

Biologic Oxidation

BIOMEDICAL IMPORTAN

Chemically, **oxidation** is defined as the removal of electrons and **reduction** as the gain of electrons. Thus, oxidation is always accompanied by reduction of an electron acceptor. This principle of oxidation-reduction applies equally to biochemical systems and is an important concept underlying understanding of the nature of biologic oxidation. Note that many biologic oxidations can take place without the participation of molecular oxygen, eg, dehydrogenations. The life of higher animals is absolutely dependent upon a supply of oxygen for **respiration**, the process by which cells derive energy in the form of ATP from the controlled reaction of hydrogen with oxygen to form water. In addition, molecular oxygen is incorporated into a variety of substrates by enzymes designated as **oxygenases**; many drugs, pollutants, and chemical carcinogens (xenobiotics) are metabolized by enzymes of this class, known as the **cytochrome P450 system**. Administration of oxygen can be lifesaving in the treatment of patients with respiratory or circulatory failure.

Cytochrome oxidase is a hemoprotein widely distributed in many tissues, having the typical heme prosthetic group present in myoglobin, hemoglobin, and other cytochromes (Chapter 6). It is the terminal component of the chain of respiratory carriers found in mitochondria and transfers electrons resulting from the oxidation of substrate molecules by dehydrogenases to their final acceptor, oxygen. The enzyme is poisoned by carbon monoxide, cyanide, and hydrogen sulfide. It has also been termed cytochrome *a*₃. It is now known that

cytochromes *a* and *a3* are combined in a single protein, and the complex is known as **cytochrome *aa3***. It contains two molecules of heme, each having one Fe atom that oscillates between Fe³⁺ and Fe²⁺ during oxidation and reduction. Furthermore, two atoms of Cu are present, each associated with a heme unit.

Flavoprotein enzymes contain **flavin mononucleotide (FMN)** or **flavin adenine dinucleotide (FAD)** as prosthetic groups. FMN and FAD are formed in the body from the vitamin **riboflavin** (Chapter 45). FMN and FAD are usually tightly—but not covalently—bound to their respective apoenzyme proteins. Metalloflavoproteins contain one or more metals as essential cofactors.

Examples of flavoprotein enzymes include **L-amino acid oxidase**, an FMN-linked enzyme found in kidney with general specificity for the oxidative deamination of

the naturally occurring L-amino acids; **xanthine oxidase**, which contains molybdenum and plays an important role in the conversion of purine bases to uric acid (Chapter 34), and is of particular significance in uricotelic animals (Chapter 29); and **aldehyde dehydrogenase**, an FAD-linked enzyme present in mammalian livers, which contains molybdenum and nonheme iron and acts upon aldehydes and N-heterocyclic substrates. The mechanisms of oxidation and reduction of these enzymes are complex. Evidence suggests a two-step reaction

DEHYDROGENASES CANNOT USE OXYGEN AS A HYDROGEN ACCEPTOR

There are a large number of enzymes in this class. They perform two main functions:

(1) Transfer of hydrogen from one substrate to another in a coupled oxidation-reduction reaction

These dehydrogenases are specific for their substrates but often utilize common coenzymes or hydrogen carriers, eg, NAD⁺. Since the reactions are reversible, these properties enable reducing equivalents to be freely transferred within the cell. This type of reaction, which enables one substrate to be oxidized at the expense of another, is particularly useful in enabling oxidative processes to occur in the absence of oxygen, such as during the anaerobic phase of glycolysis (Figure 17–2).

(2) As components in the **respiratory chain** of electron transport from substrate to oxygen

These dehydrogenases use **nicotinamide adenine dinucleotide (NAD⁺)** or **nicotinamide adenine dinucleotide phosphate (NADP⁺)**—or both—and are formed in the body from the vitamin **niacin** (Chapter 45). The coenzymes are reduced by the specific substrate of the dehydrogenase and reoxidized by a suitable electron acceptor (Figure 11–4). They may freely and reversibly dissociate from their respective apoenzymes. Generally, NAD-linked dehydrogenases catalyze oxidoreduction reactions in the oxidative pathways of metabolism, particularly in glycolysis, in the citric acid cycle, and in the respiratory chain of mitochondria. NADP-linked dehydrogenases are found characteristically in reductive syntheses, as in the extramitochondrial pathway of fatty acid synthesis and steroid synthesis—and also in the pentose phosphate pathway.

Other Dehydrogenases Depend on Riboflavin

The flavin groups associated with these dehydrogenases are similar to FMN and FAD occurring in oxidases.

They are generally more tightly bound to their apoenzymes than are the nicotinamide coenzymes. Most of the riboflavin-linked dehydrogenases are concerned with electron transport in (or to) the respiratory chain (Chapter 12). **NADH dehydrogenase** acts as a carrier of electrons between NADH and the components of higher redox potential (Figure 12–3). Other dehydrogenases such as **succinate dehydrogenase, acyl-CoA dehydrogenase, and mitochondrial glycerol-3-phosphate dehydrogenase** transfer reducing equivalents directly from the substrate to the respiratory chain (Figure 12–4). Another role of the flavin-dependent dehydrogenases is in the dehydrogenation (by **dihydrolipoyl dehydrogenase**) of reduced lipoate, an intermediate in the oxidative decarboxylation of pyruvate and α -ketoglutarate (Figures 12–4 and 17–5). The **electron-transferring flavoprotein** is an intermediary carrier of electrons between acyl-CoA dehydrogenase and the respiratory chain

HYDROPEROXIDASES USE HYDROGEN PEROXIDE OR AN ORGANIC PEROXIDE AS SUBSTRATE

Two type of enzymes found both in animals and plants fall into this category: **peroxidases** and **catalase**. Hydroperoxidases protect the body against harmful peroxides. Accumulation of peroxides can lead to generation of free radicals, which in turn can disrupt membranes and perhaps cause cancer and atherosclerosis.

Peroxidases Reduce Peroxides Using Various Electron Acceptors

Peroxidases are found in milk and in leukocytes,

platelets, and other tissues involved in eicosanoid metabolism (Chapter 23). The prosthetic group is protoheme.

In the reaction catalyzed by peroxidase, hydrogen peroxide is reduced at the expense of several substances that will act as electron acceptors, such as ascorbate, quinones, and cytochrome *c*. The reaction catalyzed by peroxidase is complex, but the overall reaction is as follows:

In erythrocytes and other tissues, the enzyme **glutathione peroxidase**, containing **selenium** as a prosthetic group, catalyzes the destruction of H₂O₂ and lipid hydroperoxides by reduced glutathione, protecting membrane lipids and hemoglobin against oxidation by peroxides (Chapter 20).

Catalase Uses Hydrogen Peroxide as Electron Donor & Electron Acceptor

Catalase is a hemoprotein containing four heme groups. In addition to possessing peroxidase activity, it is able to use one molecule of H₂O₂ as a substrate electron donor and another molecule of H₂O₂ as an oxidant or electron acceptor.

Under most conditions in vivo, the peroxidase activity of catalase seems to be favored. Catalase is found in blood, bone marrow, mucous membranes, kidney, and liver. Its function is assumed to be the destruction of hydrogen peroxide formed by the action of oxidases.

Peroxisomes are found in many tissues, including liver. They are rich in oxidases and in catalase. Thus, the enzymes that produce H₂O₂ are grouped with the enzyme that destroys it. However, mitochondrial and microsomal electron transport systems as well as xanthine oxidase must be considered as additional sources of H₂O₂

OXYGENASES CATALYZE THE DIRECT

TRANSFER & INCORPORATION OF OXYGEN INTO A SUBSTRATE MOLECULE

Oxygenases are concerned with the synthesis or degradation of many different types of metabolites. They catalyze the incorporation of oxygen into a substrate molecule in two steps: (1) oxygen is bound to the enzyme at the active site, and (2) the bound oxygen is reduced or transferred to the substrate. Oxygenases may be divided into two subgroups,

Monooxygenases (Mixed-Function Oxidases, Hydroxylases) Incorporate Only One Atom of Molecular Oxygen Into the Substrate

The other oxygen atom is reduced to water, an additional electron donor or cosubstrate (Z) being necessary for this purpose.

Cytochromes P450 Are Monooxygenases Important for the Detoxification of Many Drugs & for the Hydroxylation of Steroids

Cytochromes P450 are an important superfamily of heme-containing monooxygenases, and more than 1000 such enzymes are known. Both NADH and NADPH donate reducing equivalents for the reduction of these cytochromes (Figure 11–5), which in turn are oxidized by substrates in a series of enzymatic reactions collectively known as the **hydroxylase cycle** (Figure 11–6). In liver microsomes, cytochromes P450 are found together with **cytochrome b5** and have an important role in detoxification. Benzpyrene, aminopyrine, aniline, morphine, and benzphetamine are hydroxylated, increasing their solubility

and aiding their excretion. Many drugs such as phenobarbital have the ability to induce the formation of microsomal enzymes and of cytochromes P450.

Mitochondrial cytochrome P450 systems are found in steroidogenic tissues such as adrenal cortex, testis, ovary, and placenta and are concerned with the biosynthesis of steroid hormones from cholesterol (hydroxylation at C22 and C20 in side-chain cleavage and at the 11^β and 18 positions). In addition, renal systems catalyzing 1 α - and 24-hydroxylations of 25-hydroxycholecalciferol in vitamin D metabolism—and cholesterol 7 α -hydroxylase and sterol 27-hydroxylase involved in bile acid biosynthesis in the liver-

SUPEROXIDE DISMUTASE PROTECTS AEROBIC ORGANISMS AGAINST OXYGEN TOXICITY

Transfer of a single electron to O₂ generates the potentially damaging **superoxide anion free radical** (O₂

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the destructive effects of which are amplified by its giving rise to free radical chain reactions (Chapter 14).

The ease with which superoxide can be formed from oxygen in tissues and the occurrence of **superoxide dismutase**, the enzyme responsible for its removal in all aerobic organisms (although not in obligate anaerobes) indicate that the potential toxicity of oxygen is due to its conversion to superoxide.

Superoxide is formed when reduced flavins—present, for example, in xanthine oxidase—are reoxidized univalently by molecular oxygen.

or be removed by superoxide dismutase.

In this reaction, superoxide acts as both oxidant and

reductant. Thus, superoxide dismutase protects aerobic organisms against the potential deleterious effects of superoxide. The enzyme occurs in all major aerobic tissues in the mitochondria and the cytosol. Although exposure of animals to an atmosphere of 100% oxygen causes an adaptive increase in superoxide dismutase, particularly in the lungs, prolonged exposure leads to lung damage and death. Antioxidants, eg, α -tocopherol (vitamin E), act as scavengers of free radicals and reduce the toxicity of oxygen (Chapter 45).