Reactive forms of oxygen
Free radicals
Oxygen-negative influence on humane organism:

Most exposed to oxygen are the lungs and eyes. Breathing pure oxygen under pressure of 1 atm for 6 hours leads to appreciable respiratory disorders.

Lung damage is leading to:

- Damage of pulmonary alveolus.
- Necrobiosis of pulmonary alveolus epithelium
- Increased production of collagen
- Lungs fibrosis

An alveolus (plural: alveoli, "little cavity") is an anatomical structure that has the form of a hollow cavity.
Oxygen - negative influence on humane organism

In 1940 a new dangerous disease was discovered - retinopathy of prematurity. It has been shown that the cause is excessive supply of oxygen to premature babies in incubators. Its occurrence is sufficient during 10 days of contact with the atmosphere containing 35-40% oxygen.

The results are:

- Fibrosis leading to blindness in infants as a result of:
  - contraction of veins in eye
  - necrobirosis of retina blood vessels
  - creating of new blood vessels which grow into vitreous body and as a result causing tear of retina.

- Deafness as a result of bleeding to inner ear
- Damage of central nerves system
Oxygen - negative influence

Negative influence on plants:
- Decreases development of chloroplasts
- Decreases life of seeds
- Decreases development of root system
- Stimuli leafs aging
- Increases probability of growth anomaly
Oxygen – pharmacological application

- CO intoxication
- Cells’ hypoxemia
- Cancer therapy
- MS
- Some lung diseases

Hypoxemia is generally defined as decreased partial pressure of oxygen in blood, sometimes specifically as less as 60 mmHg (8.0 kPa) or causing hemoglobin oxygen saturation of less than 90%.
Certain organ systems are predisposed to greater levels of *oxidative or nitrosative stress*. 

Organ systems most susceptible to damage are:

- **the pulmonary system** (exposed to high levels of oxygen)
- **the brain** (exhibits intense metabolic activity but has lower level of endogenous antioxidants),
- **the eyes** (constantly exposed to damaging UV light),
- **circulatory system** (victim of fluctuating oxygen and nitric oxide levels)
- **reproductive systems** (at risk from the intense metabolic activity of sperm cells).

Nearly every organ system can be found to have an Oxidative or Nitrosative “weak point”. With the current understanding that free radicals can act as cell signaling or “messenger” agents it is likely that they also play a role in normal cellular function as well as various disease etiologies.
Oxygen’s free radicals

**Free radical** - atom or molecule be able to exists by itself, which possess one or more unpaired electrons.

Usually very reactive, can react with other atoms very quickly.

- Biological free radicals are highly unstable molecules that have electrons available to react with various organic substrates.

- Free radicals react with key organic substrates such as lipids, proteins, and DNA. Oxidation of these biomolecules can damage them, disturbing normal functions and may contribute to a variety of disease’s states.
Molecular oxygen is an oxidant - in a reaction with organic compounds oxidizes them by taking electrons. Oxygen is then reduced.

\[ \text{O}_2 + 4e^- + 4 \text{H}^+ \rightarrow 2 \text{H}_2\text{O} \]

Unfortunately, the oxygen molecule not always undergoes, four-electron reduction reduction. Therefore:

\[ \text{O}_2 + e^- \rightarrow \text{O}_2^- \cdot \]

\[ \text{O}_2 + 2e^- + 2\text{H}^+ \rightarrow \text{H}_2\text{O}_2 \]

\[ \text{O}_2^- + e^- + 2\text{H}^+ \]

\[ \text{O}_2 + 3e^- + \text{H}^+ \rightarrow \text{OH}^- \cdot \]

Total reduction, not always happens

ROS

Superoxide

Hydrogen peroxide

Hydroxyl radical
**Reactive Oxygen Species (ROS)**

It’s a term collectively describing radicals and other non-radical reactive oxygen species.

These intermediates may participate in reactions giving rise *to free radicals* or *are damaging* organic substrates.

*ROS in living organisms include the following forms:*

**Radicals:**
- Hydroxyl $\text{OH}\cdot$
- Superoxide $\text{O}_2\cdot^-$
- Nitric Oxide $\text{NO}\cdot$
- Thyl $\text{RS}\cdot$
- Peroxyl $\text{RO}_2\cdot$
- Lipid peroxy $\text{LOO}\cdot$

**Non-radicals:**
- Peroxynitrite $\text{ONOO}^-$
- Hypochloric acid $\text{HOCl}$
- Hydrogen Peroxide $\text{H}_2\text{O}_2$
- Singlet Oxygen $1\Delta g$ ($^1\text{O}_2$)
- Ozone $\text{O}_3$
- Lipid peroxide $\text{LOOH}$
Reactive Nitrogen Species (RNS) - are nitrogen-based radical molecules that can act to facilitate nitrosylation reactions.

**RNS include:**

<table>
<thead>
<tr>
<th>Compound</th>
<th>Formula</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nitrous oxide</td>
<td>NO•</td>
</tr>
<tr>
<td>Nitrosyl cation</td>
<td>NO⁺</td>
</tr>
<tr>
<td>Peroxynitrite</td>
<td>OONO⁻</td>
</tr>
<tr>
<td>Peroxynitrous acid</td>
<td>ONOOH</td>
</tr>
<tr>
<td>Dinitrogen trioxide</td>
<td>N₂O₃</td>
</tr>
<tr>
<td>Nitroxyll anion</td>
<td>NO⁻</td>
</tr>
<tr>
<td>Nitrous acid</td>
<td>HNO₂</td>
</tr>
<tr>
<td>Nitryl chloride</td>
<td>NO₂Cl</td>
</tr>
</tbody>
</table>
FREE RADICAL REACTIONS

**Initiation** – this is a type of the reaction in which FREE RADICALS ARE CREATED FROM MOLECULES NOT BEING FREE RADICALS

*Initiation* of free radical reaction can start at:

- **radiolysis** – decay of water molecules and substances dissolved in water under ionized radiation
- **photolysis** – UV or visible light radiation leads to initiation, ionization or decay of compound
- **sonolysis** of organic compounds – *ultrasound* directed to water solution creates reactive form of oxygen
- one-electron redox reactions
- **homolysis** – decay of initiators' molecules
Elementary process of free radical reaction involves the following steps:

I. INITIATIONS:

\[ H_2 + O_2 \overset{\text{HEAT}}{\rightarrow} 2 \text{OH}^\cdot \]

*Hydroxyl radical*

II. PROPAGATION: (in this reaction sum of free radicals does not change, only atom-carrier changes)

\[ 2 \text{OH}^\cdot + H_2 \rightarrow H_2O + H^\cdot \]

(There are several types of propagation reactions)

III. TERMINATION

\[ R_1^\cdot + R_2^\cdot \rightarrow R_1 - R_2 \]
FREE RADICAL REACTION

II. PROPAGATION – it is exchange of unpaired electron carries, with no change of total amount of free radicals

a) **Transfer** of atoms or group of atoms

\[
\begin{align*}
Q^\cdot + RH & \rightarrow QH + R^\cdot \\
H_3C – CH_2OH + \cdot OH & \rightarrow \begin{cases} 
H_2C – CH_2OH \\
H_3C – \cdot CH_2OH \\
H_3C – CH_2O^\cdot 
\end{cases} + H_2O
\end{align*}
\]
II. PROPAGATION

b) **Addition** of free radical to molecule

\[
Q^* + R - C = C - R_1 \rightarrow R - C - C^* - R_1
\]
II. PROPAGATION

c) **β-elimination** - bond destroying in β position in respect to single electron located in a molecule being transferred to free radical.

\[ R - C - C \rightarrow R + C = C \]
II. PROPAGATION

D) SINGLE ELECTRON REDOX REACTIONS

\[ R^\cdot + Q \rightarrow R + Q^\cdot \]

E) Intramolecular transformation (from less stable, more stable radicals are created)

\[ (\text{Ar})_3C-\cdot\text{CH}_2 \rightarrow (\text{Ar})_2C^\cdot-\text{CH}_2 - \text{Ar} \]

*Ar = rest of aromatic compound*
III. Termination - reaction between two free radicals, ending the propagation process

\[ R_1^* + R_2^* \rightarrow R_1 - R_2 \]

Termination - usually between two free radicals ends the propagation process.

Free radicals termination of important macromolecules may lead to a custom new covalent bond e.g. protein-protein, protein-lipid, protein-nucleic acid that could compromise the biological activity of at least one of the substrates of the reaction.
Natural (cell) sources of free radicals

- **Lysosome**
  - mieloperoxidases
- **Mitochondria**
- **Cytoplasm**
  - Oxidases: hemoglobin, xanthine, riboflavin
- **Oxidases transporting electrons**
  - cytochrome P-450
- **Cell membrane**
  - Prostaglandin synthase, oxidases membrane **NADPH**
Natural (cell) sources of free radicals

**Endogenous** ROS source are numerous biochemical processes running **under physiological conditions**.

- One of the most efficient sources of ROS is the respiratory chain, and many reactions with the participation of oxyreductases.
- Another important source is the microsomal electron transport chain, the cytochrome P-450, participating in the metabolism of xenobiotics.
Natural (cell) sources of free radicals

- Under physiological conditions, 1-5% $O_2$ in the body is converted to ROS which are necessary to normal life processes. They are involved in intracellular signaling, modulate the expression of genes activate transcription, proliferation, apoptosis, control of intracellular homeostasis of calcium ions, are involved in the induction of inflammatory processes and regulate the activity of some enzymes (SOD).

- Intracellular ROS source are mainly the products of reduced by molecular oxygen compunds and many enzymatic reactions.
Other than cell source of free radicals

- Radiation: UV, ionization

- Condition connected with differentiated amount of atmospheric oxygen:
  - Hyperbaric chambers
  - Condition with decreased amount of oxygen

- Environmental pollution such as:
  - ozone, sulphur dioxide
  - nitrate oxide,
  - cigarette smoke

- Food components contaminated with pesticides, preservatives

- Some medicine and chemicals became free radicals in process of enzymatic reaction:
  - paracetamol
Other than cell source of free radicals

- Factors causing physical and emotional stress.
- Xenobiotics in home environment: cosmetics, textile dyes, insulation materials, solvents, wood protecting chemicals, etc.
Hydrogen peroxide – source of free radicals

✓ Undergoes disproportionate reaction:

$$
\text{H}_2\text{O}_2 + \text{H}_2\text{O}_2 \xrightarrow{\text{Metal ions}} 2 \text{H}_2\text{O} + \text{O}_2
$$

✓ Oxidizes -thio, -indol, phenol, -thioester groups

✓ Easily penetrates through the membrane cells

✓ Oxidizes transient metal ions:

$$
\text{Fe}^{+2} + \text{H}_2\text{O}_2 \rightarrow \text{OH}^\cdot + \text{OH}^- + \text{Fe}^{+3}
$$

$$
\text{Cu}^{+1} + \text{H}_2\text{O}_2 \rightarrow \text{OH}^\cdot + \text{OH}^- + \text{Cu}^{+2}
$$
Superoxide anion \( O_2^- * \) more reactive than oxygen
exists in balance with

Perhydroxyl radical \( \text{HO}_2^* \)
(protonated form, in the lysosomes and near cell membranes)

**Disproportionation reactions:**

\[
\begin{align*}
O_2^- * + O_2^- & \rightarrow H_2O_2 + O_2 \\
\text{HO}_2^* + \text{HO}_2^* & \rightarrow H_2O_2 + O_2 \\
\text{HO}_2^* + O_2^- & \rightarrow H_2O_2 + O_2
\end{align*}
\]
Superoxide anion/perhydroxyl radical - similarities and differences

<table>
<thead>
<tr>
<th></th>
<th>Superoxide anion</th>
<th>Perhydroxyl radical</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Reactivity</strong></td>
<td>fairly large</td>
<td>stable</td>
</tr>
<tr>
<td><strong>Reactivity with anions</strong></td>
<td>difficult</td>
<td>easy</td>
</tr>
<tr>
<td><strong>Membrane transport</strong></td>
<td>difficult</td>
<td>easy</td>
</tr>
<tr>
<td><strong>Initiation of lipid peroxidation</strong></td>
<td>not possible</td>
<td>easy</td>
</tr>
</tbody>
</table>
Hydroxyl radical – source of free radicals

✓ one of the strongest oxidizing agent
✓ has low energetic barrier
✓ has low reaction specificity

Main reactions:

1. Detaching hydrogen atom from alkanes

\[
\text{H}_3\text{C} - \text{CH}_2\text{OH} + \text{OH}^\bullet \rightarrow \begin{cases} 
\text{H}_2\text{C}^\bullet - \text{CH}_2\text{OH} \\
\text{H}_3\text{C} - \text{•CHOH} \\
\text{H}_3\text{C} - \text{CH}_2\text{O}^\bullet 
\end{cases} + \text{H}_2\text{O}
\]

2. Addition to double bond (e.g. oleic acid)

\[
\text{H}_3\text{C} - (\text{CH}_2)_7 - \text{HC} = \text{CH} - (\text{CH}_2)_7\text{COOH} + \text{OH}^\bullet \rightarrow \\
\text{H}_3\text{C} - (\text{CH}_2)_7 - \text{HC} - \text{CH} - (\text{CH}_2)_7\text{COOH}
\]

\[\text{OH}\]
Other active forms of oxygen

- Allotropic form of oxygen – ozone $\text{O}_3$ – present in atmosphere, absorbs UV radiation, very toxic.

- Nitrate oxide $\text{NO}^+$ formed in cells with nitric oxide synthetasis. In cigarette fume oxidizes to nitric dioxide radical, taking part of process generating carbon and i ROO· radicals.

- $^1\text{O}_2$ - excited oxygen is formed during peroxide decay. Exposed to light oxidizes cholesterol, damages aminoacids: histidine, methionine, tyrosine, cysteine.

In *addition reaction* for alkenes and multi-unsaturated fatty acids creates peroxides.
Other active forms of oxygen

- **Peroxynitrous acid, HONO$_2$** - in reactions catalyzed by metal ions, react with -SH groups of proteins. Donor of NO$_2$ group in the nitration reaction of unsaturated fatty acids.

- **Hypochlorous acid, HOCl** - produced mainly by neutrophils in a reaction catalysed by myeloperoxidase (MPO), oxidizes thiol groups (-SH) of peptides and GSH (reduced glutathione).
Nitrate oxide NO$^\cdot$

- Combination of:
  - Oxygen atom with eight electrons and
  - Nitrate atom with seven electrons

- Reacts with proteins containing:
  - Fe-S centers
  - transient metal ions
  - hem groups

- Unstable in presence of oxygen

$$2\text{NO}^\cdot + \text{O}_2 \rightarrow 2\text{NO}_2^\cdot$$

In human organism is created with participation of enzymes
Dioxide nitrate $\text{NO}_2^\cdot$

$$2\text{NO}^\cdot + \text{O}_2 \rightarrow 2\text{NO}_2^\cdot$$

Reacts with unsaturated compounds creating free radicals on carbon atom

$$\text{NO}_2^\cdot + R - \text{C} = \text{C} - R_1 \rightarrow R - \text{C} - \text{C}^\cdot - R_1$$

$R_1 = \text{Organic radicals created in reaction with oxygen:}$

- peroxide radicals $\text{ROO}^\cdot$
- alkoxy radicals $\text{RO}^\cdot (+ \text{NO}_2)$

**In water disproportionate reaction**

$$2\text{NO}_2^\cdot + \text{H}_2\text{O} \rightarrow \text{NO}_2^- + \text{NO}_3^- + 2\text{H}^+$$
Peroxynitrite – it is not a free radical !!!

Nitric oxide reacts with superoxide:

\[
\dot{\text{NO}} + \dot{\text{O}}_2^- \rightarrow \text{ONOO}^-
\]

It has short half-life - 1s

It has strong oxidizing properties:

- reacts with the thiol groups of proteins
- reacts with polyunsaturated fatty acids
- nitrates tyrosine residues in proteins (reaction catalysts - the transition metals)
- inhibits the activity of cytochrome oxidase and other components of the respiratory chain
- reacts with \( \text{HCO}_3^- \) anion creating radical carbonate \( \text{HCO}_3^- \) *
Influence of reactive forms of oxygen on cells:

- Oxidation of low molecular compounds
- Collagen degradation
- Hemoglobin oxidation
- Inactivation of enzymes and transporting proteins
- Damage in DNA, ribose degradation
- Chromosomes damage
- Peroxidation of membrane’s lipids
- Modification of cell’s antigen properties
- Transformation of cancer cells
- New mutation
- Erythrocyte lyses
Influence of reactive forms of oxygen on human organism

- Heart diseases, heart attack
- Atherosclerosis
- Autoaggression diseases:
  - Rheumatologic joint inflammation
  - Autoimmune diseases
  - Diabetes
  - Muscular dystrophy
- Flue complications
- Eye diseases – cataract, retinopatia, glaucoma
- Neurological diseases – Alzheimers’, Parkinsons’, Down syndrome, multiple sclerosis (MS)
- Stomach and duodenum ulcer
Increased production of free radicals  
**OXIDATIVE STRESS**

- **Oxidative stress** is caused by an imbalance between the production of reactive oxygen and a biological system's ability to readily *detoxify* the reactive intermediates or easily *repair* the resulting damage.

- Oxidative stress occurs when the generation of ROS in a system exceeds the system’s ability to neutralize and eliminate them.

- The imbalance can result from a *lack of antioxidant capacity* caused by disturbance in production, distribution, or by an over-abundance of ROS from an environmental or behavioral stressor. If not regulated properly, the excess ROS can damage a cell’s lipids, protein or DNA, inhibiting normal function.

- Oxidative stress has been implicated in a growing list of human diseases as well as in the aging process.

- **Antioxidants** should prevent oxidative stress.
Reperfusion

Reperfusion injury refers to damage of tissue when blood supply returns to the tissue after a period of ischemia.

The absence of oxygen and nutrients from blood creates a condition in which the restoration of circulation results in inflammation and oxidative damage through the induction of oxidative stress rather than restoration of normal function.

Ischemia – restriction in blood supply
Reperfusion

*Example*: after a heart attack an immediate goal is to quickly open blocked arteries and reperfuse the heart muscles. Early reperfusion minimizes the extent of heart muscle damage and preserves the pumping function of the heart.

Reperfusion exists at the following conditions:

- ✓ Obstructed blood vessels
- ✓ Organ transplantation
- ✓ After surgery
- ✓ Limb numbness
- ✓ Long lasting physical effort
Defense systems against free radicals

I. Prevention of hydroxy radical creation
   a. removing reactions’ substrates: hydrogen peroxide and anion-radical
   b. bonding of transient metal ions (limiting continuation of Fenton’s reaction)

II. Inactivation of free radicals by:

   - Enzymatic systems
   - Low molecular antioxidants
   - Repair of damaged genetic material

III. „The third line of defense“ - the elimination or repair of damaged material (genetic)
Defense systems against free radicals

**Proteins defending against oxygen’s free radicals:**
The most important are contained in the erythrocyte enzyme systems such as:

1. **Superoxide dismutase (SOD)** – catalyzes superoxide to oxygen
   
   \[
   \begin{align*}
   O_2^\cdot + O_2^\cdot - + 2H^+ & \underset{\text{SOD}}{\rightarrow} H_2O_2 + O_2 \\
   \end{align*}
   \]

2. **Catalase (CAT)**

   *CAT acts when large quantities of H\textsubscript{2}O\textsubscript{2} are created*

   \[
   \begin{align*}
   2 H_2O_2 & \underset{\text{CAT}}{\rightarrow} 2H_2O + O_2 \\
   \end{align*}
   \]
Defense systems against free radicals

Proteins defending against oxygen’s free radicals:

3. Glutathione peroxidase (GSH-P)

Glutathione disulfide

Disulfide bridge

HSe-CH₂-CH-COO⁻ +NH₃

GSH-Px (selenium-dependent enzyme)

\[ 2 \text{GSH} + \text{H}_2\text{O}_2 \rightarrow \text{GSSG} + 2 \text{H}_2\text{O} \]

Glutathione disulfide

\[ \text{protein} – \text{SH} + \text{GSSG} \rightarrow \text{protein} – \text{S} – \text{SG} + \text{GSH} \]

\[ \text{protein} + \text{GSSG} \rightarrow \text{protein} + 2 \text{GSH} \]

Disulfide group can be oxidised to disulfide bridge

protein disulfide

Disulfide bridge
Defense systems against free radicals

Proteins defending against oxygen’s free radicals

4. **Glutathione reductase** (GR)

\[
\text{GSSG} + \text{NADPH} + \text{H}^+ \xrightarrow{\text{glutathione reductase}} 2 \text{GSH} + \text{NADP}^+
\]

- uses NADPH as a reducing agent (electron emitter)
- it is present in the cytosol and mitochondria
- main function - maintaining normal levels of GSH in cells by converting GSSG to GSH

[The NADPH oxidase (nicotinamide adenine dinucleotide phosphate-oxidase) is a membrane-bound enzyme complex that faces the extracellular space.]
Defense systems against free radicals

III. Low molecular weight antioxidants

1. Hydrophilic antioxidants:
   - Ascorbic acid – vitamin C
   - Uric acid
   - glutathione

2. Hydrophobic antioxidants:
   - Vitamins E, A
   - Carotenoids (organic pigments such as chloroplast and chromoplasts) - xanthophil
   - Lycopene
Glutathione (GSH)

- the most common low molecular weight thiol compound in nature;
- possesses free thiol group
- it occurs in all prokaryotic and eukaryotic cells
- thiol group (SH) of reduced GSH can rapidly oxidize and oxidized form is created - disulphide glutathione

\[ 2 \text{GSH} + \text{H}_2\text{O}_2 \rightarrow \text{GSSG} + 2 \text{H}_2\text{O} \]
Low molecular weight hydrophilic antioxidant

- Ascorbic acid – vitamin C

Has very reductive properties in respect to:

- hydroxyl radical,
- hydrogen peroxide,
- peroxide radicals

In high concentration destroys free radicals and regulates redox potential

In small concentration can lead to pro-oxidative properties in Fenton's reaction which was demonstrated in vitro cultures of tumor cells
Low molecular weight antioxidant system

Ascorbic acid – vitamin C

- in the body fluids is almost completely dissociated (99%) and occurs in the form of ascorbate anion (AH-)

- ascorbate (AH-) has reducing properties: with oxidizing agents by the one-electron reduction reaction can form a free radical ascorbyl (A* -), with low chemical reactivity

- in two electron reduction reaction dehydroascorbic acid (DHA) is formed

- DHA is unstable and may decay to 2,3-dioxo-L-gulonic acid which is susceptible to further oxidation, leading to the formation of oxalic acid and L-threonic acid
H.J.H Fenton discovered in 1894 that several metals have a special oxygen transfer properties which improve the use of hydrogen peroxide. Actually, some metals have a strong catalytic power to generate highly reactive hydroxyl radicals (OH•).

**Fenton reaction:**

\[
\text{Fe}^{2+} + \text{H}_2\text{O}_2 \rightarrow \text{Fe}^{3+} + \text{OH}^+ + \text{OH}^-
\]

This is the iron-salt-dependent decomposition of dihydrogen peroxide, generating the highly reactive hydroxyl radical. Addition of a reducing agent, such as ascorbate, leads to a cycle which increases the damage to biological molecules.
Uric acid - the final product of purine nucleotide degradation in *humans and great apes*

- combines Fe ions (discontinuation of Fenton reaction)
- reacts with oxidants
- urinary radical is stable

In *other mammals*, the final product of purine nucleotide metabolism secreted in the urine is allantoin.
Low molecular weight hydrophobic antioxidant

Bilirubin

• in mammals Bilirubin is produced by the degradation of part of the porphyrin heme

• heme degradation leads to creation of biliverdin (greenish color), which is then reduced to bilirubin with the enzyme - biliverdin reductase and NADPH (proton donor)
Low molecular weight **hydrophobic antioxidant**

Bilirubin as an antioxidant:

- it is one of the antioxidants present in plasma and in cell membranes
- protects against peroxidation linoleic acid (one molecule of bilirubin protects at least 100 molecules of acid)
- deactivates singlet oxygen
- coupled with glucuronate reacts with an acid HClO
- reacts with peroxide radicals:

\[ \text{ROO}^\cdot + \text{Brb} \rightarrow \text{ROOH} + \text{Brb}^\cdot \]

\[ \text{Brb}^\cdot + \text{ROO}^\cdot \rightarrow \text{Brb-OOR} \]

\[ \text{Brb}^\cdot + \text{O}_2 \rightarrow \text{Brb-OO}^\cdot \]
Retinol (vitamin A)
• the antioxidant properties of vitamin A are due to the presence of the conjugated C = C bonds system in the side chain of the molecule

• main form of vit. A is β-trans-retinol - in plasma associated with LDL
• scavenger of singlet oxygen and LOO• radical

Low molecular weight hydrophobic antioxidant
Tocopherols : vitamin E

- scavenges of secondary organic free radicals
- participates in the termination of lipid peroxidation
- reacts rapidly with organic peroxide radicals
- reacts with singlet oxygen (I$^{st}$ line of defense)
Low molecular weight hydrophobic antioxidant

- **carotenoids:**
  - α-carotene
  - β-carotene
  - γ-carotene
  - Lycopene

- **xantophylls** (oksycarotenoides)
  - lutein
  - zeaxanthin
  - cryptoxanthin
Carotenoids:

- Belong to isoprenoid group and are present in over several hundreds compounds, in which 50 are in food.
- Differ from each other by double bond location and presence of acyclic ring in molecule.
- α, β i γ -caroten have provitamin-A activity and are included in nutritious components of food.
- Lycopene is natural substance with no-nutritious properties.
Low molecular weight hydrophobic antioxidant

Xanthophylls:

- oxygen derivatives of carotenes (hydroxyl, ketone, aldehyde, carboxylic).
- They are created during carotenoids oxidation with enzymes.
- Deactivates singlet oxygen.
- They react with organic radicals, arising in the process of lipid peroxidation.
- They are, in addition to carotenoids, in all plant tissues (accounting for 60-70% of the carotenoid compounds) and animals.
Low molecular weight hydrophobic antioxidant

Xanthophylls cont.:

- Lutein - 3,3'-dihydroxy derivative of α-carotene - is present in virtually all tissues of green plants
- Zeaxanthin - isomer of lutein - isolated from corn seed
- Cryptoxanthin - orange dye in peppers and oranges
- Fucoxanthin - a pigment involved in photosynthesis of kelp
- Capsorbin and capsanthin - the main dyes of peppers
- Astaxanthin - dye armor crustaceans, sea urchins, feathers and bird legs
Colorful antioxidants

- **Tomato** - carotenoids, especially lycopene, vitamin C

- **Spinach, kale, beetroot** - vit. B, folic acid (lowers homocysteine levels), phytochemicals - lutein, zeaxanthin

- **Broccoli** - phytochemicals (indol-3-carbinol), β-carotene, vitamin C, dietary fiber. Cruciferous plants reduces the incidence of breast, stomach, colon cancer

Colorful antioxidants

- **Nuts** - vit. E, mono- and polyunsaturated improve levels of "good" cholesterol. Italian nuts contain ellagic acid and initiate the process of apoptosis.

- **GREEN TEA** - polyphenols called catechins, (Antioxidant power 100 times greater than vit. C), stops the supply of nutrients to tumors at an early stage of development.

- **Red wine** - polyphenols, among others, resveratrol increase the level of HDL.

- **Fish** - Salmon, Mackerel, fish oil.
The End