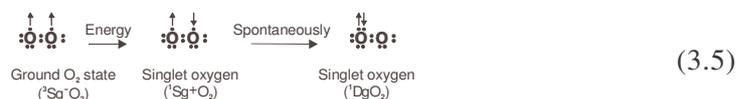
When in the $^1O_2$ molecule the spin restrictions are abolished, its oxidizing ability is greatly increased (Knox and Dodge, 1985).

Different types of ROS can be easily interconverted forming a complex (Elstner and Osswald, 1994). Some transition metals which have unpaired electrons in their electron configuration, such as Fe, Cu, Mn, can intermediate in transferring single electrons and, by promoting monoelectron transfers to O_2, in interconvertion and oxidoreduction of high reactivity molecules (ROS) (Hippeli *et al.*, 1999).
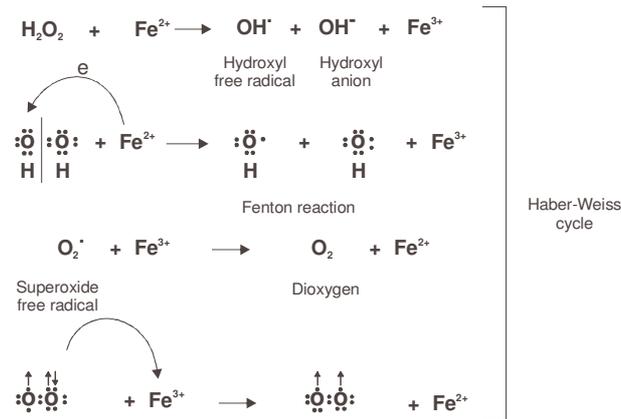
Fig. 3.2. Fenton reaction and Haber-Weiss cycle. In Fenton reaction H_2O_2 undergoes heterolytic fission. By acceptance of one electron from Fe^{2+} the one moiety of H_2O_2 is reduced to hydroxyl free radical (OH*), and Fe^{2+} is oxidized to Fe^{3+}. The other moiety of H_2O_2 is the hydroxyl anion (OH⁻). In the next step Fe^{3+} is reduced to Fe^{2+} by accepting one electron from superoxide free radical ($O_2^{·-}$), the latter being oxidized to O_2. (After Edreva, 2005)

In one of those, known as the Fenton reaction, there is a component of the Haber-Weiss cycle, in which an active role in the conversion of H_2O_2 to OH plays Fe^{+2} (Fig. 3.2). This type of damage is one of the central events which produce the most reactive free radicals known in biological systems (Grant and Loake, 2000). The conversion of superoxide free radical to O_2 and H_2O_2 reduces Fe^{+2} to Fe^{+3} and can be enzymatically catalysed by SOD (superoxide dismutase) or provided spontaneously. This type of reaction in which one electron acts as an electron donor ant another acts as an electron acceptor is called dismutation (Fig. 3.3).

It was found that ROS (Tab. 3.1) are intrinsic for cell functioning, and are present at a low level even at stationary conditions in cells. They can play also signal functions at narrow concentrations (van Breusegem *et al.*, 2001, Neil *et al.*, 2002).

Protective or signalling factors depend on the equilibrium between ROS production and scavenging at the same time. In the situation of uncontrolled ROS production and non sufficient scavenging, there may arise oxygen toxicity.

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Table 3.1. Reactive oxygen species (After Bartosz, 1995)

Name	Structure
Singlet oxygen	$^1O_2$
Ozone	O_3
Hydroperoxyl radical	HO_2
Superoxide radical anion	$O_2^{\cdot-}$
Hydrogen peroxide	H_2O_2
Hydroxyl radical	$^{\cdot}OH$
Nitric oxide	$NO^{\cdot}$
Nitric dioxide	$NO_2^{\cdot}$
Peroxynitrous acid	$O=N-OOH$
Peroxynitrite	$O=N-OO^-$
Hypochlorous acid	$ClOH$
Hypochlorite	ClO^-
Hypobromous acid	$BrOH$
Hypobromite	BrO^-
Hypoiodous acid	IOH
Hypoiodite	IO^-
Hypothiocyanous acid	$S=C=N-OH$
Hypothiocyanite	$S=C=N-O$
Alkoxyl radical	$RO^{\cdot}$
Peroxyl radical	$ROO^{\cdot}$
Peroxide (= hydroperoxide)	$ROOH$
Acyloxyl radical	$R-\overset{\overset{\displaystyle O}{\|}}{C}-O^{\cdot}$
Acylperoxyl radical	$R-\overset{\overset{\displaystyle O}{\|}}{C}-OO^{\cdot}$
Aryloxyl radical	$ArO^{\cdot}$
Arylperoxyl radical	$ArOO^{\cdot}$
Semiquinone radical	$H-Ch^{\cdot}$
Semiquinone radical anion	$Ch^{\cdot-}$
Epoxide	$-\overset{\displaystyle O}{\underset{\diagup\diagdown}{C}}-C-$

Different types of scavengers: enzymatic and non-enzymatic, were found in plants, which operate in chloroplasts or in the situation of biotic and non-biotic stressors (Blokhina *et al.*, 2003).

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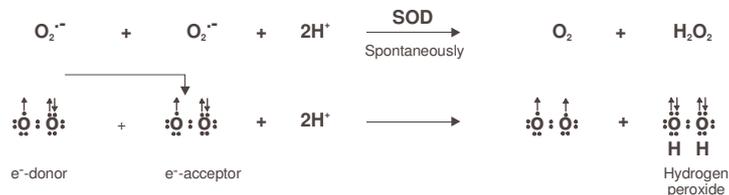
Fig. 3.3. Superoxide free radicals in dismutation reaction (From Edreva, 2005)

There are three main types of ROS producers in plants:
- in chloroplasts and mitochondria as an electron-transport chain (ETC)
- some peroxidases and oxidases (NADPH oxidase, NADH oxidase, xanthine oxidase, lipooxygenase, glycolate oxidase, amine oxidase, etc.)
- photosensitizers such as chlorophyll molecules (Dat *et al.*, 2000).

The main ROS producers and scavengers in relation to chloroplasts are presented in Table 3.2.

Table 3.2. Main ROS producers and scavengers related to chloroplasts (After Edreva, 2005)

Producers	ROS	Scavengers' Enzymatic	Non-enzymatic
ETC in PSI	O_2^-	SOD	Ascorbic acid, glutathione (hydrophilic),
ETC in PSII	H_2O_2	APX, TdPX	carotenoids, a-tocopherol (lipophilic)
	OH		
Triplet chlorophyll	$^1O_2$		
Photorespiration	$H_2O_2^b$ CAT^b		

Note: APX: ascorbate peroxidase; CAT: catalase; ETC: electron transport chains; PSI, PSII: photosystem I, photosystem II; ROS: reactive oxygen species; SOD: superoxidedismutase; TdPX: thioredoxin peroxidase.

" Extrinsic polypeptides and Mn clusters in the water splitting complex of PSII were recently reported as O_2^- scavengers (Zhang *et al.*, 2003).

[b] Located in peroxisomes.

Oxygen is activated mainly during photosynthesis reactions (called type I and II) by photodynamic chlorophyll excitation (Hippeli *et al.*, 1999). Production of $^1O_2$ in chloroplasts is presented in Figure 3.4. In the chlorophyll molecule in the ground state (S_o), by adsorption of light energy, one electron is ejected to a highly energetic state (S_2^*). This electron reaches lower energetic state (S_1^*) by losing energy and after that triplet chlorophyll T^* is formed. This changes of energetic state are connected with reversal of electron spin – in this way triplet state of chlorophyll is generating, which in contact with oxygen in ground state produces reversal spin of one electron in O_2 *i.e.* singlet oxygen ($^1O_2$) is formed when triplet state of chlorophyll returns to ground state (S_o) (Niyogi, 1999).
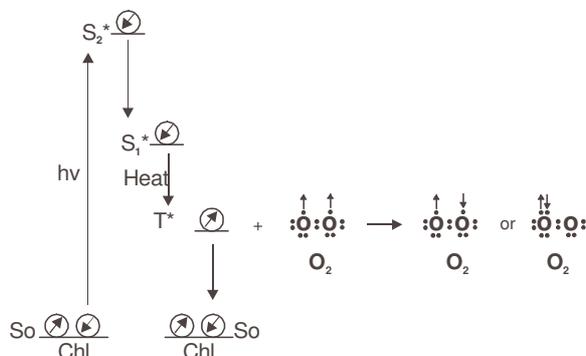
Fig. 3.4. Generation of singlet oxygen ($^1O_2$) through interaction of O_2 with triplet state chlorophyll (T^*), (After Edreva, 2005)

During photosynthesis processes, the ETC in chloroplasts operate in $O_2^{\cdot}$ rich environment. To keep ROS level under control, different scavengers are revealed. An important role in leakage of electrons to O_2 may occur as the easy exchange electrons. This reaction is possible in the presence of transition metals, such as Fe in ferredoxin, or by quinone with facilities of electron transfers. This last reaction is reversible (Fig. 3.5).
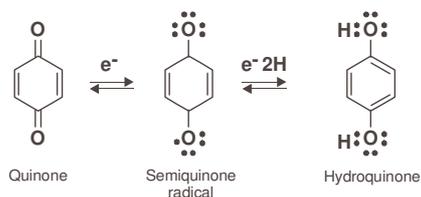
Fig. 3.5. Reduction of quinone to semiquinone radical by transfer of one electron and when one-electron reduction with protonisation with 2 protons to hydroqinone (After Edreva, 2005)

Photodamage and photoprotection exist in chloroplasts when intense light produces a flux of excess electrons, leading to the electron flow from the excited PS centres to NADP which is reduced to NADPH. This results in increased formation of ROS and, as a consequence, in damage of the photosynthetic system. In this situation, overproduction of NADPH and decreasing $NADP^+$ takes place and can interfere with the Calvin cycle (Vranova *et al*., 2002).

Highly reactive singlet electrons which are produced have a strong effect on the pigment-protein complex in chloroplasts that involves damaging the chloroplasts by the attack of double bond-containing compounds (such as unsaturated fatty acid or chlorophylls). This reaction is effected in the destruction of the chloroplast membranes and, as a consequence, may cause destructive changes in

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Calvin cycle enzymes, Fe^{+2} – containing enzymes, Mn clusters and proteins with aromatic amino acids such as tyrosine (Slooten *et al.*, 1998, Zhang *et al.*, 2003).

Table 3.3. Damaging effects of ROS on molecular targets in chloroplasts (After Edreva, 2005)

ROS	Targets
$OH\cdot$	All loci
$^1O_2$	enes, dienes: membrane lipid peroxidation, chlorophyll destruction
H_2O_2	-SH: Calvin cycle enzyme inactivation cross-linking: proteins (D_1, D_2) Fe^{2+} oxidation: metal-containing enzymes damage to Mn cluster in PSFI
$O_2^{\pm}$	enes, dienes: membrane lipid peroxidation, chlorophyll destruction aromatic amino acids: Tyosine residues destruction in D_1 protein ROS: reactive oxygen species

ROS: reactivate oxygen species

Protective mechanisms that operate in chloroplasts are both enzymatic and non-enzymatic in nature:
- ascorbate-glutathione cycle for scavenging of H_2O_2 to H_2O and O_2 without producing another ROS,
- the tioredoxin system-2 cystein peroxiredoxins-glutathione peroxidases for protecting tiol modulation of protein,
- non-enzymatic $OH^\bullet$ scavenging by ascorbate, tocopherols and glutathione,
- non-enzymatic $^1O_2$ scavenging to prevent or minimize the production of triplet chlorophyll, the source for $^1O_2$ generation by carenoids (by specificity of iso-prening chain, containing numerous double bonds), i.e delocalized π electrons:

$$- CH = C - CH = CH - \longleftrightarrow \ = CH = C = CH = CH = \qquad (3.6)$$

- formation of alternative oxidases for decreasing of ROS production. This type of protective system is going on through Q_A, Q_B, cytochromes and plastocyanins:

$$\begin{array}{c} \qquad\qquad e^- \nearrow O_2 \longrightarrow O_2^{\cdot -} \\ PSII \xrightarrow{e^-} \text{Plastoquinones} \longrightarrow PSI \\ \qquad\qquad e^- \searrow AOXs \xrightarrow{e^-} O_2 \longrightarrow H_2O \end{array} \qquad (3.7)$$
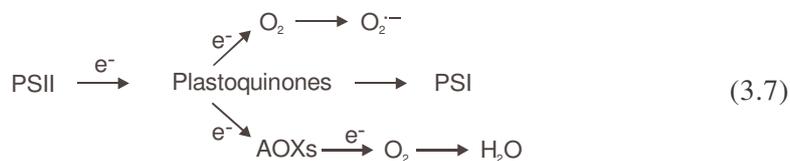
Alternative oxidases (AOX_s) can catalyse the reduction of O_2 directly to H_2O excluding the formation of partially reduced ROS.
- ROS scavenging during water-water cycle by dissipation of excess photons in chloroplasts evoked by heat or light (Asada, 1999):

$$^-2H_2O \longrightarrow 4e^- \longrightarrow PSI \longrightarrow 2e^- \longrightarrow 2O_2 \longrightarrow 2O_2^{\cdot-}$$

$$\downarrow 2e^- \qquad\qquad\qquad \downarrow SOD$$

$$DHAsc \longrightarrow Asc \quad + \quad H_2O_2 + O_2 \qquad\qquad (3.8)$$

$$2H_2O + DHAsc$$
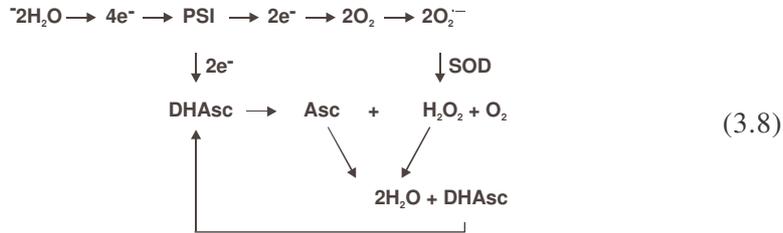
- protective strategy through photorespiration pathway by lowering O_2 concentration, production and re-assimilation of CO_2, production of H_2O_2 (accompanied by scavenging of H_2O_2 by catalase), regeneration of $NADP^+$ and recycling of phosphoglycolate to phosphoglycerate, when light energy is in excess.
- decreasing the light-driven formation of ROS by photoinhibition as a feedback mechanism by donor-side and acceptor side during overproduction of electrons. This type of strategy is effective in the early stage of photooxidative processes (Anderson *et al.*, 1997).

The schematic representation of the intermediates between water and oxygen and with related redox potentials of particular transitions are shown in Figures 3.6 and 3.7.
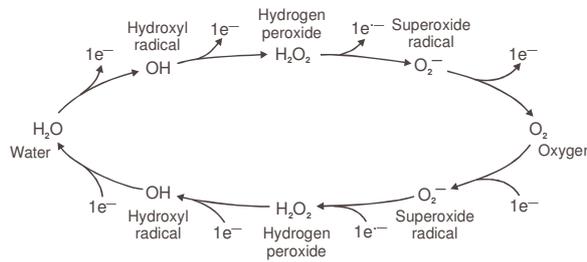
Fig. 3.6. Schematic representation of the intermediates between water and oxygen (After Bartosz, 1995)
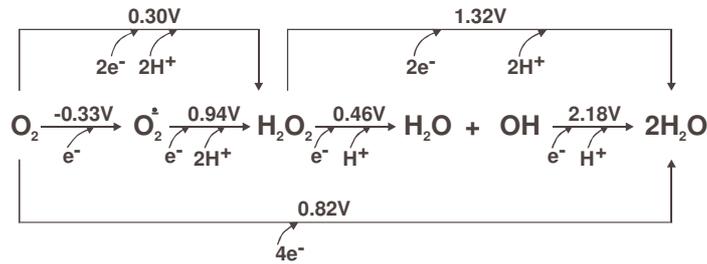
Fig. 3.7. Schematic representation of the intermediates between water and oxygen with related redox potentials of particular transitions (After Bartosz, 1995)

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4. OXYGEN CYCLE AND BALANCE

Free oxygen (O_2) is one of the most oxidizing substances known, and electron transfer from reduced substances to O_2 molecules is connected with the release, during aerobic metabolism, of large amounts of free energy which allows the elaborate structure and activity of higher organisms (Schlesinger, 1997). The major constituents of all living organisms are C, H, O, N, P (constituting 95% of the biosphere). Perhaps the most important balance in the biosphere exists between the two elements of life: carbon and oxygen. Now there is evidence that oxygen is derived from life.

The net production of O_2 over the geological time is balanced stoichiometrically by the storage of reduced organic carbon (1.56×10^{22} g) as a result of autotrophic photosyntesis and sedimentary pyrite (4.97×10^{21} g S) according to oxidation of reduced crystal minerals. For oxidation of every mol of FeS_2 nearly 2 moles of O_2 are consumed from the atmosphere, and annual burial of pyrite consumes about 20% of the oxygen in our atmosphere (Fig. 4.1).
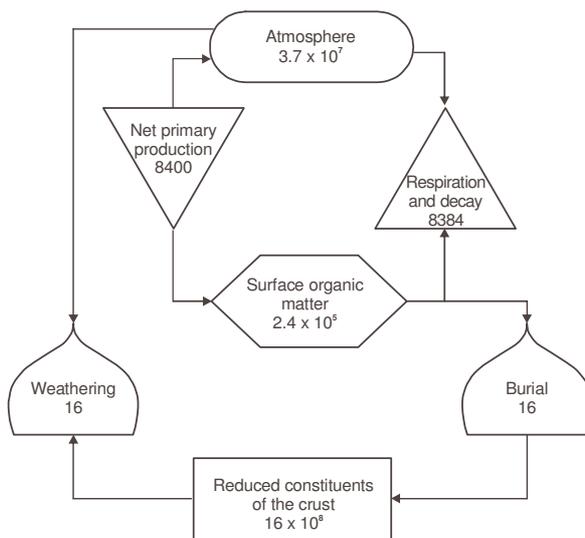
Fig. 4.1. A simple model for the global biogenochemical cycle of O_2. Data are expressed in units of 10^{12} moles of O_2 per year or the equivalent amount of reduced compounds. Note that a small misbalance in the ratio of photosynthesis to respiration can result in a net storage of reduced organic materials in the crust and an accumulation of O_2 in the atmosphere. (From Schlesinger, 1997, modified)

The atmospheric oxygen (3.7 x 10^7 units of 10^{12} moles of O_2) is only a small fraction of total oxygen production during geological time. For all organisms, large fluctuation of atmospheric oxygen would have important implications for the physiology, morphology and evolution on the world (Graham *et al.,* 1995). In our time, the pool of atmospheric oxygen is well buffered. There exists equilibrium between the consumption of O_2 during aerobic respiration (8384 units of 10^{12} moles of O_2 yr^{-1}) and the photosynthetic production (8400 units of 10^{12} moles of O_2 yr^{-1}).

Higher level of atmospheric oxygen can increase the rate of aerobic respiration in marine and oceanic sediments, and increase the adsorption of phosphorus to iron minerals in sediments, consequently lowering nutrient availability and net production of biomass in the sea (van Cappellen and Ingall, 1996). Changes in the concentration of atmospheric oxygen were noted during the past 500 million years, within the range from 15 to 35% (Lane, 2003). At the highest values of oxygen content, a large amount of organic matter was buried in sediments.

The oxygen cycle, like the carbon cycle, is mostly described on the basis of annual fluxes (Fig. 4.1). The annual fluctuation of atmospheric O_2 is about $\pm 0.0020\%$ in the average concentration of 20.946% (Keeling and Shertz, 1992). The mean time of residence for O_2 molecule in the atmosphere is about 4000 years. Annual production and consumption of oxygen is difficult to measure for the reason of the large amount of O_2 in the atmosphere as an independent check. Examination was done with isotopic $\delta^{18}O$ (+23.5%0) in mixture in atmospheric O_2. The gross primary production was established on the level > 180 x 10^{15} g C yr^{-1} on land and about 140 x 10^{15} g C yr^{-1} in the oceans. In photosynthesis process, no discrimination between oxygen isotopes of water were noted, which means that isotopic composition of released O_2 is the same in the case of plants growing in sea or in soil water. Respiration process discriminates among oxygen isotopes with preference of ^{16}O isotopes (Bender *et al*., 1994).

The oxygen cycle is directly combined with the biogeochemical nitrogen cycle. Annual circulation of N on land is assumed to be 1200 x 10^{12} g and 8000 x 10^{12} g in the oceans. About half of annual N circulation from land and about 15% from the oceans is derived by plants as nitrates and returned in ammonium form, when about 3% of annual production of photosynthesis oxygen is consumed in the nitrification reactions.

For the reason of its reactivity, oxygen is present in many geologic cycles and is included in the turnover of elements such as C, S, P, N, Mn, Fe, and others (Fig. 4.2).
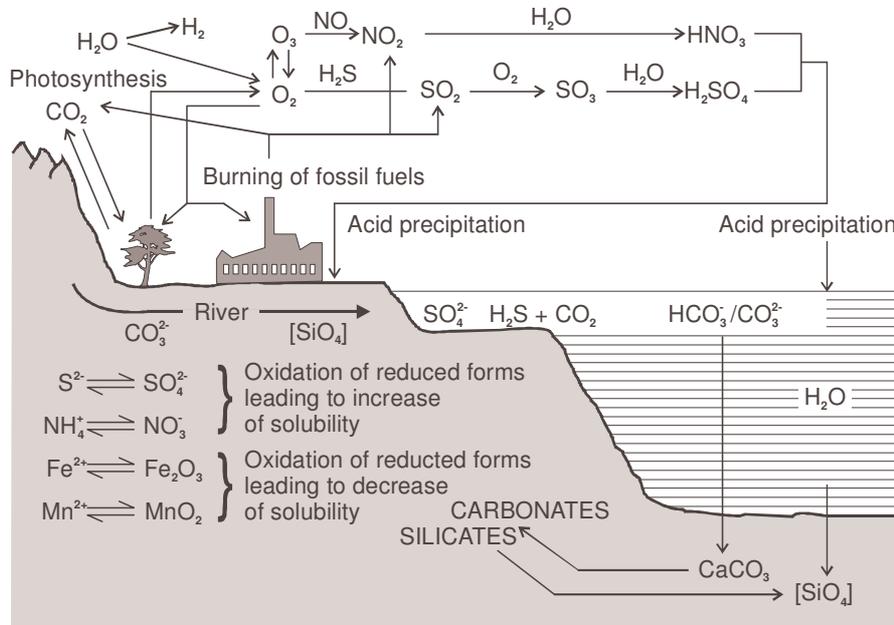
Fig. 4.2. Forms of oxygen appearance in its turnover cycle (From O'Neil *et al.*, 1993, modified)

The mayor P pool is found in soil and in unweathered rocks while the majority of nitrogen is found in the atmosphere with the turnover time of 10^7 years. The turnover of N and P is ecosystems com be substantially modified by human activity (*e.g.* application of fertilizers) what may influence their global cycling.

5. ATMOSPHERIC OXYGENOLOGY

Oxygenology of atmosphere is related to the problems of oxygen distribution, production, transport, absorption, turnover in the atmosphere and losses to the cosmic space, as well as to the formation, distribution and reactivity of ozone in the troposphere and stratosphere. Within the atmospheric oxygenology, such research areas as tropospheric oxygenology, stratospheric oxygenology etc. can be distinguished.

5.1. Structure and stratification of the atmosphere

The total mass of the atmosphere can be estimated to be 5.2×10^{21}g (Weast *et al.*, 1989); of this the mass of oxygen is estimated as 1.18×10^{21} g.

The atmospheric properties and behavior are influenced by several factors, the most important of them being the gravitation field which prevents the atmospheric gases from escaping into space. Gravity compresses the atmosphere into a shallow layer above the Earth's surface, causes stratification of mass, affects atmospheric circulation, and vertical translocation of water in the form of precipitations. Other factors affecting the atmospheric behaviour are solar radiation (affecting the photochemistry of the atmosphere), exchange of energy with the environment, transformation of one form of energy into another, Earth rotation, radiant energy transfer and properties of the particular atmospheric components themselves.

According to average temperature changes, the atmosphere is divided into several layers: troposphere, tropopause, stratosphere, stratopause, mesosphere, mesopause, thermosphere, thermopause and exosphere. Troposphere, which means a "turning sphere", is the lowest layer extending from the Earth surface to a height of about 8 km in polar regions and about 16 km in tropical regions. It is characterized by temperature decrease with altitude, the lapse rate being on average 6.5 K km^{-1}. The troposphere contains most of the atmospheric mass and the phenomena known as the weather. Tropopause is the upper boundary of the troposphere and is characterized by a minimum temperature. The atmospheric pressure in the tropopause is about 100 hPa. Above the tropopause there is the stratosphere (layered sphere) with the temperature increasing with height due to absorption of solar UV radiation by ozone. The upper boundary of the stratosphere, called the stratopause (with more or less defined temperature maximum), lies at a height of about 50 km and is characterized by pressure of about 1 hPa. The layer above the stratopause is called the mesosphere (where ozone heating diminishes), which is characterized by a decrease

of temperature with height. The second temperature minimum is reached in the mesopause at a height of about 85 km and the pressure of about 0.01 hPa. The region from the tropopause to the height of 85 km is called the middle atmosphere. Above this height, kinetic temperature increases steadily again in the thermosphere. Above the thermosphere and demarcated by the thermopause there is an essentially isothermal region called the exosphere.

The height profiles of global mean pressure, temperature, mean molar weight and density of atmosphere are presented in Figure 5.1. As it can be noticed, the global mean pressure and density decrease with height almost exponentially. Pressure decreases from about 1000 mb (10^5 Pa) at sea level to about only 10% of this value at 15 km height, which means that about 90% of the atmosphere mass is beneath this level. Pressure decreases by another factor of 10 for each additional 15 km of height. Air density decreases from the surface value of about 1.2 kg m^{-3} in the same way to the altitude of about 100 km. Due to this the mean free path of atmospheric gases increases exponentially with height from about 10^{-7}m to about one meter at 100 km. Above this level the mean free path is too high for turbulent eddies to occur and diffusive transport becomes the dominant mechanism for vertical transport. The well mixed region below 100 km is called the homosphere, the transition zone from the turbulent to diffusive transport at an average height of about 100 km is called the homopause which is known also as the turbopause. The region above the homopause and below 500 km is known as the heterosphere, in which the flow of air is nearly laminar. Molecular diffusion stratisfies the components so that concentration of the heaviest species O_2 decreases with altitude more rapidly than second heaviest species N_2. Because of this stratification the composition of the heterosphere varies with altitude, which is reflected by the decrease of the mean molecular mass. Although the diffusive separation is responsible for the heterosphere stratification, an important role is played also by photodissociation. Ultraviolet radiation dissociating molecular oxygen provides and important source of atomic oxygen at those altitudes and due to this O becomes the dominant form of oxygen not far after the homopause.

In the homosphere and heterosphere the molecules interact strongly through frequent collisions. Above the critical level of the upper heterosphere limit the molecular collisions are very rare so that a significant part of the molecules move out into space. The molecules follow ballistic trajectories and mostly are captured by the Earth's gravitational field and return to denser atmosphere along parabolic trajectories. Some of the molecules have velocities high enough to escape from the gravitation field of the Earth to the deep space and are lost. The escape velocity for the Earth is about

11 km s^{-1} and is the same for all the molecules irrespective of the molecular mass. But because of equipartition of energy among molecules, lighter molecules have higher velocities and escape into space more readily than heavier molecules. On the basis of Boltzmann distribution of molecular velocities (Salby, 1996) it was found that for the temperature of 1000 K occurring normally at the level of 500 km, for atomic oxygen the most probable velocity is 1.02 km s^{-1}, while the fraction of atoms having velocities higher than the escape velocity is very low, of about 10^{-45}. According to this calculation, the time necessary for all the O molecules initially present at a height of 500 km to escape from the Earth's gravitational field is more than 10^{46} s, which is much higher than the 4 billion years of the Earth existence.
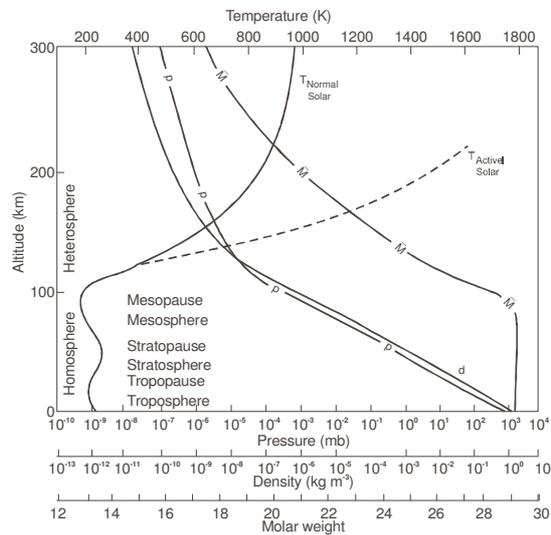
Fig. 5.1. Global – mean pressure (p), temperature (T), mean molar (M) weight and density(d) of atmosphere (source – US Standard Atmosphere, 1976)

It is obvious that the heavier molecules are captured by the Earth's gravitational field even more efficiently, while those lighter – less efficiently. In the case of molecular hydrogen, the most probable velocity is 4.08 km s^{-1} and 0.01% of molecules have a velocity higher than the escape velocity. So a significant fraction of H molecules present initially at the critical altitude of 500 km is lost during one day. It explains why hydrogen continually produced by photodissociation of water occurs in the Earth's atmosphere at very low concentrations. It should be added that under disturbed conditions the temperatures at the critical height can be substantially higher, which makes the escape of H molecules even faster (Salby, 1996).

5.2. Composition of the troposphere

The atmospheric composition inside the troposphere is presented in Table 5.1.

Table 5.1. Atmospheric composition in the troposphere with the tendencies of changes with height (Compiled from Salby, 1996, and Weast *et al.*, 1989)

Constituent	Molecular mass (kg kmol^{-1})	Tropospheric mixing ratio	Vertical distribution (mixing ratio)	Controlling processes
N_2	28.134	0.7808	Homogeneous	Vertical mixing
O_2	31.9988	0.2095	Homogeneous	Vertical mixing
H_2O[1]	18.01534	≤0.030	Decreases sharply in troposphere; increases in stratosphere; highly variable	Evaporation, condensation, transport; production by CH_4 oxidation
Ar	39.938	0.00934	Homogeneous	Vertical mixing
CO_2[1]	44.00995	345 ppmv	Homogeneous	Vertical mixing; production by surface and anthropogenic processes
Ne	20.183	18.18 ppmv	Homogenous	Veritcal mixing
O_3[1]	47.9982	10 ppmv[2]	Increases sharply in stratosphere; highly variable	Photochemical production in stratosphere; destruction at surface transport
He	4.0026	5.24 ppmv	Homogenous	Vertical mixing
CH_4[1]	16.04303	1.7 ppmv	Homogeneous in troposphere; decreases in middle atmosphere	Production by surface processes; oxidation produces H_2O
Kr	83.80	1.14 ppmv	Homogenous	Vertical mixing
H_2	2.01594	500 ppbv	Homogenous	Vertical mixing
N_2O[1]	44.1334	350 ppbv	Homogeneous in troposphere; decreases in middle atmosphere	Production by surface and anthropogenic processes; dissociation in middle atmosphere; produces NO transport
Xe	131.30	87 ppbv	Homogenous	Vertical mixing
CO[1]	28.0104	70 ppbv	Decreases in troposphere; increases in stratosphere	Anthropogenic production and by oxidation of CH_4
NO	30.0664	0.1 ppmv[2]	Increases vertically	Production by dissociation of N_2O catalytic destruction of O_3
CFC-11[1]		0.2 ppbv	Homogeneous in troposphere; decreases in stratosphere	Industrial production; mixing in troposphere; photodissociation in stratosphere
CFC-12[1]		0.3 ppbv		

[1] Radiatively active; [2] Stratospheric value

Because of its variability, the composition of the atmosphere is usually expressed as the U.S. Standard Atmosphere (1976) being an idealized, steady-state representation of the Earth atmosphere from the surface to 1000 km as it is as-

sumed to exist in a period of moderate solar activity. The air is assumed to be dry and, at heights below 86 km, to be well mixed. The mean molecular mass of the atmosphere is 28.96 till the height of 85 km and then decreases due to change of the molar composition, reaching 14.33 g mol^{-1} at 500 km height and 3.94 at 1000 km height (U.S. Standard Atmosphere, 1976).

It should be added that ozone in the troposphere is formed only under the conditions of contamination of air with NO_2, which happens mainly in the urban air. The photolysis of NO_2 leads to the formation of ozone. Normal ozone content in the troposphere due to its transport from the stratosphere is about 2 pphm (parts per hundred millions) and it constitutes about 10% of the total atmospheric ozone (Wayne, 2002). The concentrations normally found in polluted troposphere, as for example in Los Angeles smog during eye irritation, are of the order of 20 pphm. Air containing 1 ppm of ozone has a distinct odor accompanied by headache and its inhalation causes severe irritation of upper respiratory system, lungs and sometimes can cause fatal pulmonary edema. The effect of ozone is connected with generation of free radicals in tissues (Manahan, 1993).

In Poland maximum admissible concentration of ozone for 8 h exposition of humans is 110 μg m^{-3} (about 5 pphm).

5.3. Composition of the middle atmosphere

5.3.1. Molecular oxygen and oxygen containing compounds

The main oxygen species in the atmosphere are molecular oxygen O_2, atomic oxygen O and ozone O_3. The species of oxygen and of its compounds can be divided into neutral species, ionic species and radicals. The dynamics of their vertical distribution depends mainly on photochemical reactions induced by solar radiation and on vertical and horizontal transport within the atmosphere.

The vertical distribution of oxygen neutral species is shown, together with such oxygen compounds as H_2O, OH, NO and NO_2, in Figure 5.2. This figure presents a summary of the trends to be expected in normal atmosphere. However, the detailed structure of the atmosphere varies in a predictable way with latitude, time of day, season and position in the eleven-year cycle of solar activity. As it can be noticed, the concentration of molecular oxygen O_2, expressed in number of molecules per unit volume (this concentration unit as number density is used in atmospheric chemistry and physics due to temperature and pressure variations with altitude) decreases with height. The number density of atomic oxygen below the height of 50 km is lower by several orders of magnitude than that of O_2, but at the height of about 100 km the number density of atomic

oxygen reaches maximum and becomes equal to that of molecular oxygen. Above this level atomic oxygen prevails, and at the height of 140 km the number density of molecular oxygen constitutes only about one per cent of that of atomic oxygen which becomes the main constituent of the atmosphere, more abundant than total nitrogen forms.
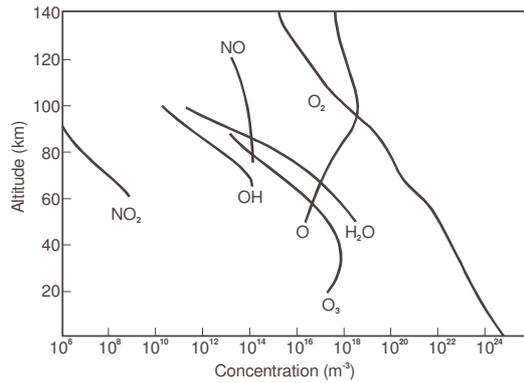
Fig. 5.2. Representative concentration profiles (in density of molecules per cubic meter) of neutral species of oxygen and of oxygen compounds in the homosphere (Modified from McEwan and Phillips, 1975)

5.3.2. Ozone

Ozone is a very important component of the stratosphere. Its abundance can be expressed in three different ways: as mixing ratio (up to 10 ppmv), as "absolute" concentration expressed as number density of molecules (up to 5×10^{18} m^{-3}), and as the total ozone or column abundance in Dobson Units (DU). One DU represents 0.01 mm ozone column under standard temperature and pressure conditions. The values of ozone column abundance range from 200 DU in the tropical zone to more than 450 DU at high latitudes, which corresponds to 2.0-4.5 mm of pure ozone layer under normal conditions (Wayne, 2002).

Ozone is formed of molecular oxygen due to the presence of UV radiation. Oxygen molecules O_2 are converted due to photodissociation into atomic oxygen:

$$O_2 + hv \rightarrow 2O \tag{5.1}$$
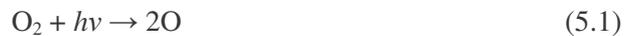
Atomic oxygen O can recombine with O_2 (in the presence of a third molecule, *e.g.* N_2, needed to carry off the excess of energy liberated by the recombination) to form ozone in the thermomolecular reaction:

$$O_2 + O + M \rightarrow 2O_3 + M \tag{5.2}$$

Ozone formed in this reaction is dissociated by UV radiation according to the reaction:

$$O_3 + h\nu \rightarrow O_2 + O \qquad (5.3)$$

In the presence of abundance of third bodies, atomic oxygen produced by the latter reaction recombines almost immediately with O_2 to form ozone. Thus, the three above reactions form a kind of closed cycle without net loss of components and with the only result in the form of absorption of solar UV radiation. This set of reactions protects the Earth's surface from UV radiation harmful for living organisms. It should be stressed that we owe this protection to the layer of 2-4.5 mm of total ozone under normal conditions!

The zonal volumetric mixing ratios of ozone are presented in Figure 5.3. It should be stressed that the mixing ratio reaches its maximum at a height of about 30 km, but due to pressure drop with altitude the highest ozone density is in the lower stratosphere at altitudes of 10-20 km. The highest mixing ratios reaching 10 ppmv occur in the tropics, characterized by the largest flux of solar UV radiation and photodissociation of O_2.
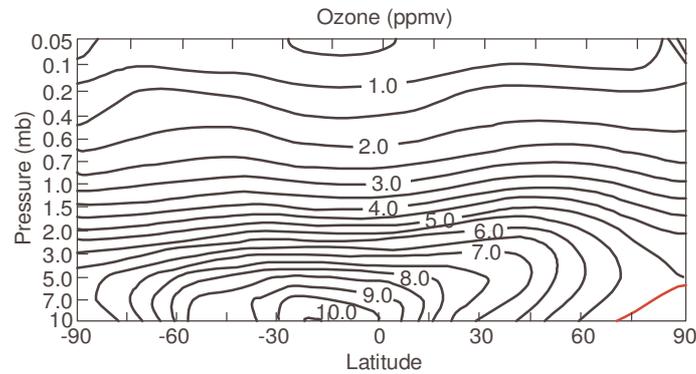
Fig. 5.3. Zonal volumetric mixing ratios of ozone (in ppmv) averaged over January-February 1979 versus latitude and pressure obtained from the Limb Infrared Monitor of the Stratosphere on board of Nimbus – 7 (Modified from Salby, 1996)

The column abundance of ozone being its integral of its absolute concentration over height depends on the solar activity. The mean column abundance of ozone versus latitude and month for the records prior to 1980 is presented in Figures 5.4 and 5.5. Values of column abundance range from 240 DU to more than 460 DU. Although most of stratospheric ozone is produced in the tropics, the

column abundance is the greatest in the middle and high latitudes. It should be added that global distribution of ozone on individual days is very dynamic and that the circulation of air plays an important role in the distribution of ozone. Ozone in the troposphere is quickly destroyed due to its high solubility in the precipitation water and subsequent involvement in numerous oxidation reactions. Thus the troposphere is a sink for stratospheric ozone.
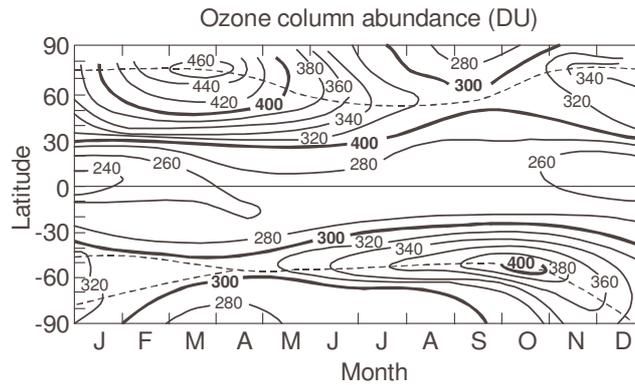
Fig. 5.4. Mean column abundance of ozone or total ozone (in Dobson Units) as a function of latitude during one year based on the records before 1980. (Modified from Salby, 1996)
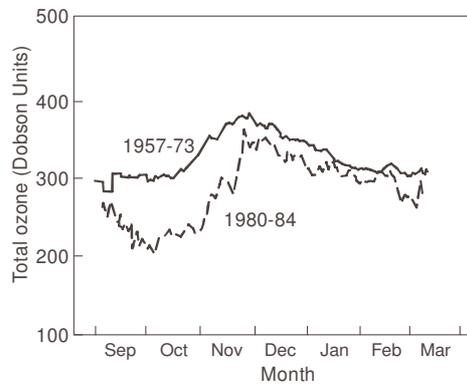
Fig. 5.5. Seasonal cycle of total ozone over Halley Bay, Antarctica, based on the historical record since 1957 and on years since the appearance of Antarctic ozone hole (After Salby 1996, modified)

It should be emphasized that ozone is catalytically destroyed by chlorine atoms produced by the decomposition of chlorofluorhydrocarbons. The following reactions then take place:

$$CFCl_3 + h\nu \rightarrow CFCl_2 + Cl \qquad\qquad (5.4)$$

$$Cl + O_3 \rightarrow ClO + O_2 \qquad\qquad (5.5)$$

$$ClO + O \rightarrow Cl + O_2 \qquad\qquad (5.6)$$

then the next reaction is:

$$O_3 + O \rightarrow 2O_2 \qquad\qquad (5.7)$$

This closed cycle leaves the same amounts of Cl + ClO, so one Cl atom can destroys many ozone molecules (Salby, 1996). The decomposition is also catalized by NO, NO_2, H^+, HO·, HOO·, Cl, Br and BrO (Bailey *et al.*, 2002, Manahan, 1993). Ozone is also decomposed on solid surfaces of metal oxides and salts produced by rocket's exhaust (Manahan, 1993)

Mixing ratios of ozone and chlorine monoxide along a flight into the Antarctic polar-night vortex are shown in Figure 5.6.
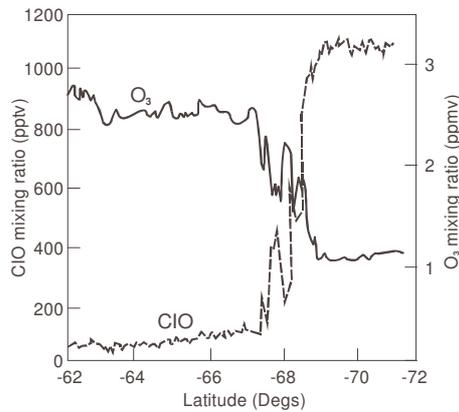
Fig. 5.6. Mixing ratios of ozone (solid line) and chlorine monoxide (dashed line) along a flight path into the Antarctic polar-night vortex. (After Salby, 1996, modified)

5.3.3. Oxygen-dependent other radiatively active trace gases

The vertical distribution of volume mixing ration of radiatively active trace gases, which are called greenhouse gases(IPPC, 1996), is shown in Figure 5.7. As it can be seen, only the mixing ratio for carbon dioxide is constant in the homosphere, while that for other gases changes with altitude.
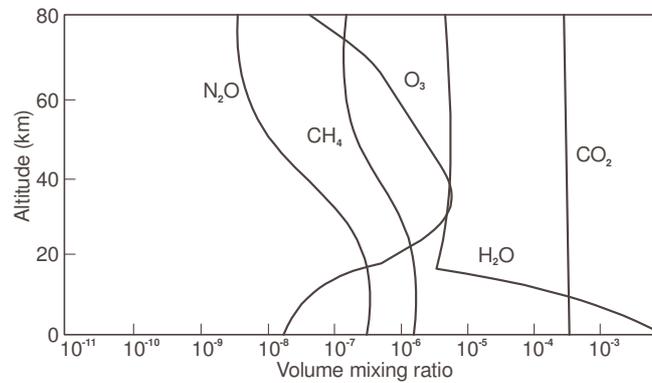
Fig. 5.7. Mixing ratios of natural radiatively active trace species as functions of altitude. (After Salby, 1996, modified)

With the exception of ozone, the mixing ratio of which shows (as described previously) a maximum at the height of about 30 km, the volume contribution of other gases decreases with altitude. In the case of water vapor, the decline takes place in the troposphere and above it the mixing ratio remains fairly constant. Methane and nitrous oxide are stable in the troposphere, and due to this their mixing ratios here remain constant and tend to decrease above it. Contribution of methane decreases in the stratosphere due to its oxidation leading to the formation of water vapour, which explains the increase of the mixing ratio of the latter in the stratosphere. The tropospheric concentration of methane has been increasing during the last several decades of years due to human activity.

Nitrous oxide N_2O (as well as nitric oxide NO) is also relevant to the photochemistry of ozone. In the stratosphere nitrous oxide decreases with altitude due to its dissociation being a primary source of stratospheric NO which, like free chorine, destroys ozone catalytically. The atmospheric nitrous oxide concentration tends to increase steadily during recent years as a result of human activity.

5. 4. Ionic oxygen species in the heterosphere and the exosphere

In the heterosphere and the exosphere there are ionic species of oxygen and other gases due to photolysis. The typical distribution of their concentration for a solar minimum during daytime is presented in Figure 5.8. As it can be noticed, above the height of 100 km the total concentration of the ionic species expressed here by the electron density increases with height, reaches a maximum (of the order of 5x 10^{11} electrons per cubic meter) at a height of about 250 km and then

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decreases again to about 10^{10} m^{-3}. The positive counterions are dominated by the O^+ ion which in the height interval from 200 to 500 km is the main positive ion. At lower heights, 100-150 km, the dominating positive ions are O_2^+ and NO^+.
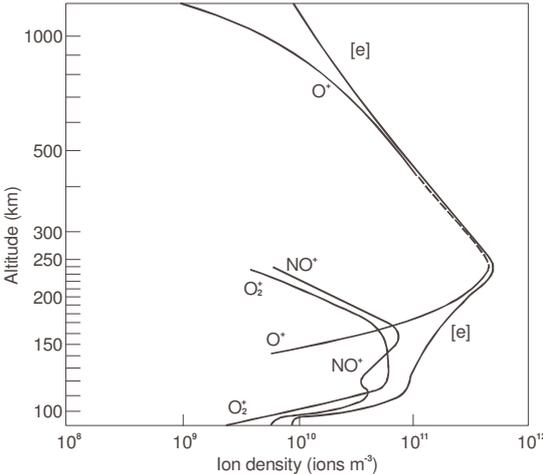
Fig. 5.8. Typical distribution of electrons and dominating positive ions in the daytime ionosphere during solar minimum (modified from McEwan and Phillips, 1975)

6. SOIL OXYGENOLOGY

Oxygenology of the Earth crust relates to the processes of distribution, supply, uptake, transport and the role of oxygen within the external part of the Earth crust and especially in the pedosphere, being its most active part. Here we can define some special research fields such as: 1° – oxygenology of wetlands (natural, agricultural and constructed); 2° – oxygenology of drylands (natural and agricultural); 3° – oxygenology of anthropogenic systems (mines, landfills, recultivated areas, waste water treatment plants, storage sites for agricultural materials etc.) (*e.g.* Kang *et al.*, 2002; Pawłowska, 1999; Pawłowska and Stępniewski, 2004; Pawłowska *et al.*, 2003; Stępniewska *et al.*, 2002; Stępniewski *et al.*, 2002; Stępniewski and Pawłowska, 1996; Stępniewski and Rożej, 2000; Stępniewski and Stępniewska, 2000; Stępniewski and Zygmunt, 2000a, b; Whalen *et al.*, 1990). This chapter is concerned with the oxygenology of pedosphere, as other aspects are beyond of the scope of this book.

6.1. Soil respiration

The main components of soil oxygen demand are the microbial respiration of soil and the respiration of plant roots. The contribution of the respiration of mezofauna as well as the contribution of chemical reactions is considered to be negligible and is usually neglected. Thus, the soil oxygen demand is used as synonymous for the soil respiration rate. Soil respiration rate is often expressed also as carbon dioxide production, as under aerobic respiration consumption of oxygen by volume is equivalent to that of carbon dioxide produced. The intensity of root respiration is usually by two orders of magnitude higher as compared to the soil microbial respiration. Thus, the roots present themselves as threads of high oxygen demand on the background of the much less active soil matrix. Soil oxygen demand decreases with soil depth because both its components, *i.e.* microbial respiration and root density (and thus root respiration), decrease with depth.

Microbial respiration of soil is affected by temperature, water content, and oxygen availability as well as the availability of organic carbon and nutrients. Microorganisms performing the mineralization processes in soil with respect to their temperature requirements are divided into cryophilic (temperature optimum $< 20^o$C), mesophilic (temperature optimum 20^oC to 40^oC) and thermophilic (temperature optimum $> 40^o$C) organisms. As soil contains a mixture of different groups, the overall microbial respiration increases 2-3 times with the increase of temperature by 10^oC until the temperatures of break of metabolic processes (about 60-70^oC).

The relationship of microbial respiration versus water content shows a maximum corresponding to optimum availability of water and oxygen. The increase of soil water content is accompanied by the increase of respiration due to increase of water availability for microbes, but after the maximum a subsequent decrease is observed due to limitation of oxygen transport within the soil. The position of the maximum depends on the porosity providing sufficient amounts of oxygen to the entire bulk of the soil. Microorganisms are divided, according to their water requirements (Gliński and Stępniewski, 1984, 1985), into hygrophilic (disappearance of activity at pore water tension of –7.1 MPa), mesophilic (disappearance of activity within the pore water pressure interval from –7.1 to –30 MPa) and xerophilic (disappearance of activity at pore water pressures < –30 MPa). The shape of the curve of oxygen uptake versus soil water content is related to the dependence of respiration on oxygen content or, more precisely, on its availability.

According to oxygen requirements, soil microorganisms are divided into obligate anoxic microorganisms (traditionally called obligate anaerobes), facultative anoxic organisms (or facultative anaerobes) and oxic organisms (traditionally called aerobes). A special group is constituted by microaerophylles with oxygen optimum of about several percent. Aerobic microorganisms utilize oxygen as a terminal acceptor of electrons from the cytochrome oxidase and as a material for biosynthesis of sterols and unsaturated fatty acids as well as for oxidation of aromatic ring. The oxygen uptake q by a single cell of an oxic microorganism is described by Michaelis-Menten equation:

$$q = q_{max} \, C/(C + K_M), \qquad\qquad (6.1)$$

where C is the concentration of oxygen, K_M is the Miachaelis constant defined as the concentration of oxygen when $q = 0.5 \, q_{max}$, and q_{max} is the maximum respiration when the reaction is not limited by oxygen availability. The first measurements of K_M performed for several strains of bacteria by Longsmuir (1954) gave the values from 1.1×10^{-8}M to 3.57×10^{-6} M, and similar values (2.7 to 4.4×10^{-6} M) were obtained with four soil suspensions by Greenwood (1962).

Soil microbial respiration rate under field conditions usually ranges form 0.1 to 10 mg $m^{-3}s^{-1}$ (Gliński and Stępniewski, 1995)

Root respiration is affected by internal factors of the plant itself such as physiological age of the root tissue, root dimensions, type of the plant and its development stage as well as by external factors such as temperature, soil moisture content, nutrient availability. It should be added that the presence of plant roots in the soil increases also microbial respiration of soil due to production of root exudates being an easily available carbon source for microorganisms. As it was already mentioned

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respiration rate of the root tissues is about two orders of magnitude of that of the soil itself and ranges from 10 to 50 mg m^{-3}s^{-1} (Gliński and Stępniewski, 1995). Thus the presence of plant roots in soil can double the respiration of the soil.

Respiration of a cropped field is a sum of microbial respiration of the soil and of the respiration of the plant roots. Oxygen uptake under field conditions is of order of several tons of oxygen per hectare and year (Gliński and Stępniewski, 1985). Diurnal dynamic of soil respiration under field conditions (Fig. 6.1) shows a maximum respiration in the afternoon and minimum in the morning before the sun rise. The annual dynamics of soil respiration under field conditions of a moderate climate zone is characterized by a maximum in summer time (Fig. 6.2). In the later case the presence of plant canopy elevated the respiration rate by more than by hundred per cent.
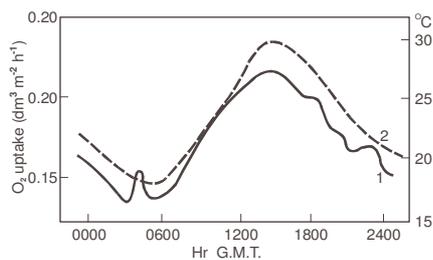
Fig. 6.1. Diurnal dynamics of soil respiration rate as measured by oxygen uptake rate (Modified from Currie, 1975)
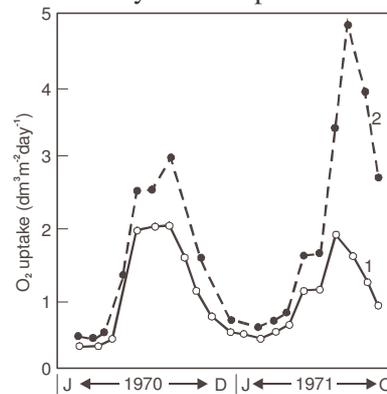
Fig. 6.2. Annual dynamics of soil respiration as measured by oxygen uptake rate in a uncropped (1) and cropped (2) soil under moderate climate conditions of England (Modified from Currie, 1975)

6.2. Gas transport in soil

The exchange of considerable amounts of gases (several to several tens mega grams per hectare) between the soil and the atmosphere takes place due to pressure gradient (mass flow) and the concentration gradient (diffusion flow). Both these kinds of flow may take place in the soil pores as well as in the plant tissues (internal gas exchange). The relative contribution of the two fundamental mechanisms of gas exchange, as well as that of the two flow pathways (*i.e.*, through the soil and through the plant), may vary within broad limits. Thus the contribution of the mass flow is important for the soils with very deep ground water table (of the order of several meters)

due to atmospheric pressure fluctuations, as well as for landfills due to both atmospheric pressure fluctuations and to pressure gradients induced by the biogas formation. The diffusive gas flow through the soil is of primary significance for mesophytes, while that through the plant tissues for the aquatic species.

6.2.1. Mass flow

The flow of gas through a porous medium due to pressure gradient may be of a laminar or a turbulent character. Under natural soil conditions, we deal usually with laminar flow (characterized by the Reynold's number Re less than 1; Currie, 1970). In the landfill cover soil the flow may be often of turbulent character due to higher pressure gradients.

Laminar flow of gases within a porous medium is described by Darcy's equation:

$$\frac{dV}{dt} = -A\frac{k}{\eta}\frac{dp}{dx} \qquad (6.2)$$

where: dV/dt = volumetric rate of gas flow, m^3s^{-1}, A= cross-section area of the porous medium under consideration, m^2; η = dynamic viscosity of the gas, $kg\ m^{-1}s^{-1}$; dp/dx = pressure gradient, $Pa\ m^{-1} = kg\ m^{-2}s^{-2}$; k = gas permeability, m^2.

Factors likely to induce the mass flow in soil are: variations in soil temperature, fluctuations of atmospheric pressure, changes of soil water content and wind action. Of these factors, the variation in the atmospheric pressure can play an important role, resulting in similar variation in the pressure of the soil air. McCool and Bouyoucos (quoted after Currie, 1970), as early as 1924, noted, by placing a barometer in the soil at a depth of 3 m, that pressure changes of the soil air followed those in the atmosphere without any visible delay.

Since the day-night changes in the atmospheric pressure reach no more than 4%, the amount of air exchanged in the soil profile does not exceed 4% of the volume of air contained in the soil. This means that in a profile of uniform porosity, underlain by an impervious horizon (ground water table, compacted layer, bedrock) at a depth of, *e.g.*, 1.5 m, only a thin 6 cm surface layer is involved in the exchange with fresh atmospheric air. The depth of this layer is frequently less due to the decrease in the air-filled porosity with the depth of the soil profile. In the case of soils in which the limitation of air-filled porosity occurs at considerable depths (*e.g.*, soils derived from loess with ground water at a depth of more than 10 m), the aeration of the profile due to variations in the atmospheric pressure may reach much deeper layers, even down to several tens of centimetres, which is significant for the root systems of most plants.

The values of air permeability in soil are within the range 0.01-500 x 10^{-12} m^2. They depend on the content of air-filled pores, on their diameter, continuity and tortuosity which is affected by soil water content and bulk density. The air permeability increases with soil water potential and with air filled porosity of the soil, as illustrated in Figs 6.3 and 6.4. The threshold value of pore water pressure at which the porous body becomes permeable to air is called the water entry pressure.
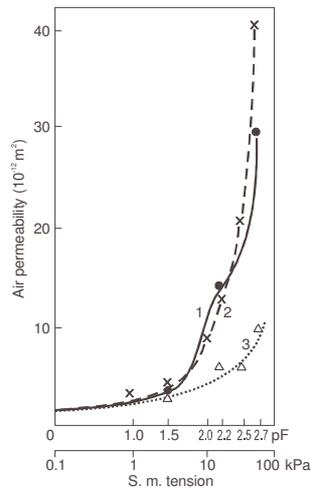
Fig. 6.3. Air permeability of a sandy loam Cambisol of different bulk densities (1 – 1.09 Mg m^{-3}, 2 – 1.21 Mg m^{-3}, and 3 – 1.41 Mg m^{-3}) versus soil moisture tension (from data of Turski *et al.*, 1978)
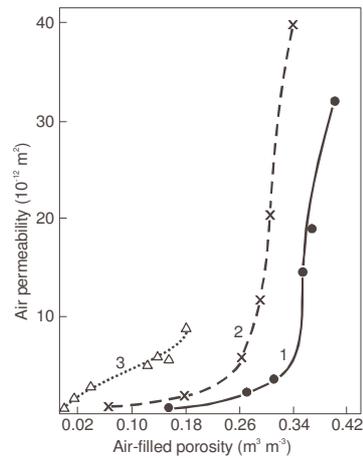
Fig. 6.4. Air permeability of a sandy loam Cambisol of different bulk densities (1 – 1.09 Mg m^{-3}, 2 – 1.21 Mg m^{-3}, and 3 – 1.41 Mg m^{-3}) versus air-filled porosity of the soil (from data of Turski *et al.*, 1978)

6.2.2. Diffusion

The basic mechanism of gas exchange in the soil is the concentration diffusion. This is a continuous process of gas transport in the direction of decreasing concentration.

Diffusion flow, unlike mass flow, is not dependent on the presence of external factors, but it is induced solely by the gradient of concentration being a result of respiration processes.

In considering the process of gas diffusion in the soil it is necessary to distinguish macrodiffusion (*i.e.*, the diffusion in the whole soil profile disregarding the heterogeneity of soil in the microscale), the diffusion in particular crumbs, micro-

diffusion through water films surrounding plant roots, and diffusion through the plants themselves.

6.2.2.1. Principles of gas diffusion in porous media

The diffusion flow (f_x) of any diffusing agent through a unit surface area of the medium in which the diffusion takes place, in a unit of time, in a uniaxial system, according of the first Fick's low, is proportional to the gradient of concentration (dC/dx) constituting the driving force of the flow, and to the diffusion coefficient D (the dimensions of which are the square of length per unit of time) characterizing the mobility of the agent diffusing in a given medium:

$$fx = - D \, dC/dx \qquad (6.3)$$

Oxygen concentration C in Eq. (6.3), applied to oxygen diffusion in soil, refers to the unit soil air volume. It is in this sense that the symbol C will be used throughout the present text. The coefficient D is related to the soil as a whole (and not to the pores filled with air), and the flow f_x is related to a unit surface area of the soil. This is emphasized in order to avoid confusion when comparing publications by various authors. If, in Eq. (6.3), the oxygen concentration in soil air C is replaced by the content of oxygen in the soil unit volume G, we get:

$$f_x = -D_G \frac{dG}{dx} \qquad (6.4)$$

where:

$$G = C(E_g + \alpha_B \theta) \qquad (6.5)$$

and, E_g and θ are the contents of air and water, respectively, expressed as fractions of the soil volume, and α_B is Bunsen's coefficient of solubility.

Coefficient D_G (defined by Eq. (6.4) is related to coefficient D (defined by Eq. (6.3) as follows:

$$D_G = \frac{D}{E_g + \alpha_B \theta} \qquad (6.6)$$

In the case of a dry soil or of a soil fully saturated with water, we have:

$$D_G = \frac{D}{E_O} \qquad (6.7)$$

where E_o is total porosity of the soil. In this case D_G is the coefficient of diffusion in soil pores only.

In a soil partly saturated with water, for gases of low solubility in water (*e.g.*, O_2), an approximated relationship can be applied with the exception of very high moisture contents:

$$D_G \approx \frac{D}{E_g} = D^*$$
(6.8)

where: D^* (frequently called the apparent diffusivity) has the physical sense of the diffusion coefficient in air-filled pores alone.

The concentration of a gas in water C_θ is related to its partial pressure P by the Henry's law:

$$C_\theta = \alpha_w P$$
(6.9)

where: a_w is solubility coefficient of the gas in water.

The relationship between the gas concentration in air (C) and its partial pressure is similar in form:

$$C = \alpha_a P$$
(6.10)

where: a_a means "solubility" in the gaseous phase, which is known to be 1/RT, where R = universal gas constant and T = absolute temperature.

Combining Eq. (6.3) with Henry's law, we obtain the formula for diffusion flow expressed by means of the gradient of partial pressure:

$$f_x = -K \frac{dP}{dx}$$
(6.11)

where K is known as the diffusion constant related to diffusion coefficient as follows:

$$K = \alpha_a D \frac{E_g + \alpha_B \theta}{E_O}$$
(6.12)

Equations (6.3) and (6.11) are two forms of Fick's first law which is analogous to Darcy's law describing the flow of liquids and gases under the effect of the pressure gradient, or to Fourier's law describing the conduction of heat due to temperature gradient.

The diffusion constant of gas in air, K_o, is, therefore:

$$K_o = \alpha_a \, D_o \qquad (6.13)$$

and the diffusion constant in water, K_w, is:

$$K_w = \alpha_w \, D_w \qquad (6.14)$$

where D_o and D_w, and α_a and α_w are the coefficients of diffusion and solubility of the considered gas, in air and water, respectively. By combining Eqs (6.13) and (6.14) we get:

$$K_w = \frac{D_w \, \alpha_w}{D_o \, \alpha_o} \; K_o \qquad (6.15)$$

and, having considered that:

$$\frac{\alpha_w}{\alpha_a} = \alpha_B \qquad (6.16)$$

we obtain:

$$K_w = \alpha_B \, \frac{D_w}{D_o} \, K_o \qquad (6.17)$$

As can be seen from Eq. (6.11), the gas diffusion in water, at the same gradient of partial pressure, takes place $a_B D_w / D_o$ times slower than in air.

In the case of oxygen it is approximately 300,000 times slower, and in the case of carbon dioxide it is about 10,000 times slower.

The dependence of diffusion coefficients on temperature and pressure is described by the following equation:

$$D_{T2P2} = D_{T1P1} \, \frac{P_1}{P_2} \, (\frac{T_2}{T_1})^{1.5} \qquad (6.18)$$

where D_{T1P1} and D_{T2P2} = diffusion coefficients at pressure and temperature values indicated by the subscripts.

The conservation law says that the change in the quantity of gas ∂G diffusing in a unit of soil volume during time ∂t is equal to the change taking place in flow ∂f_x on the diffusion path ∂x minus the consumption (*e.g.* O_2) or plus the production (*e.g.*, CO_2, C_2H_4) of the gas q in the soil, according to the equation:

$$\frac{\partial G}{\partial t} = -\frac{\partial f_x}{\partial x} \pm q \tag{6.19}$$

Taking into account that the amount of gas G in a unit soil volume is equal to the sum of its contents in the liquid and gaseous phases of the soil, *i.e.*, $G = C\,(E_g + \alpha_B\theta)$, and combining Eqs (6.19) and (6.3), we obtain:

$$\frac{\partial\left[C(E_g + \alpha_B\theta)\right]}{\partial t} = \frac{\partial\,(D\frac{\partial C}{\partial x})}{\partial x} \pm q \tag{6.20}$$

where C = concentration of the diffusing component in the soil air, α_B — Bunsen's solubility coefficient of the gas, θ — soil moisture content, x — diffusion path, q — respiratory activity related to the soil volume unit, and E_g — air-filled porosity of the soil.

In the case of gases which are not soluble in the soil solution ($\alpha_B = 0$), Eq. (6.20) simplifies to:

$$\frac{\partial(C \cdot E_g)}{\partial t} = \frac{\partial\,(D\frac{\partial C}{\partial x})}{\partial x} \pm q \tag{6.21}$$

In a soil profile all the parameters considered may be affected both by the depth (x) and time (t), which results from the heterogeneity of the profile and from the variability of the properties of the soil with time. If we express this effect mathematically, we obtain:

$$\frac{\partial}{\partial t}\left\{C\left[E_g(\theta,t,x) + \alpha_B\,\theta(t,x)\right]\right\} = \frac{\partial}{\partial x}\left[D(x,t,\theta)\frac{\partial C}{\partial x}\right] \pm q(x,t,\theta,C) \tag{6.22}$$

This general diffusion equation is the starting point for the consideration of specific situations.

If D, E, q, and θ are constant, what happens *e.g.*, in a particular horizon of the soil profile, it is possible to apply a simplified equation. If we consider that D (within the range of applicability of Fick's law) does not depend on the concentration of gas, and if we neglect the solubility of the gas in the liquid phase of the soil (which is possible, *e.g.*, for oxygen, for which at a temperature of 20°C the value of α_B is 0.033), this equation will be simplified to the form:

$$\frac{\partial C}{\partial t} = \frac{D}{E_g} \frac{\partial^2 C}{\partial x^2} \pm \frac{q}{E_g} \qquad (6.23)$$

or

$$\frac{\partial C}{\partial t} = D^* \frac{\partial^2 C}{\partial x^2} \pm q^* \qquad (6.24)$$

where $D^* = D/E_g$ and q^* — q/E_g are apparent diffusivity and respiration, respectively. For spatial diffusion of oxygen this equation, in any system of coordinates, has the form:

$$\frac{\partial C}{\partial t} = \frac{D}{E_g} \nabla^2 C \pm q^* \qquad (6.25)$$

where $\nabla^2 = \mathrm{div\ grad}$.

In the perpendicular system of coordinates x, y, z:

$$\nabla^2 = \frac{\partial^2}{\partial x^2} + \frac{\partial^2}{\partial y^2} + \frac{\partial^2}{\partial z^2} \qquad (6.26)$$

while in a spherical system of coordinates r, θ, φ:

$$\nabla^2 = \frac{1}{r^2} \frac{\partial}{\partial r}\left(r^2 \frac{\partial}{\partial r}\right) + \frac{1}{r^2 \sin\theta} \frac{\partial}{\partial \theta}\left(\sin\theta \frac{\partial}{\partial \theta}\right) + \frac{1}{r^2 \sin^2\theta} \frac{\partial^2}{\partial \varphi^2} \qquad (6.27)$$

where the relationship between the perpendicular and spherical coordinates is given as:

$$\begin{aligned} x &= r\sin\theta\cos\varphi \\ y &= r\sin\theta\sin\varphi \\ z &= r\cos\varphi \end{aligned} \qquad (6.28)$$

For diffusion in a cylindrical system of coordinates ρ, φ, z, Eq. (6.26) has the form:

$$\nabla^2 = \frac{1}{\rho} \frac{\partial}{\partial \rho}\left(\rho \frac{\partial}{\partial \rho}\right) + \frac{1}{\rho^2} \frac{\partial^2}{\partial \varphi^2} + \frac{\partial^2}{\partial z^2} \qquad (6.29)$$

where the relationship between the coordinates is given as:

$$x = \rho \cos \varphi$$
$$y = \rho \sin \varphi \qquad (6.30)$$
$$z = z$$

Under steady state conditions, Eqs (6.23) and (6.24) assume the form of Poisson's equation for uniaxial diffusion:

$$\frac{\partial^2 C}{\partial x^2} = \pm \frac{q}{D} = \pm \frac{q^*}{D^*} \qquad (6.31)$$

In the absence of sources or sinks of the diffusing gas (*e.g.*, no respiratory activity, *i.e.*, q = 0), Eq. (6.23) is reduced to the form:

$$\frac{\partial C}{\partial t} = \frac{D}{E_g} \frac{\partial^2 C}{\partial x^2} \qquad (6.32)$$

which is known as Fick's second law and in the case of a steady state diffusion (dC/dt = 0), to an equation of the Laplace type:

$$\frac{\partial^2 C}{\partial x^2} = 0 \qquad (6.33)$$

6.2.2.2. Macrodiffusion (Diffusion within the soil profile)

Gas diffusion coefficient in soil

Coefficient of gas diffusion in soil is lower as compared to that in free atmosphere. It depends on the kind of gas, its pressure and temperature, and on the amount of air-filled pores, and also on their continuity and shape. These in turn are determined by spatial arrangement of soil particles and by moisture content.

Usually, the diffusive properties of a soil medium are characterized by means of the relative diffusion coefficient D/D_o, where D = gas diffusion coefficient in soil and D_o = diffusion coefficient of the same gas in atmospheric air, under the same pressure and temperature conditions. This way of expressing the gas diffusion coefficient in soil is convenient as its value does not depend on the temperature, pressure, nor the kind of the diffusing gas.

The dependence of D/D_o on soil moisture tension and compaction is presented in Figure 6.5. The D/D_o value in soil is usually below 0.2 and it increases,

in a curvilinear manner, with increase in the soil moisture tension, and decreases rapidly with soil compaction level.
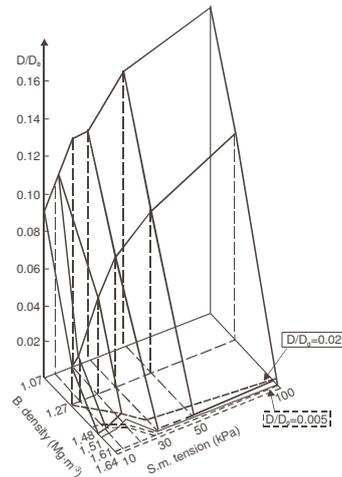
Fig. 6.5. Dependence of relative gas diffusion coefficient in a loamy textured Phaeosem on soil moisture tension and bulk density (Modified from Stępniewski, 1981)

The gas diffusion coefficient in a porous medium like soil does not depend on the size of the capillaries or pores (unlike to the air permeability which is related to the square of the pore radius), provided that the pore diameters are greater than the mean free path of the molecules. As long as this condition is satisfied, the effects of the collision of gas molecules with the capillary walls have no real significance. It can be assumed that this limit is reached at pore diameters of 0.10 μm. In the case of soil, pores of that size are emptied of water at soil moisture tension levels over 3 MPa (pF > 4.5), *i.e.*, at moisture contents below the wilting point. Thus we can conclude that in aeration macro pores (above 30 μm) as well in the mezopores (30-0.2 μm) containing usually plant available water, the pore size does not affect the diffusion rate.

Therefore, it is the gradual transition from normal diffusion to diffusion of rarefied gas that seems to be the main reason for the decrease in the diffusive effectiveness of small pores which become emptied of water at high values of the soil moisture tension.

The relationship of D/D_o to E_g, is usually curvilinear (Fig. 6.6) and it is described by an empirical power model in the following form:

$$\frac{D}{D_o} = \gamma E_g^{\mu} \tag{6.34}$$

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where γ and μ are empirical coefficients characterizing the material under investigation.
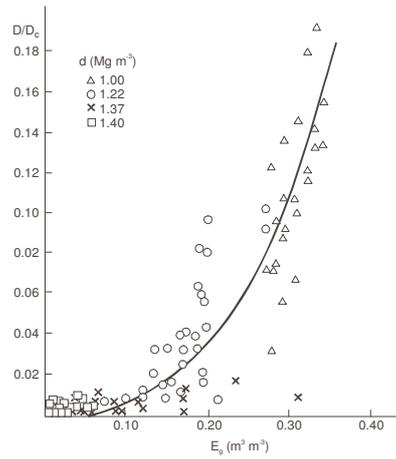
Fig. 6.6. Relationship of D/D_o to air-filled porosity E_g, of silt Fluvisol at different bulk densities (Modified from Stępniewski, 1981)

Calculated oxygen distribution in soil

Solution of the diffusion equations for oxygen under steady state conditions gives, in the simplest case of a homegenous monolayer profile with constant q and D, the following equation for oxygen distribution (C) with depth x:

$$C = C_o - \frac{q\,(2Lx - x^2)}{2D} \qquad (6.35)$$

where: C_o denotes oxygen concentration at soil surface, and L is the depth of the biologically active layer (Gliński and Stępniewski, 1985; Kowalik, 1973).

Oxygen concentration at the lower boundary of the biologically active layer C_L, that is for x = L, can be calculated from the equation:

$$C_L = C_o - \frac{qL^2}{2D} \qquad (6.36)$$

From this equation we can calculate the critical value of respiratory activity q' at which the oxygen concentration at the lower boundary of the biologically active layer reaches a minimum value C' limiting the activity of microorganisms or of plant roots:

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$$q' = \frac{2D(C_o - C')}{L^2} \qquad (6.37)$$

Moreover, we can calculate the critical value of the diffusion coefficient D' which ensures oxygen concentrations above C' in the whole profile down to depth L:

$$D' = \frac{qL^2}{2(C_o - C')} \qquad (6.38)$$

If the desirable minimum oxygen concentration C' at a depth L is zero (eg. for beginning of anoxic processes), then Eqs (6.37) and (6.38) are simplified to the formulas:

$$q'_{max} = \frac{2C_o D}{L^2} \qquad (6.39)$$

and

$$D'_{min} = \frac{qL^2}{2C_o} \qquad (6.40)$$

where: q'_{max} is the maximum respiration activity allowing for the presence of oxygen in the entire biologically active layer till the depth L at the diffusion coefficient D, and D'_{min} is the maximum gas diffusion coefficient allowing for the presence of oxygen in the entire biologically active layer till the depth L at a given respiratory activity q.

In the case when not all the biologically active profile is oxygenated ($q > q'_{max}$), the depth of the oxic zone L_{an} (identical with the depth of the anoxic zone) is given by the formula:

$$L_{an} = \sqrt{\frac{2C_o D}{q}} \qquad (6.41)$$

The values of the oxygenation depth calculated from Eq. (6.41) (Gliński and Stępniewski, 1985) for respiration values normally occurring in soil (0.1 to 10 mg $m^{-3}s^{-1}$), and for the diffusion coefficient/constants likely to occur in soil, range from less than one millimeter (for compacted soils saturated with water, characterized by very high respiratory activities) to several meters (for dry loose soils of low respiratory activities).

Three examples of oxygen concentration distribution in soil calculated form Eq. 6.35 are presented in Figure 6.7.
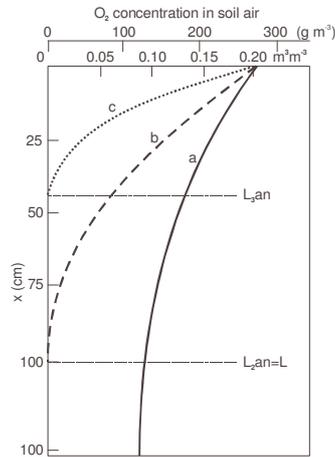
Fig. 6.7. Oxygen distribution in a homogenous soil profile of respiratory activity q = 0.6 mg m^{-3} s^{-1} at different oxygen diffusion coefficient D values: (a) D = 2 x 10^{-6} m^2 s^{-1} > D'$_{min}$; (b) D = 1 x 10^{-6} m^2 s^{-1} = D'$_{min}$; (c) D = 0.2 x 10^{-6} m^2 s^{-1} < D'$_{min}$. (From Stępniewski, 1975)

Curve "a" presents a situation in which oxygen diffusion coefficient is higher than the critical value. In other words, it means that the whole profile is oxygenated and the oxygen concentration at the bottom of the biologically active zone is above zero.

Curve "b" illustrates the case when oxygen concentration decreases to zero at the bottom of the biologically active zone. Diffusion coefficient equals to the critical value D'$_{min}$.

Curve "c" shows a situation when only part of the biologically active zone is oxygenated. The oxygen diffusion coefficient is below the critical value D'$_{min}$.

6.2.2.3. Diffusion within soil aggregates

The first to consider the problems of the oxygenation of aggregates were Currie (1961) and Greenwood (1961).

The equation of diffusion Eq. (6.26) in the case of a dry crumb spherical in shape, assuming it is isotropic, *i.e.*, the diffusion coefficient within the aggregate D$_c$ and its respiratory activity q are uniform in all directions within the internal space of the aggregate, has the form:

$$E_g \frac{\partial C}{\partial t} = \frac{1}{r^2} \frac{\partial}{\partial r}(D_c r^2 \frac{\partial C}{\partial r}) + q \qquad (6.42)$$

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where r = distance from the center of the aggregate and E_g, C, t, q, and D_c are the air-filled porosity, oxygen concentration in the gaseous phase, time, respiratory activity of the aggregate, and diffusion coefficient in the aggregate, respectively.

The solution of this equation, under steady-state conditions and with q constant in time (Gliński and Stępniewski, 1985), has the form:

$$C_r = C_R - \frac{q}{6D_c}(R^2 - r^2)$$ (6.43)

where R = aggregate radius, C_r = oxygen concentration at a distance r from the center of the aggregate, and C_R = oxygen concentration on the surface of the aggregate.

This equation permits the calculation of critical oxygen concentration on the surface of the aggregate C_R at which C_r at the center of the aggregate drops to zero:

$$C_R' = \frac{qR^2}{6D_c}$$ (6.44)

and of the critical value of aggregate radius R' for given values of D_c, C_R, and q:

$$R' = \sqrt{\frac{6C_R D_c}{q}}$$ (6.45)

The value R' determines the radius of the biggest aggregate that can still be fully oxygenated under these conditions.

In the case when the inner part of the aggregate of a radius r_{an} is anoxic, i.e., $C_r = 0$ for $0 < r < r_{an}$, then the oxygen concentration distribution in the outer, oxygenated part of the aggregate, i.e., in the range $r_{an} < r < R$, is described by the equation:

$$C_r = C_R - \frac{q}{6D_c}\left[R^2 - r^2 - 2r_{an}^3(\frac{1}{r} - \frac{1}{R})\right]$$ (6.46)

Substituting $C_r = 0$ for $r = r_{an}$, we get:

$$\frac{6C_R D_c}{q} = R^2 - 3r_{an}^3 + \frac{2r_{an}^3}{R}$$ (6.47)

Equations (6.43) and (6.46) have been verified experimentally by Greenwood and Goodman (1967) through direct measurements of oxygen concentration in soil aggregates. The agreement obtained was satisfactory.

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Oxygen concentration in water and air, its solubility and diffusion characteristics are shown in Tables 6.1 and 6.2 and in Figure 6.8.

Table 6.1. Oxygen concentration at NP and different temperatures and partial pressure in water saturated atmospheric air and in water equilibrated with air (From Gliński and Stępniewski, 1985)

Temp. °C	Partial pressure in moist air kPa	Conc. in moist air			Conc. in water	
		$m^3\,m^{-3}$	$g\,m^{-3}$	$mol\,m^{-3}$	$g\,m^{-3}$	$mol\,m^{-3}$
0	21.10	0.2082	297.5	9.30	14.6	0.456
5	21.03	0.2076	291.3	9.09	12.6	0.397
10	20.97	0.2070	285.4	8.92	11.3	0.352
15	20.86	0.2060	279.0	8.72	10.0	0.315
20	20.74	0.2047	272.5	8.52	9.13	0.284
25	20.56	0.2030	265.7	8.30	8.22	0.257
30	20.33	0.2007	258.4	8.16	7.52	0.233

Table 6.2. Solubility coefficient, diffusion coefficients, and diffusion constants of oxygen in air and water, at different temperatures (From Gliński and Stępniewski, 1985)

Temp. °C	Solubility coeff.					Diffusion coeff.		Diffusion constants	
	In air at NP		In water			In air	In water	In air	In water
	α_a		α_w		α_B	D_o	D_w	K_o	K_w
	$\times 10^{-2}\,g$ $m^{-3}\,Pa^{-1}$	$\times 10^{-4}\,mol$ $m^{-3}\,Pa^{-1}$	$\times 10^{-2}\,g$ $m^{-3}\,Pa^{-1}$	$\times 10^{-4}\,mol$ $m^{-3}\,Pa^{-1}$	$m^3\,m^{-3}$	$\times 10^{-5}$ $m^2\,s^{-1}$	$\times 10^{-9}$ $m^2\,s^{-1}$	$\times 10^{-9}\,mol$ $m^{-1}\,s^{-1}\,Pa^{-1}$	$\times 10^{-13}\,mol$ $m^{-1}\,s^{-1}\,Pa^{-1}$
0	1.95	4.43	3.34	7.59	1.713	1.38	1.15	6.11	8.73
5	1.92	4.35	2.78	6.30	1.450	1.43	1.30	6.22	8.20
10	1.88	4.28	2.33	5.29	1.238	1.48	1.46	6.33	7.72
15	1.85	4.20	1.99	4.51	1.075	1.53	1.63	6.43	7.35
20	1.82	4.13	1.71	3.89	0.942	1.59	1.77	6.57	6.89
25	1.79	4.06	1.48	3.36	0.829	164	1.92	6.66	6.45
30	1.76	4.00	1.30	2.95	0.738	1.70	2.08	6.80	6.14

$\alpha_B = \alpha_w/\alpha_a$ (Bunsen's absorption coeff. = gas solubility per unit vol. at a given temp. and NP (normal pressure) = 101.3 kPa
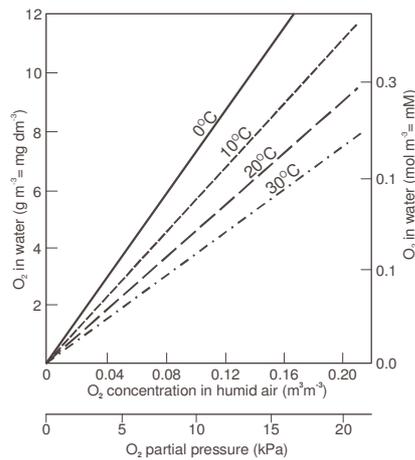
Fig. 6.8. Equilibrium oxygen concentration in water as a function of oxygen concentration in air saturated with water vapour, at normal pressure (101.3 kPa), at different temperatures (From Gliński and Stępniewski, 1985)

6.2.2.4. Microdiffusion

The concentration of oxygen in the soil is insufficient to describe plant reaction as its availability to plants is dependent on other factors.

In 1952 Lemon and Erickson (1952) proposed an indirect measure of the potential availability of oxygen for plant roots. They designed an electrode simulating oxygen uptake by plant roots. The method consists in the measurement of the amount of oxygen diffusing to the surface of a platinum electrode where it is reduced electrochemically. The platinum wire is therefore a model of a root absorbing oxygen, and the intensity of oxygen flux to the electrode indicates the maximum amount of oxygen that would be available for a root placed in the same spot as the electrode.

The value of the oxygen reduction current on the Pt electrode in a situation where the intensity of that current depends only on the rate of oxygen diffusion to the electrode surface from the surrounding medium is expressed as oxygen diffusion rate (ODR):

$$ODR\,[\mu g\,m^{-2}\,s^{-1}] = 8.29 \cdot 10^{-5}\,\frac{i}{A}\,\frac{[\mu A]}{[mm^2]} \tag{6.48}$$

where: i = current in μA, A = area of the electrode in mm^2.

The moisture content of the soil determines the effective thickness of the water film surrounding the root, and in the combination with the structural characteristics of the soil, the value of the effective diffusion coefficient in the film. This in turn, together with oxygen concentration in the soil air, determines the value of ODR which affects the plants. The ODR values in soils vary, most frequently, within the range from 0 to 200 $\mu g\,m^{-2}\,s^{-1}$.
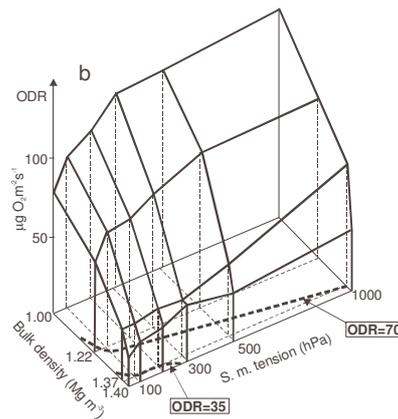
Fig. 6.9. ODR dependence on bulk density and moisture tension of heavy Fluvisol

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They decrease with an increase in soil moisture content, and therefore increase with an increase in the moisture tension and air-filled porosity of the soil. They also decrease with an increase in the soil bulk density, which is shown in Figure 6.9.

6.3. Oxygen distribution in soil air

Oxygen distribution in a real soil is the resultant effect of oxygen demand and of the gas transport processes described above. An example of oxygen distribution within a wetland soil is presented in Figure 6.10.
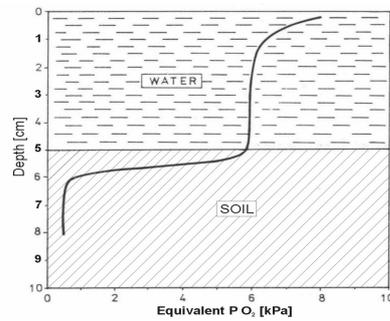
Fig. 6.10. Oxygen distribution in a flooded soil (Modified from Patrick and Mikkelsen, 1971)

As it can be noticed, oxygen concentration drops rapidly to zero at a depth of one centimetre below the soil-water interface. An example of oxygen distribution in a meadow soil is presented in Fig. 6.11.
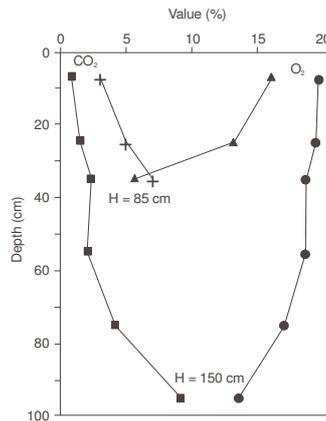
Fig. 6.11. Oxygen and carbon dioxide distribution in a gley meadow soil (Garbów, near Lublin, Poland) at two groundwater levels on 1971.06.19 (ground water level H = 85 cm) and on 1971.09.04 (H = 150 cm) (Unpublished data of Stępniewski)

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As we can notice, the sum of oxygen and carbon dioxide concentrations in the soil atmosphere is approximately equal to 20%, which indicates the domination of oxic respiration.

The latter curve is typical for arable soils, where oxygen concentrations are usually within 10 and 20% by volume, and carbon dioxide concentrations are between 0.1 and 10% by volume. Annual dynamics of soil oxygen concentration under moderate climate conditions is characterized by a summer minimum corresponding to maximum carbon dioxide concentration (Fig. 6.12).
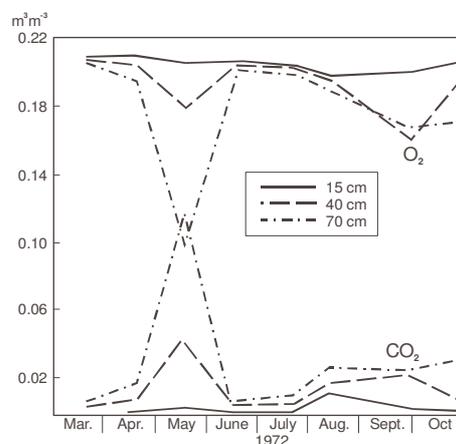
Fig. 6.12. Seasonal dynamics of oxygen and carbon dioxide concentration in soil air at three different depths A– 15 cm, B– 40 cm and C– 70 cm. Data from a study in lysimeters (in Poland) filled with sandy loam soil under winter wheat vegetation in 1972. (From Stępniewski, 1977)

6.4. Soil redox processes

The exhaustion of the soil oxygen pool takes place in situation of non equilibrated demand by oxygen supply. The plants start to suffer from oxygen stress, the root system perishes and, finally, the whole plant dies (Gliński and Stępniewski, 1985; Stępniewski and Gliński, 1985; Bennicelli, 2000).

The time of oxygen exhaustion in soil depends on the size of oxygen pool and the actual oxygen consumption rate. After the exhaustion of oxygen in soil, a sequence of redox processes, comprising the reduction of nitrates, then manganese and iron oxides, and finally sulphates, followed by methane fermentation begins (Gliński *et al*., 1992; Stępniewska, 1986; Stępniewska *et al*., 1997). The oxidized forms of nitrogen, manganese, iron, etc are used as final acceptors of electrons.

The redox status of the soils can be expressed by redox potential (Eh) and by

the ability of the soil components to stabilize Eh at a given level, which is described by such a parameter as a redox capacity or soil redox resistance (R_c) to reduction (Gliński and Stępniewska, 1986; Stępniewska *et al.*, 2004).

6.4.1. Definition of Eh

Redox potential (Eh) expresses the tendency of an environment to receive or to supply electrons in solution. Electron transfer between donors and acceptors is involved in any redox reactions causing oxidation states of the two components (Stumm and Morgan, 1970, 1996).

In a water sample or in a soil solution the redox potential is determined by the amount of dissolved oxygen and its consumption by organisms. In oxic environments a higher redox potential is observed because oxygen present easily accepts electrons. In environments rich in oxygen, heterotrophic organisms capitalize on the use of O_2 as a powerful electron acceptor. Electrons are generated in the soil from the metabolism of reduced organic compounds which are oxidized to CO_2 (Reddy and Graetz , 1980).

Electron free energy scale for mole of electrons can be expressed in ΔG (Joule) or in E (Volts) as well as in pE (dimensionless). The energy gained in the transfer of 1 mole of electrons from oxidants to H_2, expressed in volts, is the redox potential Eh. Redox potential measurement is made with the use of inert (eg. platinum) and reference (saturated calomel or silver-chloride) electrodes. Redox potential Eh for the redox reaction:

$$aA + bB \leftrightarrow cC + Dd \qquad (6.49)$$

is expressed by Nernst equation, as follows:

$$E_h = Eo - (0.0592/n) \cdot \log Q \qquad (6.50)$$

where $Q = [(C)^c (D)^d]/[(A)^a (B)^b]$, n = number of electrons transferred in reaction and Eo is the standard electrode potential at 25°C in which all substances are at 1.0 M concentrations at pH = 7 in equilibrium.

Standard Redox Potential (Eo) expresses a tendency of a reducing agent to donate electrons when Q=1.

6.4.2. Concept of pE

Redox status of electrochemical systems is often expressed in terms of pE, which is derived from the equilibrium constant of the oxidation-reduction reaction (Tab. 6.1.).

For any reaction:

$$Ox + e^- + H^+ \leftrightarrow Red + H_2O \tag{6.51}$$

the equilibrium constant, K, is determined by:

$$Log\ K = \log[Red] - \log[Ox] - \log[e^-] - \log[H^+] \tag{6.52}$$

where: Ox is the oxidized form of the substrate and Red is the reduced product of the reaction.

If we assume that the concentrations of oxidized and reduced species are equal then:

$$pE + pH\ = \log K \tag{6.53}$$

Table 6.3. Differences in concepts and assumptions in the definitions of pH and pE (After Thorstensson, 1984)

pH	pE
$H^+_{(aq)}$ concentrations > 0	e^- concentrations = 0
Electrodes respond to $H^+_{(aq)}$ from solutes and solvent	Electrodes respond to electron transfer from solutes
$H_{2(g)} = 2H^+_{(aq)} + 2e^-$(Pt) approaches reversibility	$Red_{(aq)} = Ox_{(aq)} + e^-$(Pt) is generally reversible
$10^{-15} < mH^+_{(aq)} < 10$	$m[e^-]_{(aq)} = 0;\ 10^{-65} < a[e^-]_{(aq)} < 10^{-35}$
In pure water, pH = 7; $mH^+_{(aq)} = 10^{-7}$	In pure water, pE = 5.9; $pE_{(aq)} = 55$
$H^+_{(aq)}$ is weak oxigizing agent; it can be reduced to $H_{2(g)}$ in aqueous solution	$e^-_{(aq)}$, once formed is a stronger reducing agent than metallic Na

Because the sum of pE and pH is constant, if one of them increases, the other must decline. When a given reaction occurs at lower pH, it means that it will occur at higher redox potential or pE.

The value of redox potential can be converted to pE according to the formula:

$$pE = \frac{Eh}{2.303\,(RT/F)} \tag{6.54}$$

where: R = universal gas constant (8.31447 J mole^{-1} K^{-1}), F is Faraday's constant (96500 C mole^{-1}), T is the temperature in Kelvin, and 2.303 is a constant.

The use of pE makes calculations simpler than the use of Eh because every tenfold change in the activity ratio causes a unit change in pE. But in performing electrochemical measurements the electromotive force (in volts) is being measured.

6.4.3. Effects of pH on redox potential

Between redox potential and pH there exists a strong relationship. In natural waters or soil solutions which are in a highly dynamic state with regard to oxidation-reduction reactions the values of Eh and pH are rather far from the thermodynamic equilibrium. In the absence of biochemical catalysts, most oxidation-reduction reactions under these conditions have a tendency to be much slower than acid-base reactions (Sposito, 1989).

For example at pH 5, when equilibrium between Fe^{2+} and Fe^{3+} is established the redox potential is about +400 mV.

$$Fe^{2+} + 3\ H_2O \leftrightarrow Fe(OH)_3 + 3H^+ + e^- \tag{6.55}$$

At higher pH, the reaction equilibrium will shift to the right so Fe^{3+} will prevail in neutral and alkaline conditions. At lower pH, such as the acid anoxic waters of peat bogs, Fe^{2+} will prevail.

The redox potential (Eh) for the reaction: $O_2(Aq) + 4H^+ + 4e^- = 2H_2O$, taking place in any water solution containing soluble oxygen (for example: <u>8 mg dm^{-3} O_2</u>= $10^{-3.6}$ M= 32 g mol^{-1}) at pH values of 5, 6, 7, 8 and 9 would be as follows: Eh_5= 0.921 V, Eh_6= 0.862 V, Eh_7 = 0.802 V, and Eh_9 = 0.684 V.

Changes in temperature also influence Eh, but not to the same extent as pH. A rise in temperature by 1°C will decrease Eh by 0.0016 V.

Under natural conditions the values of pE range from –6 to +12, while those of pH from 3 to 9 and those of Eh: from –600 to +400 mV (Fig. 6.13).
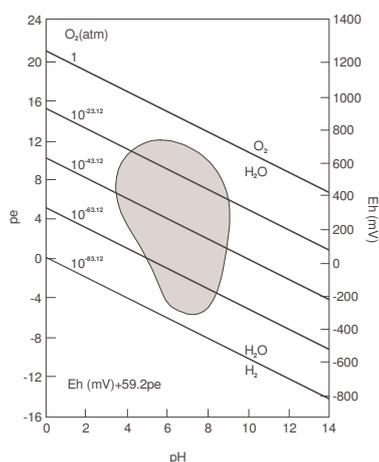
Fig. 6.13. The range of Eh found in natural environments (After Schlesinger, 1997, modified)

6.4.4. Redox transformations

Some macroelements such as C, N, O, S, Mn, Fe, and trace elements or contaminants such as As, Se, Cr, Hg, U, Mo, V, Cu, Ag, Pb can exist (Sposito, 1989) in a number of oxidation states in near-surface geologic environments (Fig. 6.14.).
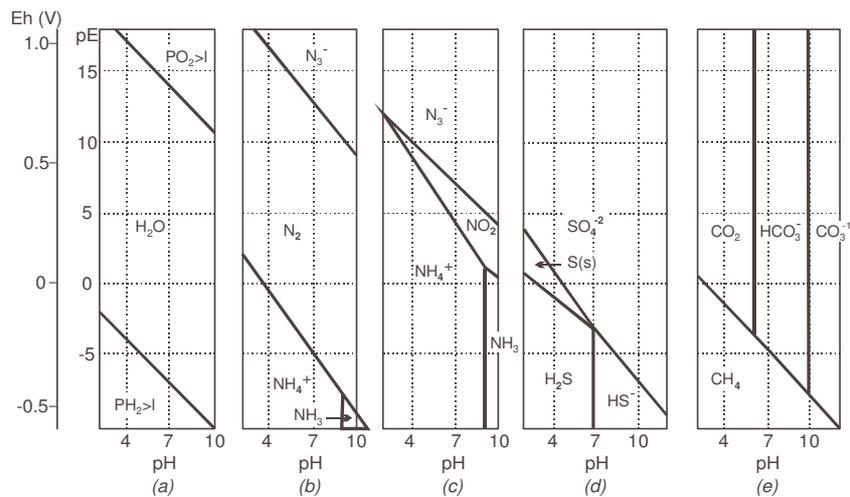
Fig. 6.14. The pE-pH diagrams for most biologically important elements (at 25°C): (a) Oxygen and hydrogen equilibrium with water; (b) and (c) Nitrogen oxidation states with regard to N_2; (d) Sulfur species stable for assumed conditions. (e) Thermodynamically possible existence of C compounds (After Schlesinger, 1997, modified)

6.4.5. Redox processes in the environment

In different elements of the environment the following values of redox potential are found:
- well-oxygenated water – Eh ~ 0.802 V
- oxygenated soils and surface waters – Eh ~ 0.560 V
- anaerobic waters or soils – Eh <~ 0.300 V

Soils and sediments that resist changes in their redox potentials are said to be well poised. The term poising relates to stabilization of redox status, while the term buffering capacity is related to the ability of stabilization of the pH of the system (Fig. 6.15).

As long as the system is exposed to the atmosphere, O_2 will maintain a high redox potential. In the absence of O_2, a decline in redox potential and the reduction of oxidized forms (nitrate, Mn^{4+}, Fe^{3+} and SO_4^{2-}) takes place.

The driving force causing a decrease in the redox potential is the consumption of oxygen by microbial respiration and the use of organic matter as a carbon source. All aerobic organisms must have O_2 to survive. Facultative anaerobes can tolerate periods of aerobic conditions.
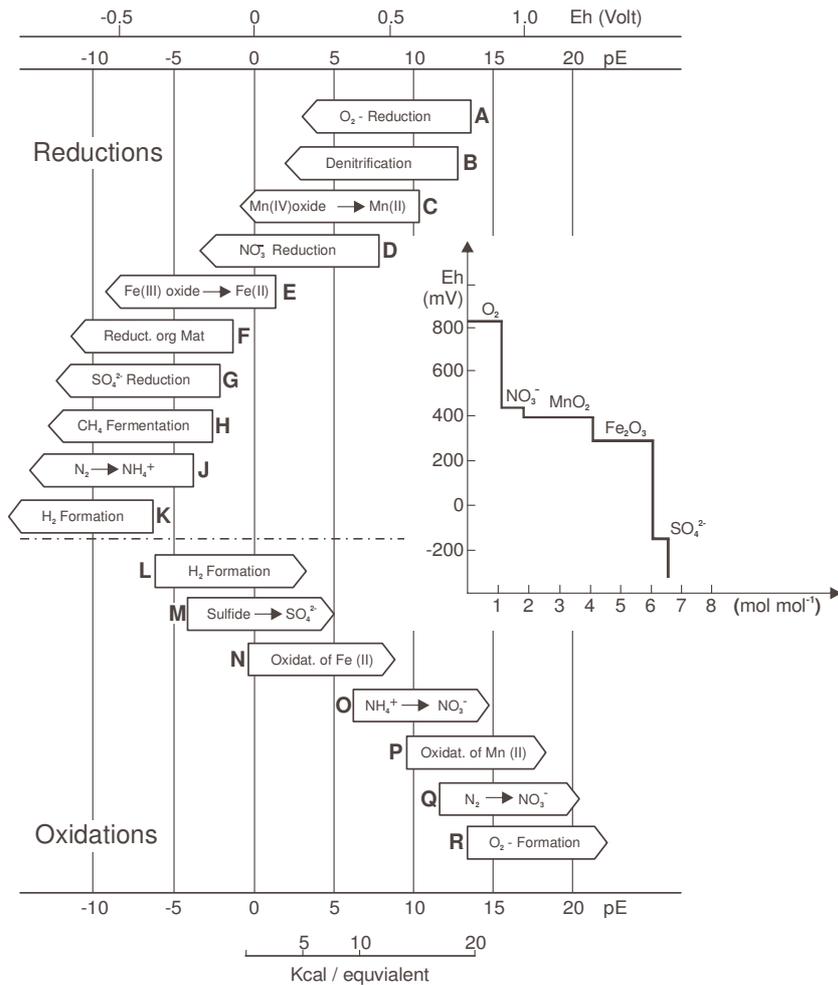
Fig. 6.15. Redox reactions which occur during wetting and rewetting of soils. (After Schlesinger, 1997, modified). The inset presents the Eh levels of particular compounds and their oxygen equivalents (width) *i.e.* number of moles of a given compound substituting as electron acceptor one mole of oxygen.

After O_2 is depleted by aerobic respiration, denitrification occurs within the redox potential interval from +750 to 300 mV, dependently on pH. Denitrifying bacteria use nitrate as an alternative electron acceptor during the oxidation of organic matter. Below a redox potential of +500 mV, when nitrate is depleted, the reduction of Mn^{4+} begins. Reduction of manganese is performed also by facultative anaerobes.

Obligate anaerobes at Eh < –50 mV perform the reduction of Fe^{3+}. Due to this, the appearance of reduced Fe^{+2} ions is used as the indicator of the transition from mildly reducing to strongly reducing conditions.

As it can be noticed from Figure 6.15 the oxidation processes proceed with some asymmetry versus redox potential. This is reflected by higher Eh (pE) values for the same reaction under oxidation conditions. This asymmetry is confirmed by the plots presented in Figure 6.16. The Figure presents ODR, Eh and reduced forms of manganese and iron 14 days of flooding and the subsequent period of drainage and drying.
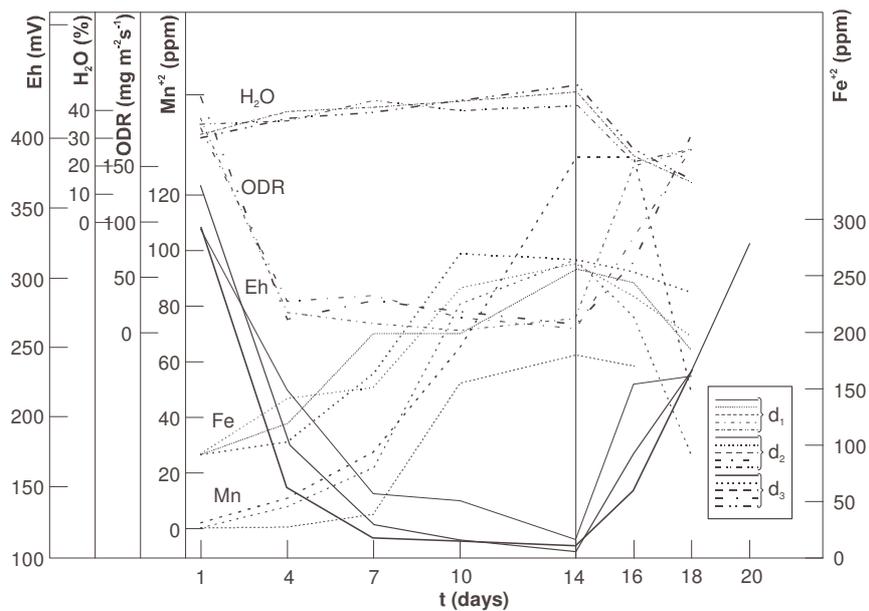
Fig. 6.16. Changes in ODR, Eh, Mn^{+2}, Fe^{+2} and moisture content during 14 days flood incubation of a Cambisol developed from loess at bulk densities: $d_1=1.2$; $d_2=1.35$; $d_3=1.5$ g cm^{-3} and subsequent drainage and air – drying (From Gliński *et al.*, 1986)

Seasonal fluctuations of water table or water level in surface waters may expose a previously flooded soil to the air and the boundary between oxidized and reduced

conditions will shift downward in the profile (Januszek, 1987). Due to that, products of previous reduction reactions become substrates for oxidizing bacteria.

Denitrification is enhanced when seasonal periods of aerobic conditions stimulate the mineralization and nitrification of organic nitrogen, which makes nitrate more available for denitrifiers when the water level later rises. Generally, pH and Eh (pE) are considered the master geochemical variables controlling the geochemical reactions of elements in geologic and aquatic systems (Schlesinger, 1997).

6.4.6. Redox capacity – redox resistance

The redox capacity r (eq/L) at any Eh can be defined as the quantity of strong reductant which must be added to reduce the Eh by 1 volt (Nightingale, 1958).

$$r = dC/ d(Eh) \qquad\qquad (6.56)$$

or, as the time needed to decrease redox potential (Eh) to a given value: to +400 mV corresponding to beginning of nitrate decomposition, or to +300 mV corresponding to the beginning of manganese and iron reduction.

Redox buffering capacity can help to interpret important reactions that control pH of natural waters (van Breemen and Wielmaker, 1974) and sediments (Hutcheon *et al.*, 1993).

The status of progress in the redox process is expressed by the redox potential. Soil redox properties are especially important in the effective use of fertilizers and the environmental impact of agriculture in expected climatic change, which will intensify the redox process and, among other things, mobilize the accumulated toxic substances (CTB – Chemical Time Bomb; Stigliani, 1991) (Figs 6.17 and 6.18).
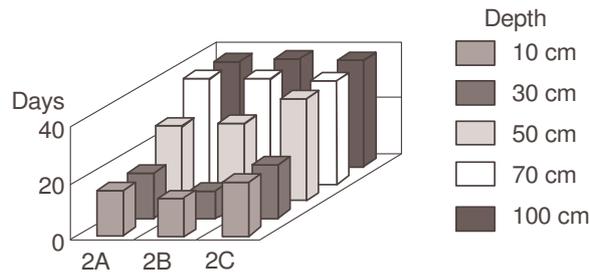
Fig. 6.17. Sum of days with Eh <400 mV after flood irrigation of a willow (*Salix viminalis*) plantation on Histosol with waste water. 2A – control field, 2B – field irrigated with 60 mm of waste-water, 2C– field irrigated with 120 mm of water (Unpublished data of Stępniewska and Nosalewicz)
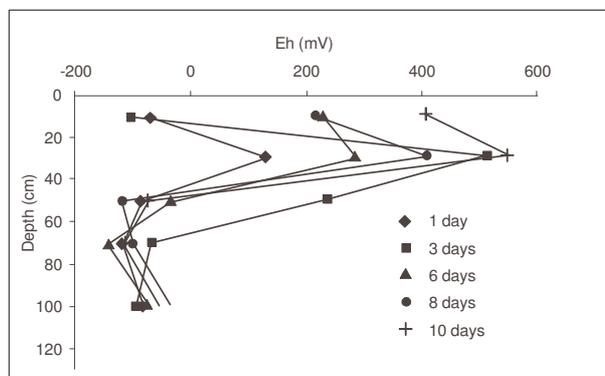
Fig. 6.18. Distribution of Eh values in soil with depth after flood-irrigation of a Histosol with waste water after different period of time from the onset of flooding (Unpublished data of Stępniewska and Nosalewicz)

6.4.7. Release of P

When conversion of agricultural lands to wetlands takes place under the conditions of saturation with water, an increase of P concentration in soil solution is observed. Fluctuations of water table in wetlands can lower the soil redox potential and influence the content of reactive P in soil. In this transformation, temperature and microbial activity play an important role (Figs 6.19-6.21). The effect of Eh on P solubility was first reported by Patrick (1964) who demonstrated that the decrease of insoluble ferric phosphate was directly related to the reduction of iron. In noncalcareous soils release of P is connected with reduction of Fe(III) or Mn(IV) and takes place between 300 mV (pH5) and 100 mV (pH 7).
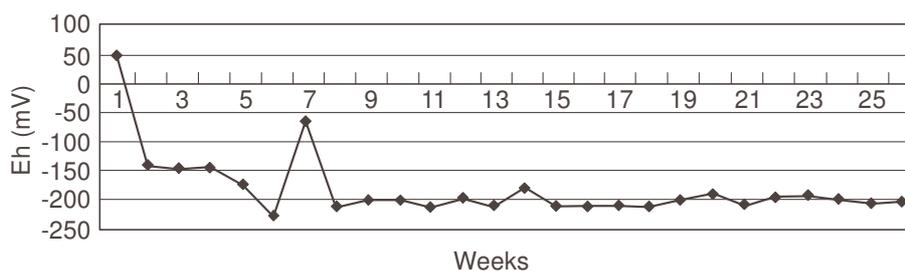
Fig. 6.19. Redox potential changes after flood-incubation of a Histosol from Orłowskie peatland in Poleski National Park (From Stępniewska and Szafranek, 2004)

The behaviour of P in soil is connected with positive feedbacks related to its availability.

This positive feedback occurs because:

- under anaerobic conditions in soil and sediments the release of phosphorus takes place
- more dissolved P results in more biomass production
- more biomass production enhances anoxic conditions

In calcareous system apatite equlibrium may be responsible for controlling P solubility. In this case, the solubility of Ca-bound phosphates such as apatite, tricalcium phosphate, octacalcium phosphate and brushite or monetite is controlled by pH rather than by Eh.
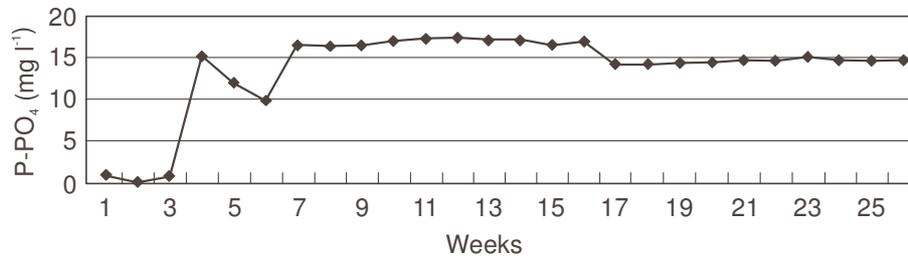
Fig. 6.20. Changes of P-PO$_4$ concentration after flood-incubation of a Histosol of Orłowskie peatland in Poleski National Park (From Stępniewska and Szafranek, 2004)
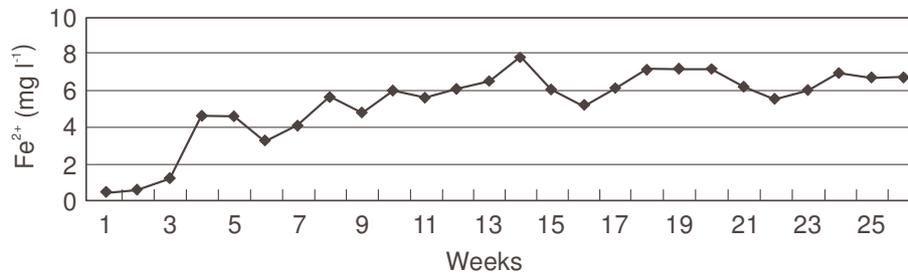
Fig. 6.21. Concentration of Fe^{+2} after flood-incubation of a Histosol from Orłowskie peatland in Poleski National Park (From Stępniewska and Szafranek, 2004)

6.4.8. Eh and stability of pesticides

Behavior and transport of pesticides in a soil profile is connected with their solubility, availability to microbes and with climatic conditions *i.e.* water content and temperature. It was shown that a period of oxygen inhibition could be sur-

vived by the corresponding soil microorganisms and the activity can be acceler-
ated when oxygen returns again. The effect of redox potential appears to be an
important screening parameter to assess their environmental risks (Vink, 1997)
(Fig. 6.22).
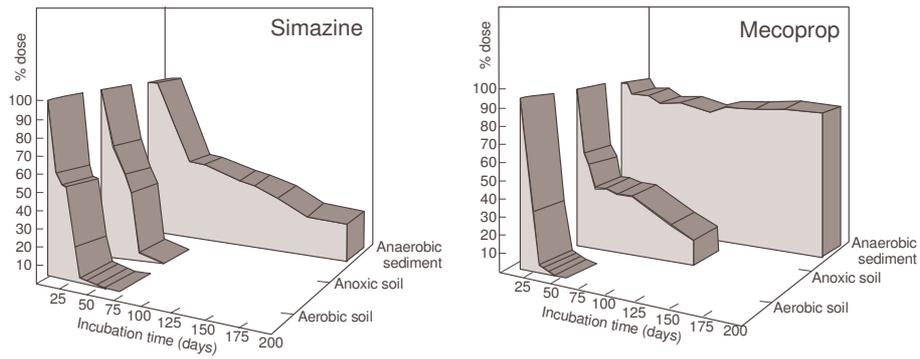
Fig. 6.22. .Stability of simazine and mecoprop in oxic soil (Eh= +330mV), low oxic soil (Eh=
+180 mV) and anaerobic lake sediment (Eh= –120mV (After Vink, 1997, modified)

7. AQUATIC OXYGENOLOGY

Aquatic oxygenology comprises all aspects of oxygen in aquatic medium, such as: oxygen production, distribution, transport, demand, seasonal dynamics, relationships with other elements dissolved in water (methane, nitrates, phosphates), emission to the atmosphere.

Oxygen status in aquatic medium has a specific character depending on the kind of the medium – oceanic, marine, lake or river. All these media are characterized by their own water stratification, movement (upper, deep and bottom circulation, mixing), chemical and physical properties (dissolved organic matter, salinity, methane, phosphate, nitrate, pH, temperature, pressure and density), as well as living organisms and their biological activity.

Oceans and seas are characterized by great masses of water movement caused by currents and wind affecting water ventilations. Similar phenomena, but on a smaller scale, concern rivers. Lakes are calmer.

In the literature, three water masses are identified in oceans and deep seas:

Central Water (CW) which represents a water mass, typically occupying depths from 200 m to the deep salinity minimum layers.

Deep Water (DW) which resides in between the deep salinity minimum and the top of the bottom adiabatic mixed layer.

Bottom Water (BW) which concerns homogenous bottom mixed layer.

There is also another category distinguished – Pore Water (PW) existing in bottom sediments.

As far as oxygen in water is concerned, such terms are used as:
- dissolved oxygen (DO),
- chemical oxygen demand (COD),
- biochemical oxygen demand (BOD),
- sediment oxygen demand (SOD),
- oxygen utilization rate,
- biological oxygen utilization,
- apparent oxygen utilization (AOU),
- oxygen consumption rate,
- oxygen saturation,
- pore water oxygen.

7.1. Oxygenology of oceans

Oceans cover about 2/3 of Earth surface. They differ in size, amount of water and specificity resulting from their localization on the globe.

Examining oxygen properties of oceans, some elements are considered such as: water masses partition, existing currents, water circulation and its upwelling and mixing with other masses.

Oxygen distribution in ocean and sea waters is affected on a large scale by currents and their special kind – cascades.

Ventilation of oceanic intermediate and abyssal layers is made by dense water cascades flowing off continental shelves. These cascades, originating from cooling, evaporation, freezing and salinization on a shallow shelf, spill over the shelf edge and may create near-bottom gravity currents. Sixty one such cascades have been identified around the World Ocean. In most cases, cascades are characterized by their own cross-slope temperature and salinity sections and they deliver colder and fresher water to the deep ocean.

This problem is widely described by Ivanov *et al.* (2004).

Oxygenation of deep water, connected with its currents, is well shown on the example of the subtropical Southwest Indian Ocean by Donohue and Toole (2003) and van Aken *et al*. (2004).

Donohue and Toole (2003) present results obtained from 5 stations (sections) with 54 positions situated in the Mozambique Basin, Mozambique Channel, Madagascar Basin and Mascarene Basin. Measurements of the potential temperature of water, its salinity, silicate, nitrate, phosphate concentration, dissolved oxygen and density, were measured and their distribution maps were elaborated.

Salinity reduces the solubility of oxygen in water – by about 20% in normal sea water as compared to its amount in fresh water (cf. Tab. 6.1 and 6.2; Fig. 6.8).

Solubility of oxygen is affected by temperature, and increases considerably in cold water. It is also affected by the atmospheric partial pressure (Wetzel, 1983) according to Henry's Law.

An example of a map of dissolved oxygen distribution along one of the investigated sections is shown in Figure 7.1.

For the same Southwest Indian Ocean, van Aken *et al.* (2004) present results obtained from investigation on the circulation of deep water with its chemical tracers (silica, phosphate, nitrate concentrations and the apparent oxygen utilizations – AOU).

Apparent oxygen utilizations (AOU), often used in oceanography [μmol kg^{-1}], is expressed as:

$$AOU = [O_{2sat}] - [O_2] \tag{7.1}$$

where O_{2sat} is the saturation value of oxygen concentration [O_2] in balance with the atmosphere.
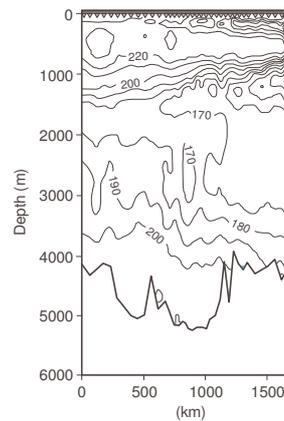
Fig. 7.1. Distribution of dissolved oxygen in μmol kg^{-1} along the Madagascar Basin section (From Donohue and Toole, 2003, modified)

The data gathered from 48 regional station clusters showed variation in AOU within 100 μmol kg^{-1} depending on the cluster localization. AOU was positively correlated with dissolved silica, phosphate and nitrate concentration (Fig. 7.2).
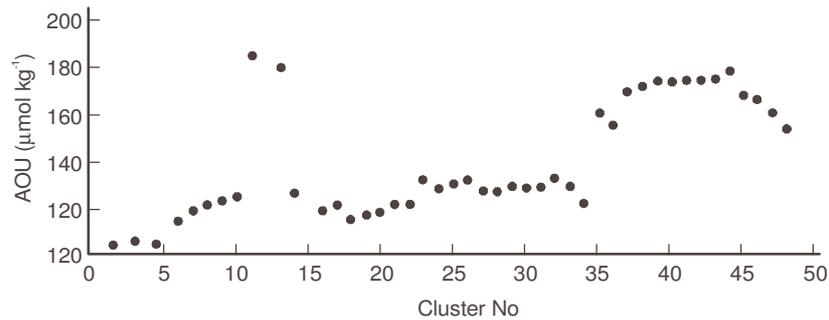
Fig. 7.2. Development of the apparent oxygen utilization (AOU) for successive station clusters (From van Aken *et al.*, 2004, modified)

Some anomalies are noted in oxygen deficiencies at deep water levels.

Braga *et al.* (2004) describe oxygen and silica anomalies within the Guinea Basin at the depth of around 4000 m, where concentrations of oxygen were < 220 μmol kg^{-1} and silica > 60 μmol kg^{-1}. These concentrations were anti-correlated (Fig. 7.3).
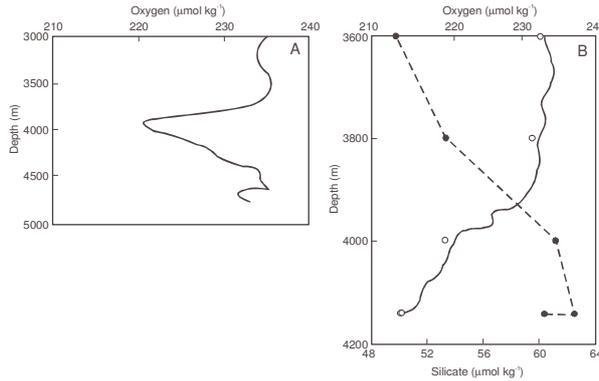
Fig. 7.3. Examples of oxygen vertical profile from continuous measurements with the oxygen minimum at 4100 m depth (A) and oxygen (solid line) and silica (dashed line) profiles (in the range of 3600-4200 m) (B) (From Braga *et al.*, 2004, modified)

The results obtained confirm that the origin of these anomalies is from re-mineralization of terrestrial suspended silica, clay and organic matter carried by the Congo River and deposited through its submarine channel to the depth of 4000 m (Figs 7.4 and 7.5).
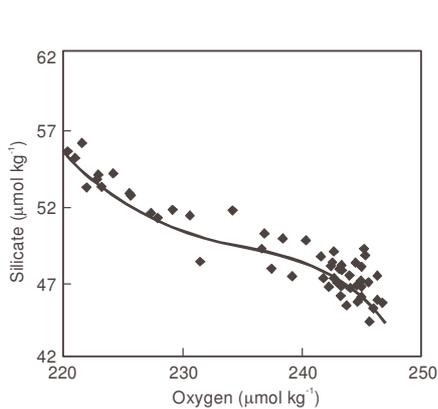
Fig. 7.4. Oxygen versus silicate diagram within the 4000-4200 m depth range (From Braga *et al.*, 2004, modified)
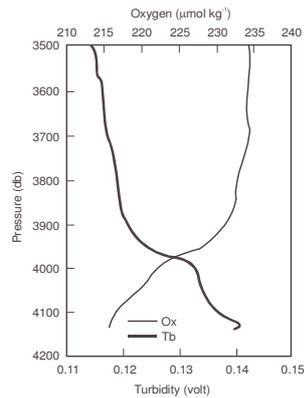
Fig. 7.5. An example of oxygen (thin line) and superimposed turbidity (thick line) profiles (From Braga *et al.*, 2004, modified)

Within deep-ocean water there appear near-bottom cold water anomalies like those found in the Pacific near Hawaii (Lucas *et al.*, 2001). They consist in temperature decrease of 0.035ºC, salinity increase of 0.003, and dissolved oxygen increase of 7 µmol kg^{-1} (Fig. 7.6).
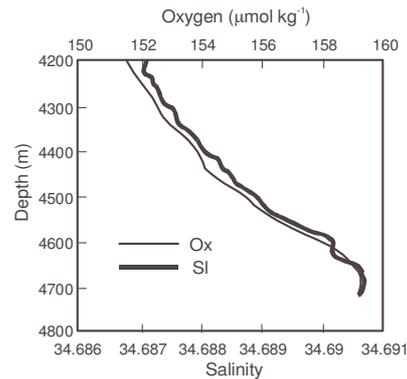
Fig. 7.6. Dissolved oxygen (Ox) and salinity (Sl) along one of the cruises (78) (From Lucas *et al.*, 2001, modified)

7.2. Marine oxygenology

It is not possible to separate strictly sea waters from ocean waters. Climatic conditions (*e.g.* monsoonal wind intensity) and currents cause upwelling of oceanic waters to sea and mixing them.

As an example one can indicate the upper ocean diapycnal mixing in the north western Weddell Sea (Muench *et al.*, 2002). The dense deep- and bottom-water formation of the Arctic Ocean originates within the Weddell Sea a tidal wave whose mean current speed is 5-10 cm s^{-1} and the area-averaged energy (heat) fluxes through the pycnoline are 2-10 Wm^{-2}.

Vertical profiles of water potential temperature, potential density and dissolved oxygen concentration along a transect with a distance of 600 km were elaborated by a number of stations. Data concerning dissolved oxygen concentration during August, 1997, are shown in Figure 7.7.

Between the North Atlantic and the Arctic Ocean there is the Nordic Seas area being the main passageway between these two Oceans. For this area hydrographic, temperature, nutrient and oxygen data were collected by Blindheim and Rey (2004) and presented for Greenland, Norwegian, Lofoten and west Bear Island Basins.
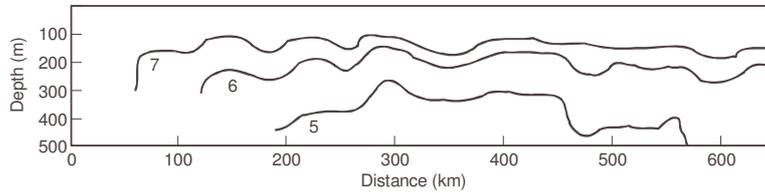
Fig. 7.7. Distributions of dissolved oxygen concentration (ml l^{-1}) along a transect extending from the Antarctic Peninsula into the central Powel Basin during August, 1997 (From Muench *et al.*, 2002, modified)

Some examples concerning salinity, dissolved oxygen concentration and distribution in deep waters are shown in Figures 7.8 and 7.9. Salinity of waters is defined since 1978 as the electrical conductivity ratio of sea water to a standard KCl solution. Ratios have no units in comparison to earlier used ppt or ‰ (grams of salt per liter of water) (Wikipedia). Fluctuations in these components are to indicate the increasing influence of the Arctic Ocean Deep Water on the Nordic Seas water.
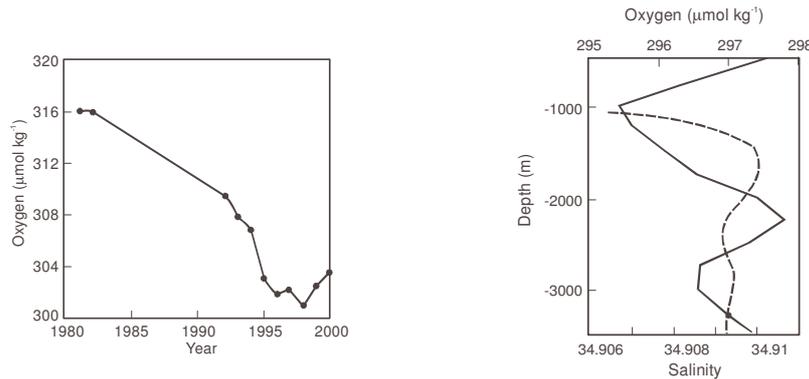
Fig. 7.8. The dissolved oxygen concentration at 2500 m in the central Greenland Basin from years 1981-2000 (From Blindheim and Rey, 2004, modified)

Fig. 7.9. An example of dissolved oxygen concentration (solid line) and salinity (dashed line) at depths greater than 500 m in one of examined positions within the Norwegian Basin in July, 1994 (From Blindheim and Rey, 2004, modified)

The oxygen saturation and consumption rate at the entrance to the Baltic Sea during the period from 1975 to 2000 as a result of the influence of water exchange and temperature were examined by Rasmunssen *et al.,* [15] They used for their calculations the temperature-normalised consumption rate constants (µM 5°C) using the model:

$$\frac{dC(t)}{dt} = -\mu C(t) - \mu_o \tag{7.2}$$

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where the left side of the equation is the consumption rate, and the first term on the right side is a first order reaction term, and the second term is a zero-order reaction term.

They also introduced ventilation as the time elapsed since a submerged water parcel was the last part of the surface water mass.

It was found that the oxygen saturation and the consumption period of oxygen display a significant seasonal variation (Fig. 7.10) depending on the seasonal temperature. It is short (25-50 days) during winter, when the temperature is 3-6ºC, and it is prolonged to 75-100 days in summer months, when the temperature reaches 11-14ºC.
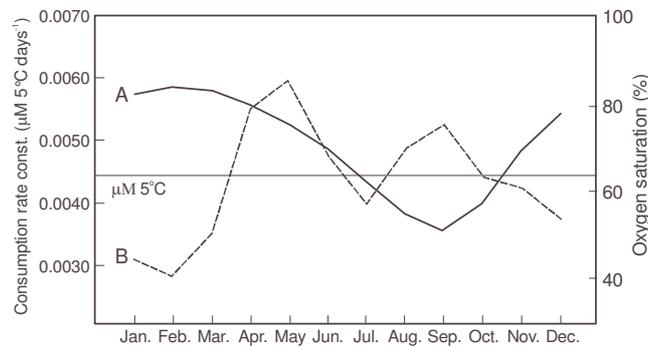
Fig. 7.10. Averaged seasonal variations in oxygen saturation (A) and consumptions (B) observed in the period 1975-2000 (From Rasmunssen *et al.*, 2003, modified)

Oxygen deficiencies in aphotic bottom waters are explained by oxygen consumption through the mineralization of organic matter transported to the bottom waters from the euphotic zone.

The East Japan Sea, which is a small mid-latitude marginal sea in the western Pacific, exhibits a vertical profile of dissolved oxygen (Fig. 7.11) similar to the open ocean (Muench *et al.*, 2002). A drastic decrease in the dissolved oxygen concentration of the deep waters noted in the last decades gives basis for the assumption that the deep water of the East Sea will become anoxic within a few hundred years. This was confirmed by Kang *et al.* (2004) but with the time limit until the year 2040 when the current structural change will replace the Bottom Water before it becomes hypoxic.

They estimated the oxygen utilization rates (OUR) for water masses (CW, DW and BW) as 2.1, 1.1 and 0.8 μmol kg^{-1} yr^{-1}, respectively. These values are an order of magnitude higher than the world ocean average of about 0.1 μmol kg^{-1} yr^{-1}.

An example how oxygen concentration and temperature in sea water affect living organisms is shown in Figure 7.12.

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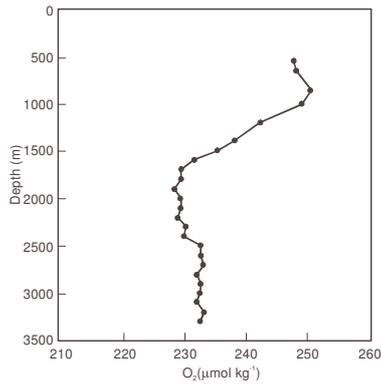
Fig. 7.11. Typical dissolved oxygen profile of the Japan Basin in 1996 (After Kang *et al.*, 2004, modified)
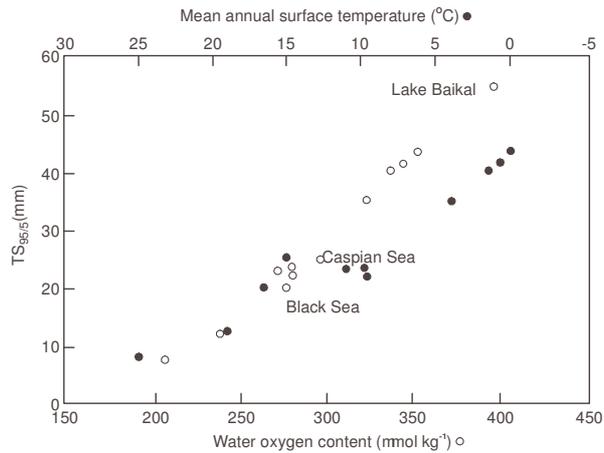
Fig. 7.12. Correlation between body length of amphipods in millimetres (given as an index of average length, $TS_{95/5}$) and temperature ($\bullet$) and oxygen concentration ($\circ$). Lake Baikal, the Caspian Sea and the Black Seas are outliers from the temperature curve because they are blackish or freshwater, rather than saline. Oxygen dissolves more rapidly in freshwater, so the correlation is restored by plotting the length of amphipods against the dissolved oxygen concentration. (From Lane, 2003, modified)

7.2.1. Oxygen processes in marine sediments

Oxygen processes in marine sediments are induced by microbial metabolism, mainly by the microbial oxidation of the particulate organic mater (POM) which

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is incorporated in the sediments. In the deep-sea, the oxidation occurs mainly within the upper few centimetres of the oxic zone of sediments by utilization of dissolved molecular oxygen.

During sediments metabolism, carbon and nitrogen incorporated in the POM in the euphotic zone of the ocean are released to the sediment pore water as their dissolved inorganic species.

Papadimitriou *et al.* (2004) investigated the oxidation of POM during the sediment metabolism at the depth below 1000 m of the eastern north Atlantic. They found a continuous decrease with depth in the utilization of oxygen during the microbial oxidation of POM (Fig. 7.13).
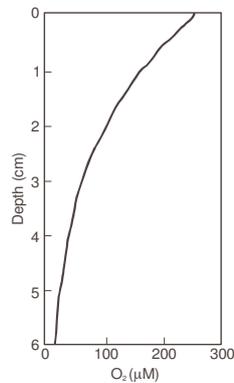
Fig. 7.13. In situ O_2 microelectrode profile in sediment (From Papadimitriou *et al.*, 2004, modified)

Dezileau *et al.* (2002) explained the increase of authigenic uranium content in sediments of the Southern Ocean during glacial periods with a decrease in the oxygen concentration in deep waters with a simultaneous increase in the lateral transport of organic matter and a process of diagenesis. The role of the bottom-water oxygen concentration in the transformation of benthic N as NH_4^+ is shown by Farias *et al.* (2004) on the example of the continental shelf region off central Chile, one of the widest and most productive areas of the eastern South Pacific. The sediments of this area act as a large NH_4^+ source or sink closely connected with the amount of labile organic carbon reaching the sediments. The results obtained by the authors were discussed in relation to the temporal variability of the oceanographic conditions in the area and in relation to the seasonality of upwelling and the occurrence of the 1997-1998 El Niño events and the biogeochemical consequences of that variability. It was found that the near-bottom dissolved oxygen concentrations were maximum in wintertime in the period of 1997-2001

(16.5-50 μM) and minimum in summertime (2-9.4 μM). The balance between net NH_4^+ production and potential nitrification did not support the observed NH_4^+ fluxes. The sediments acted as a large sink for NO_3^- produced by nitrification.

Oxygen, together with temperature, plays an important role in methane inventory in marine sediments as its called icy solid hydrate (clathrate) (Buffett and Archer, 2004) (Fig. 7.14). Such clathrate is formed bellow water depth of less than 250 m but more often at 600 m.
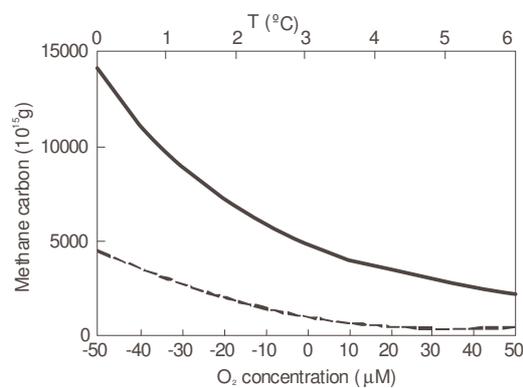
Fig. 7.14. Predicted changes in steady-state clathrate methane inventory in response to expected seafloor temperature changes due to greenhouse effect (dashed line) and resulting changes in O_2 concentration near the seafloor (solid line) (From Buffet and Archer, 2004, modified)

Organic carbon content in sediments is responsible for the concentration of oxygen in the overlying water. It was found that even a small (40 μM) decrease in oxygen concentration is sufficient to cause an increase in methane concentration by a factor of 2.

As an increase in oxygen concentration, parallel to temperature (Fig. 7.14), lowers the clathrate inventory in the sediment, future warming will cause substantial changes in this inventory, *eg.* reducing it to 15% of its present value when temperature increases by 3ºC.

7.3. Lymnooxygenology (Oxygenology of lakes)

Oxygen is a fundamental constituent of lakes. It is essential to the metabolism of all aerobic aquatic organisms, responsible for their distribution, behaviour, and growth.

As fresh waters of lakes are not too dynamic, the supply of dissolved oxygen from the atmosphere undergoes intensive consumptive metabolism. It governs nutrient availability and the resultant growth of many organisms.

Oxygen concentration in lakes is based on physical control of its diffusion, mixing and saturation. It becomes highly variable vertically, horizontally and

seasonally. The limnitic or meromictic types of lakes create an additional variability of the condition for oxygen status.

Possible vertical distribution in oligotrophic and eutrophic lakes during the four seasons is shown in Figure 7.15.
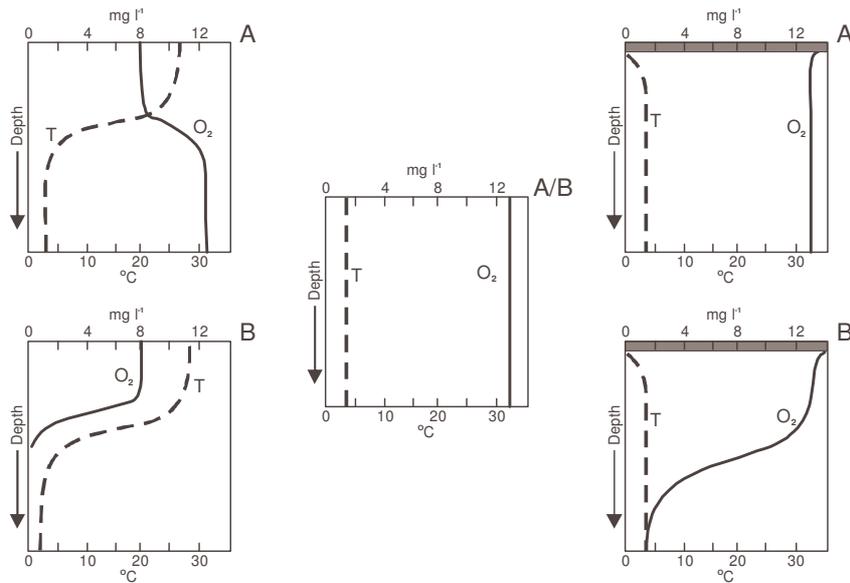
Fig. 7.15. Idealized vertical distribution of oxygen concentration (solid line) and temperatures (dashed line) during the four seasonal phases (summer –left, winter – right, spring and fall – center) of an oligotrophic (A) and an eutrophic (B) dimictic lakes (From Wetzel, 1983, modified)

The kind of lake (oligotrophic, eutrophic) and temperature didtribution affect the oxygen seasonal stratification and create so called orthograde or clinograde oxygen profiles. These profiles are characteristic for summer oxygen stratification.

In the oligotrophic lake, the oxygen concentration in epilimnion (a top water layer) decreases with the increase of temperature. The decrease of temperature in the metalimnion (medium part of water depth) and hypolimnion (bottom water) cause an increase of oxygen concentration. Such an oxygen curve when the whole profile is aerobic is termed orthograde.

In the hypolimnion of the productive eutrophic (with high nutrient content and organic production) lakes, the oxygen content rapidly drops due to oxidative reactions. This process in highly eutrophic lakes passes quickly, within a few weeks. The oxygen concentration curve in which the hypolimnion is anaerobic is termed the clinograde.

The biological oxidation of organic matter and oxygen consumption by animals, plants and bacterial respiration is responsible for oxygen losses from the hypolimnion. It was found that bacterial decomposition of oxygen is greater and much more intensive at the sediment-water interface than in the free water. The interface zone very rapidly becomes anaerobic during summertime. Diffusion in this zone from surrounding layers occurs slowly but may be forced by vertical turbulence, horizontal translocations, and currents that move along the basin sediments.

In the open water, also chemical and photochemical oxidation of dissolved organic matter appears, increasing the consumption of dissolved oxygen. The oxidizable organic matter may come from natural processes as well as from agricultural activities, sewage and industry.

The mechanism of hypolimnetic oxygen depletion depends on the kind of lake. In large and deep lakes, bacterial respirations of phytoplanktonic organic matter dominate. In shallow lakes with large terrestrial inputs of organic matter, benthic decomposition of this matter dominates.

Further on, in stained bog lakes with substantial inputs of dissolved humic compounds, chemical oxidation may assume greater significance.

Spring and fall do not show summer stratification of lakes (Fig. 7.15).

In winter time, bellow ice formation, the exchange of oxygen with the atmosphere is stopped. Only in eutrophic lakes, respiration and chemical oxidation increase with depth similarly to summer stratification (Fig. 7.15). It is a result of continuation of photosynthetic production of organic matter bellow ice- and snow-covers.

Malve *et al.* (2005) estimated the winter respiration in a hyper-eutrophic shallow (3.2 m average depth) Lake Tuusulanjärvi in Finland under ice-cover periods in 1970-2003. They gathered observations of oxygen concentrations and temperature at ice-cover from 30 years. These winter time observations showed a statistically significant negative trend throughout the study period, indicating a decrease in respiration rate (Fig. 7.16).

On the basis of above mentioned observations, the authors elaborated a dynamic nonlinear model of winter respiration in Lake Tuusulanjärvi as the total consumption of oxygen in the lake [$mg\,m^{-3}\,d^{-1}$], which includes both the consumption in the lake water and on the bottom sediment. The model can be used for predicting the winter oxygen regime in a given year in the future. It allows also to predict needed artificial oxygenation efficiency that will prevent fish mortality in the lake.

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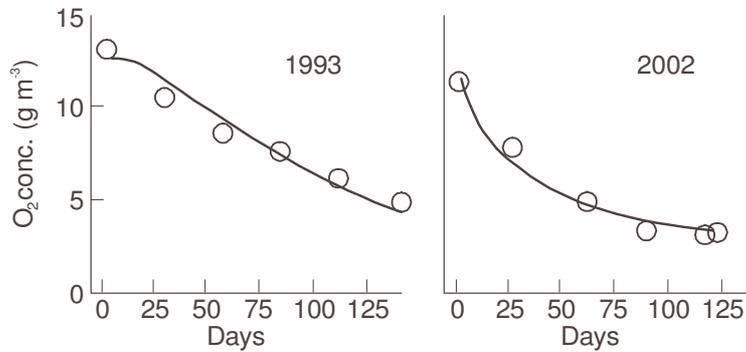
Fig. 7.16. Observed averaged oxygen concentrations at ice-covered period in years 1993 and 2002 (After Malve *et al.*, 2005, modified). The x-axis is time from start of the ice-cover period

Horizontal variations in oxygen content are rapid in winter under ice-cover in the surface strata. It can be seen *e.g.* in Green Lake in Michigan (Wetzel, 1983) in which, under ice-cover, methane and hydrogen are effectively oxidized by bacteria, which causes a severe reduction in oxygen content (Fig. 7.17).
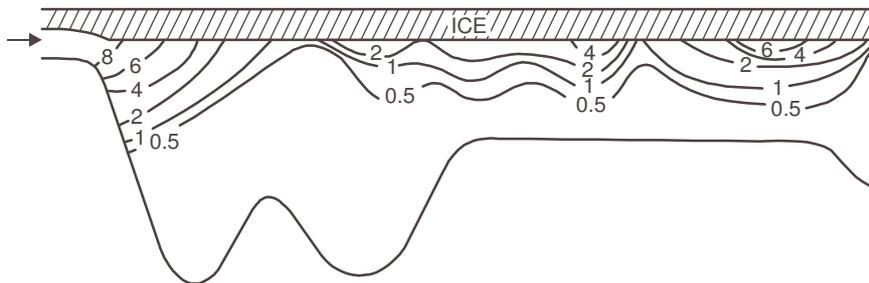
Fig. 7.17. Horizontal isopletes of oxygen concentrations (mg l^{-1}) during a two-week period in Green Lake, Michigan, February 8. 1943 (From Wetzel, 1983, modified)

Horizontal variations in the distribution of dissolved oxygen are more intensive in summer periods and under ice-cover. The oxygen regime differs between littoral zone and the open water, especially in eutrophic lakes where diurnal fluctuations in oxygen concentrations of the epilimnion are often similar to those of the littoral zone.

In consideration of the vertical and horizontal distribution of dissolved oxygen phenomena in lakes, oxygen maxima and minima ought to be mentioned.

Metalimnetic oxygen maxima are often observed in many lakes at the depth between 3 and 10 m, but in very clear lakes even at 50 m (Fig. 7.18). They are

extremely pronounced with supersaturated values above 200% and may reach nearly 36 mg O_2l^{-1} (400% of saturation). The maxima are produced by algal populations which grow well at low temperatures and low light intensity.
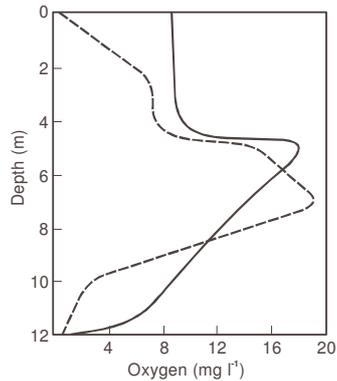
Fig. 7.18. Metalimnetic oxygen maximum, showing a positive heterograde curve (solid line) in relation to rate of phytoplanktonic photosynthesis PS (dashed line) in Lawrance Lake, Michigan, July 20. 1971 (From Wetzel, 1983, modified)

Metalimnetic oxygen minima also appear, but they are less frequent than the maxima, and are associated with metalimnetic reduction processes. In some cases, in certain productive lakes, biogenic oxidation of methane by methane-oxidizing bacteria can result in metalimnetic oxygen minima.

Special conditions for oxygen profiles occur in permanently meromictic lakes, supplied with saline water, that deplete oxygen intrusions.

Examples of very long (historical) and very short (daily) fluctuations of oxygen in lakes are presented by D'Autilia *et al.* (2004) and Watanabe *et al.* (2004).

The first one – long term – was based on analysis of 10 m long sediment core from one of the oldest and largest lakes in the world – Lake Baikal. The water between glacial and interglacial periods was characterized by a decrease of dissolved oxygen concentrations at sediment surface caused by weakening of deep water ventilation, low vertical mixing and low nutrient concentrations in these periods.

An example of the short-time dissolved oxygen dynamics in shallow water of the lagoon of Fogliano, approximately 100 km south of the city of Rome (Italy), is shown in Figure 7.19. The fluctuations in the concentration of dissolved oxygen were in the range of 2-11.5 mg l^{-1}. Dissolved oxygen was measured in July, 1999, within an interval of 30 s for 5-7 days. Problems of actual, absolute and relative oxygen deficits of the lake waters are widely discussed in Chapter 9 of Limnology Book (Wetzel, 1983).

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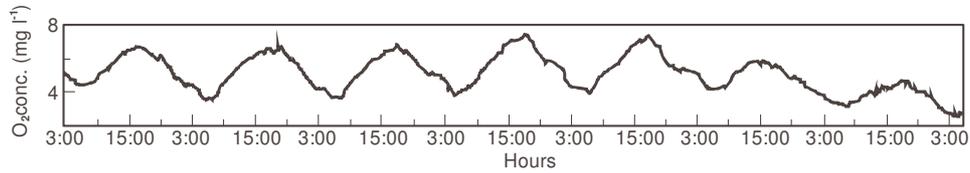
Fig. 7.19. An example of oxygen short time evolution in water of the lagoon of Fogliano (From D'Autilia *et al.*, 2004, modified)

7.4. Potamic oxygenology (Oxygenology of rivers)

Dissolved oxygen concentration in lowland rivers, similarly to lakes, is very sensitive for higher aquatic life.

The variability in dissolved oxygen (DO) in rivers is very great and is caused by the influence of many factors (Fig 7.20).

Among the main sources of DO the following are mentioned (Cox, 2003):

- Atmosphere.
- Enhanced aeration at weirs.
- Photosynthetic oxygen production (P).
- Income from tributaries.
  The main sinks are:
- Oxidation of organic material and other reducing substances in the water.
- Degassing of oxygen in supersaturated water.
- Respiration by aquatic plants (R).
- Oxygen demand exerted by river bed sediments.

The above processes may be expressed by simple mathematical terms such as: (Cox, 2003):

$$\frac{dM}{dt} = M_i = M_o + (P - R) + C_R - BOD - SOD - C_D \pm \Delta S \qquad (7.3)$$

where: t is the time, M_i is the mass flux of DO entering the water body, M_o is the mass flux leaving, C_R represents the aeration and re-aeration processes, BOD is the biochemical oxygen demand representing the oxidation of organic material, SOD is the sediment oxygen demand, C_D represents degassing of oxygen and ΔS represents changes in the water body due to transport from external sources.

The processes that influence DO in lowland rivers are so complicated that they require mathematical models that can simulate them.

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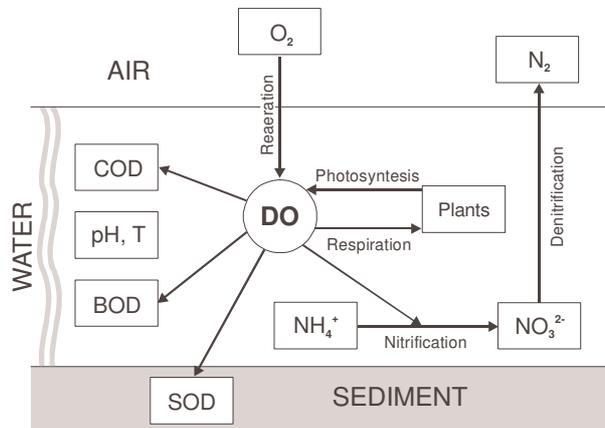
Fig. 7.20. A scheme of main processes influencing the concentration of dissolved oxygen (DO) in rivers. COD – chemical oxygen demand, BOD – biochemical oxygen demand, SOD – sediment oxygen demand (After Cox, 2003, modified)

A review of such models is presented by Cox (2003). A lot of processes are included in those models, such as:

- the removal of BOD by sedimentation or adsorption;
- the addition of BOD by the re-suspension of bottom sediments or by the diffusion of partially decomposed organic matter from the bed sediments into the water above;
- the addition of BOD by local runoff;
- the removal of oxygen from the water by the action of gases in the sediments;
- the removal of oxygen by the respiration of plankton and fixed plants;
- the addition of oxygen by the photosynthesis of plankton and fixed plants;
- the addition of oxygen by atmospheric reaction;
- the redistributions of BOD and DO by the effect of dispersion, particularly when the polluting load varies suddenly;
- the removal of oxygen by nitrifying bacteria;
- the oxygen demand of the carbonaceous and nitrogenous wastes in the water;
- the oxygen demand of the bottom deposits;
- an immediate COD;
- the oxygen required for plant respiration;
- the oxygen produced by plant photosynthesis;
- the oxygen gained from atmosphere re-aeration.

8. PLANT OXYGENOLOGY

Plant oxygenology is a part of biooxygenology whitch has been defined as oxygenology of living organisms *i.e.* oxygenology of biota. It is focused on the effect of oxygen availability in the environment on living organisms as well as on the studies of transport, absorption, and the role of oxygen within the organisms themselves. Within biooxygenology, such areas as microbial oxygenology, phytooxygenology (plant oxygenology), and zoological oxygenology (zooxygenology) have been distinguished. From the above areas only plant oxygenology is presented in this book.

8.1. The Plants Kingdom and oxygen supply

Green plants are critical to other life forms because they form the basis of almost all food webs. Most plants are autotrophic, creating their own food using water, carbon dioxide, and light through photosynthesis. Also, the photosynthesis process produces all the essential life-supporting oxygen that is required by all living oxic organisms. Plants are divided into several kingdoms: Plantae, Protista, and Fungi. All plants are multicellular and eukaryotic (*i.e.*, each cell possesses a membrane-bound nucleus that contains the chromosomes). Plants are independent in their nutritional needs (autotrophic) and store their excess food in the form of macromolecules of starch.

Most aquatic plants occur in the kingdoms of Plantae and Protista. They grow under water, *i.e.* obligate submersed. But most aquatic plants fall into the group referred to as amphibious plants which are capable of growth both in and under water (submerged) and out of water (emersed), with only their root wet or damp.

Aquatic plants are plants that require a water environment to complete all or most of their life cycle. Based upon growth form, these plants can be divided into four types: emergent/emersed, submerged, floating-leaf, and free-floating.

Emergent/emersed plants extend above the water surface in shallow areas of lakes, ponds, and ditches. They have relatively rigid stems and do not rely on the water for support. Leaves of this group of plants are essentially like typical leaves of herbaceous angiosperms. The leaves may be amphistomatic (stomata on both surfaces) and have well developed mesophyll to take advantage of the abundant sunlight. Cattails (*Typha spp.*), buttercups (*Ranunculus spp.*), wetland irises (*Iris spp.*), are a few examples of emergent aquatic plants.

Submerged aquatic plants have flexible stems and leaves, are rooted in the sediments, and are completely covered by water. Leaves of underwater plants are often highly dissected to increase the surface area to permit rapid diffusion of

carbon dioxide into the chloroplasts of the cell. Examples of submerged aquatic plants are: water buttercups, water milfoils (*Myriophyllum spicatum*), bladderwort (*Utricularia spp.*). Rooted-floating leaf or floating-leaf plants have their roots in the mud or muck while their leaves float on the water.

Rooted-floating plants lack stem rigidity and depend on the water for support. These plants usually extend out of the water like emergent plants but have floating leaves. Plants such as bur-reeds (*Sparganium erectum* L.), water plantains (*Alisma* spp.), and arrow-heads (*Saggitaria saggittifolia*) are examples of rooted-floating plants.

Free-floating plants obtain their nutrients directly from the water, since they are not rooted to the soil or muck. Examples of free-floating aquatic plants or macrophytes are water hyacinth (*Eichornia crassipes*), water-lettuce (*Pistia stratiotes*), duckweed (*Lemna spp.*) and Mosquito fern (*Azolla spp.*). Floating leaves tend to be much broader, without much lobbing, and remain flat on the water to take advantage of the sun. Stomatas are present on the upper leaf surface for gas exchange, and the upper leaf surface tends to have a very prominent epidermis or cuticle to permit water to roll off or prevent growth of epiphytic algae that interfere with photosynthesis on the leaves. Floating leaves often have lacunae (air chambers) to provide buoyancy, and sclerids (hard cells) within the mesophylls to provide toughness to leaves and prevent them from collapsing.

Algae are a group of predominantly aquatic, photosynthetic organisms. They range in size from the tiny flagellate *Micromonas* that is 1 μm in diameter to the giant kelp *(Macrocystis pyrifera)* that reaches 60 meters in length. The various major algal groups, such as the green algae, brown algae, and red algae, are now placed in the kingdom "Protista" because they lack one or more of the features that are characteristic of plants. Like all green plants, algae use photosynthesis to form organic food molecules from carbon dioxide and water, and provide oxygen to the atmosphere as a byproduct of photosynthesis.

In the ocean, oxygen is produced as a byproduct of photosynthesis by phytoplankton (single celled sea plants) and algae (multicelled sea plants). Although individual algae are much larger than plankton, the latter have a larger biomass and so produce the most oxygen.

Considering the Earth is more than 70% water and phytoplankton are found throughout the ocean, it is not surprising that they make up 90% of the Earth's oxygen production. The oxygen produced by phytoplankton is released as a gas. Some of this is absorbed back into the ocean, but most flows into the atmosphere. From there it becomes available for use by all oxygen breathing organisms.

8.2. Terrestrial plants and oxygen requirements

8.2.1. Why terrestrial plants need oxygen?

Plants are obligate aerobic organisms which perform respiration processes. Oxygen is essential because it acts as the terminal electron acceptor in the oxidative phosphorylation pathway, which provides the vast majority of ATP for cellular metabolism by regenerating NAD^+ from NADH. Oxygen is also required in several important cellular pathways, including haem, sterol and fatty-acid biosynthesis. The provision of sufficient oxygen to internal tissues is a fundamental physiological challenge in multicellular organisms (Geigenberger, 2003).

From the biochemical point of view, plant respiration can be defined as the sum of glycolisis, the oxidative pentose phosphate pathway, the tricarboxylic acid (TCA) or Krebs cycle, mithochondrial electron transport, oxidative phosphorylation, and related reactions. Physiologically, the definition of respiration will be the non-photorespiratory CO_2 release (photorespiration being associated with photosynthesis), though photorespiration can contribute directly to mitochondrial electron transport (Amthor, 2000). Plants respire about half of the carbon available from photosynthesis after photorespiration.

8.2.2. Oxygen transport within plant roots

The transport of oxygen from the soil to the respiring tissue of the plant root is caused by the radial diffusion of oxygen from the root surface to its center (Gliński and Stępniewski, 1985). In turn, part of the carbon dioxide produced in the roots diffuses outwards, in a direction opposite to the radial movement of oxygen, and, part is transported upwards with the transpiration stream.

From the point of view of plant adaptation to anoxic conditions, the most significant process is the longitudinal diffusion of oxygen from stems to roots. It is determined by the presence of intercellular spaces filled with air. In the case of mesophytes, the exchange of oxygen and carbon dioxide between the internal air system of the plant and the atmosphere takes place through stomata and lenticels.

Radial diffusion of oxygen from the soil into a root of radius R, having a respiratory activity q and a diffusion coefficient inside the root tissue D_i, the root being surrounded by a layer of water saturated soil of effective thickness d with an effective oxygen diffusion coefficient D_e (Fig. 8.1) has been presented in the book by Gliński and Stępniewski (1985).

The general equation of diffusion, assuming the absence of longitudinal gradients and neglecting the respiratory activity of the soil moisture film surrounding the root, for oxygen in the root tissue in a cylindrical coordinate system for steady-state conditions, has the form:

$$D_i \left[\frac{d^2 C_i}{dr^2} + \frac{1}{r} \frac{dC_i}{dr} \right] - q = 0 \qquad (8.1)$$

and for the soil water film surrounding the root:

$$D_e \left[\frac{d^2 C_e}{dr^2} + \frac{1}{r} \frac{dC_e}{dr} \right] = 0 \qquad (8.2)$$

where C_i and D_i denote the concentration and diffusion coefficient of oxygen within the root, respectively; C_e and D_e denote the same parameters in the soil water film surrounding the root, and q denotes the respiratory activity of the root.
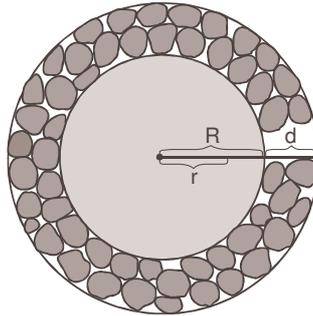
Fig. 8.1. Schematic cross-section of a plant root of radius R, surrounded by soil saturated with water of effective thickness d; r = distance of a root element under consideration from the root axis (After Gliński and Stępniewski, 1985, modified)

8.2.3. The growth-maintenance respiration paradigms

For understanding the respiration of ecosystems, modelling the part of respiration is critical. Plant respiration refers only to the dark respiration, and the root respiration is a part of the belowground respiration. Models of root, leaf and stem respiration are usually referred to as plant respiration models. McCree (1970) and Thornley (1970) first portioned plant respiration into growth respiration (construction respiration) and maintenance respiration components. Johnson (1983) improved the two-component model to

a three-component model by adding a term for ion uptake, because uptake of anions, particularly nitrate, can be costly, representing a third functional component of respiration (Amthor, 2000). A general paradigm that relates respiration to any number of individual processes that it supports, as biosynthesis of new biomass, translocation of photosynthate from source to sink, uptake of ions from the soil solution, assimilation of N (including N_2) and S into organic compounds, protein turnover, and cellular ion-gradient maintenance was introduced by Amthor (2000).

8.2.4. Oxygen exchange in plants

Virtually all to date studies of respiratory and photosynthetic gas exchange in plants under atmospheric conditions have used infra-red CO_2 analysis as the measurement system. This is because the change in CO_2 content of air brought about by the plant is proportionally far greater than the change in O_2 content and therefore is much easier to measure accurately.

A leaf in a leaf chamber may cause a decline in the CO_2 content of the atmosphere from 350 ppm to 330 ppm, a difference of 5.7% between the reference and sample gas. If one molecule of O_2 is produced for every CO_2 fixed, the corresponding increase in the O_2 content of the atmosphere would be from 209 000 ppm to 209 020 ppm, a change of only 0.0001%. Despite the difficulties of measuring such small changes in O_2, differential O_2 analyzers have been developed that are capable of resolving down to +1 ppm O_2 against a background of air (Willms *et al.*, 1997, 1999; Cen *et al.*, 2001; Bloom *et al.*, 2002).

It may be asked why the researcher would choose to measure respiration and photosynthesis by O_2 exchange when CO_2 exchange measurements are easier and more accurate, especially when the CO_2 exchange rate (CER) and O_2 exchange rate (OER) are often assumed to be equal. The reactions determining O_2 exchange and CO_2 exchange rates are linked in both photosynthesis and aerobic respiration, but they are separated by many other oxidative and reductive reactions. If all the electrons released from water splitting in PSII are used for reduction of CO_2 to carbohydrate, 1 mole of CO_2 is fixed in Calvin Cycle for every mole of O_2 produced by PSII, and the photosynthetic quotient (PQ = –OER/CER), where gas evolution rate is positive and consumption rate is negative, equals 1.0. However, if electrons are drawn from photochemical reactions to other reductive sinks (*e.g.* nitrate assimilation), more O_2 will be produced than CO_2 consumed, and PQ will be greater than 1.0.

Similarly, respiratory quotient (RQ = –CER/OER) is equal to 1.0 only when carbohydrate is being respired aerobically. Because fat is far more reduced than carbo-

hydrate, a greater amount of O_2 is required for its oxidation, so that less CO_2 is produced per unit of O_2 consumed, and RQ falls significantly below 1.0 (Hunt, 2003).

Measuring higher-plant respiration is difficult, but in any case, as shown in Table 8.1, respiration is a large component of a plant's seasonal or annual C balance, ranging from less than 50% of photosynthesis in many crops to 65-70% in some tropical and boreal trees and coastal marshes (Amthor, 2000).

Table 8.1. Estimates of annual or seasonal respiration ranges as a fraction of annual or seasonal photosynthesis (*i.e.* the balance of photosynthetic carboxilations with photorespiratory decarboxilation) in intact ecosystems (mol C m^{-2} ground year^{-1}) (Modified from Amthor, 2000)

Ecosystem	Species or climate	Respiration/photosynthesis
Crop	Alfalfa	0.35-0.49
	Maize, rice, and wheat	0.3-0.6
Grassland	Shortgrass prairie	0.34-0.51
	Tallgrass prairie	0.61-0.65
Forest	Tropical moist	0.66-0.75
	Temperate	0.32-0.72
	Subalpine	0.61-0.72
	Boreal	0.64-0.77
Coastal salt marsh	Temperate	0.69-0.77
Tundra	Arctic	0.5

8.3. Consequences of soil oxygen deficiency

The influence of soil oxygen on plants comprises direct and indirect effect (Gliński and Stępniewski, 1985). The former are connected with the physiological effects of oxygen and carbon dioxide which are basic components of soil air. The latter are connected with the numerous changes occurring in the soil under the influence of oxygen and carbon dioxide. None of these changes is without an effect on the plants.

The indirect effects comprise the influence of both O_2, and CO_2 on redox processes in the soil, on its acidity and on the living organisms within it. All these factors influence the availability of nutrients for the plants as well as their well-being in general. A scheme of these mutual interrelations is presented in Figure 8.2.

The extreme case in the soil-air to plant relation is that in which the soil is submerged and the soil air is almost completely replaced by water. Such conditions may be permanent (*e.g.*, in bog or marsh soils), intermittant (*e.g.*, in rice soils), or sporadic and of short duration.

As to the plant's tolerance to soil submergence, we have a whole spectrum of plants – from hygrophites adapted to permanent flooding (*e.g.*, rice) to mesophytes among which significant differentiation exists. Hence, among these plants there are

those moderately tolerant to flooding (most grasses and cultivated plants), as well as those very sensitive to it so that after only a few days of flooding they die away (*e.g.*, tobacco, peas). Xerophytes have been investigated to only a limited extent. Even if their root system is sensitive to oxygen deficiency, the fact is that some of them can live with a reduced root system, being capable of increasing their resistance to all stresses, and among them to an oxygen deficiency stress.
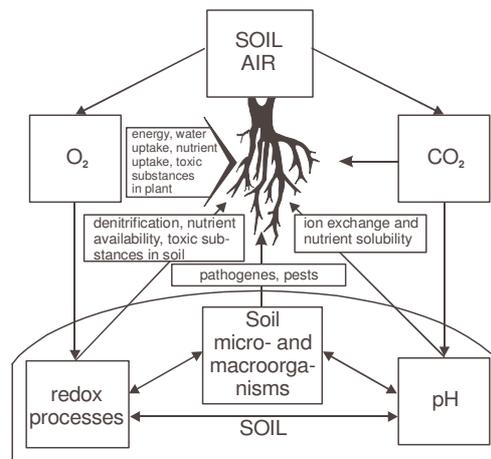
Fig. 8.2. A schematic of direct and indirect effects of soil oxygen on plants (After Gliński and Stępniewski, 1985)

Obviously, plant tolerance to root flooding is relative because it also depends on other factors. One of them is the stage of plant development. Thus in the case of cereal plants (wheat, oats, and barley) we find they are most sensitive to soil flooding in the stem elongation stage and much less so in the earlier stages.

One of the essential negative consequences of soil flooding is oxygen deficiency in the tissues of the submerged plants. Oxygen deficiency affects the intensity and the direction of a number of physiological and biochemical reactions and induces oxidative stress in the plant cells (Zakrzhevsky *et al*., 1995; Yan *et al*., 1996; Bennicelli *et al*., 1998, Blokhina *et al*., 1999; Neil *et al*., 2002).

Under optimal conditions, the content of reactive oxygen species (ROS) maintains, with the help of antioxidative defence system, at a level which is safe for the organism (Larson, 1988). Enzymes of superoxide dismutase and ascorbate – glutathione pathway eliminate the excess of H_2O_2 in chloroplasts, in the cytoplasm and in non photosynthesizing tissues (Foyer and Halliwell, 1976). Under

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stress conditions, the formation of ROS can exceed the antioxidative potential of the cell and cause an oxidative damage (Halliwell, 1984).

The plant capability to activate the defence system against oxidative destruction may be a key link in the mechanism of plant tolerance to unfavourable conditions. Changes in the activity level of one or more antioxidative enzymes are connected with the plant resistance to stressor action (Allen, 1995).
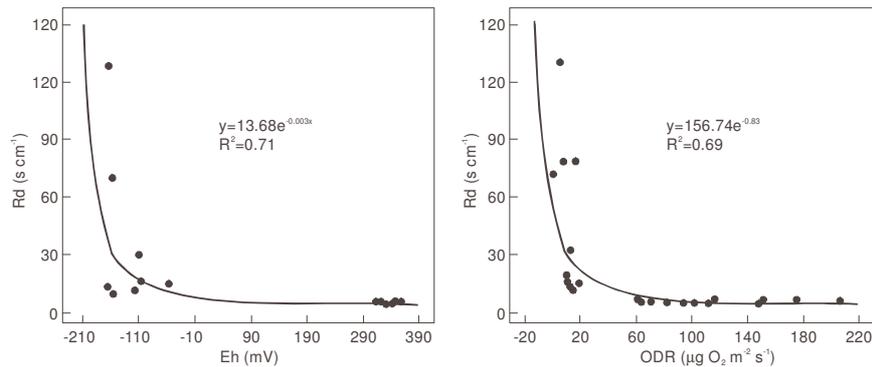
Fig. 8.3. Leaf stomatal resistance (R_D) of *Pisum sativum* L. as a function of Eh and ODR of the soil after 12 days of oxygen stress (After Bennicelli, 2002)

The response of stomatal resistance (R_d) of some plants (*Pisum sativum* L., *Zea mays* L., *Triticum aestivum* L., *Triticale* L.,) is the reaction to unfavourable conditions through hypoxic to anoxic soil indicated by redox potential (Eh) and oxygen diffusion rate (ODR) (Figs 8.3-8.6). Oxygen deficiency may occur in ecosystems in which plant organs are surrounded by water, particularly stagnant water. Both temporary and continuous flooding occurs in flood plains, marshes and irrigation areas throughout the world. Agricultural soils may become also saturated with water (waterlogging) (Gibbs and Greenway, 2003).

The oxygen availability from soil aeration pores (air-filled porosity – Eg) is one of the environmental factors affecting plant respiration rates. When water fills the pore space in soil, the rate at which oxygen can diffuse from the atmosphere through the soil is greatly reduced, and anaerobic (or reduced as opposed to oxidized) conditions result in several hours to several days. The diffusivity of gases in water is 10^4 times slower than in air (Gliński and Stępniewski, 1985). During transient flooding, waterlogging or microbial activity, the soil can be rapidly oxygen-deprived and oxygen stress conditions will arise in the roots, affecting the growth and distribution of terrestrial plants and leading to major reductions in crop yield (Kozłowski, 1984; Gliński and Stępniewski, 1985; Drew, 1997).
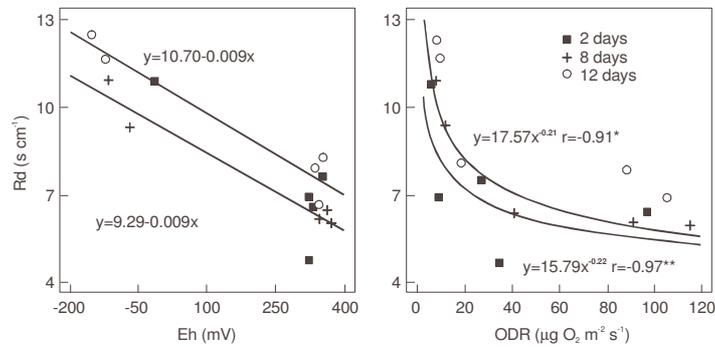
Fig. 8.4. Leaf stomatal resistance (R_D) of *Zea Mays* L. as a function of Eh and ODR of the soil after 2, 8 and 12 days of oxygen stress (After Bennicelli, 2002)
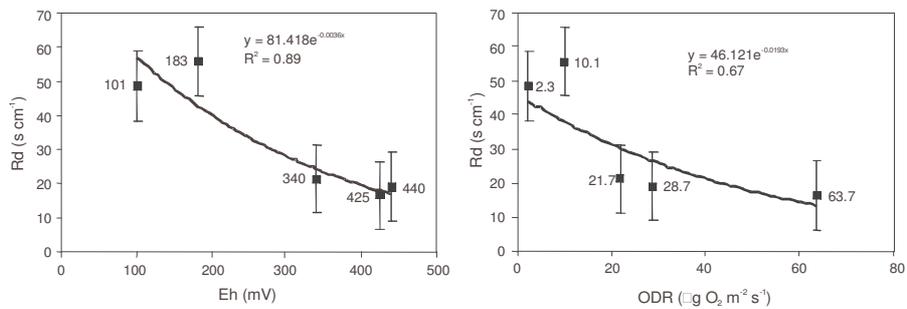
Fig. 8.5. Changes of stomatal diffusive resistance (Rd) in relation to ODR and Eh (*Triticum aesti-vum* L.). Mean value ± standard deviation (After Bennicelli, 2002)
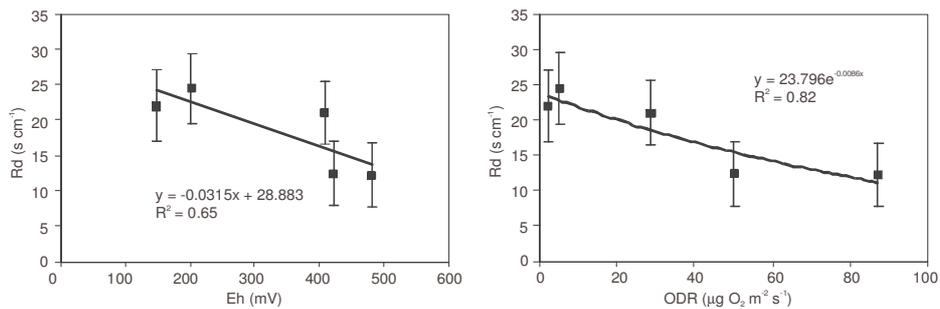
Fig. 8.6. Changes of stomatal diffusive resistance (Rd) in relation to ODR and Eh (*Triticale*). Mean value ± standard deviation (After Bennicelli, 2002)

Under anoxic conditions, when cytochrome oxidase activity becomes oxygen limited [0.013% oxygen; (Drew, 1997)], ATP formation through oxidative phosphorylation is inhibited and ATP has to be produced by fermentation. This impairs cellular metabolism and function because the efficiency of ATP formation is sharply reduced. The respiration of one molecule of hexose equivalent produces up to 39 molecules of ATP, whereas the fermentation of such a molecule provides a maximum of just three molecules of ATP. In addition, fermentation results in falling cytosolic pH, the induction of glycolysis, and the accumulation of lactate and ethanol (Drew, 1997).
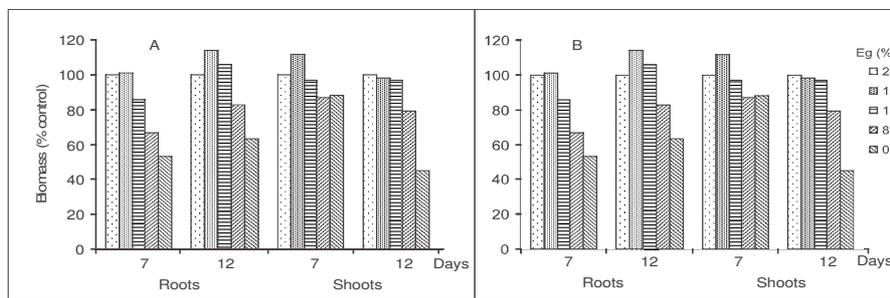
Fig. 8.7. Increase of root and shoot biomass (%) *Triticale* – CZR 1406 (A) and *Triticum aestivum* var. Rosa (B) after 7 and 12 days of oxygen stress at 5 levels of soil air-filled porosity (Eg). Mean value ± standard deviation (After Bennicelli, 2002)
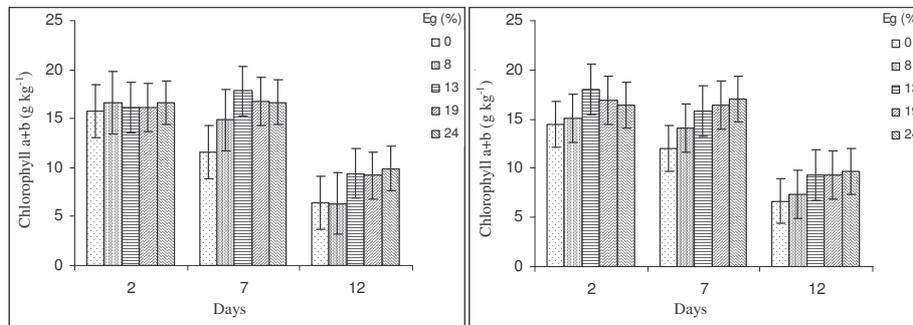
Fig. 8.8. Chlorophyll a+b content (*Triticale* – CZR 1406 and *Triticum aestivum* var. Rosa) at 5 values of air-filled porosity (Eg). Mean value ± standard deviation. (After Bennicelli, 2002)

Bennicelli *et al*. (1998) and Bennicelli (2002) found that plant response to soil aeration conditions (as evaluated by biomass production (Fig. 8.7), chlorophyll a+b content (Fig. 8.8), antioxidant system status – expressed by super oxide dismutase (SOD) activity (Figs 8.9 and 8.10) as well as the advancement of destructive processes (Hunter *et al*., 1983) (assessed by malondialdehyde – MDA content

– Figs 8.11 and 8.12), were related to soil air filled porosity. Similar dependences of growth and oxidative processes to soil aeration parameters were also shown for pea known as a flood intolerant plant (Zakrzhevsky *et al.*, 1995).
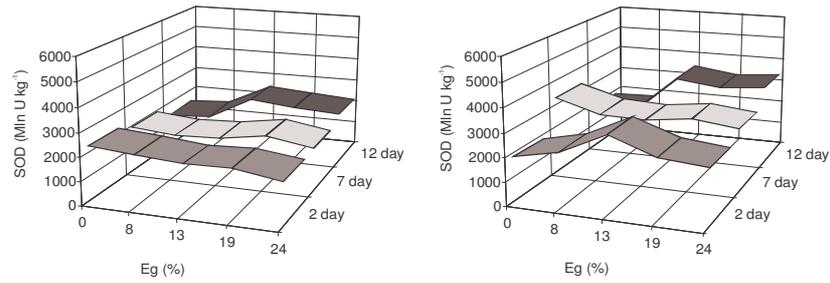
Fig. 8.9. SOD activity in roots *Triticale* – CZR 1406 and *Triticum aestivum* var. Rosa after 2, 7, 12 days of oxygen stress at 5 levels of air-filled porosity (Eg) (After Bennicelli, 2002)
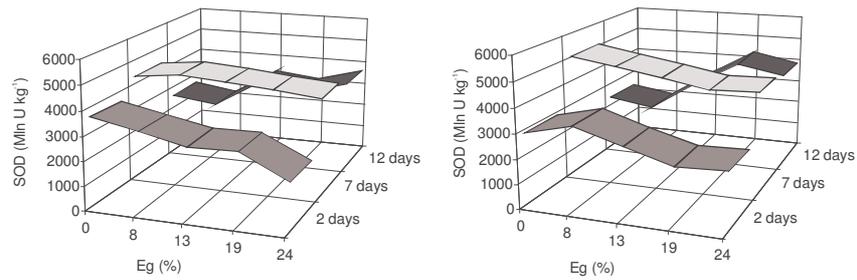
Fig. 8.10. SOD activity in leaves *Triticale* – CZR 1406 and *Triticum aestivum* var. Rosa at the same experimental conditions as in Fig. 8.9 (After Bennicelli, 2002)
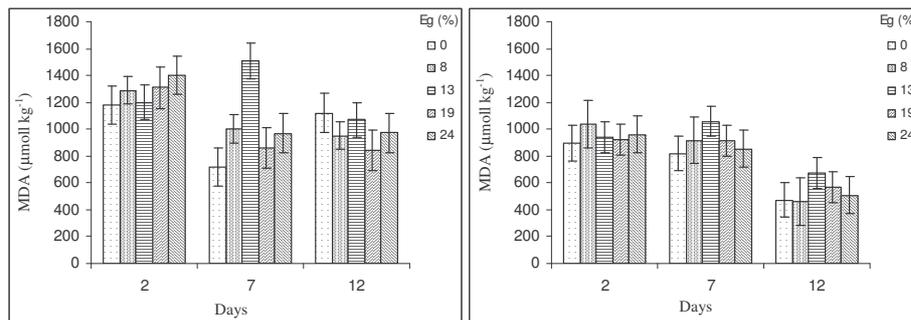
Fig. 8.11. MDA level in roots of *Triticale* – CZR 1406 and *Triticum aestivum* var. Rosa after 2, 7 and 12 days of oxygen stress at 5 levels of air porosity (Eg). Mean value ± standard deviation (After Bennicelli, 2002)
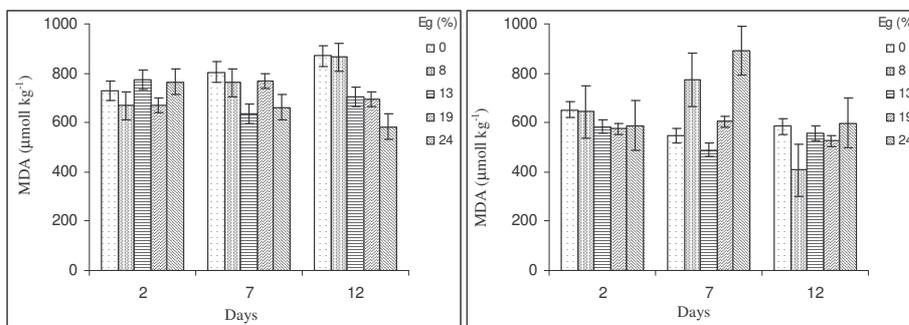
Fig. 8.12. MDA level in leaves of *Triticale* – CZR 1406 and *Triticum aestivum* var. Rosa after 2, 7 and 12 days of oxygen stress at 5 levels of air porosity (Eg). Mean value ± standard deviation (After Bennicelli, 2002)
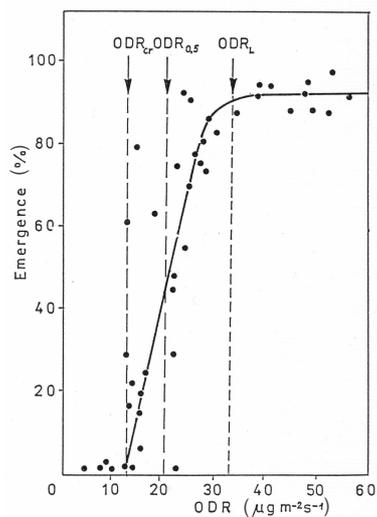
Fig. 8.13. Final emergence of oats versus oxygen diffusion rate (ODR) (After Gliński *et al.*, 1984)

The dependence of emergence of oats seedlings versus oxygen availability (as measured by the indicator called oxygen diffusion rate ODR) is presented in Fig. 8.13. It can be noticed that the response has a typical character with limiting (L) and critical (Cr) values of ODR.

8.3.1. How do plants sense the lack of soil oxygen?

To understand how individual plants will respond to flooding, it is crucial to understand what changes will result in the soil environment due to lack of oxygen.

Animals have evolved specialized respiratory organs, circulation systems, and oxygen-carrying pigments to allow the efficient delivery of oxygen to metabolically active tissues, and to buffer against rapid changes in the rate of oxygen use and resulting changes in the internal oxygen tension. In contrast, plants lack efficient systems for oxygen delivery (Geigenberger, 2003), so that soil waterlogging must be perceived by plants to trigger a cellular signal transduction pathway leading to physiological and morphological changes. In this way, the rhizosphere physical stress (flooding) must be converted into a plant biochemical signal (Bennicelli, 2002).

The identity and the exact mechanism(s) of oxygen sensing remain unclear, although the sensor is most probably a haem (prosthetic group with a central iron atom) or a protein that encloses a haem cofactor (Dat *et al.*, 2004).

8.3.2. Changes in soil chemical properties during soil flooding

An approach to a valuable understanding of the original plant signals reaction is to identify the temporal sequence of soil physicochemical changes and correlate them with plant responses (Dat *et al.*, 2004).

The first event that will take place is in fact the increased presence of H_2O conducing to soil water saturation. The mechanisms which trigger the response are often presumed by-products of root zone flooding (*i.e.* decline in O_2 level). Soil water saturation has dramatic consequences for gas diffusion processes, as gases diffuse 10,000 faster in air than in water. Consequently, one of the main effects of flooding is a lower pool of available O_2, heightened by the rhizosphere microbial respiration in the submerged plant part. Anoxic conditions will develop, leading to a reduction in ATP production and a consequent decrease in root metabolism. The decline in available energy can subsequently reduce other active cellular processes, such as osmotic adjustment, nutrient uptake or regulation of cytoplasmic pH (Gliński and Stępniewski, 1985; Dat *et al.*, 2004).

Three types of soil-plant cell related oxygen conditions are distinguished based on the level of O_2 in the root environment (Tab. 8.2) (Bennicelli, 1992).

During soil flooding there exist different levels of O_2 deficiency and, below a certain threshold concentration, O_2 may not only become the main limiting factor for normal plant development, but probably the prime signal triggering the response. Recent findings suggest the possibility that although cells may not di-

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rectly sense changes in surrounding H_2O levels, internal cellular sensors may detect water homeostasis. Recently, the identification of an *Arabidopsis* trans-membrane hybrid-type histidine kinase which functions as an osmosensor (Urao *et al.*, 1999) could have opened up new routes to explore how flooded root cells detect changes in water homeostasis.

Table 8.2. The soil-plant cell related oxygen stress conditions (From Bennicelli, 1992)

Aeration – Status Relationships		Soil	Plant cell
a	Normoxia	The oxygen supply from atmos-phere to soil pores, roots and microbial fraction is enough to satisfy their requirements	Conditions under which aerobic respira-tion and metabolism proceed normally and most of the ATP is generated via oxidative phosphorylation
b	Hypoxia	Inside the soil aggregates there are aerobic and anaerobic sites simultaneously	Conditions under which the reduction in available O_2 starts to become a limiting factor for ATP production through oxida-tive phosphorylation
c	Anoxia	As a result of long-term inter-rupted atmosphere oxygen diffu-sion, the rhizosphere becomes anaerobic (critical oxygen con-centration – COC – is reached)	Conditions under which ATP is only pro-duced through glycolysis, as no more O_2 is available. As a result, there is a reduction in protein synthesis and other adverse effects on cell division and elongation.

The possibility that a cell uses several cellular indicators of water availability (*i.e.* changes in turgor, cell volume or membrane area, and changes in cell wall-plasma membrane connections or protein-ligand interactions) as potential signalling agents is not novel in cell biology. Osmosensors have long been identified in bacteria and yeast. In plants, soil water saturation drastically alters sap osmotic potential and turgor pressure within a few hours (Jackson *et al*., 1996). Therefore, osmosensors could rapidly perceive and transmit changes in cell water homeostasis and trigger an adap-tive response. However, as with many other stresses, flooding will induce a multitude of other soil physicochemical changes which can serve as environmental signals.

8.3.3. Changes in N, P and Fe availability

Changes affecting soil chemical characteristics during flooding include varia-tions in soil pH and redox potential (Eh) (Gliński, Stępniewski, 1985). As soil becomes reduced, iron and iron oxides can also become reduced, leading to a modification of proton (*i.e.* pH) and cation balances. This process is influenced by the partial pressure of CO_2 which will buffer carbonate, thus lowering pH. The pH modifications may not directly affect plant growth, however undesirable effects

through aluminium or manganese phytotoxicity, calcium deficiency, reduced mineralisation, or reduced turnover of soil organic matter, will drastically alter plant metabolism (Probert and Keating, 2000).

Changes in soil Eh will also accompany the above mentioned transformations. Redox potential will affect the charge conditions of certain clay minerals and may alter their cation exchange capacity. Reduction conditions will also increase metal solubilisation under acidic pH.

The availability of nutrients is influenced to a large extent by the soil redox status (Patrick and Mikkelsen, 1971). A decline in soil redox potential will result in the release of cations through adsorption of ferrous iron on the exchange complexes, whereas phosphorous will be released by dissolution of oxides. NO_3^- will turn into NH_4^+ upon reduction, and NH_4^+ may then be fixed on the soil's cation exchange complexes. The decrease in soil redox potential usually causes decrease in plant nitrogen and subsequent phosphorous contents. In contrast, soil iron and manganese availability will increase, as ferric and manganic forms are reduced to soluble ferrous and manganous forms (Gliński and Stępniewski, 1985).

Sudden increase or restriction of availability of various elements will drastically affect root metabolism. There are numerous studies on the adverse effects of increased levels of phosphorous (P), potassium (K), copper (Cu) and iron (Fe^{3+}) combined with decreased bioavailability of nitrogen (N), sulphur (S) and zinc (Zn) on plant growth (Drew, 1997).

Finally, slowed diffusion of gases in water will hasten the accumulation of potentially phytotoxic by-products of anaerobic metabolism, such as ethanol, lactic acid, CO_2, N_2, H^+, and methane. These may accumulate intracellularly and/or be released in the soil solution and adversely alter soil chemical properties. Several experiments have tentatively tried to address the possibility that the accumulation of some of these by-products may explain some of the anatomical changes observed during flooding (*i.e.* aerenchyma development).

However, other processes (*i.e.* calcium fluxes and ethylene accumulation) have recently been identified in the induction and development of these changes. Therefore, the possibility that these by-products are rapidly perceived as "flooding signals" by the plant is probably trivial.

Because of the alterations in their availability, nutrients such as nitrate, sulphate and iron can act as signals that can not only be perceived by the plant but also trigger molecular responses (Lopez-Bucio *et al.*, 2003). Plant roots move towards nutrients and water, and away from phytotoxic molecules, by sensing signals that notify them of the presence or absence of specific stimuli. The pri-

mary site for perception of underground signals is the root tip, as cells within the apical meristem respond to such diverse signals as electricity, light, ions and water (Aiken and Smucker, 1996). Several root responses to nutrient changes are similar to those commonly observed during flooding. For instance, lateral root initiation and growth are stimulated under low P concentrations (Lopez-Bucio *et al.*, 2002) as well as during submergence.

Nutrient availability and/or restriction are sufficient signals for altering gene expression and thus potential candidates for stimulating some of the flooding responses (Dat *et al.*, 2004).

8.3.4. Potential O_2 – redox sensors: hemoglobin

An inside plant cell oxygen sensing pathway must possess an oxygen sensor capable of detecting changes in ambient O_2 concentrations and triggering a signalling cascade leading to specific transcription factors which in turn will mediate the cellular response. Hemoglobins were identified in the plant kingdom; their widespread presence and long evolutionary history suggest a major role for them in the life of plants (Chaparro-Giraldo *et al.*, 2000; Wittenberg and Wittenberg, 1990).

Hemoglobin is characterized by high O_2 affinity and reversible combination with O_2 in the ferrous state. There are cytoplasmic proteins in higher plants (Sowa *et al.*, 1998; Wittenberg and Wittenberg, 1990). Two types of hemoglobins have been identified in plants: one symbiotic (leghemoglobin), the other not (non-symbiotic hemoglobin) (Tab. 8.3.).

The first was originally isolated in leguminous species and is present mainly in nodules where it helps oxygen transport. The non-symbiotic hemoglobin (Hb) is induced in tissues exposed to hypoxia stress (Taylor *et al.*, 1994).

Table 8.3. The possible roles of non-symbiotic hemoglobin (Hb) under hypoxic conditions (From Dat *et al*., 2004, modified)

	The plant cell hypoxia/anoxia status and the non-symbiotic hemoglobin (Hb) roles:
a	It serves as O_2 carrier to help preserve mitochondrial respiration under anoxic conditions
b	It acts as an electron-transfer protein
c	It serves as an O_2 sensor capable of regulating gene expression under anoxia
d	It sustains glycolytic metabolism in stressed tissues

The high affinity to O_2 (Duff *et al.*, 1997) indicates that the free protein will remain oxygenated at O_2 concentrations below those at which anaerobic processes are activated and makes it an ideal sequester-O_2 molecule under anoxic environments (Sowa *et al.*, 1998).

These results in combination with those obtained with barley (Taylor *et al.*, 1994) support the idea that hemoglobin utilizes the available cellular O_2 to maintain ATP homeostasis.

8.3.5. Nitric oxide

Nitric oxide (NO) is also an ubiquitous signal in plants where inorganic forms of oxidized nitrogen are sources of NO formation in plants. Under certain conditions, NO can be produced by enzymatic and non-enzymatic reduction of nitrate and nitrite. Furthermore, the non-enzymatic formation of NO is favoured at acidic pH. Thus, NO formation in the apoplast depends on acidic pH and on extracellular nitrite, conditions that occur during anaerobiosis. Generation of NO by various species depends on CNR (cytosol nitrate reductase) with nitrite as a substrate (Rockel *et al.*, 2002).

Nitrite can accumulate within the cells only when they are under anaerobic conditions (Botrel *et al.*, 1996) – common root cell conditions during soil oxygen stress due to flooding.

Furthermore, the spatial accumulation of NO is correlated with initiation of cell death in alfalfa roots exposed to hypoxia conditions (Dordas *et al.*, 2003).

The biochemistry of NO involves reactions with free radicals, and reversible binding to metalloproteins or to specific protein amino acid residues. Coupled with hemoglobin and redox homeostasis, NO could be one of the prime signals during flooding, as recently suggested by Dordas *et al.*, (2003) and Durner and Klessig (1999).

8.3.6. Sensing pH changes

The decline in cell pH is one of the earliest cellular events following root hypoxia. Several studies using pH sensitive dyes have shown that changes in cellular pH proceed cell death and aerenchyma formation (Drew *et al.*, 2000). In addition, changes in pH have been linked to ABA in regulating stomata resistance and proposed as a signal during both drought and flooding stress (Wilkinson, 1999). The discovered role of cytosolic pH in regulating water channel proteins (aquaporins) of the plasma membrane indicates a signalling role for cytosolic pH during hypoxia (Tournaire-Roux, 2003).

8.3.7. Mitochondria as sensing sites of oxygen lack

Due to their tight dependence on O_2 homeostasis for normal functioning, mitochondria may play a crucial part in sensing changes in environmental O_2 levels. In fact, O_2 is the final electron acceptor at the terminal end of the respiratory chain

and this process generates reactive oxygen species (ROS) as by-products of respiratory metabolism. The increasing evidence that redox homeostasis is essential in regulating plant stress responses, combined with the emerging evidence that mitochondrial disruption is an early trigger in aerenchyma development, are further indication for redox-derived signalling during soil oxygen stress. Hypoxia plant cell conditions lead to alteration in redox status of the respiratory chain and cause upstream accumulation of electrons confirming the importance of mitochondria during the anoxia response (Chandel *et al.*, 1996; Chandel *et al.*, 2000).

As a result, photosynthesis is reduced, thus considerably altering plant metabolism and growth (Pezeshki, 2001), as such as respiration is impaired because of extremely sensitive to changes in ambient O_2 levels mitochondria, as they are the prime cellular O_2 consumer.

8.3.8. Plant growth regulators and oxygen stress

Flooding characterizes changes in the level of several plant growth regulators.

Ethylene, the most studied, is involved in the stimulation of shoot extension, aerenchyma development and adventitious root initiation. Its biosynthesis increases rapidly (within 4 h) during hypoxia in several species (Drew *et al.*, 1979; Gliński and Stępniewski 1985; Lorbiecke and Sauter, 1999). Ethylene production rate in leaves of flooded plants closely correlates with the induction of 1-amino-cyclopropane-1-carboxylic acid (ACC) synthesis and the accumulation of ACC in the roots (Kende, 1993; Olson *et al.*, 1995).

In tomato plants exposed to flooding, ACC is rapidly synthesized in the roots, and transported to the shoot within 6-12 h (Shiu *et al.*, 1998). Once in the leaves, ACC is converted by ACC oxidase to ethylene, causing epinasty.

Roots under anoxic conditions accumulate ACC, as long as enough ATP is available. Therefore, ACC is transported as a positive message from an environment where oxidation (catalyzed by ACC oxidase) is impossible (the root) to an environment that permits oxidation reactions (the shoot).

Indeed, the biosynthesis of ethylene is inhibited under anoxic conditions because the conversion of ACC to ethylene by ACC oxidase requires oxygen (Peng *et al.*, 2001). Therefore, the increased ethylene production during hypoxia is caused by increased levels of ACC as a result of enhanced ACC synthase activity. In non-aquatic plants, ethylene is often associated with shoot inhibition, leaf wilting and curling – typical flooding responses. In contrast, ethylene is implicated in the promotion of shoot elongation in some semi-aquatic plants. Ethylene is also

considered one of the prime signals required for adventitious root growth in deepwater rice (Lorbiecke and Sauter, 1999; Azuma *et al.*, 1995) and for aerenchyma formation in maize roots (Voesenek *et al.*, 1993).

This role is supported by the inhibition of aerenchyma formation when either the ethylene biosynthesis pathway or the ethylene receptors are blocked (Kende, 1993).

The role of ethylene in plant responses is crucial, but how and where it is positioned in the signalling cascade is still unknown.

In addition to the convincing example of root-to-shoot ethylene signalling events during flooding, there is increasing interest in the role played by abscisic acid (ABA), gibberellic acid (GA), auxin (IAA), and cytokinin (CK).

Foliar ABA concentrations are increased transiently in tomato plants (Else *et al.*, 1995), and a doubling in ABA concentration was measured in the xylem sap of de-topped *Phaseolus vulgaris* during flooding (Newman and Smit, 1991). In addition, exogenous ABA applications increased anoxia tolerance in maize and *Arabidopsis* (Ellis *et al.*, 1999; Hwang and Van Toai, 1991). Synergism between IAA and ethylene has been proposed during adventitious root formation. Adventitious root development at the base of the shoot is an important adaptation to flooding and is initiated soon after submergence. There is also evidence for an increase in GA concentration and sensitivity to ethylene during flooding (Raskin and Kende, 1984; Rijnders *et al.*, 1997). In fact, the synergism between ethylene and GA is believed to increase the responsiveness of rice internodes to GA. When GA inhibitors are applied to rice seedlings, ethylene and submergence-induced growth are inhibited (Raskin and Kende, 1984).

Finally, CK may also participate in the cross-talk during responses to flooding, as sunflower xylem sap CK levels are remarkably reduced following 24 h of flooding (Burrows and Carr, 1969). Soil inundation is believed to reduce CK production by lowering O_2 concentrations at the site of CK production, the root apical meristem.

8.3.9. Plant acclimation to environmental oxygen stress

Plants have evolved a wide range of characteristic responses that appear to reduce the impact of oxygen depletion in the soil. The conditions prevailing in wetlands are an example of such an extreme environment since the highly water-saturated soils exclude oxygen, one of the fundamental requirements for plant life. Oxygen starvation in these soils arises from an imbalance between the slow diffusion of gases in water compared with air, and the rate that oxygen is consumed by

microorganisms and plant roots. The outcome is that flooded soil quickly becomes devoid of oxygen at depths below a few millimetres.

Several acclimations to such conditions can sometimes be found together. For example, plants may develop morphological and biochemical features that are either constitutive or are induced by the flooding event. Several anatomical responses facilitate internal transport of oxygen by diffusion or sometimes by mass flow. This permits underground organs to avoid developing anaerobic interiors.

Of particular importance is the development of aerenchyma (gas-filled channels that can interconnect throughout much of the plant). This creates a low resistance network for the transport of gases from well-aerated aerial shoots to organs engulfed by anaerobic surroundings (Visser and Bögemann 2003)**.** The effectiveness of aerenchyma can be increased by the formation of gas-tight barriers in the epidermis and exodermis in roots that inhibit radial loss of oxygen from roots to the surrounding oxygen-deficient soil (Aschi-Smiti *et al.*, 2003; Colmer, 2003).

When floodwater deepens sufficiently to inundate the shoots as well as the roots, stress on the plants is much magnified. The extra stress arises because the influx of aerial carbon dioxide for photosynthesis is largely prevented. Only a relatively small group of well-adapted aquatic or amphibious species can survive total submergence of the shoot system for long at growing temperatures. The principal strategy for survival is to shorten the period of total submergence by means of a strong increase in the shoot elongation rate that reunites the shoot with air. In most cases this growth requires oxygen, is regulated by a build-up of the plant hormone ethylene, and is mediated via expression of expansion genes (Voesenek *et al.*, 2003; Vriezen *et al.*, 2003). In contrast, a small number of species (*e.g. Potamogeton pectinatus*) are also able to escape by means of accelerated vertical extension growth even in the complete absence of oxygen and independently of ethylene. Taken together, these acclimations help individual plants to survive through improved access to oxygen achieved by accelerated upward shoot growth.

In *Rumex palustris*, the shoot acclimation to submergence consists in keeping the internal oxygen pressure in the petioles above the critical oxygen concentration (COC) for aerobic respiration (Mommer *et al.*, 2004). Even species that are susceptible to poorly aerated conditions possess metabolic and molecular responses that lengthen survival time from a few hours to several days.

All plant species synthesize so-called anaerobic proteins that enable an oxygen-independent energy-generating metabolism to proceed where fermentable substrates are available (Subbaiah and Sachs, 2003). In better-adapted species with large respirable reserves, these fermentation pathways can sustain survival under

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water for many months, and are the means by which aquatic perennials cope with seasonal winter flooding in cool latitudes.

A further group of plants, typical of arctic regions, is able to withstand total anoxia for long periods, even as green plants (Crawford *et al.*, 2003). This long-term metabolic tolerance is seen as an adaptation to ice encasement or submergence in melt water that is common in artic regions which comprise a fifth of the Earth's land surface. The mechanism may involve a highly controlled down-regulation of almost all aspects of metabolism.

Prevention of the build-up of potential phytotoxins is another mechanism that enhances plant survival under flooded conditions. A specific type of hemoglobin (phytoglobin) may play such a role by detoxifying nitric oxide formed during hypoxia of root tissues. Reactive oxygen species (ROS) are possible phytotoxins affecting flooded plants. Low oxygen concentrations and the re-oxygenation that occurs upon retreat of floodwater together favour the generation of ROS. Protection mechanisms against ROS involving chemical and enzymic antioxidant systems are essential traits of flood-tolerant plants (Blokhina *et al.*, 2003), helping to protect lipids and other macromolecules from oxidative damage.

8.3.10. Soil oxygen availability to plants

When evaluating the requirements of crops with respect to soil aeration, one should distinguish between long-term effects of adverse aeration and the effects of temporary flooding. In the first case, there is a change in environment which permanently limits the metabolic activity and development of the root system, impeding the uptake of nutrients, whereas in the second case the effects of a short-term O_2 deficiency or an excess of CO_2 are the main injurious factors. The latter case will be far more complicated to assess because damage done by temporary anaerobic conditions will to a large extent depend on plant species, growing stage, temperature, and the duration of waterlogging. Most agricultural crops grow well if the O_2 levels in soil air are between 5 and 50% and CO_2 levels are below 10% (Gliński and Stępniewski, 1985).

On the interactions of soil oxygen and water stresses upon navy beans pay attention was done by Smucker (1975).

The soil oxygen stress can be determined by monitoring the changes occurring in the physical status of the soil: air-filled porosity (Eg), composition of soil air, permeability to oxygen, and physicochemical parameters such as: oxygen diffusion rate (ODR), redox potential (Eh), as well as the biological parameters such as biological oxygen demand (BOD_5) and dissolved oxygen (DO) (Gliński and Stępniewski, 1985).

Purely on the basis of diffusion in the gaseous phase of soils, minimum air porosity for plant growth can be set. Values between 8 and 15% are mentioned in literature (Wesseling and van Wijk, 1957; Grable and Siemer, 1968). Miller and Johnson (1964) found a lower limit for adequate aeration when 20% of total pore space was air filled.

Critical air porosities will depend on the texture and structure of the soil. When water infiltrates into dry soil, it first enters and fills the smaller pores inside soil crumbs and decreases air-filled porosity without appreciably reducing gaseous diffusivity. Further additions of water begin to fill the larger pores between the crumbs, causing a decrease in diffusivity both by decreasing air phase and by making air-filled pores discontinuous. Possibly critical air porosity can be found by determining the bubbling pressure or air entry value (Grable and Siemer, 1968). In sandy soils this point is found at a suction of 10 to 30 cm, whereas in clay soils the suction amounts to 80 to 150 cm (Gliński and Stępniewski, 1985). Although for specific soils fairly good relationships between air porosity and yield can be found, these relations cannot be extrapolated to other soil types since the specific activity (O_2 use rate) is then not taken into account.

Stolzy and Letey (1964) reviewed literature on plant response to measured O_2 diffusion rate (ODR) values. They concluded that roots of many plants do not grow in soils with ODR values of 35 $\mu g\ m^{-2}s^{-1}$ or less. For germination and emergence of seeds, minimum ODR values in the order of 70-140 $\mu g\ m^{-2}s^{-1}$ are mentioned.

In general, ODR values decrease with water-table depth. On the other hand, McIntyre (1970), reviewing research with platinum micro electrodes, arrived at the conclusion that it is not known what, if any, are the critical values of O_2 flux for root or plant growth. While correlations between root growth and ODR have been established for particular soils and experimental conditions, these correlations cannot, in general, be extrapolated to plant growth in soils differing from those studied.

Low O_2 levels in soils are not necessarily the result of long-term waterlogging. They may occur temporarily, immediately following rainfall or irrigation in slowly draining soils. A decrease in O_2 content may be accentuated under such conditions if readily decomposable organic matter is present or soil temperature is high. As reported by Gliński and Stępniewski (1985), pea's seedlings *(Pisum sativum* L.) after 1 day of waterlogging just prior to the blooming stage reduced final yields by one-third.

From the review given by Grable (1966), it is clear that the effects of temporary O_2 deficiency caused by flooding depend not only on plant species, but also

on the physiological stage of growth, the time and duration of waterlogging, the light intensity, and the temperature and fertility of the soil.

Recently, high sensitivity to soil oxygen stress and the break down of the defence system was shown by Bennicelli (1992) in *Triticale,* on the basis of metabolic plant parameters, where the limiting values between normoxic and anoxic soil conditions such as Eg = 13%, ODR = 27 μg O_2 m^{-2} s^{-1}, and Eh = 400 mV were observed.

8.3.11. Plant response to super-atmospheric oxygen levels

Kader and Ben-Yehoshua (2000) showed in their review that oxygen concentration greater than 21 kPa (induced through high O_2 atmospheres or hyperbaric atmospheres) may influence post-harvest physiology and quality maintenance of fresh horticultural perishables either directly or indirectly via altered CO_2 and C_2H_4 production rates. Fridovich (1986) showed that increased concentrations of O_2 around the plant result in higher levels of free radicals (ROS) that can damage plant tissues. In some plant organs, cyanide-resistant respiration is enhanced by elevated O_2 atmospheres. Ripening of mature-green fruits was slightly enhanced by exposure to 30-80 kPa O_2, but levels above 80 kPa retarded their ripening and caused O_2 toxicity disorders in some fruits, such as bitterness of carrots and russet spotting on lettuce.

9. HUMAN OXYGENOLOGY

Human oxygenology comprises such topics as: oxygen demand, oxygen transport through the blood circulation system and through the skin, anoxic zones within the organism, response to oxygen deficiency and hyper atmospheric oxygen concentration for human ventilation.

In this chapter only some aspects of the above mentioned topics are marked.

Oxygen is of vital importance for human life through the metabolism and functioning of all cells in the body. If we stop with its inhalation, we die after a few minutes (Lane, 2002). An adult man with a weight of 70 kg inhales about 250 ml of oxygen during one minute. The construction of our body ensures a continuous supply of oxygen to each of 15×10^{12} cells of the organism. According to Lane (2000), "Within the lungs, oxygen is taken up by hemoglobin which is packed tightly in the red blood cells circulating through the capillaries. In these capillaries, hemoglobin is usually 95 per cent saturated with oxygen. The pressure exerted by oxygen is about 100 mm Hg. As the blood is transported around the body, hemoglobin gives up its oxygen and so the oxygen pressure begins to fall. As blood leaves the heart, the oxygen pressure has already fallen to about 85 mm Hg; in the arterioles it falls further to about 70 mm Hg; and in the capillary networks in our organs to about 50 mm Hg. Here the saturation of hemoglobin is about 60 to 70 per cent. Oxygen dissociates from hemoglobin and diffuses into the individual cells from the capillaries down a concentration gradient. This gradient is maintained by the continuous removal of oxygen by respiration. In most cells, the oxygen pressure is approximately 1-10 mm Hg. In the final stage, oxygen is sucked into the mitochondria, where active respiration lowers levels even further. The oxygen pressure inside the mitochondria is typically less than 0.5 mm Hg".

Oxygen transport is well described by Stroev (1989) "Diffusion of oxygen from the alveoli pulmonis to the blood is effected owing to the alveolar-capillary partial oxygen pressure drop which is: 13.83 kPa (p_{O2} in alveoli) – 5.98 kPa (p_{O2} in lung capillaries) = 7.85 kPa. Oxygen that has passed through the capillary wall dissolves in the blood plasma and then, across the erythrocytic membrane, penetrates into the erythrocytes to become bound to hemoglobin. One gram of hemoglobin is capable of binding 1.34 ml of oxygen. Taking into account that the blood concentration of hemoglobin is 140-160 g l^{-1}, one will have easily estimated that 1 litre of blood binds a maximum of 180 to 210 ml of oxygen (providing that all available hemoglobin is saturated with oxygen). The quantitative

measure for hemoglobin-bound oxygen in blood is called the *oxygen capacity of blood*. It chiefly depends on the hemoglobin concentration in blood.

During respiration under normal atmospheric conditions, hemoglobin is never saturated completely with oxygen, but to some 95-97% (to attain complete saturation, the O_2 percentage in the inspired air should be equal to 35% as compared to the usual 21%). It follows, therefore, that under normal atmospheric pressure, the oxygen concentration in blood (in the form of oxyhemoglobin) is about 200 ml l^{-1}. A small amount of oxygen (about 3 ml l^{-1}) remains dissolved in blood plasma. Thus, hemoglobin, through binding a 60-fold excess of oxygen, as compared to simple dissolution of O_2 in blood plasma, provides for a 60 times more efficient transport of oxygen to the tissues; otherwise, the bloodstream rate should have been increased by a factor of 50-60. For this reason, the human organism cannot exist without hemoglobin.

Saturation of hemoglobin with oxygen depends upon the partial oxygen pressure. This relationship is represented by a sigmoid curve (S-curve) whose shape, as has been noted previously, is defined by mutual influence of the hemoglobin subunits on their ability to bind oxygen. Owing to this relationship, the oxygen supply of tissues can be ensured at small drops in the partial oxygen pressure: from 13.03 kPa in lung capillaries to 3.99-5.32 kPa in tissue capillaries.

The erythrocytes posses a special regulatory mechanism which varies the affinity of hemoglobin to O_2. These facilities are provided by 2,3-bisphosphoglycerate. Its low concentration in lung capillaries increases the affinity of hemoglobin to O_2 and favours the formation of oxyhemoglobin, *i.e.* the dissociation of oxyhemoglobin at low 2,3-bisphosphoglycerate level is suppressed. An increase in the production of this agent leads to a reverse situation: when 2,3-bisphosphoglycerate binds to the regulatory sites of hemoglobin molecules, it makes the hemoglobin-to-O_2 affinity decrease and facilities the uptake of oxygen by tissues. Actually, this process is observed in the tissue capillaries.

At the arteriolar end of the blood capillary, oxygen diffuses through the capillary wall into the intercellular medium and then enters the cells. Initially, the plasma-dissolved oxygen diffuses, then dissociation of oxyhemoglobin takes place; the released oxygen passes across the erythrocyte membrane, becomes dissolved in blood plasma and transported to the cells' interior. The delivery of oxygen to peripheral tissues leads to a drop in the partial oxygen pressure at the venular end of the capillary and to a loss in oxyhemoglobin concentration from 97% (at the arteriolar end) down to 65-75% (at the venular end). The arteriolar-venular difference in oxygen concentration along the capillary length gives a general idea

of the oxygen requirement of tissues: the higher this oxygen concentration difference, the greater the requirement for oxygen in a given tissue. The release of oxygen from oxyhemoglobin at the arteriolar capillary end is also dependent on oxygen consumption in oxidative reactions within the cells. If, because of certain reasons, the oxygen consumption in tissues becomes smaller, then, owing to its sparing solubility in a liquid medium, oxygen stops to be delivered from the arterial blood channel. In this case, a drop in oxygen arteriolar-venular difference and a high oxyhemoglobin pressure at the venular end are observed.

Muscular organs contain myoglobin which is active in binding oxygen and assists oxyhemoglobin in "unloading" in blood capillaries. Presumably, the mechanical activity of the muscular tissue stands in need of special molecules of myoglobin type, capable of providing an oxygen reserve for continuous oxygen supply to the mitochondria, even in the contingency of a capillary bloodstream stoppage. Probably, non-muscular organs and tissues are also in possession of yet to be discovered oxygen-binding compounds. The acidic proprieties of hemoglobin during oxygen transport are liable to variations. Hemoglobin is a weaker acid than oxyhemoglobin, and for this reason, as oxyhemoglobin is formed, the medium in lung capillary becomes more acidic:

$$HHb + O_2 \rightarrow Hb.O_2 + H^+ \tag{9.1}$$

As the blood pH value is lowered (*e.g.* due to the accumulation of acidic ketone bodies in diabetes mellitus or prolonged starvation), the saturation of hemoglobin with oxygen in the lungs is seen to diminish. In return, the release of oxygen from oxyhemoglobin in tissues is facilitated. An increase in pH leads to the opposite effect".

At normal atmospheric pressure, oxygen accounts for a fifth of breathing air. Breathing oxygen at high concentration is toxic; at about 2 atmospheres of pressure it causes convulsions and even death.

The concentration of oxygen in vein blood is 53 μmol l^{-1} (Bartosz, 1995).

A lack of oxygen in the air causes anoxia and resultant asphyxia. Four stages of oxygen deficit in blood are distinguished (Tab. 9.1).

The most sensitive to anoxia are centres of the cerebral cortex.
The positive role of oxygen in human life is well documented in literature. Also, there is a rich literature concerning the harmful effect of oxygen when it undergoes gradual reduction to create reactive forms when full oxygen reduction creates water molecules during the process of respiration.

Table 9.1. Stages of oxygen deficit in blood (After Seńczuk, 1990, modified)

Stage	Oxygen content in the air %	Symptoms
I	16-12	Increased breathing and as a consequence loss of CO_2 in blood and increase of blood pH
II	12-10	Fast breath and increased fatigue.
III	10-6	Nausea and vomiting, danger of death.
IV	below 6	Apnoea and death.

Some effects of reactive forms of oxygen on cells and their components (Bartosz, 1995):
- Oxidation of low-molecular weight compounds (glutathione, ascorbate, nicotinamide adenine nucleotides).
- Degradation of collagen, loss of ability to gelastinise.
- Depolymerization of hyaluronic acid.
- Deterioration of functions of lung surfactans.
- Oxidation of hemoglobin.
- Inactivation of enzymes.
- Inactivation of transport proteins.
- Proteoglycan synthesis disorders.
- DNA strand ruptures, damage to DNA bases, ribose degradation
- Damage to chromosomes.
- Membranes lipid peroxidation.
- Lysis of erythrocytes.
- Inhibition of oxidative phosphorylation in mitochondria.
- Intracellular Ca^{+2} homeostasis disorder
- Cytoskeleton structure disorders (polymerization of actin, disruption of microfilaments).
- Modification of antigen properties of cells.
- Aggregation of blood platelets.
- Changes in cell morphology (membrane blebbing).
- Mutations.
- Neoplastic transformations of cells.

According to Lane (2002), "Life is not free from the restrictions imposed by the odd chemistry of oxygen, and we too must supply electrons one at a time in order to tap into its reactivity. Cells contrive to break down the oxidation of food into a series of tiny steps, each of which releases a manageable quantity of energy

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that can be stored in a chemical form as ATP. Unfortunately, at each of these steps there is the risk of single electrons escaping from their shackles and joining with oxygen to form superoxide radicals. The continual production of superoxide radicals by calls means that, despite the emotive associations of radiation sickness, breathing oxygen carries a qualitatively similar risk.

Estimates suggest that, at rest, about 1 or 2 per cent of the total oxygen consumed by cells escapes as superoxide radicals, while during vigorous exercise this total might rise to as much as 10 per cent. Lest these figures sound trivial, we should remember that we consume a large volume of oxygen with each breath. An average adult, weighing 70 kilograms, gets through nearly a quarter of a litre of oxygen every minute. If only 1 per cent of this leaks away to form superoxide radicals, we would still produce 1.7 kilograms of superoxide each year. From superoxide we can go to produce peroxide and hydroxyl radicals".

The reactive forms of oxygen react with components of cells, modifying and damaging them. As a result, a lot of diseases appear, such as arteriosclerosis, diabetes, cancers.

All aspects of reactive forms of oxygen and their role for living organisms are widely discussed in a book by Bartosz (1995).

The role of oxygen for humans was last emphasized during the Academia Europaea – Klaus Tschira Foundation workshop on "Reactive Oxygen Species in Health and Disease" held in Heidelberg, 10-12.03.2005.

Ma *et al.* (2004) describe the role of hypoxia in human gene expression: "Hypoxia, the reduction of oxygen supply, results in adaptationally appropriate alterations in gene expression through the activation of hypoxia-inducible factor 1 (HIF-1) to overcome any shortage of oxygen. Thyroid hormones are required for normal function of nearly all tissues, with major effects on oxygen consumption and metabolic rate. Thyroid hormones have been found to augment the oxygen capacity of the blood by increasing the production of erythropoietin (EPO) and to improve perfusion by vasodilation through the augmented expression of adrenomedullin (ADM). Because the hypoxic expression of both genes depends on HIF-1, we studied the influence of thyroid hormone on HIF-1 activation in the human hepatoma cell line HepG2 under normoxic and hypoxic conditions. We found that thyroid hormones increased HIF-1alpha protein accumulation by increasing HIF-1alpha protein synthesis rather than attenuating its proteasomal degradation. HIF-1alpha expression directly correlated with augmented HIF-1 DNA binding and transcriptional activity of luciferase reporter plasmids, whereas HIF-1beta levels remained unaffected. Knocking down HIF-1alpha by short interfering

RNA (siRNA) clearly demonstrated that thyroid hormone-induced target gene expression required the presence of HIF-1. Although an increased association of the two known coactivators of HIF-1, p300 and SRC-1, was found, thyroid hormone did not affect the activity of the isolated COOH-terminal transactivating domain of HIF-1alpha. Increased synthesis of HIF-1alpha may contribute to the adaptive response of increased oxygen demand under hyperthyroid conditions."

Water – the simple hydrogen oxide – is ranked together with oxygen among the most remarkable compounds important for biological activity of living organisms. Water contains oxygen in its structure and also in its molecular form dissolved in it. Most of the molecular oxygen involved in processes of the human organisms is bound to hemoglobin and other transport agents.

Stroev (1989) says: "Water content in the human body, depending on the age, is subject to variation within a range of 45-75% of the total body mass. All water is distributed over three spaces: inside cells, outside cells, and within closed cavities. Most water (from 30 to 45%) is contained in the cell interior; outside the cells, water is distributed among extracellular fluids (12-16%), blood plasma (about 5%), and lymph (2%). The intracavitary water content is rather small (about 1-3%); this water makes part of cerebrospinal, intraocular, pericardial, synovial (in joint cavities) fluids, etc. As to the composition of dissolved therein substances, it resembles the extracellular fluid".

The amount of water received by living organism is smaller than that excreted, which is connected with the creation of its molecules during the cellular oxidation.

10. A LOOK AHEAD

The proposed concept of oxygenology as a new scientific discipline within the environmental sciences emphasizes the exceptional role of oxygen in different elements of the environment: atmosphere, hydrosphere, litosphere and biosphere and especially in human health. This book, being an outline of the subject presents only selected examples of this role with a particular emphasis on soil oxygenology and brief presentation of other research fields.

Taking into consideration the above mentioned aspects as well as expected increase of the importance of the oxygenological issues in the near future, we would like to indicate, among others, the need of:
– elaboration of a comprehensive and interdisciplinary book gathering the available knowledge on the subject
– creation of a journal of oxygenology accelerating the exchange of information
– organization of conferences on oxygenology in order to exchange the experience
– creation of an international institute of oxygenological research for thematic integration of research and trainings towards practical applications of a current oxygenological knowledge.

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