Peroxides are known contaminants of nonionic surfactants (19). Peroxide levels from different sources of polysorbate 80 ranged from less than 1 mEq/kg to more than 27 mEq/kg. Peroxide levels increased upon storage at ambient temperatures probably due to headspace oxygen and/or the container/closure interface allowing ingress of air. Peroxides in polysorbate can result in oxidative degradation of proteins. Improvements have been made in the manufacturing of polysorbate, for example, certified peroxide-free polysorbates are now readily available.

Electron paramagnetic resonance (EPR) spectroscopy has been used to determine the binding stoichiometry of the surfactant to the protein and, thus, what potentially is the optimal amount of surfactant to use to stabilize the protein against surface denaturation and other physical instability reactions (47).

CYCLODEXTRINS

Cyclodextrins are cyclic (α-1,4)-linked oligosaccharides of α-D-glucopyranose containing a relatively hydrophobic central cavity and hydrophilic outer surface. Cyclodextrins come in a wide variety of structural derivatives, the most common being α-, β-, and γ-cyclodextrins, which consist of six, seven, and eight glucopyranose units, respectively. Two parenteral cyclodextrins are Encapsin™, a hydroxylpropyl-β-cyclodextrin, and Captisol™, a sulfobutylether-β-cyclodextrin. They have been used widely for increasing the solubility stability, and bioavailability of small drug molecules (see chap. 6). Peptides and proteins can also be stabilized in cyclodextrin complexes. β-cyclodextrins at a 25-fold excess stabilized leucine enkephalin against enzymatic degradation in sheep nasal mucosa (48). Hydroxypropyl-β-cyclodextrin at a 1% concentration was shown to enhance the reconstituted solution stability of keratinocyte growth factor (49) and several other proteins in solution (50). Glucagon will form inclusion complexes with γ-cyclodextrin in acidic solution that results in enhancement of glucagon's physical and chemical stability (51).

ALBUMIN

Serum albumin is a widely used stabilizer in protein formulations for minimizing protein adsorption to glass and other surfaces (Table 8-4). Albumin preferentially competes with other proteins for binding sites on surfaces, but why this is so is not clear.

Because albumin is a natural protein, concerns have been raised about potential contamination of albumin with human prion protein that is thought to be the infectious agent in bovine spongiform encephalopathy (BSE). Indeed, the use of animal-source excipients (and this includes not only albumin, but also glycerol and polysorbate 80) is no longer practiced. The development of synthetic (e.g., recombinant HSA) versions of these materials has eliminated concerns over potential disease transmission.

OTHER PHYSICAL COMPLEXING/STABILIZING AGENTS

PEG is a common co-solvent for solubilizing small nonproteinaceous molecules and may minimize the aggregation of several peptides and proteins (52). PEG modification of proteins for sustained-release purposes has seen wide application. The concentration of PEG needs to be

Table 8-4 Some Examples of Commercial Protein Dosage Forms Containing Human Serum Albumin

Generic	Brand®	% HSA in product
Alglucerase	Ceredase	1.0
Erythropoietin	Epogen	0.25
Interferon Alpha-2a	Roferon-A	0.5
Interferon Alpha-2b	Intron-A	0.1 (after reconstitution)
Urokinase	Abbokinase	5.0 (after reconstitution)
Alpha-1-Proteinase	Aralast	0.5 (after reconstitution)
Antihemophilic factor	Recombinate	1.0 (after reconstitution)
Botulism toxin	Myobloc	0.05
Streptokinase	Streptase	2.0 (after reconstitution)
Hyaluronidase	Halozyme	0.1