complementarity determining regions (CDRs) where binding to the target occurs. Examples of this are Herceptin® where an isomerization of Asp H102 in the CDR results in an 80–90% decrease in activity and an anti-IgE mAb where an isomerization of Asp L32 on one chain results in a decrease of 60% activity and on both chains a reduction in activity of 85% relative to the mAb with unaltered Asp residues.

Oxidation

Oxidation of proteins is the next most common degradation pathway where amino acid residues in proteins susceptible to oxidation include Met, Tyr, His, Trp, and Cys. Oxidation may be mediated by oxidants such as peroxides, exposure to light, metals, and ionizing radiation. Oxidation can also occur in the absence of oxidants, a process referred to as auto-oxidation. However, the direct reaction between ground-state molecular oxygen and proteins is extremely slow and does require the presence of catalysts. In particular, exposure to light and transition metal ions may accelerate the oxidation process and be the major factors responsible for apparent auto-oxidation.

As shown for deamidation/isomerization several factors such as solvent exposure, conformation, and primary sequence can impact the rate of oxidation for the same residue at different positions in the polypeptide chain. As an example, in human relaxin, a small protein pregnancy hormone closely related to human insulin, the rate of oxidation of the two Met residues in the B chain appears to be sequential where one Met oxidizes before the other during photooxidation (Cipolla & Shire, 1991). This was also shown for oxidation by hydrogen peroxide where the determined rate of oxidation at Met 25 was faster than at Met 4 (Nguyen, Burnier, & Meng, 1993). However, in the case of metal-catalyzed oxidation the rate of oxidation at Met 4 was faster than at Met 25 (Li et al., 1995). Inspection of the crystallographic structure for human relaxin shows that this observation may be explained by the close proximity of the Met 4 to potential metal-binding sites (Asp and Lys).

The oxidation of the different amino acid residues results in the generation of different species. The common degradation products include Met sulfoxide from Met, 2-oximidazoline, Asp and Asn from His, *N*-formyl kynurenine and kynurenine from Trp, Tyr–Tyr cross-links and DOPA from Tyr, and sulfenic acid, sulfinic acid, and disulfide scrambling from Cys (Li, Schoneich, et al., 1995a). Since the different susceptible residues yield different reaction products it is worthwhile to discuss mechanisms of oxidation for the different residues.

Mechanisms of oxidation

The various mechanisms for oxidation have been reviewed extensively (Berges, Trouillas, & Houee-Levin, 2011; Chu et al., 2004; Cleland et al., 1993; Hiller, Masloch, Gobl, & Asmus, 1981; Li, Schoneich, et al., 1995a; McDermott, Chiesa, Roberts, & Dillon, 1991; Pattison, Rahmanto, & Davies, 2012; Sharma, 2013; Sharma & Graham, 2010; Stadtman, 1990; Stadtman & Berlett, 1998; Stadtman & Levine, 2000; Swallow, 1960) and a brief summary of mechanism for each oxidizing residue in proteins follows.