(Yalkowsky and Valvani 1977) and dynamic methods (Schroeder and DeLuca 1974; Yalkowsky et al. 1983; Cox et al. 1991; Davio et al. 1991; Irwin and Iqbal 1992). Static methods simply involve mixing of a formulation with another liquid, as opposed to injecting the formulation into a flowing stream of another liquid, that is, dynamic methods, to mimic more closely the actual injection process. These techniques allow formulation optimization to occur before animal testing is initiated.

One must also remember that precipitation can occur on dilution with other intravenous fluids. Investigation of dilution in common parenteral vehicles, for example, normal saline, 5% dextrose solution, or likely coadministered drug products, should be conducted for solubility and stability purposes before recommending such procedures. Phlebitis, hemolysis, and pain on injection are also issues for parenteral formulations containing cosolvents.

Hemolysis induced by cosolvents has been extensively studied (Cadwallader 1963; Banziger 1967; Oshida et al. 1979; Osigo et al. 1983; Fort et al. 1984; Howard and Gould 1985; Reed and Yalkowsky 1985, 1986; Cherng-Chyi Fu 1987; Obeng and Cadwallader 1989; Ward and Yalkowsky 1992; Krzyzaniak et al. 1996, 1997). In these references, various means of evaluating hemolysis are proposed. Reed and Yalkowsky (1985) used an *in vitro* method of determining hemolysis versus a reference formulation composed of 10% ethanol, 40% propylene glycol, and 50% water. They determined the LD_{50}, the concentration of cosolvent to induce hemolysis in 50% of healthy red blood cells, of various cosolvents. The measured LD_{50} values are expressed as total volume percent of cosolvent in whole blood. A higher LD_{50} indicates a relatively less hemolytic cosolvent. Reed and Yalkowsky determined LD_{50} values of 39.5% dimethyl isosorbide, 37.0% dimethylacetamide, 30.0% polyethylene glycol 400, 21.2% ethanol, 10.3% of the reference formulation, 5.7% propylene glycol, and 5.1% dimethyl sulfoxide. Therefore, although propylene glycol is commonly used in parenteral formulations, it is one of the more hemolytic cosolvents. Both Reed and Yalkowsky (1985) and Cadwallader (1963) have determined that the addition of sodium chloride to 40% propylene glycol solutions reduces or prevents hemolysis of red blood cells. On the basis of data summarized earlier by Reed and Yalkowsky (1985) and data by Fort et al. (1984), ethanol and polyethylene glycol 400 are less hemolytic alternatives to propylene glycol.

Muscle damage or myotoxicity is a concern when injecting cosolvents intramuscularly (Brazeau 1989a,b, 1990a,b, 1992). Brazeau and Fung (1989a) used an *in vitro* model to assess myotoxicity to see if there were any correlation with dielectric constant, apparent pH, surface tension, or viscosity. Aqueous mixtures of propylene glycol, ethanol, and polyethylene glycol 400 were studied. Muscle damage was assessed by release of creatine kinase. Brazeau and Fung (1989b) determined that no single property or combination of properties could predict the degree of muscle damage that occurred. They postulated that muscle damage might be due to complex interactions of a given cosolvent with the biochemical processes occurring in skeletal muscle. Further investigation by Brazeau and Fung (1990a) indicated that polyethylene glycol 400 in mixed cosolvent systems might have a protective effect on the myotoxicity generated by intramuscular injections. Studies where diazepam was injected intramuscularly in different cosolvent systems with varying degrees of producing muscle damage did not seem to affect the bioavailability of this compound.

Inflammation by subcutaneous injection of cosolvents has not been published on extensively. Radwan (1994) used an *in vivo* screening model to study the effect of various oils and cosolvents in oils on increasing skin fold thickness of rats after subcutaneous injection. With the exception of benzyl alcohol, ethyl oleate, and phospholipon 100, most excipients showed only minor inflammatory responses.

In multiuse parenteral products that require the use of a preservative system, studies have shown that preservative effectiveness can be influenced by the cosolvents. Darwish and Bloomfield (1995) determined that the presence of a cosolvent produced an increase in the preservative activity of methyl and propyl paraben because of the increase in solubility afforded by the presence of cosolvents. Further studies revealed that cosolvents also influence preservative efficacy by causing cell membrane damage in and of themselves (Darwish and Bloomfield 1997).

Since cosolvents can produce unwanted effects when used parenterally, investigation of other means of solubilizing compounds may be worthwhile if pain, inflammation, or phlebitis