Met oxidation

Formation of Met sulfoxide has frequently been observed in proteins and is the most common oxidation reaction in proteins. Generally three mediators of oxidation, chemical oxidants such as peroxides, light, and metals, convert Met to Met sulfoxide. The mechanism for chemically induced oxidation is believed to involve the transfer of oxygen from the oxidant peroxide via a nucleophilic substitution reaction where the sulfur on the Met residue interacts with the oxygen atom of the peroxide. This reaction, which is acid catalyzed, proceeds through an intermediate as shown in Figure 3.6(a). The formation of Met sulfoxide by exposure to light proceeds by interaction with singlet oxygen forming an unstable intermediate persulfoxide, which then oxidizes a second Met to form Met sulfoxide (Figure 3.6; Sysak, Foote, & Ching, 1977). Oxidation of Met by metals is very complex and involves catalysis by the metal ion to produce reactive oxygen species such as O_2^-, H_2O_2, $^1O_2$, and OH (Stadtman, 1990). Schöneich et al. identified the main reactive oxygen intermediates that were responsible for Met oxidation as peroxides at $pH<7$ and other intermediates such as iron-bound hydroperoxide or site-specifically generated reactive oxygen species at $pH>7$ (Li, Schoneich, et al., 1995b). The latter likely goes via a mechanism whereby the reactive oxygen species are formed near metal-binding sites on the protein and react with residues that are in close proximity. This observation is supported by the fact that oxygen radical scavengers have little effect on metal-catalyzed oxidation in proteins.

Met oxidation in mAbs

As in most proteins oxidation of Met in mAbs occurs frequently. In one of the first mAbs to be commercialized, OKT3 (an IgG_2), the major degradation route was oxidation of Met residues during storage at 5 °C (Kroon et al., 1992). Light exposure at 27 °C of another mAb, Herceptin®, formulated as a liquid, had oxidation at Met 255 and 431 in the Fc region after storage at 30 and 40 °C (Lam, Yang, & Cleland, 1997).

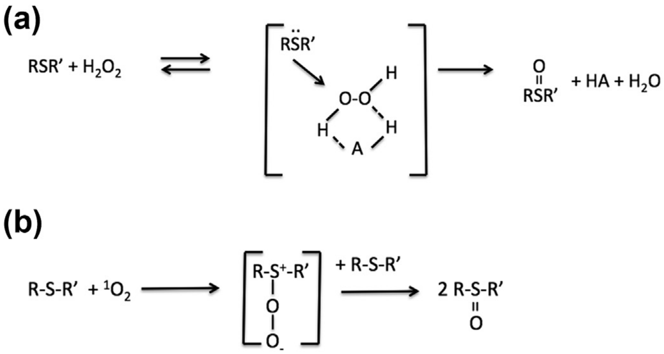

Figure 3.6 (a) Acid–base catalysis of Met to Met sulfoxide by hydrogen peroxide (HA = acid). (b) Photooxidation of Met to Met sulfoxide by singlet oxygen.
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