Cysteine Sulfenic Acid

Oxidation is rarely used as a method for the modification of cysteine in protein as, for example, is alkylation; oxidation of cysteine is usually required for measurement (1/2 Cys) by amino acid analysis (1). Oxidation is a consequence of exposure of cysteine to solvent. Early understanding of the function of cysteine and cystine in proteins suggested that cysteine was found only in intracellular proteins and disulfide bonds found in extracellular proteins (2). While there is a definite difference between intracellular proteins and extracellular proteins (3) the difference is not absolute in that while intracellular proteins do not contain disulfide bonds, some 10% of extracellular proteins contain sulfhydryl groups which, if exposed, would be susceptible to oxidation.

Cysteine sulfenic acid (3-sulfenylalanine) is the initial product in the oxidation of cysteine (it could be equally as well argued that cystine is the initial product) but can also be derived from the hydrolysis of cystine oxides such as cystine thiosulfonate. While organic sulfenic acids are unstable in solution, cysteine sulfenic acid is proteins is somewhat stable although there is tendency to move to high oxidation states (cysteine sulfinic acid) or to form mixed disulfides (4,5). It is noted that, of all the cysteine derivatives, only cysteine sulfenic acid has not been isolated (6,7). The presence of cysteine sulfenic acid in proteins has been demonstrated by mass spectrometry (8) and there are a variety of assay techniques which are available (9). The most common assays for cysteine sulfenic acid use either reaction with dimedone (10) or thionitrobenzoic acid (11). Other novel reagents have been developed (12). Cysteine sulfenic acid can be reduced to cysteine with hydrogen sulfide or with dithiothreitol (13). In these latter studies (13), the physical recovery of cysteine was not demonstrated but rather the recovery of peroxidase activity from enzyme which had been inactivated with hydrogen peroxide or peroxynitrite. A novel finding was the formation of a mixed disulfide between the oxidized enzyme and 2-mercaptoethanol. The formation of a mixed disulfide between protein-bound sulfenic acid and glutathione is an important feature of redox regulation (14,15). The presence of cysteine sulfenic acid in proteins is becoming increasingly frequent with the improvement of analytical methods (4). One particular striking example is glyceraldehyde-3-phosphate dehydrogenase where oxidation of the active site cysteine to cysteine sulfenic acid converts the dehydrogenase activity to acyl phosphatase activity (16-18). An upsurge in recent interest in cysteine sulfenic acid has resulted from its role in redox regulation (19-22). Cysteine sulfenic acid has also been described in albumin (9,23-25) and transerythrin (9). It is somewhat surprising that the work of Le Gaillard and Dautrevaux (26) on transcortin (corticosteroid binding globulin) found in human blood plasma has not received greater consideration. Transcortin contains two cysteine residues which are not present in a disulfide bond (27). Le Gaillard and Dautrevaux (26) reported that one of the two cysteine residues in transcortin appeared to oxidized to cysteine sulfenic acid. The presence of an oxidized sulfhydryl group in transcortin that could be reduced with dithiothreitol was confirmed subsequently by Defaye and coworkers (28). Later work showed that one cysteine, CYS60, was important in binding steroid and may be the residue oxidized to cysteine sulfenic acid (29). However, the role of the cysteine and the importance of oxidation is not clear as inhibition with sulfhydryl reagents appears to depend on prior treatment with reducing agent. It is possible that there are other plasma proteins such as factor VIII, which have free sulfhydryl groups may also contain cysteine sulfenic acid. The modification of cysteine sulfenic acid in the AhpC of alkyl hydroperoxide reductase (*Salmonella typhimurium*) by either iodoacetate or *N*-ethylmaleimide has been reported (30).

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