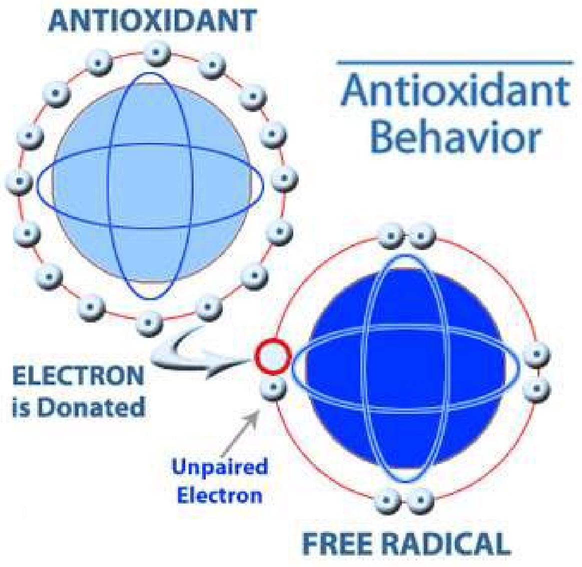
In the name of God

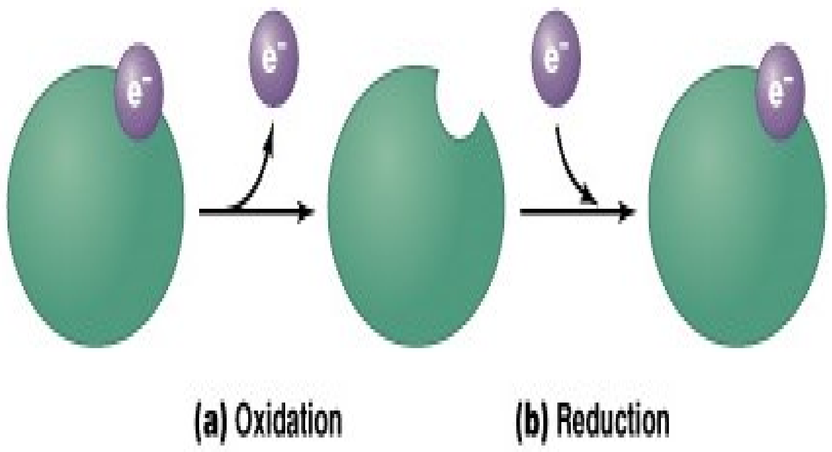
Antioxidants

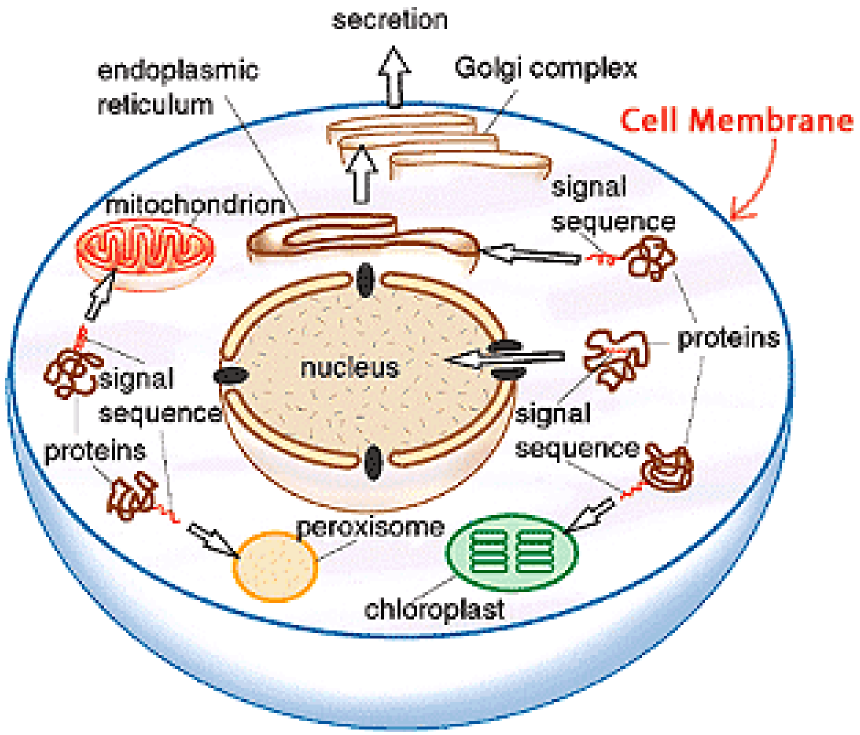
An **antioxidant** is a molecule capable of inhibiting the oxidation of other molecules. Oxidation is a chemical reaction that transfers electrons or hydrogen from a substance to an oxidizing agent. Oxidation reactions can produce free radicals. In turn, these radicals can start chain reactions and damage cells.

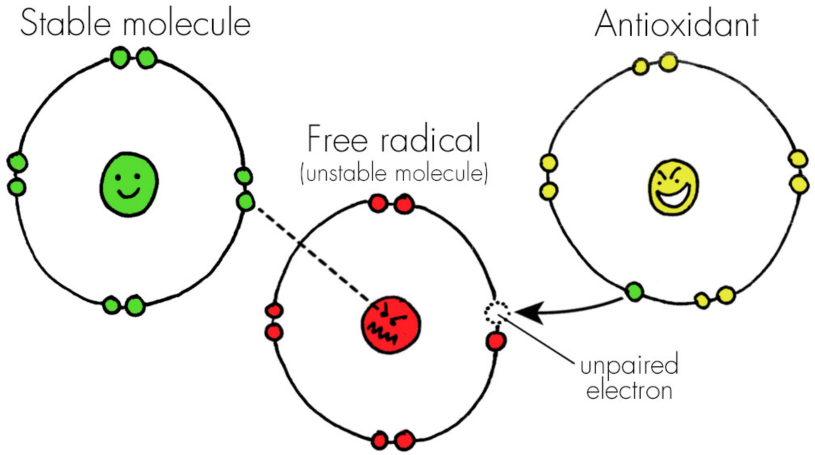
Antioxidants terminate these chain reactions by removing free radical intermediates and inhibit other oxidation reactions.

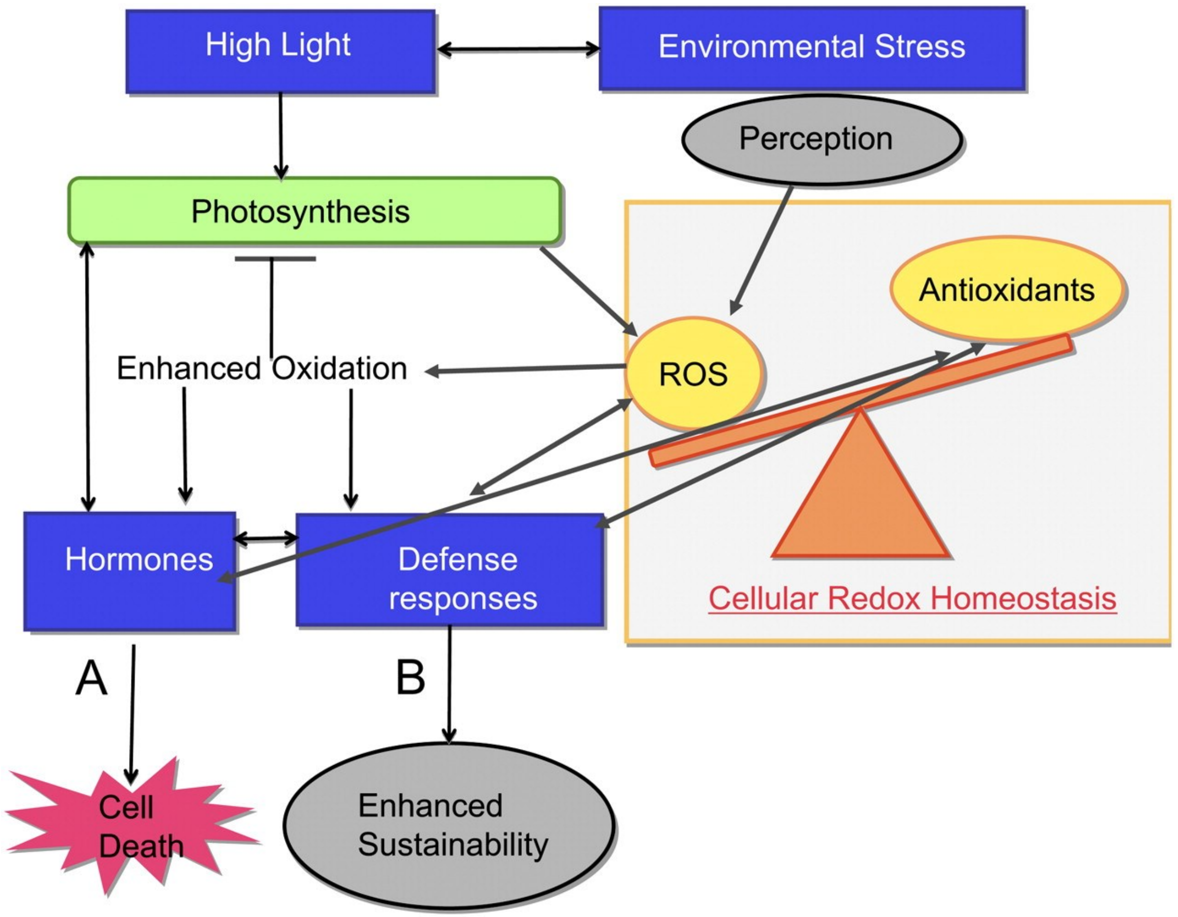


Term	Definition
Oxidation	Gain in oxygen Loss of hydrogen Loss of electrons
Reduction	Loss of oxygen Gain of hydrogen Gain of electrons
Oxidant	Oxidizes another chemical by taking electrons, hydrogen, or by adding oxygen
Reductant	Reduces another chemical by supplying electrons, hydrogen, or by removing oxygen

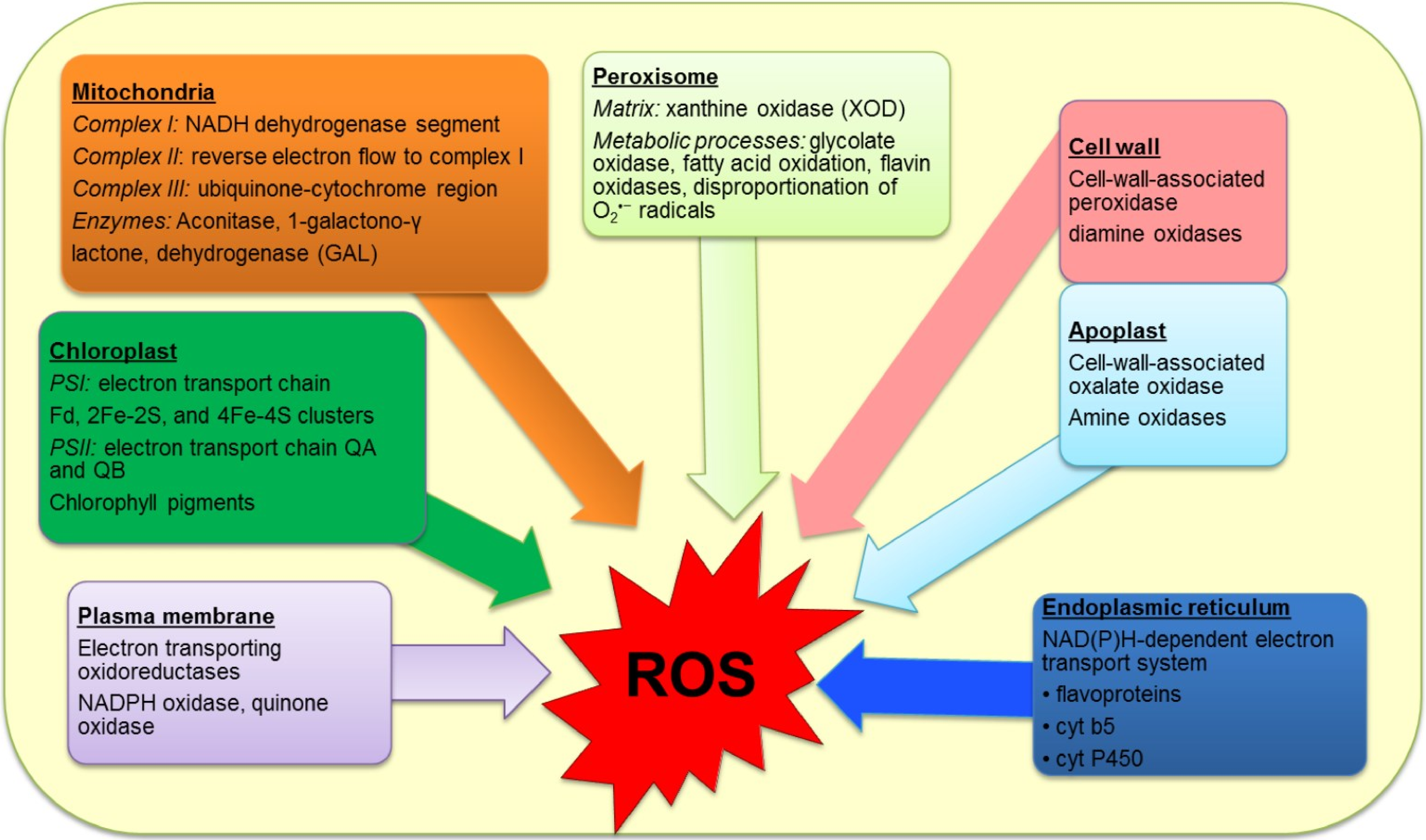








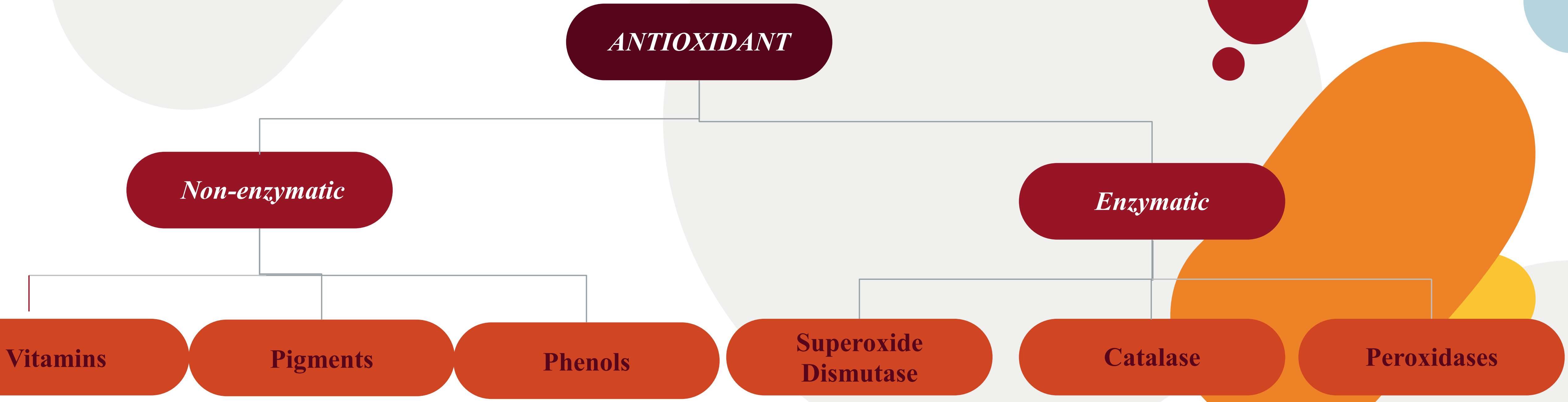
Both chloroplasts and mitochondria are major sources of ROS production either under normal growth conditions or during exposure to various stresses.



Mitochondria
Complex I: NADH dehydrogenase segment
Complex II: reverse electron flow to complex I
Complex III: ubiquinone-cytochrome region
Enzymes: Aconitase, 1-galactono-γ lactone, dehydrogenase (GAL)
Peroxisome
Matrix: xanthine oxidase (XOD)
Metabolic processes: glycolate oxidase, fatty acid oxidation, flavin oxidases, disproportionation of $O_2^{\bullet-}$ radicals
Cell wall
Cell-wall-associated peroxidase
diamine oxidases
Chloroplast
PSI: electron transport chain
Fd, 2Fe-2S, and 4Fe-4S clusters
PSII: electron transport chain QA and QB
Chlorophyll pigments
Apoplast
Cell-wall-associated oxalate oxidase
Amine oxidases
Plasma membrane
Electron transporting oxidoreductases
NADPH oxidase, quinone oxidase
ROS
Endoplasmic reticulum
NAD(P)H-dependent electron transport system
• flavoproteins
• cyt b5
• cyt P450

Harvested horticultural crops undergo postharvest stress conditions somewhat similar to stresses that plants experience in the field. These include wounding or bruising from the harvesting and sorting process, water stress from water loss in storage, temperature stress from cold storage conditions, and anaerobic stress from controlled or modified atmosphere storage. These stresses are superimposed on the normal ripening or senescence process of the fruit or vegetable, which also produce ROS in the tissue. Therefore, to minimize the potentially deleterious effects of storage conditions, ripening, and senescence, it is important to maintain the antioxidative processes in the tissue.

Mode of action



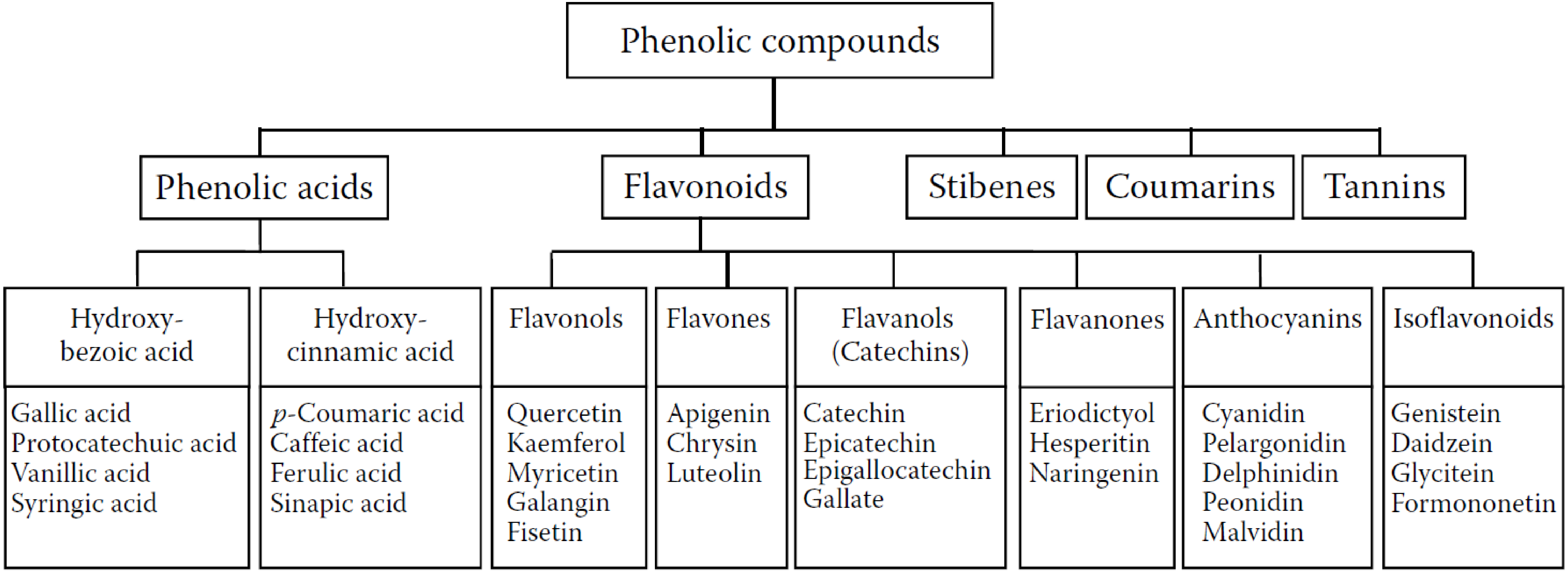
Solubility

- Antioxidants
  - Lipid-soluble
    - Tocopherols
    - Carotenoids
  - Water-soluble
    - Glutathione
    - Ascorbate
    - Flavenoids

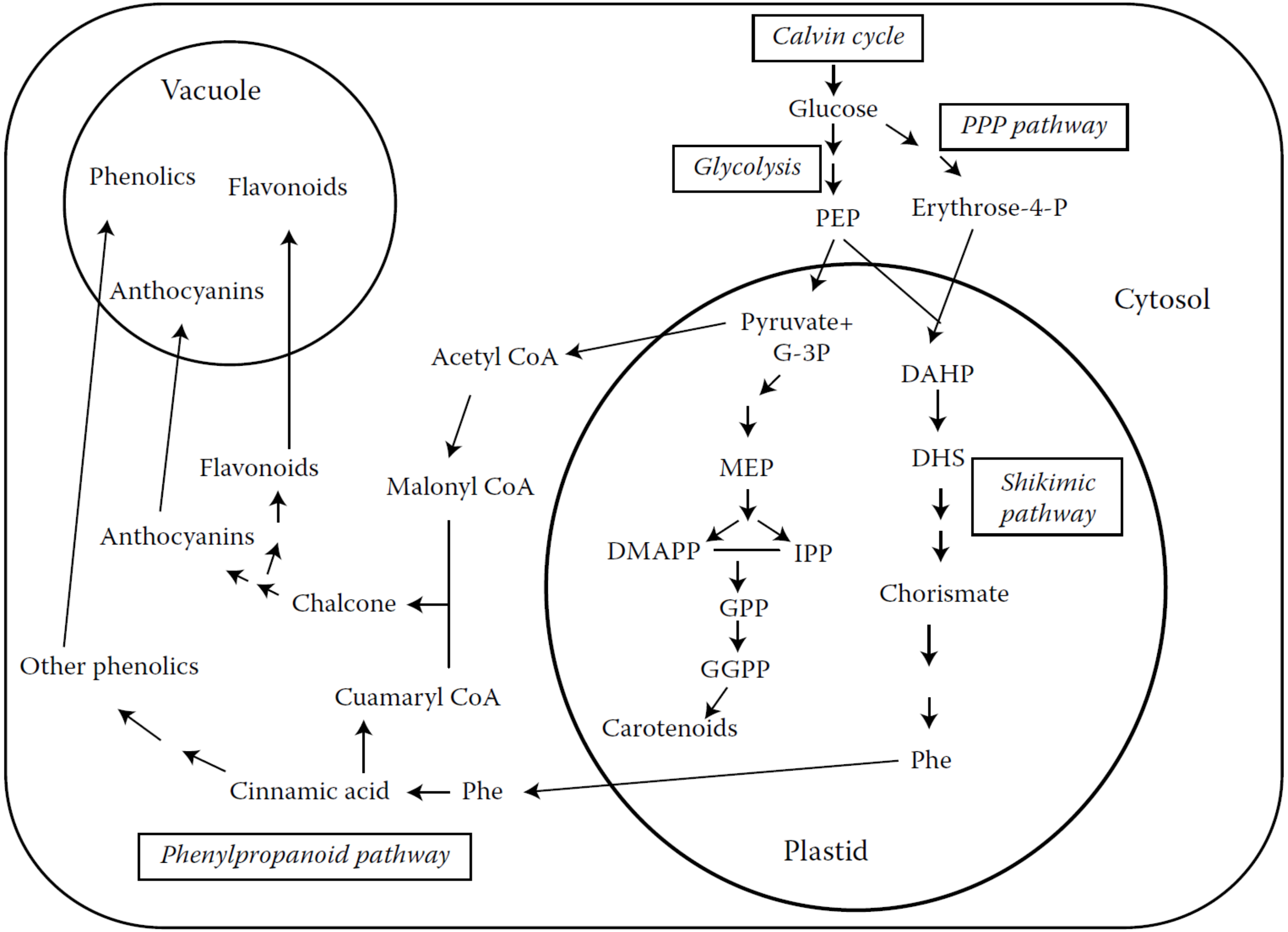
Phenolics

Plants contain various phenolic compounds exhibiting a good source for antioxidants. The activities depend on their chemical structures and the experimental systems.

Phenolics such as monophenolics, hydrolyzable tannins, tannins, and flavonoids are ubiquitously found in fruits and vegetables, whereas mammalians cannot produce such chemicals. For example, pineapple fruit contains eight main phenolic compounds, that is, gallic acid, gentisic acid, syringic acid, vanillin, ferulic acid, sinapic acid, isoferulic acid, and o-coumaric acid.


Phenolic compounds
Phenolic acids
Flavonoids
Stibenes
Coumarins
Tannins
Hydroxy-bezoic acid
Gallic acid
Protocatechuic acid
Vanillic acid
Syringic acid
Hydroxy-cinnamic acid
p-Coumaric acid
Caffeic acid
Ferulic acid
Sinapic acid
Flavonols
Quercetin
Kaemferol
Myricetin
Galangin
Fisetin
Flavones
Apigenin
Chrysin
Luteolin
Flavanols (Catechins)
Catechin
Epicatechin
Epigallocatechin Gallate
Flavanones
Eriodictyol
Hesperitin
Naringenin
Anthocyanins
Cyanidin
Pelargonidin
Delphinidin
Peonidin
Malvidin
Isoflavonoids
Genistein
Daidzein
Glycitein
Formononetin

Biosynthesis and localization of some antioxidants in a plant



Functional Group	*Alternative Name*	*Major Example*	*Predominant Color*
Tetrapyrroles	Porphyrin derivatives	Chlorophylls	Green
Tetraterpenoids	Carotenoids	Carotenes	Yellow–red
		Xanthophylls	Yellow
O-Heterocyclic compounds (flavynium ion)	Flavonoids	Anthocyanins	Blue–red
		Flavonols	Yellow–white
		Flavones	White–cream
Quinones	Phenolic compounds	Tannins	Brown–red
N-Heterocyclic compounds	Indole derivatives	Betalains	Yellow–red

	Carotenoids Content (μg/100 g Fresh Weight)				
	Oxygenated Carotenoids		Nonoxygenated Carotenoids		
Produce	*Lutein*	*Cryptoxanthin*	*Lycopene*	*α-Carotene*	*β-Carotene*
Banana	0–37	–	–	0–157	0–92
Guava	270	–	769–1816	–	102–2669
Mango	100	0–1640	–	–	300–4200
Jackfruit	–	–	37–111	–	40–772
Orange	64–350	14–1395	–	0–400	0–500
Papaya	20–820	60–1483	2080–4750	0–60	71–1210
Pineapple	–	–	–	–	171–476
Tomato	40–1300	–	21–62273	–	36–2232
Watermelon	0–40	62–457	2300–7200	0–1	44–324

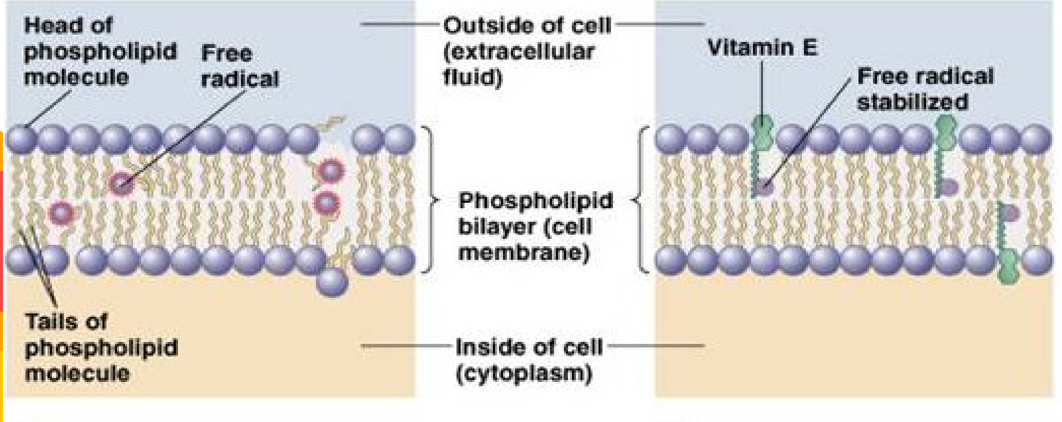
Carotenoids

In most plant tissues, carotenoids occur in the chloroplast membranes and help prevent oxidative damage from ROS generated by the absorption by chlorophyll of more light energy than can be transferred to the photosynthetic electron transport chain.

Chloroplast membranes are rich in the polyunsaturated fatty acid linolenate which can easily become peroxidized. Carotenoids are very powerful quenchers of ROS and at relatively low concentration -carotene can effectively protect membrane lipids from Oxidation. In this case, the carotenoids are concentrated in chromoplasts, not chloroplasts, and it is unclear how much they contribute to antioxidative protection of the plant tissue, although they may be important for nutritional health.

Tocopherols

- Tocopherols are amphipathic molecules; the hydrophobic phytl tail is located in a membrane, associated with the acyl chains of fatty acids or their residues, whereas the polar chromanol head group lies at the membrane cytosol interface where it can interact with other cytosolic molecules.
- Maintenance of high–tocopherol levels in fruit tissue can help prevent injury from water or temperature stress by preventing membrane damage. There is evidence that the ratio of vitamin E to vitamin C (tocopherol to ascorbate) changes during senescence. During leaf aging, -tocopherol transiently increases in membranes, but ascorbate decreases and the consequence is lipid oxidation.



Ascorbate

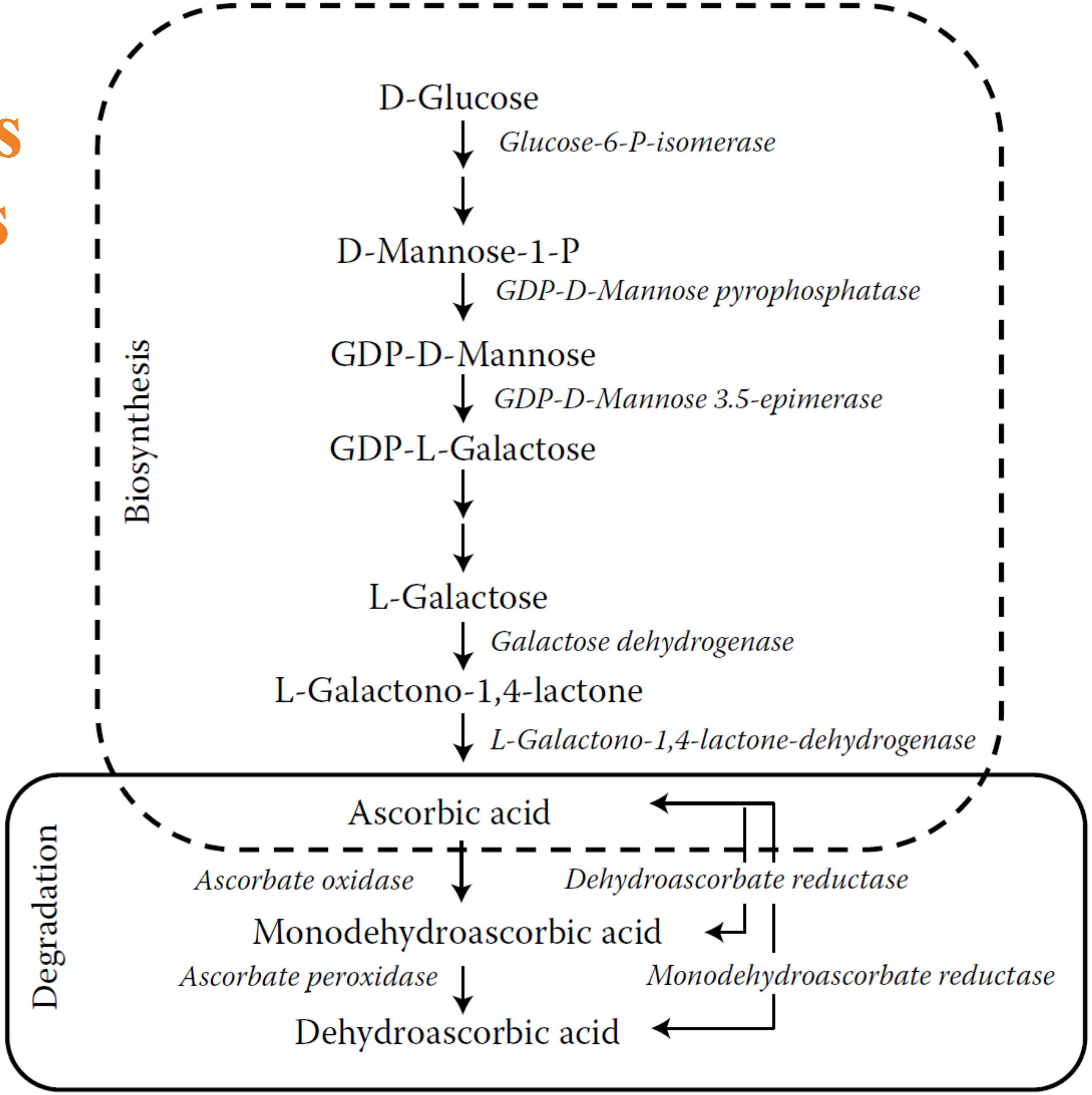
L-Ascorbic acid or vitamin C is an essential nutrient for both plant and human health since it is responsible for free radical scavenging. AsA presents naturally in many tropical fruits in which some of them contain vitamin C in abundance such as guava and gac fruit.

Ascorbate plays a major role in the prevention of peroxidative damage by scavenging ROS and, as a consequence, producing its own free radical, monodehydroascorbate.

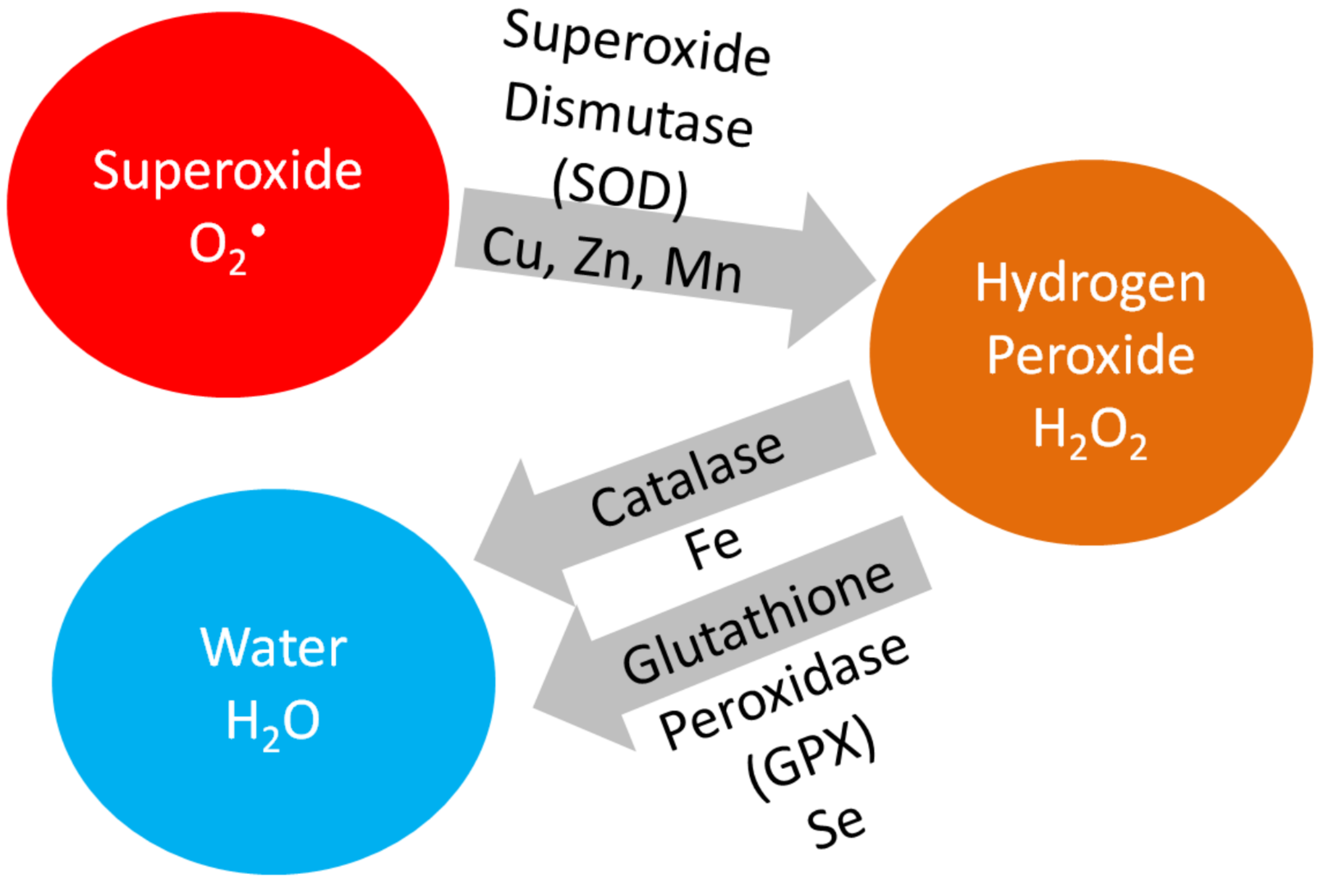
Ascorbic acid also acts against ROS in concert with other antioxidants such as glutathione in the ascorbate-glutathione cycle, and this cycle can also interact with - tocopherol.

This ascorbate-glutathione cycle is found in chloroplasts to protect against ROS generated by photosynthesis but is also found in mitochondria and the cytoplasm.

Ascorbic acid biosynthesis and biodegradation in fruits



Antioxidant enzymes

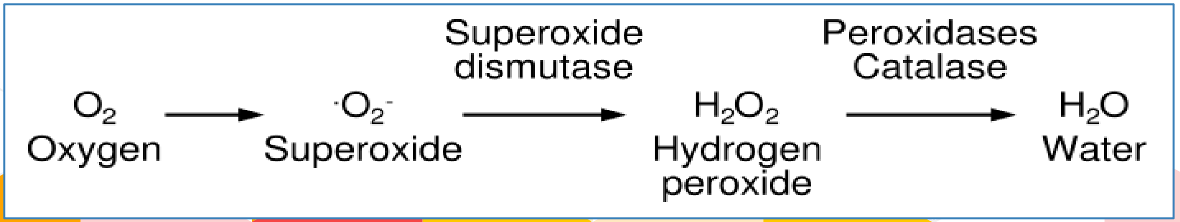


Superoxide dismutase (SOD)

Three classes of SOD activity have been identified that differ by the active site metal cofactors (Fe, Mn, or Cu/Zn). Cu/ZnSOD occurs mainly in the cytosol and chloroplast stroma of plants, whereas MnSOD occurs in the mitochondrial matrix, although a thylakoid-bound MnSOD has been reported to exist in some plants.
FeSODs are generally found in prokaryotes, but have been found in some plants such as the Cruciferae in association with the chloroplasts. In monocotyledonous plants, only choroplastic Cu/ZnSOD and mitochondrial MnSOD have been found.

SOD activity has been linked to physiological stresses such as low temperature, high intensity light, water stress, and oxidative stress.

SOD activity in tomato fruit is highest in the immature-green fruit, passes through a minimum level at the mature-green and breaker stages, and rises again at the pink stage of ripening. The change in SOD activity between immature-green and red-ripe is a 50 percent decrease, while the difference between mature-green and red-ripe is only 5 percent. Also reported that there were no major quantitative changes in total SOD activity between pre- and postclimacteric apple, banana, avocado, and tomato fruits . Therefore, SOD does not always respond to changes in physiological states of fruit.

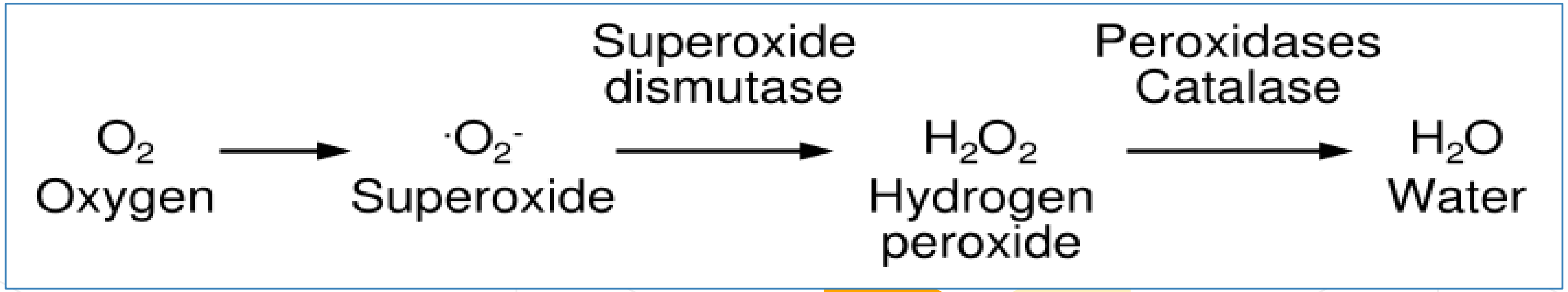


Catalase

Peroxisomes, for which CAT activity is a biological marker, are present in almost all eukaryotes.

Catalases, together with SOD and peroxidases, make up a defense system for the scavenging of O2 – and hydroperoxides.

Catalase is also found in fruits. In citrus fruits, high CAT activity has been linked to resistance of the fruits to chilling injury.



Peroxidases

PODs are heme-containing enzymes which comprises Class I enzymes from mitochondria, chloroplasts, and bacteria; Class II from fungi; and Class III from higher plants.
Cytochrome c peroxidase, glutathione peroxidase , Ascorbate peroxidase

PODs are involved in many growth-related processes, including cell wall extension, lignin biogenesis, and auxin catabolism. In addition, they are involved in stress-related processes such as wounding and disease resistance.

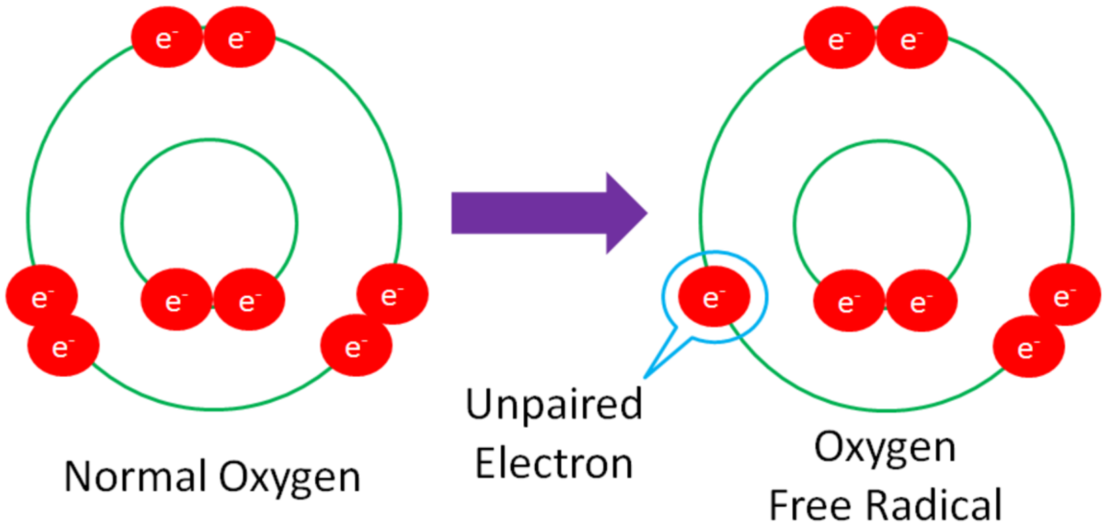
However, fruit ripening is generally viewed as a regulated senescence phenomenon where increased levels of ROS are involved. During ripening, in many cases, both enhanced POD as well as higher levels of peroxides are found .It is unclear whether higher POD activity is present to try to decrease peroxide levels, or if, in spite of higher POD activity, the peroxide levels are high because of their generation during ripening-related processes.
In tomatoes, new isoforms of POD were found to be induced by high temperature treatment, which were correlated with prevention of chilling injury.

Glutathione peroxidase enzyme

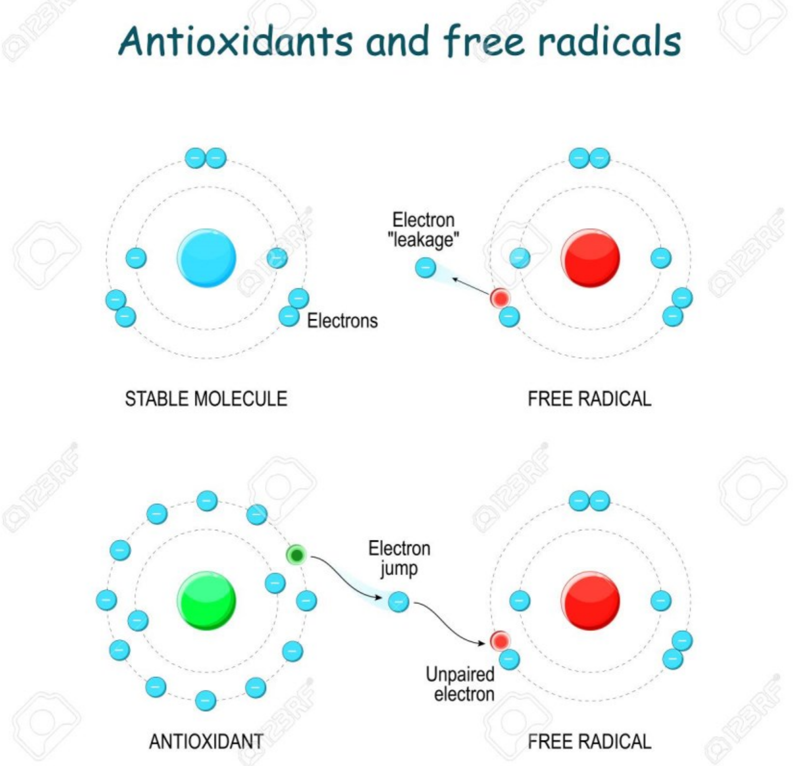
Glutathione peroxidase reduces H2O2 to H2O by oxidizing glutathione (GSH)

Form	Name	Form	Name
$O_2^{\bullet -}$	Superoxide radical	$^1O_2$	Singlet oxygen
$OH^\bullet$	Hydroxyl radical	H_2O_2	Hydrogen peroxide
$RO^\bullet$	Alkoxyl radical	$ROO^\bullet$	Alkylperoxyl radical
ROOH	Alkylhydroperoxide	ClO^-	Hypochlorite ion
$NO^\bullet$	Nitric oxide	$Fe^{4+}O$	Ferryl ion
$Fe^{5+}O$	Periferryl ion		

Free radicals, usually unstable and reactive, are molecules or atoms with an unpaired electron involved in the cell malfunction and damage.



Reactive Species	Antioxidant
Singlet oxygen $^1O_2$	Vitamin A, vitamin E
Superoxide radical (O_2^{-}•)	superoxide dismutase, vitamin C
Hydrogen peroxide (H_2O_2)	Catalase; glutathione peroxidase
Peroxyl radical (ROO•)	Vitamin C, vitamin E
Lipid peroxyl radical (LOO•)	Vitamin E
Hydroxyl radical (OH•)	Vitamin C



Antioxidants during postharvest storage

Superficial scald disorder is caused by oxidative processes that occur only in low-temperature storage, and, therefore, is one of the many manifestations of chilling injury found in fruits and vegetables.

In addition to lower ascorbate, a decrease during storage of SOD, CAT, and glutathione peroxidase activities in pears was found in a cultivar susceptible to fresh browning.

Internal browning in other fruits has also been connected to breakdowns in the tissue antioxidant defense systems.

Internal quality and the rate of senescence of fruits and vegetables in storage have been linked to antioxidants. The antioxidants delay lipid peroxidation and concomitant increase in membrane leakage associated with senescence.

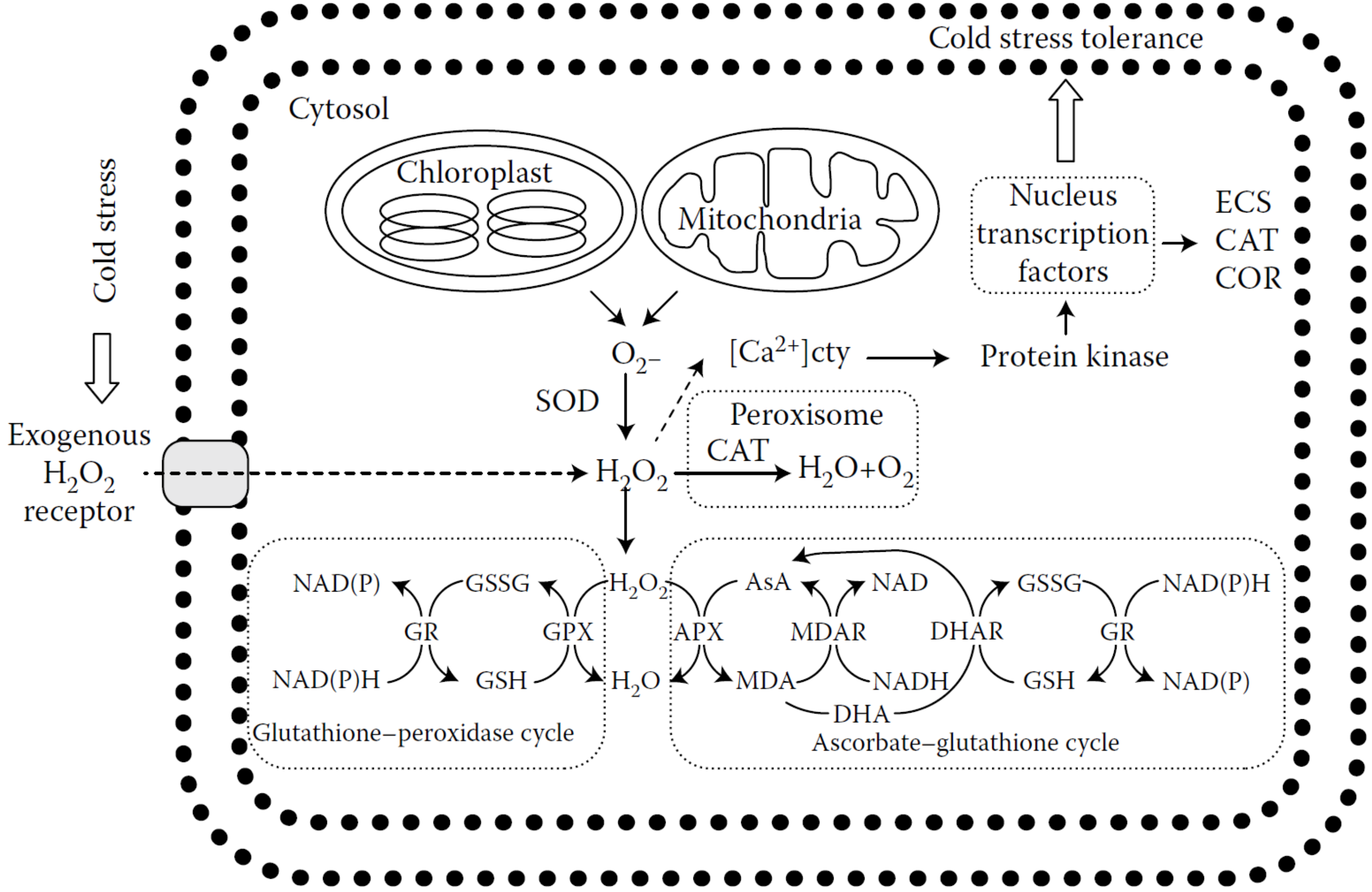
Leaves with greater antioxidant activity senesced more slowly, including yellowing from chlorophyll loss.

Losses of antioxidant enzymes in storage will vary by the type of fruit or vegetable, physical damage, storage temperature, and environment.

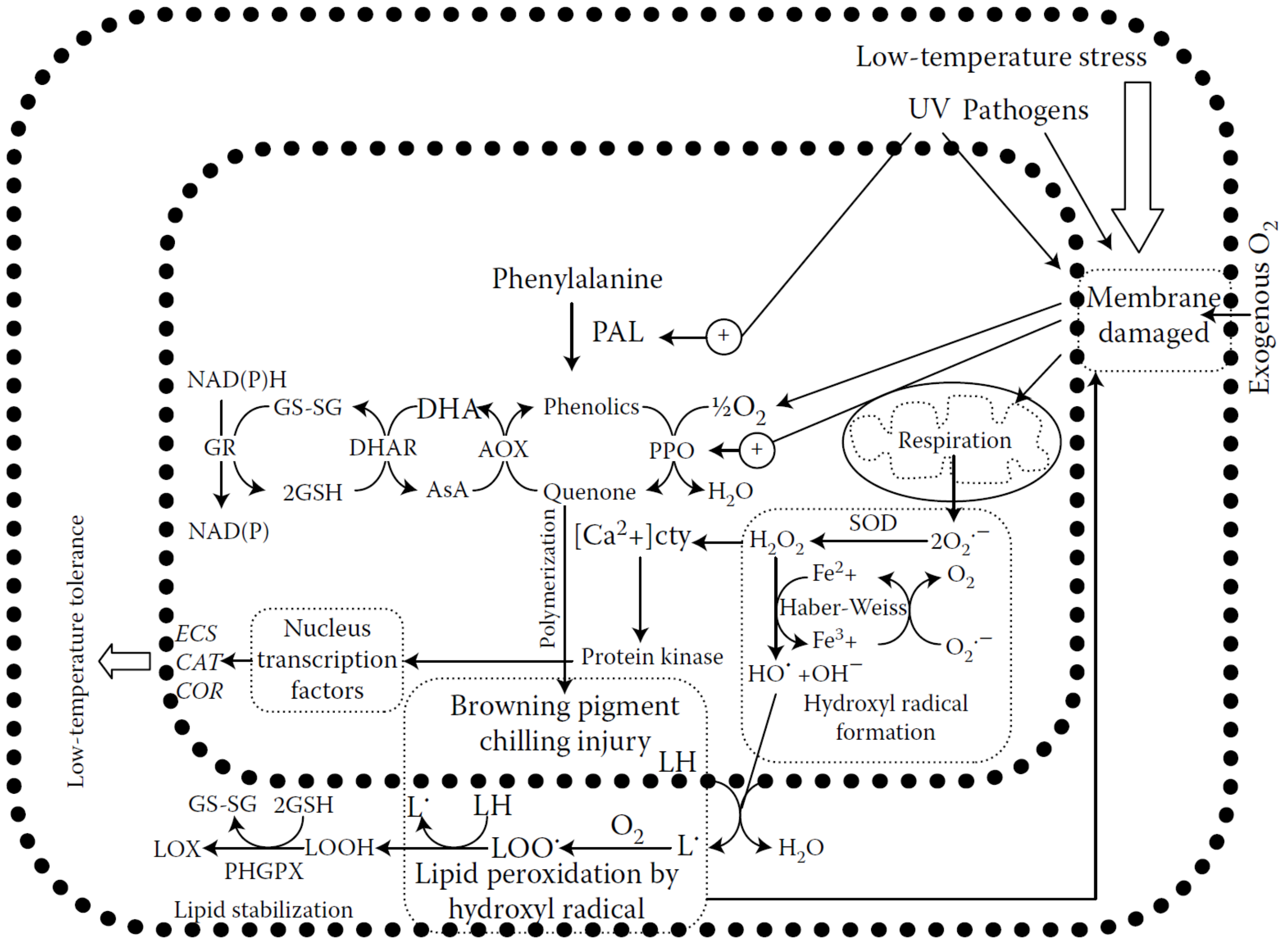
Lipid-soluble antioxidants are generally more stable in storage than are water-soluble ones; for example, carotenoids and –tocopherol are much more stable than ascorbic acid. Lipid-soluble antioxidants were found to increase during apple storage, while water-soluble antioxidants declined

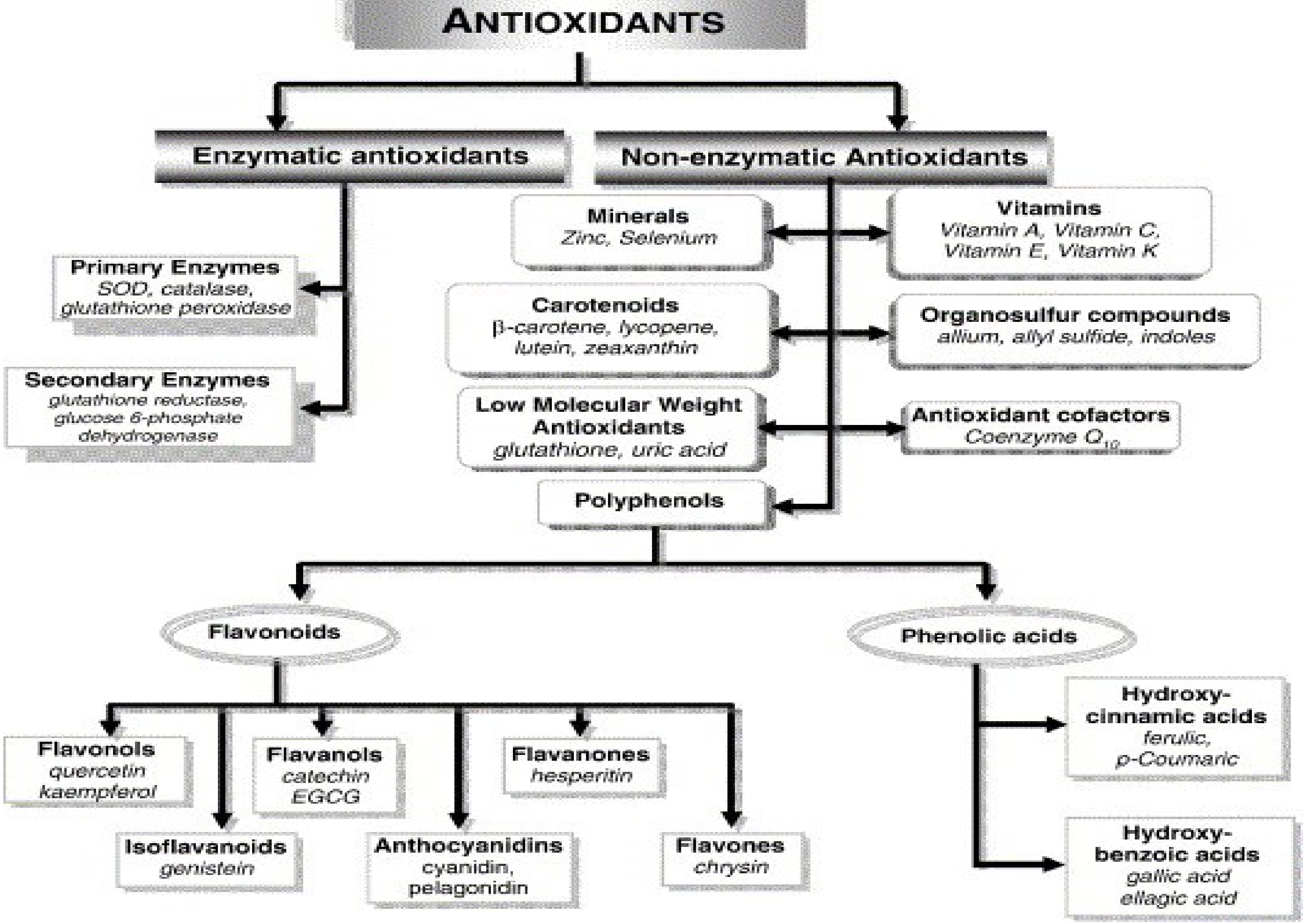
During storage there was an increase in the antioxidant capacity of strawberries and raspberries due to an increase in anthocyanins in strawberries and in anthocyanins and total phenolics in raspberries

Hydrogen peroxide scavenge system in cold stored fruits



Chilling injury and browning tolerant in fruit as a tentative scavenging mechanism of superoxide radical, hydrogen peroxide, hydroxyl radical-induced lipid peroxidation.




ANTIOXIDANTS
Enzymatic antioxidants
Non-enzymatic Antioxidants
Primary Enzymes
SOD, catalase,
glutathione peroxidase
Secondary Enzymes
glutathione reductase,
glucose 6-phosphate
dehydrogenase
Minerals
Zinc, Selenium
Vitamins
Vitamin A, Vitamin C,
Vitamin E, Vitamin K
Carotenoids
β-carotene, lycopene,
lutein, zeaxanthin
Organosulfur compounds
allium, allyl sulfide, indoles
Low Molecular Weight Antioxidants
glutathione, uric acid
Antioxidant cofactors
Coenzyme Q_{10}
Polyphenols
Flavonoids
Phenolic acids
Flavonols
quercetin
kaempferol
Isoflavanoids
genistein
Flavanols
catechin
EGCG
Anthocyanidins
cyanidin,
pelagonidin
Flavanones
hesperitin
Flavones
chrysin
Hydroxy-cinnamic acids
ferulic,
p-Coumaric
Hydroxy-benzoic acids
gallic acid
ellagic acid

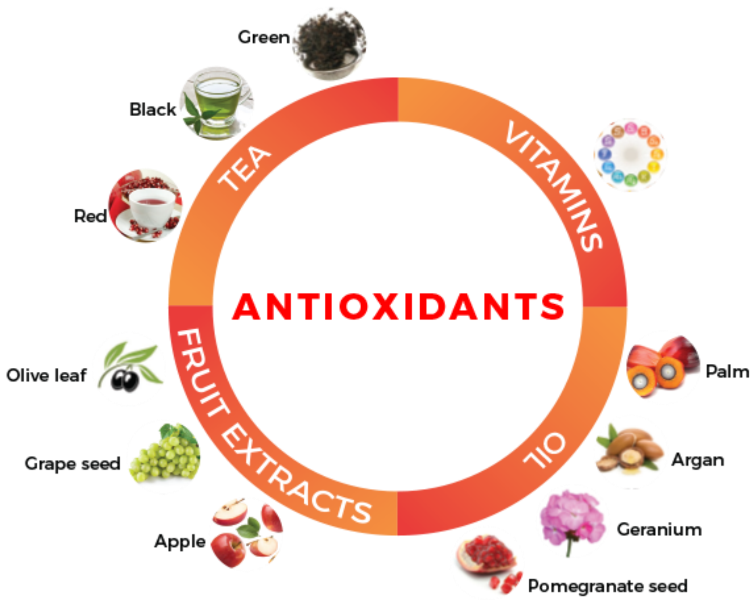
How do antioxidants work?

Antioxidants prevent oxidation of stable compounds by neutralizing free radicals.

2 methods:

Chain-breaking – when a free radical initiates a chain reaction an antioxidant molecule such as beta-carotene, vitamin C and E stabilizes the radical by providing it with the electron it needs to become neutral. In the process of neutralizing the free radical the antioxidant becomes oxidized and therefore needs to be continuously replaced.

Preventive – is the method that uses antioxidant enzymes like superoxide dismutase, catalase and glutathione peroxidase to prevent oxidation by reducing the amount of initiating free radicals that exist in the first place. They also prevent oxidation by stabilizing transition metal radicals such as copper and iron (which have positive charges).

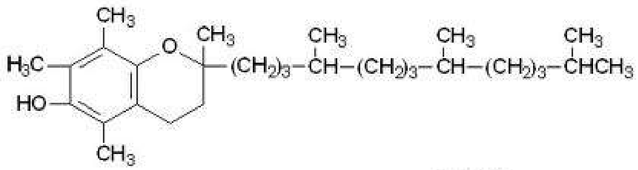


Types of Free Radicals

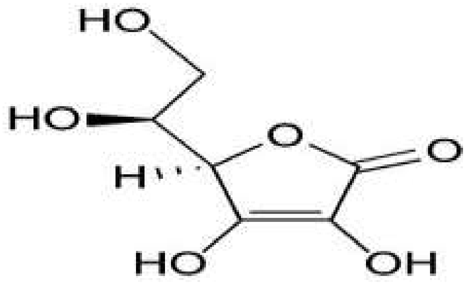
- Mainly Hydrophilic and Hydrophobic
- **Antioxidant enzymes:**
- 1. Catalase
- 2. Glutathione peroxidase
- 3. Glutathione reductase
- 4. Super oxide dismutase (both Cu-Zn and Mn)
- **Metals binding proteins:**
- 1. Ceruloplasmin
- 2. Ferritin
- 3. Lactoferrin
- 4. Metallotheinein
- 5. Transferrin
- 6. Hemoglobin
- 7. Myoglobin

- **Common antioxidants (scavengers)**
- **1. Bilirubin**
- **2. Carotenoids**
- a. Beta-carotene b. Alpha-carotene
- c. Beta-cryptoxanthin d. Lutein
- e. Zeaxanthin f. Lycopene
- **3. Flavonoids**
- a. Quercetin
- b. Rutin
- c. Catechin
- **4. Uric acids 5. Thiols (R-SH)**
- **6. Coenzyme Q10 7. Vitamin A, C, E, D.**
- **Others antioxidants**
- 1. Copper 2. glutathione (GSH)
- 3. Alpha lipoic acid 4.Manganise
- 5. Selenium 6. Zinc

Mechanism of action of antioxidants



- **Alpha tocopherol (vitamin E):**
- Prevent the peroxidation of membrane
- phospholipids, and avoids cell membrane damage
- through its antioxidant action



- **Ascorbic acid (vitamin C)**
- Scavenges free radicals and reactive oxygen molecules, which are
- produced during metabolic pathways of detoxification
- **Beta Carotene**
- Ability to quench singlet oxygen, scavenge free radicals and protect the
- cell membrane lipids from the harmful effects of oxidative degradation

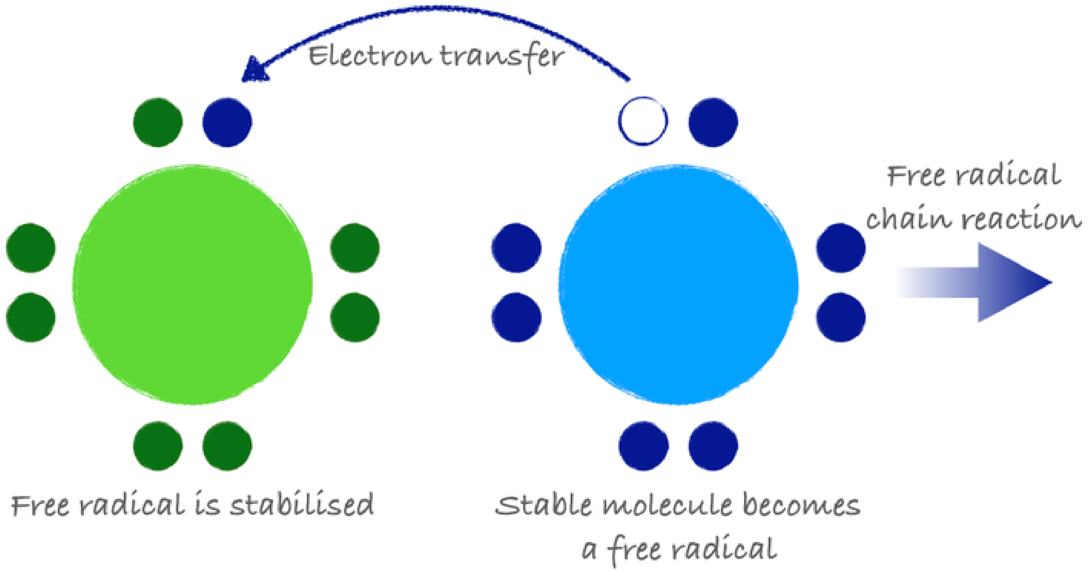
•Pro-oxidant activities

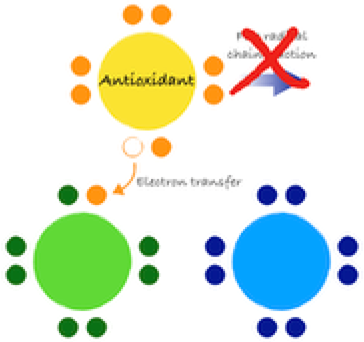
- Antioxidants that are reducing agents can also act as pro-oxidants.
- For example, vitamin C has antioxidant activity when it reduces oxidizing substances such as hydrogen peroxide, however, it will also reduce metal ions that generate free radicals through the Fenton reaction.
- $2\ Fe^{3+} + \text{Ascorbate} \rightarrow 2\ Fe^{2+} + \text{Dehydroascorbate}$
- $2\ Fe^{2+} + 2\ H_2O_2 \rightarrow 2\ Fe^{3+} + 2\ OH\cdot + 2\ OH^-$

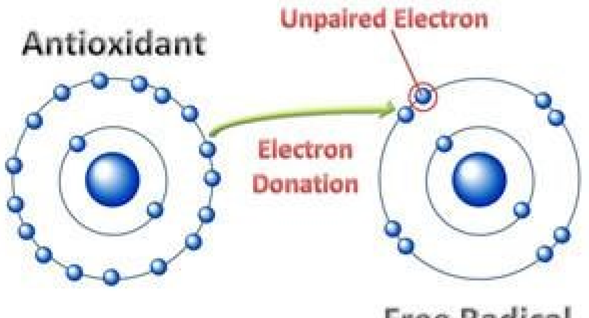
•Determining Antioxidant Activity

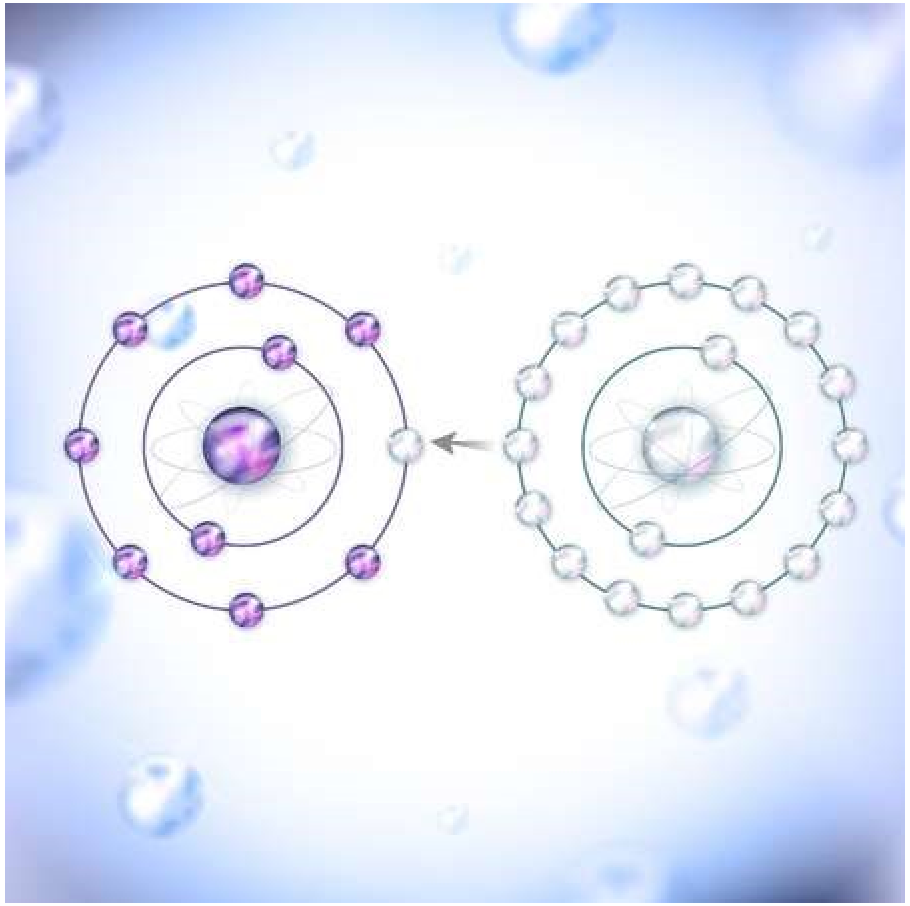
- • ORAC, Oxygen Radical Absorbance Capacity method
- • TRAP, Total Radical-Trapping Antioxidant Parameter method.
- • TEAC, Trolox Equivalent Antioxidant Capacity method
- • DPPH
- • TOSC, Total Oxyradical Scavenging Capacity method
- • PSC, Peroxyl Radical Scavenging Capacity method
- • FRAP, Ferric Reducing/Antioxidant Power method.

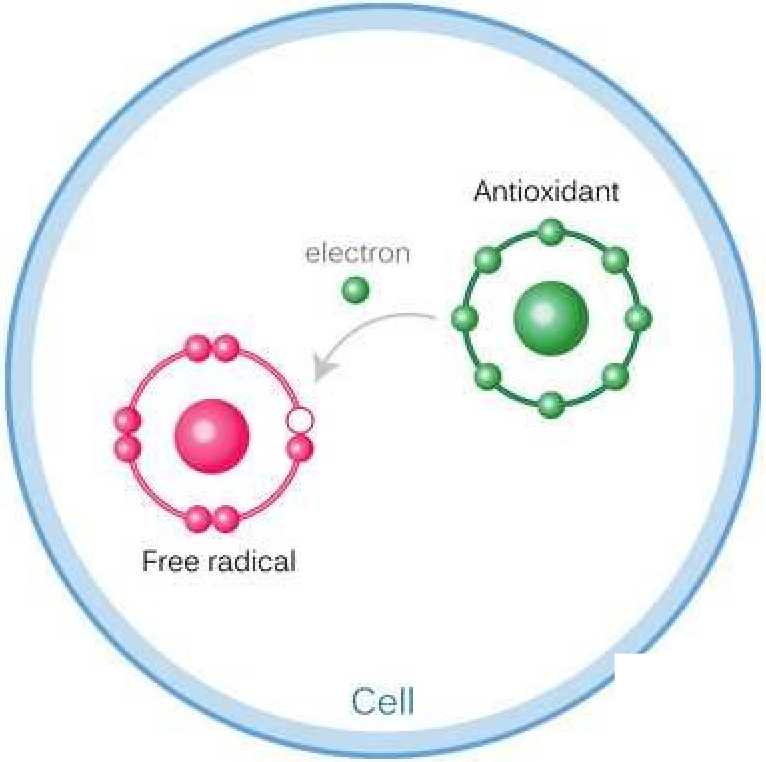
- **Mechanisms by which antioxidants may offer protection**
  - prevention of formation of free radicals
  - interception of free radicals
  - facilitating the repair
  - providing a favourable environment


Electron transfer
Free radical chain reaction
Free radical is stabilised
Stable molecule becomes a free radical

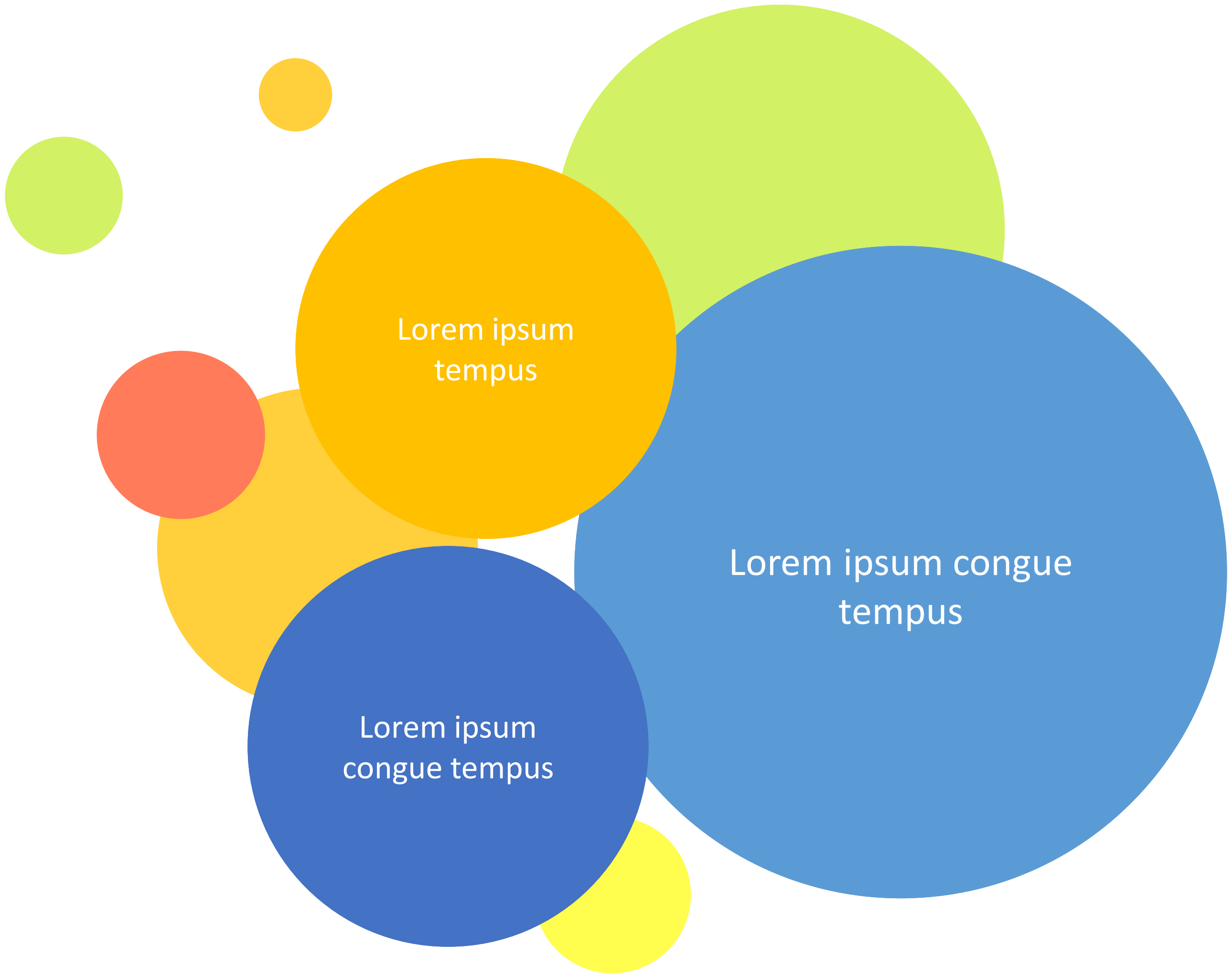

Antioxidant
Electron transfer


Antioxidant
Unpaired Electron
Electron Donation

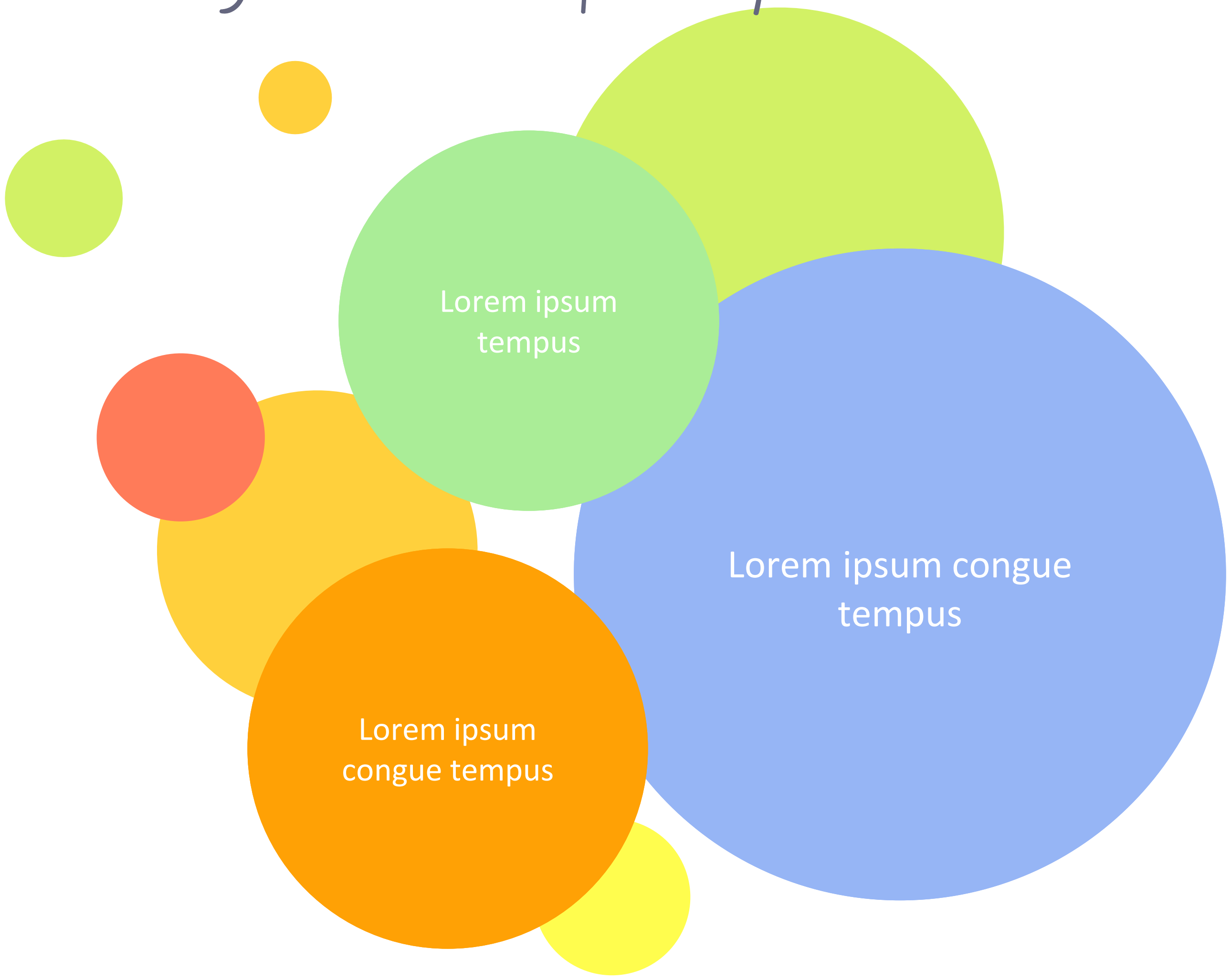



Antioxidant
electron
Free radical
Cell

Use diagrams to explain your ideas



Use diagrams to explain your ideas



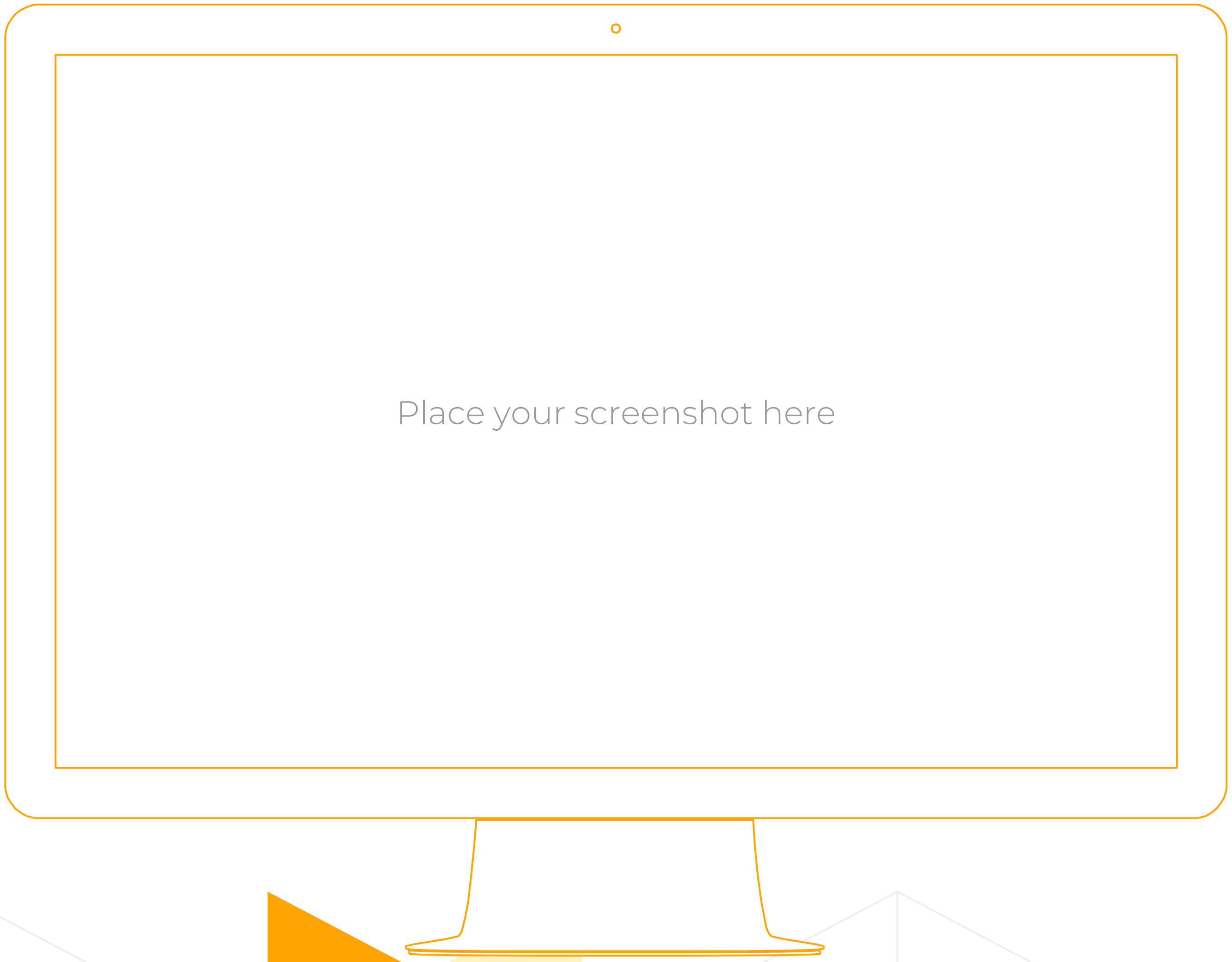
Desktop project

Place your screenshot here

Show and explain your web, app or software projects using these gadget templates.

Desktop project

Show and explain your web, app or software projects using these gadget templates.



Mobile project

Show and explain your web, app or software projects using these gadget templates.

Place your screenshot here

Mobile project

Show and explain your web, app or software projects using these gadget templates.

Our process is easy

Lorem 1

Lorem ipsum dolor sit amet, consectetur adipiscing elit. Duis sit amet odio vel purus bibendum luctus. Morbi iaculis dapibus tristique. In hac habitasse platea dictumst. Mauris convallis quam at. Morbi iaculis dapibus tristique.

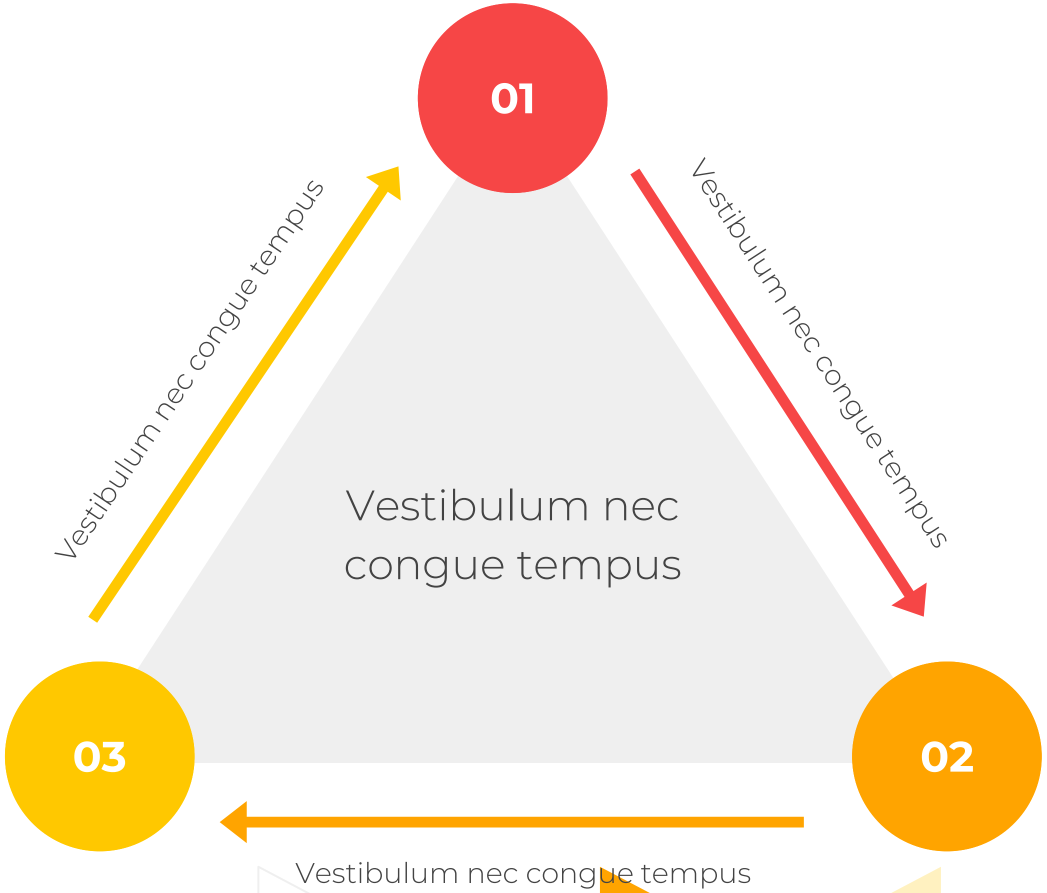
Lorem 2

Lorem ipsum dolor sit amet, consectetur adipiscing elit. Duis sit amet odio vel purus bibendum luctus. Morbi iaculis dapibus tristique. In hac habitasse platea dictumst. Mauris convallis quam at. Morbi iaculis dapibus tristique.

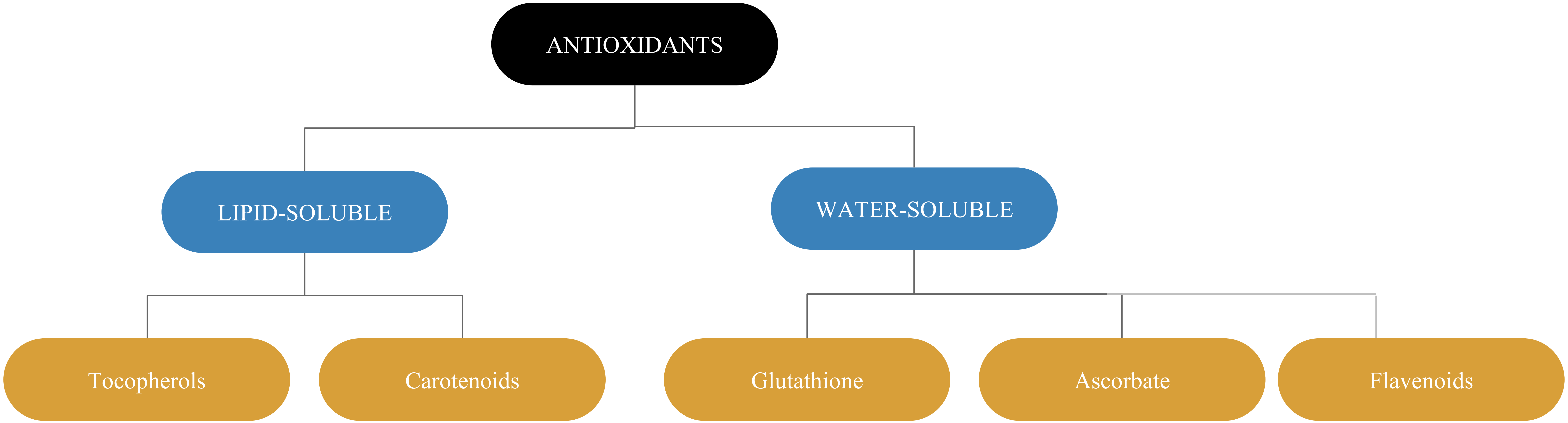
Lorem 3

Lorem ipsum dolor sit amet, consectetur adipiscing elit. Duis sit amet odio vel purus bibendum luctus. Morbi iaculis dapibus tristique. In hac habitasse platea dictumst. Mauris convallis quam at. Morbi iaculis dapibus tristique.

Use diagrams to explain your ideas



Use diagrams to explain your ideas



Presentation design

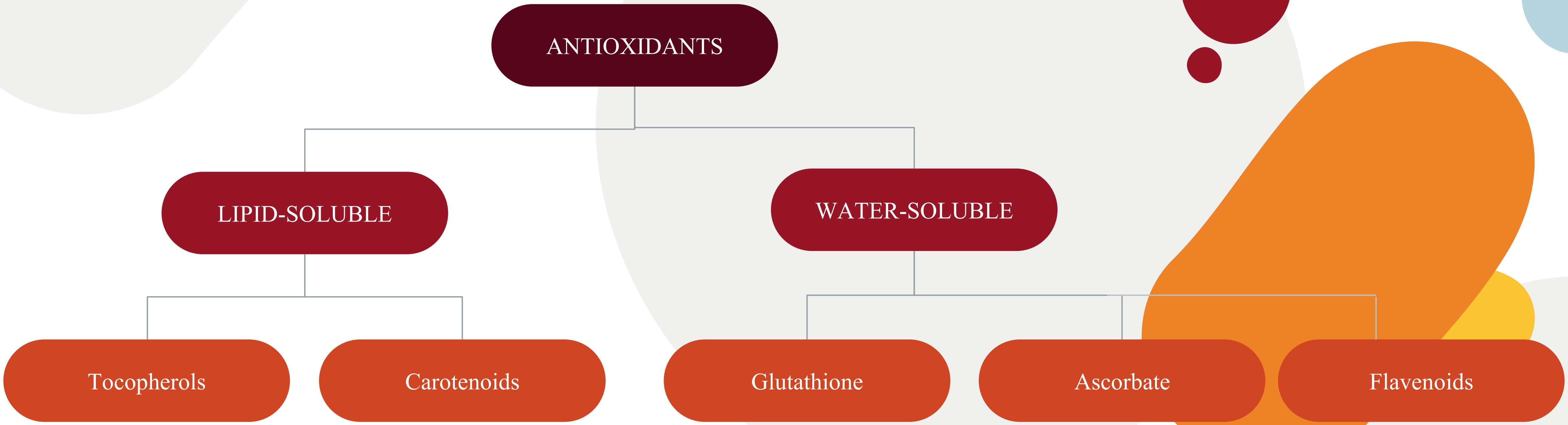
This presentation uses the following typographies:

- Titles: Zilla Slab
- Body copy: Red Hat Text

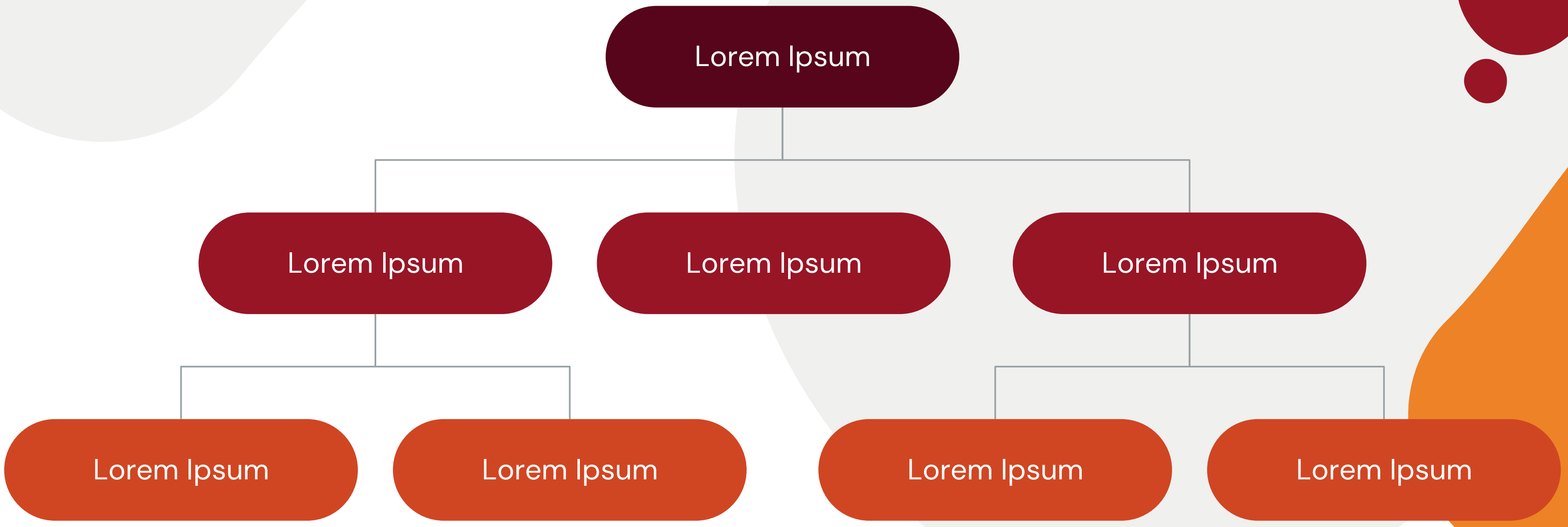
Download for free at:
https://www.fontsquirrel.com/fonts/zilla-slab
https://www.fontsquirrel.com/fonts/red-hat

You don’t need to keep this slide in your presentation. It’s only here to serve you as a design guide if you need to create new slides or download the fonts to edit the presentation in PowerPoint®

Use diagrams to explain your ideas



Use diagrams to explain your ideas



In two or three columns

Yellow

Is the color of gold, butter and ripe lemons. In the spectrum of visible light, yellow is found between green and orange.

Blue

Is the colour of the clear sky and the deep sea. It is located between violet and green on the optical spectrum.

Red

Is the color of blood, and because of this it has historically been associated with sacrifice, danger and courage.

Our process is easy

1

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

2

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

3

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

Our process is easy

1

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

2

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

3

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

In two or three columns

Yellow

Is the color of gold, butter and ripe lemons. In the spectrum of visible light, yellow is found between green and orange.

Blue

Is the colour of the clear sky and the deep sea. It is located between violet and green on the optical spectrum.

Red

Is the color of blood, and because of this it has historically been associated with sacrifice, danger and courage.

Our process is easy

1

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

2

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

3

Vestibulum congue tempus

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

Our process is easy

1 **Vestibulum congue tempus**

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

2 **Vestibulum congue tempus**

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

3 **Vestibulum congue tempus**

Lorem ipsum dolor sit amet, consectetur adipiscing elit, sed do eiusmod tempor. Donec facilisis lacus eget mauris.

- Different iso-forms can be distinguished on the basis of their metal cofactor. Normally, plants contain mitochondrial MnSOD as well as a cytosolic Cu/ZnSOD and a chloroplastic FeSOD. Hydrogen peroxide (H2O2) is eliminated by catalase and peroxidase. Catalase (CAT) is a peroxisomal enzyme requiring a reducing substrate for the reaction which is in contrast to peroxidase (POD). Ascorbate peroxidase (APX) is most important for H2O2 scavenging which operates both in cytosol and chloroplast. It utilizes AsA as a reducing substrate for an oxidation–reduction cycle known as the ascorbate–glutathione or Halliwell–Asada cycle (Figure 20.7). The other enzymes involved in this cycle include monodehydroascorbate reductase (MDAR), dehydroascrobate reductase (DHAR), and glutathione reductase (GR). Recently, a cDNA clone encoding glutathione peroxidase (GPX) has been isolated, suggesting that this protein might play an important role in H2O2 scavenging or the products of lipid peroxidation.

Antioxidants can be categorized by several methods:

- Types
- Mode of action
- Location
- Solubility
- Structural dependents
- Origin

3.) Location

INTRACELLULAR	• SOD 1 and 2, catalase, glutathione peroxidase, DNA repair enzymes
EXTRACELLULAR	• SOD 3, reduced glutathione, ascorbate,carotenoids, uric acid
MEMBRANE ASSOCIATED	• α-Tocopherol

2.) Mode of Action

PREVENTIVE	• Enzymes: superoxide dismutase , catalase, glutathione • Metal ion sequestrators: carotenoids, superoxide dismutase, catalase, glutathione, uric acid, flavenoids
SCAVENGING	• Ascorbate, carotenoids, uric acid,α-tocopherol. flavenoids, ubiquinone, thiols

4.) Solubility

WATER SOLUBLE	• Ascorbate, Uric acid, Flavenoids, thiols, Cysteine, Transferins
LIPID SOLUBLE	• α-Tocopherol, Carotenoids, bilirubin