Among the mammalian selenoenzymes, glutathione peroxidase (GPx) has attracted much attention due to its enormous importance in the defense against oxidative stress. The chemistry at the active site of GPx has been widely studied with the help of a number of small-molecule organoselenium compounds. Among these, ebselen (2-phenyl-1,2-benzisoselenazole-3(2H)-one) is currently undergoing phase III clinical trial for a number of disease states as it exhibits good antioxidant activity in vivo. However, the catalytic mechanism for the GPx activity of ebselen is not well established. In this article, different catalytic mechanisms proposed in the literature for the GPx-like antioxidant activities of ebselen and related selenenyl amides are described. Recently, it has been observed that ebselen and related compounds show poor catalytic activities in the presence of aryl or benzyl thiols as cofactors. The unusually low catalytic activities of these Sec-amide-substituted compounds have been ascribed to the undesired thiol exchange reactions that take place in the selenenyl sulfide intermediates due to the presence of strong Se···O nonbonded interactions. Furthermore, it has been shown that the extensive thiol exchange reactions can be prevented by using a dithiol as cofactor or by introducing S···N/O nonbonded interactions in selenenyl sulfide intermediates. In addition, it has been observed that the GPx activities of sec-amide-based compounds can be dramatically enhanced by the substitution at free-NH groups of the amide functionality that prevents the undesired thiol exchange reactions.
14.1 Introduction

Biochemistry of selenium in mammals emerged in 1973 with the discovery of antioxidant enzyme glutathione peroxidase (GPx) that contains selenium in the form of selenocysteine at the active site\(^1\), \(^2\). Although more than thirty selenoproteins have been identified till now, the structure and functions have not yet been clearly established for several of these proteins \(^3\), \(^4\). In eukaryotes, iodothyronine deiodinase (ID) \(^5\)-\(^8\), thioredoxin reductase (TrxR) \(^9\)-\(^14\), selenophosphate synthetase (SPS) \(^14\) and selenoprotein P (SeP) \(^15\) represent important classes of selenoenzymes in addition to the well-known glutathione peroxidase (GPx) \(^1\), \(^2\), \(^16\)-\(^19\).

Among these mammalian selenoenzymes, GPx has attracted much research attention due to its enormous importance in the defense against oxidative damage of biological components \(^20\)-\(^23\). The GPx superfamily contains four types of enzymes, the classical cytosolic GPx (cGPx), phospholipid hydroperoxide GPx (PHGPx), plasma GPx (pGPx) and gastrointestinal GPx (giGPx), all of which require selenium at their active sites to exhibit their catalytic activities \(^24\)-\(^28\). The GPx catalytic site includes a selenocysteine residue in which the selenium center undergoes a number of redox reactions for its catalytic activity. It is now clear that the active form of the enzyme is selenol (E-SeH), which reduces hydroperoxides and undergoes oxidation to the corresponding selenenic acid (E-SeOH). The E-SeOH intermediate reacts with GSH to produce a selenenyl sulfide intermediate (E-Se-SG). A second molecule of GSH attacks at the sulfur center of the E-Se-SG adduct to regenerate the active form of the enzyme (Scheme 14.1) \(^29\)-\(^31\). In the overall process, 2 equivalents of GSH are oxidized to the corresponding disulfide, while the hydroperoxide is reduced to water or alcohol. When the peroxide concentration is higher than that of the thiol co-factor, the selenium center in the selenenic acid (E-SeOH) may undergo further oxidation to the overoxidized seleninic (E-SeO\(_2\)H) or selenonic (E-SeO\(_3\)H) acids. However, these species may lie off the main catalytic cycle in the presence of the thiol co-factor.

![Scheme 14.1. Proposed catalytic cycle of GPx for the reduction of peroxides in the presence of GSH as co-factor](image-url)